2011. *Expert Rev Anti Infect Ther* 9:775-785), antibacterial activity against bacteria in biofilms, and activity in microaerophilic environments, even with high bacterial load (Azeredo J, *et al.* 2008. *Curr Pharm Biotechnol* 9:261-266), as well as the generally accepted safety of bacteriophage therapy in some parts of the world (Sulakvelidze A, *et al.*, 2001, *Antimicrob Agents Chemother.* 45(3): 649-659). Recent animal trials of bacteriophage therapy have demonstrated its potential to heal or improve skin bacterial diseases, both in internal (McVay CS, *et al.* 2007. *Antimicrob Agents Chemother* 51:1934-1938) and external applications (Soothill JS. 1994. Burns 20:209-211; Wills QF, *et al.* 2005. *Antimicrob Agents Chemother* 49:1220-1221). However, there is little published evidence supporting the use of bacteriophage to cure infections established for longer than a few hours (Ryan EM, *et al.* 2011 *J Pharm Pharmacol* 63:1253-1264).

[0008] In particular, phage cocktails may provide advantages to the use of phages individually, *e.g.*, to increase the lytic activity against a particular bacterial strain, and to decrease the possibility of emergence of bacteria resistant to an individual bacteriophage. That is, different bacteriophage can be mixed as cocktails to broaden their properties, preferably resulting in a collectively greater antibacterial spectrum of activity *e.g.*, an expanded host range, to which development of resistance is less likely. Nonetheless, to date, few phage cocktails exist with antimicrobial activity against different bacteria, possibly because of the difficulty in combining different specificities of bacteriophage while maintaining storage stability.

[0009] There is therefore a need to develop novel phage products as therapeutic and/or prophylactic agents for use *in vivo* against pathogenic bacteria. There also is a need for better treatments, particularly topical treatments, for DFIs. In particular, there is a need for bacteriophage cocktails capable of lysing bacteria responsible for DFIs, including *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and/or *Acinetobacter baumannii*. This application addresses this and other needs.

4. SUMMARY ~~OF THE INVENTION~~

[0010] ~~In one aspect, the present invention is directed to~~Provided herein are compositions comprising phage cocktails. In one aspect, there is provided a composition comprising at least two different isolated strains of bacteriophage, each of said strains having a genome that comprises a nucleic acid sequence selected from the group of consisting of SEQ ID NO:1 (F44/10), SEQ ID NO:2 (F125/10), SEQ ID NO:3 (F770/05), SEQ ID NO:4 (F510/08), and SEQ ID NO:5 (F1245/05), or a variant thereof, said variant having at least 95% sequence identity to said nucleic acid sequence

and showing antibacterial activity against at least one of *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and/or *Acinetobacter baumannii*.

In some embodiments, the ~~invention~~ disclosure provides compositions comprising at least two different isolated strains of bacteriophage, each having a genome that comprises a nucleic acid sequence selected from the group of consisting of SEQ ID NO:1 (F44/10), SEQ ID NO:2 (F125/10), SEQ ID NO:3 (F770/05), SEQ ID NO:4 (F510/08), and SEQ ID NO:5 (F1245/05),

a genome that comprises a nucleic acid sequence selected from the group consisting of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, and SEQ ID NO:4, or the variant thereof.

[0010a] In one aspect, there is provided a composition comprising at least five isolated bacteriophage strains, said strains having genomes that comprise nucleic acid sequences of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, and SEQ ID NO:5, or a variant thereof, said variant having at least 95% sequence identity to said nucleic acid sequence and showing antibacterial activity against at least one of *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and/or *Acinetobacter baumannii*.

[0011] In some preferred embodiments, the ~~invention~~ disclosure is directed to a composition comprising at least five isolated bacteriophage strains, the strains having genomes that comprise nucleic acid sequences of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, and SEQ ID NO:5, or a variant thereof, where the variant has at least 95% sequence identity to the corresponding nucleic acid sequence and shows antibacterial activity against at least one of *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and/or *Acinetobacter baumannii*. In some embodiments, the bacteriophage strains having genomes that comprise the nucleic acid sequences of SEQ ID NOs:1, 2, 4, and 5, or the variants thereof, are each present in the composition in an amount corresponding to about 10 times that of the bacteriophage strain having a genome that comprises the nucleic acid sequence of SEQ ID NO:3, or the variant thereof. In some embodiments, the composition comprises bacteriophage strains having genomes that comprise nucleic acid sequences of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, and SEQ ID NO:5.

[0011a] In one aspect, there is provided a pharmaceutical composition comprising a composition as described herein and a pharmaceutically acceptable carrier.

[0012] In another ~~aspect~~embodiment, the instant ~~invention~~ disclosure is directed to pharmaceutical compositions comprising phage cocktails, specifically, pharmaceutical compositions comprising any of the compositions described above and a pharmaceutically acceptable carrier. In some embodiments, the pharmaceutical composition is formulated for topical application. In some embodiments, the pharmaceutical composition comprises a sterile buffer, *e.g.*, a buffer comprising about 0.05 M Tris-HCl, about 0.1M NaCl, and about 10mM $MgSO_4.7H_2O$. In some embodiments, the pharmaceutical composition is contained in an ampoule.

[0013] In some embodiments, the pharmaceutical composition further comprises an additional agent, *e.g.*, an agent selected from the group consisting of an antibiotic agent, an anti-inflammatory agent, an antiviral agent, a local anesthetic agent, a growth factor, and a corticosteroid. In some embodiments, the additional agent is an antibiotic agent, *e.g.*, an antibiotic agent having antibacterial activity against *Acinetobacter baumannii*, *Pseudomonas*

aeruginosa, and/or *Staphylococcus aureus*; or an antibiotic agent having antibacterial activity against bacteria other than *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*. More specifically, in some embodiments, the additional agent is an antibiotic agent having antibacterial activity against *Staphylococcus aureus* or an antibiotic agent having antibacterial activity against bacteria other than *Staphylococcus aureus*. In some

embodiments, the additional agent is an antibiotic agent having antibacterial activity against *Pseudomonas aeruginosa* or an antibiotic agent having antibacterial activity against bacteria other than *Pseudomonas aeruginosa*. In some embodiments, the additional agent is an antibiotic agent having antibacterial activity against *Acinetobacter baumannii* or an antibiotic agent having antibacterial activity against bacteria other than *Acinetobacter baumannii*. In some embodiments, administration of the antibiotic agent comprises systemic administration.

[0014] In some embodiments, the composition is for use in treating a bacterial infection associated with an area of non-intact skin, and each of the phage strains is present in the composition in an amount corresponding to 10^3 to 10^{13} phage particles/cm^2 of the area. In some embodiments, each of the phage strains is present in the composition in an amount corresponding to 10^7 to 10^9 phage particles/cm^2 of the area.

[0014a] In one aspect, there is provided the use of a therapeutically effective amount of a pharmaceutical composition as described herein in the manufacture of a medicament for treating or preventing a bacterial infection in a subject in need thereof.

[0015] Another ~~aspect~~ embodiment of the instant ~~invention~~ disclosure is directed to methods of treating or preventing a bacterial infection in a subject in need thereof comprising administering to the subject a therapeutically effective amount of the pharmaceutical composition according to the ~~invention~~disclosure. In some embodiments, the bacterial infection is an infection by one or more of *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*. In some embodiments, the pharmaceutical composition is administered topically. In some embodiments, the subject is a mammal, *e.g.*, a human. In some embodiments, the bacterial infection is diabetic foot infection. In some embodiments, the diabetic foot infection comprises a cutaneous ulcer. In some embodiments, the bacterial infection is associated with an area of non-intact skin selected from a sore associated with cellulitis, an erysipelas lesion, a burn wound, a chronic ulcer, a decubitus ulcer, and a pressure sore. In some embodiments, the treatment comprises topically administering the pharmaceutical composition to a cutaneous ulcer associated with diabetic foot infection. In some preferred embodiments, administration follows mechanical debridement of the ulcer. In some embodiments, administration comprises use of at least one of a dressing, an instillation device, and a negative pressure wound therapy device.

[0016] In some embodiments, the pharmaceutical composition is administered every 4 hours or every 6 hours for an initial 24 hours. In some embodiments, following the initial 24 hours, the pharmaceutical composition is administered every 12 hours or every 24 hours for at least 3

additional days. In some embodiments, the pharmaceutical composition is administered every 12 hours or every 24 hours for at least 4 additional days.

41. ~~A method of treating or preventing a bacterial infection in a subject in need thereof comprising administering to said subject a~~ Use of a therapeutically effective amount of the pharmaceutical composition according to any one of claims 16-25 in the manufacture of a medicament for treating or preventing a bacterial infection in a subject in need thereof.

42. The ~~method~~ use according to claim 41, wherein said bacterial infection is an infection by one or more of *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus.*

43. The ~~method~~ use according to claim 41 or 42, wherein the pharmaceutical composition is to be administered topically.

44. The ~~method~~ use according to claim 43, wherein said subject is a mammal.

45. The ~~method~~ use according to claim 44, wherein said mammal is a human.

46. The ~~method~~ use according to any one of claims 41-45, wherein said bacterial infection is associated with a ~~chornic~~ chronic ulcer.

47. The ~~method~~ use according to any one of claims 41-46, wherein said bacterial infection is associated with a burn wound.

48. The ~~method~~ use according to any one of claims 41-46, wherein said bacterial infection is diabetic foot infection.

49. The ~~method~~ use according to claim 48, wherein said diabetic foot infection comprises a cutaneous ulcer.

50. The ~~method~~ use according to any one of claims 41-46, wherein said bacterial infection is associated with an area of non-intact skin selected from a sore associated with cellulitis, an erysipelas lesion, a decubitus ulcer, and a pressure sore.

51. ~~A method of treating or preventing a bacterial infection in a subject in need thereof comprising administering to said subject~~ Use of a therapeutically effective amount of the pharmaceutical composition according to any one of claims 26-29 in the manufacture of a medicament for treating or preventing a bacterial infection in a subject in need thereof.

52. The ~~method~~ use according to claim 51 wherein said bacterial infection is an infection by *Staphylococcus aureus.*

53. The ~~method~~ use according to claim 51 or 52, wherein said bacterial infection is diabetic foot infection.

54. ~~A method of treating or preventing a bacterial infection in a subject in need thereof comprising administering to said subject~~ Use of a therapeutically effective amount of the pharmaceutical composition according to any one of claims 30-33 in the manufacture of a medicament for treating or preventing a bacterial infection in a subject in need thereof.

55. The ~~method~~ use according to claim 54 wherein said bacterial infection is an infection by *Pseudomonas aeruginosa.*

56. The ~~method~~ use according to claim 54 or 55, wherein said bacterial infection is diabetic foot infection.

57. ~~A method of treating or preventing a bacterial infection in a subject in need thereof comprising administering to said subject~~ Use of a therapeutically effective amount of the pharmaceutical composition according to any one of claims 34-37 in the manufacture of a medicament for treating or preventing a bacterial infection in a subject in need thereof.

58. The ~~method~~ use according to claim 57 wherein said bacterial infection is an infection by *Acinetobacter baumannii.*

59. The ~~method~~ use according to claim 57 or 58, wherein said bacterial infection is diabetic foot infection.

60. ~~A method of treating or preventing a bacterial infection in a subject in need thereof comprising administering to said subject~~ Use of a therapeutically effective amount of the pharmaceutical composition according to any one of claims 38-40 in the manufacture of a medicament for treating or preventing a bacterial infection in a subject in need thereof.

61. The ~~method~~ use according to claim 60, wherein said bacterial infection is diabetic foot infection.

62. The ~~method~~ use according to claim 61, wherein ~~said treatment comprises topically administering~~ said composition is to be topically administered to a cutaneous ulcer associated with said diabetic foot infection.

63. The ~~method~~ use according to claim 62, wherein said administration follows mechanical debridement of said ulcer.

64. The ~~method~~ use according to claim 62 or 63, wherein said administration comprises use of at least one of a dressing, an instillation device, and a negative pressure wound therapy device.

65. The ~~method~~ use according to any one of claims 62-64, wherein said pharmaceutical composition is to be administered every 4 hours or every 6 hours for an initial 24 hours.

66. The ~~method~~ use according to claim 65, wherein, following said initial 24 hours, said pharmaceutical composition is to be administered every 12 hours or every 24 hours for at least 3 additional days.

67. The ~~method~~ use according to claim 66, wherein said pharmaceutical composition is to be administered every 12 hours or every 24 hours for at least 4 additional days.

68. The ~~method~~ use according to any one of claims 61-67, wherein said ~~method is~~composition is to be used in combination with a standard therapy for diabetic foot infection.

69. The ~~method~~ use according to claim 68, wherein said standard therapy is selected from the group consisting of extracellular matrix replacement therapy, moist wound therapy, negative pressure wound therapy, arterial re-vascularization therapy, hyperbaric oxygen therapy, administration of an antibiotic agent, and administration of a growth factor.

70. The ~~method~~ use according to claim 69, wherein said moist wound therapy comprises use of an adhesive-backing film, a silicone-coated foam, a hydrogel, and/or a hydrocolloid.

71. The ~~method~~ use according to claim 69 or 70, wherein said extracellular matrix replacement therapy comprises use of bio-engineered tissue.

72. The ~~method~~ use according to any one of claims 69-71, wherein said antibiotic agent has antibacterial activity against *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and/or *Staphylococcus aureus*.

73. The ~~method~~ use according to claim 72, wherein administration of said antibiotic agent comprises systemic administration.

74. The ~~method~~ use according to any one of claims 69-73, wherein said growth factor is at least one selected from the group consisting of platelet-derived growth factor, granulocyte colony-stimulating factor, epidermal growth factor, fibroblast growth factor, nerve growth factor, and vascular endothelial growth factor.

75. The ~~method~~ use according to claim 74, wherein administration of said growth factor comprises topical administration.

76. The ~~method~~ use according to any one of claims 61-75, wherein said ~~method is~~ composition is to be used in combination with a non-standard therapy for diabetic foot infection, wherein said diabetic foot infection is refractory to a standard therapy.