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Gold Nanoparticles (AuNPs): A New Frontier in Vaccine Delivery

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Prophylactic vaccination is one of the most effective interventions in medicine and is responsible for substantial decreases in morbidity and mortality by many pathogens worldwide [1,2]. Although the concept of vaccination has existed since the early 1000s, it was not until the 1700s that the first experimental proof for vaccination emerged. Edward Jenner demonstrated that exposure to cowpox virus protected individuals from infection by the related, but much more virulent, smallpox virus [3]. Today, even as our understanding of the immune system rapidly expands, the basic idea behind early strategies of vaccination still holds: protection is best achieved by activating the immune system before an exposure to an infectious pathogen.

Prophylactic vaccines are usually derived from inactivated or attenuated strains of a pathogen or from macromolecules such as proteins, glycoproteins, and polysaccharides that are synthesized by the pathogen [4-6]. These vaccines prevent or limit infection because they induce robust, specific B cell mediated antibody response and a variety of T cell mediated cellular immune responses, including development of cytotoxic cells. Cytokines generated during the early and late phase of the immune response allows communication between the immune cells as well as fine tunes the global immune response for effective elimination of the pathogen. The induction of such B and T cell mediated responses, and the subsequent generation of B and T cell memory, requires cooperation and activation of the innate immune system. Vaccines often include adjuvants to trigger the host innate immune responses by activating dendritic cells, macrophages, and other innate immune cells during early phases of the response. The importance of adjuvants in vaccination has long been recognized but how adjuvants worked remained unclear until a proposal by Charles Janeway in 1989 [7]. The focus on adjuvants and their actions quickly became an area of intense research, especially after the discovery of the Toll-like receptors in 1997 [8]. Prophylactic vaccines derived from inactivated or attenuated strains of a pathogen have natural adjuvants present in the pathogen itself. Vaccines derived from macromolecules must first be mixed with synthetic adjuvants before administration in order to boost immune responses. Currently, only a limited number of adjuvants are approved for use in humans in the United States. These include alum and a few lipid based emulsions [9]. Other adjuvants have the capability to enhance immune responses but are not currently licensed for use in vaccines, in part, due to potential toxic side effects [9]. Natural components of pathogens (i.e. LPS, muramyl di-peptide, flagellin) and nucleic acid mimics (i.e. CpG, PolyIC) are on this list [9,10]. Exposure to wild-type pathogen after vaccination recalls pathogen specific memory cells that mount a robust and rapid response, often preventing infection completely or significantly limiting its severity, duration, and spread. In the absence of vaccination, the adaptive immune response to a wild-type pathogen is slow to develop allowing the pathogen to replicate and spread quickly in the absence of pathogen-specific memory response.

Traditional vaccines are remarkably effective, but there are limitations to their production and distribution. The need for developing alternative safe and effective vaccine platforms is urgent. First, traditionally the antibody titers is considered to be a primary measure of the immunogenicity of a vaccine has traditionally been antibody titers [11]. Generating neutralizing antibody against the pathogen to prevent it from establishing an infection has proved to be a successful strategy for vaccination for many pathogens. However, it is becoming increasingly clear that T cell responses are critical at mediating clearance of many infections as well as preventing the establishment of chronic infections [12-14]. As such, the most effective vaccines should induce rapid and robust CD4 and CD8 T cell mediated immune responses. Second, traditional vaccines can be time consuming to produce. For example, the traditional seasonal influenza virus vaccine takes roughly five to six month to manufacture [15] and any pitfalls in the workflow could lengthen this time. Finally, the majority of traditional vaccines rely on cold storage, a limiting factor to distribution of vaccines across the world particularly in less or under developed nations [16].

Research in developing new vaccine platforms with the potential to elicit robust but broad B and T cell responses and mitigate other limitations of traditional vaccines is ongoing. A number of such vaccine platforms include virus like particles, liposomes, polymeric nanoparticles, and metal-based nanoparticles [17-19]. Recently, gold nanoparticle (Au-NP) based vaccines have been receiving substantial attention due to the potential advantages over traditional vaccine platforms. First, gold nanoparticles are customizable, able to be synthesized in various shapes and sizes by relatively straightforward chemical synthesis [17,20]. From an immunological perspective, this customizability has functional significance. After injection, nanoparticles smaller than 100 nm readily enter lymphatic tissues [21,22] and those less than 50 nm accumulate in multiple immune cell subsets in the secondary lymphoid compartment [23]. Localization of AuNPs to lymphatic tissues and cells is notable as it mimics what occurs during a natural infection. Pathogens and their components gain access to the lymphatic network and migrate to lymph nodes via lymphatic flow or after being internalized by resident antigen presenting cells. In the secondary lymphoid compartments, antigenic determinants (called epitopes) recognized by B cells induce antibody responses while those degraded into small peptide epitopes can be presented to and activate T cell responses. Secondly, a number of macromolecules capable of inducing broad and robust immune responses can be attached to Au-NPs by either covalent or non-covalent mechanisms [17,24]. These include antibodies [17] and other antigenic proteins [25], T cell activating peptides derived from pathogens [26,27], and nucleic acids, including siRNA [28]. Importantly, these antigenic

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determinants can be attached in the presence of adjuvants, such as CpG [29], which enhances innate immune responses induced during vaccination. Third, Au-NPs are relatively stable [20,30]. Data from Tao et al. [30] demonstrated that Au-NPs conjugated with the influenza specific peptide M2e and the adjuvant CpG could be dried into a powder and stored at room temperature, potentially breaking the cold storage chain for distribution of vaccines. Importantly, these dried Au-NPs could be easily reconstituted with water without any observable aggregation. Finally, and perhaps most importantly, Au-NPs and have a good safety profile [6,31], which is essential for the development and use of any vaccine. There is some concern that smaller Au-NPs or Au-NPs coated with certain peptides or proteins may cross the blood brain barrier when administered intravenously [32,33]; however, Au-NPs can, like traditional vaccines, be administered by injecting through subcutaneous and intradermal routes [22] and therefore mitigate this concern.

Data from numerous laboratories indicate that the shape of the particle can have a significant impact on immunogenicity [17,20]. The malleability of gold particles allow synthesis into Au-NPs of various shapes and gives added advantages to this vaccine approach. When mice were administered Au-NPs intranasally, rod shaped particles had a more prolonged effect on microglia cell activation than spheres or urchin shaped particles [34]. When microglial cells were stimulated *in vitro*, rod shaped particles induced GM-CSF and IL-1α secretion, while sphere and urchin shaped particles induced only GM-CSF and IL-1α secretion respectively. A recent study by Nikkura et al. [25] demonstrated that when Au-NPs ~40nm in size were coated with the West Nile Virus E protein, spherical particles induced higher levels of antibody and induced pro-inflammatory cytokine secretion while rod shaped particles were more efficiently internalized by professional antigen presenting cells. Finally, using a mouse model of breast cancer, Black et al. demonstrated that rod shaped Au-NPs were taken up more efficiently by tumor cells resulting in the particles being distributed throughout the tumor [35]. In contrast, sphere shaped Au-NPs were only located around the periphery of the tumor. This later finding may be useful for developing therapeutic vaccines, particularly for diseases such as multi-drug resistant tuberculosis where granulomas pose particular challenge for the immune system. Taken as a whole, the data on size and shape of Au-NPs suggest that efficient Au-NP based vaccines should be a mixture of particles of varying sizes and shapes designed to induce more broad and robust immune responses.

The most challenging aspect in developing nanoparticle based vaccines will be choosing the appropriate epitopes and/or antigens (i.e. longer peptide fragments or whole proteins) for coating Au-NPs. A large number of B and T cell epitopes have been well characterized and submitted to the Immune Epitope Database (IEDB) [36]. However, these are largely based on peptide screens and it is unclear whether these induce T cell responses during infection. Improvements on T cell epitope selection is possible by using immunoproteomic techniques to identify those epitopes that are naturally processed and presented by infected cells [37,38]. These techniques have identified novel epitopes not present in IEDB but that are presented on infected cells [37,38] and to which seropositive patients have circulating T cells [38]. Regardless of the epitope identification method used, formulating Au-NPs containing multiple epitopes/antigens to induce antibody and CD4 and CD8 T cell responses will be essential. Indeed, numerous studies have demonstrated that a broad immune response is critical for clearance of many infections, including hepatitis B [13], hepatitis C [12], and influenza [14].

In addition to choosing the appropriate targets to include in the AuNP based vaccines, ensuring the proper processing of these targets will be essential in generating a protective immune response. In order to generate robust and specific CD4 and CD8 T cell responses, internalization of pathogens and/or antigens by antigen presenting cells (APCs) is critical. A number of APC subsets, including dendritic cells and macrophages are important in this process. These cells internalize extracellular antigen and shuttle it into the endosomal compartment where it can be degraded into smaller peptide epitopes. These peptide epitopes are then loaded onto MHC class II molecules in a special vesicular compartment followed by their export and display on the cell surface. Presentation of peptide/MHC-II complexes to CD4 T cells, each bearing a specific receptor, ensures cell activation. Because Au-NPs are efficiently internalized by professional antigen presenting cells [23,25], loading Au-NPs with multiple epitope/proteins, would be sufficient to generate CD4 T cell responses but may not be as efficient at generating CD8 T cell responses since, for the majority of these antigens, targeting to the cytosolic compartment is pivotal. The proteolytic environment of the late endosome is harsh and it is possible that MHC class I epitopes loaded onto Au-NPs would be degraded before encountering MHC class I molecules and, therefore, impair CD8 T cell responses. Therefore, in order to induce the most broad and robust CD4 and CD8 T cell responses, Au-NPs and/or their associated epitopes/antigens must be targeted to the appropriate antigen presenting cell or cellular compartment to ensure optimal processing and presentation to T cells. To this end, CD8 T cell responses may be enhanced by targeting Au-NPs and associated epitopes/antigens to antigen presenting cells capable of cross presentation [39]. A number of methods can be used to promote cross presentation and thereby trigger CD8 T cell responses. First, Au-NPs coated with epitopes/antigens can be targeted to cross presenting dendritic cells by also including antibodies specific to cell surface proteins on dendritic cells that are involved in cross presentation of antigens, namely DEC-205 [40,41] or the mannose receptor [42,43]. Targeting DEC-205 may be the more attractive method as this pathway also allows for efficient MHC class II presentation [44] whereas cross presentation involving the mannose receptor seems to be specific for soluble antigen [42,43]. A second potential mechanism to target Au-NPs attached epitopes to the class I pathway is to include cell penetrating peptides (CPPs), small peptides that allow efficient delivery of molecules into the cells. Despite internalization into endocytic compartments, certain cell penetrating peptides allow escape of cargo into the cytosol [45], thereby allowing natural processing of longer peptides or proteins attached to the Au-NP to be naturally processed by the classical proteasome-class I machinery. Similar to targeting epitopes to cross presenting DCs, targeting Au-NPs to the cytosolic compartment via CPPs may enhance class II processing and CD4 T cell activation as data indicate that 30-40% of the class II restricted response during a viral infection is proteasome mediated [46,47]. Like class I restricted epitopes, class II epitopes dependent on the proteasome for processing do not survive the hard proteolytic environment of the endosome [48]. As is the case for size and shape, AuNP based vaccines should include a number of antigens targeted to the appropriate cellular compartment to ensure robust T cell activation. One can envision a vaccine formulation in which spherical Au-NPs are coated with antibody epitopes and rod shaped particles are coated with T cell epitopes that are targeted to the classical endosomal pathway (CD4 T activation), the intracellular proteasomal pathway (both CD4 and CD8 T activation), and/or cross presenting cell subsets (both CD4 and CD8 T activation). Both spherical and rod shaped Au-NPs can also be conjugated to any number of adjuvants to boost innate and adaptive responses and lead to more efficient memory B and T cell generation, thus offering enhanced protection to vaccinated individuals.

The new and improved Au-NP vaccine formulations, with different shapes and sizes of the nanoparticle, designed to direct antigens and their epitopes to different internal compartments in antigen presenting cells provides a multifaceted approach. While such an approach is likely to generate a broad and robust immune response, its potential to generate robust memory B and T cells during these early responses remains to be tested. With the number of travelers moving across the world by plane on a daily basis (estimated at 815 million in domestic and international travel in 2012; http://www.rita.dot.gov/bts/press_releases/bts016_13), the spread of infectious agents across national and international borders can occur quickly. This is illustrated by the recent emergence of Dengue virus [49] and Chikungunya virus [50,51] in the United States and the entry of Ebola virus infected individuals into the US and other countries [52,53]. In addition, the threat of new emerging diseases is ever present with the emergence of Severe Acute Respiratory Syndrome (SARS) [54-56], Middle East Respiratory Syndrome (MERS) [57,58] and a number of new influenza virus strains [59,60]. With these known and unknown dangers to human and animal health, the importance of developing a new vaccine platform cannot be overstated.

References

1. Andre FE, Booy R, Bock HL, Clemens J, Datta SK, et al. (2008) Vaccination greatly reduces disease, disability, death and inequity worldwide. Bull World Health Organ 86: 140-146.
2. Worboys M (2007) Vaccines: conquering untreatable diseases. BMJ 334 Suppl 1: s19.
3. Kennedy RB, Ovsyannikova IG, Jacobson RM, Poland GA (2009) The immunology of smallpox vaccines. Curr Opin Immunol 21: 314-320.
4. Paolo Bonanni, José Ignacio Santos (2011) Vaccine evolution. Perspectives in Vaccinology 1: 1-24.
5. Stanberry LR, Strugnell R (2011) Vaccines of the future. Perspectives in Vaccinology 1: 151-199.
6. Gregory AE, Titball R, Williamson D (2013) Vaccine delivery using nanoparticles. Front Cell Infect Microbiol 3: 13.
7. Janeway CA Jr. (1989) Approaching the asymptote? Evolution and revolution in immunology. Cold Spring Harb Symp Quant Biol 54: 1-13.
8. Medzhitov R, Preston-Hurlburt P, Janeway CA Jr (1997) A human homologue of the Drosophila Toll protein signals activation of adaptive immunity. Nature 388: 394-397.
9. Lee S, Nguyen MT (2015) Recent advances of vaccine adjuvants for infectious diseases. Immune Netw 15: 51-57.
10. O'Hagan DT, Fox CB2 (2015) New generation adjuvants--from empiricism to rational design. Vaccine 33 Suppl 2: B14-20.
11. Siegrist C (2008) Vaccine immunology In: Plotkin SA, Orenstein WA, Offit PA (Eds) Vaccines (6th Ed), Elsevier Inc.
12. Thimme R, Oldach D, Chang KM, Steiger C, Ray SC, et al. (2001) Determinants of viral clearance and persistence during acute hepatitis C virus infection. J Exp Med 194: 1395-1406.
13. Chang JJ, Lewin SR (2007) Immunopathogenesis of hepatitis B virus infection. Immunol Cell Biol 85: 16-23.
14. Wilkinson TM, Li CK, Chui CS, Huang AK, Perkins M, et al. (2012) Preexisting influenza-specific CD4+ T cells correlate with disease protection against influenza challenge in humans. Nat Med 18: 274-280.
15. WHO, UNICEF, World Bank (2009) State of the world's vaccines and immunization (3rd Edn), World Health Organization, Geneva.
16. Centers for Disease Control and Prevention (CDC) (2003) Guidelines for maintaining and managing the vaccine cold chain. MMWR Morb Mortal Wkly Rep 52: 1023-1025.
17. Cruz LJ, Tacken PJ, Rueda F, Domingo JC, Albericio F, et al. (2012) Targeting nanoparticles to dendritic cells for immunotherapy. Methods Enzymol 509: 143-163.
18. Zhao L, Seth A, Wibowo N, Zhao CX, Mitter N, et al. (2014) Nanoparticle vaccines. Vaccine 32: 327-337.
19. Bolhassani A, Javanzad S, Saleh T, Hashemi M, Aghasadeghi MR, et al. (2014) Polymeric nanoparticles: potent vectors for vaccine delivery targeting cancer and infectious diseases. Hum Vaccin Immunother 10: 321-332.
20. Shah M, Badwaik VD, Dakshinamurthy R (2014) Biological applications of gold nanoparticles. J Nanosci Nanotechnol 14: 344-362.
21. Reddy ST, Rehor A, Schmoekel HG, Hubbell JA, Swartz MA (2006) In vivo targeting of dendritic cells in lymph nodes with poly(propylene sulfide) nanoparticles. J Control Release 112: 26-34.
22. Reddy ST, van der Vlies AJ, Simeoni E, Angeli V, Randolph GJ, et al. (2007) Exploiting lymphatic transport and complement activation in nanoparticle vaccines. Nat Biotechnol 25: 1159-1164.
23. Almeida JP, Lin AY, Langsner RJ, Eckels P, Foster AE, et al. (2014) In vivo immune cell distribution of gold nanoparticles in naÃ¯ve and tumor bearing mice. Small 10: 812-819.
24. Ding Y, Jiang Z, Saha K, Kim CS, Kim ST, et al. (2014) Gold nanoparticles for nucleic acid delivery. Mol Ther 22: 1075-1083.
25. Niikura K, Matsunaga T, Suzuki T, Kobayashi S, Yamaguchi H, et al. (2013) Gold nanoparticles as a vaccine platform: influence of size and shape on immunological responses *in vitro* and in vivo. ACS Nano 7: 3926-3938.
26. Yeste A, Nadeau M, Burns EJ, Weiner HL, Quintana FJ (2012) Nanoparticle-mediated codelivery of myelin antigen and a tolerogenic small molecule suppresses experimental autoimmune encephalomyelitis. Proc Natl Acad Sci U S A 109: 11270-11275.
27. Lin AY, Lunsford J, Bear AS, Young JK, Eckels P et al. (2013) High-density sub-100-nm peptide-gold nanoparticle complexes improve vaccine presentation by dendritic cells *in vitro*. Nanoscale Res Lett 8: 72.
28. Paul AM, Shi Y, Acharya D, Douglas JR, Cooley A, et al. (2014) Delivery of antiviral small interfering RNA with gold nanoparticles inhibits dengue virus infection *in vitro*. J Gen Virol 95: 1712-1722.
29. Lin AY, Almeida JP, Bear A, Liu N, Luo L, et al. (2013) Gold nanoparticle delivery of modified CpG stimulates macrophages and inhibits tumor growth for enhanced immunotherapy. PLoS One 8: e63550.
30. Tao W, Ziemer KS, Gill HS (2014) Gold nanoparticle-M2e conjugate coformulated with CpG induces protective immunity against influenza A virus. Nanomedicine (Lond) 9: 237-251.
31. Libutti SK, Paciotti GF, Byrnes AA, Alexander HR Jr, Gannon WE, et al. (2010) Phase I and pharmacokinetic studies of CYT-609, a novel PEGylated colloidal gold-rhTNF nanomedicine. Clin Cancer Res 16: 6139-6149.
32. Wiley DT, Webster P, Gale A, Davis ME (2013) Transcytosis and brain uptake of transferrin-containing nanoparticles by tuning avidity to transferrin receptor. Proc Natl Acad Sci U S A 110: 8662-8667.
33. Cheng Y, Dai Q, Morshed RA, Fan X, Wegscheid ML, et al. (2014) Blood-brain barrier permeable gold nanoparticles: an efficient delivery platform for enhanced malignant glioma therapy and imaging. Small 10: 5137-5150.
34. Hutter E, Boridy S, Labrecque S, Lalancette-HÃ©bert M, Kriz J, et al. (2010) Microglial response to gold nanoparticles. ACS Nano 4: 2595-2606.
35. Black KC, Wang Y, Luehmann HP, Cai X, Xing W, et al. (2014) Radioactive 198Au-doped nanostructures with different shapes for in vivo analyses of their biodistribution, tumor uptake, and intratumoral distribution. ACS Nano 8: 4385-4394.
36. Vita R, Overton JA, Greenbaum JA, Ponomarenko J, Clark JD, et al. (2015) The immune epitope database (IEDB) 3.0. Nucleic Acids Res 43: D405-412.
37. Comber JD, Karabudak A, Shetty V, Testa JS, Huang X, et al. (2014) MHC Class I Presented T Cell Epitopes as Potential Antigens for Therapeutic Vaccine against HBV Chronic Infection. Hepat Res Treat 2014: 860562.
38. Comber JD, Karabudak A, Huang X, Piazza PA, Marques ET, et al. (2014) Dengue virus specific dual HLA binding T cell epitopes induce CD8+ T cell responses in seropositive individuals. Hum Vaccin Immunother 10: 3531-3543.
39. Shen L, Rock KL (2006) Priming of T cells by exogenous antigen cross-presented on MHC class I molecules. Curr Opin Immunol 18: 85-91.

40. Bonifaz LC, Bonnyay DP, Charalambous A, Darguste DI, Fujii S, et al. (2004) In vivo targeting of antigens to maturing dendritic cells via the DEC-205 receptor improves T cell vaccination. J Exp Med 199: 815-824.

41. Bozzacco L, Trumpfheller C, Siegal FP, Mehandru S, Markowitz M, et al. (2007) DEC-205 receptor on dendritic cells mediates presentation of HIV gag protein to CD8+ T cells in a spectrum of human MHC I haplotypes. Proc Natl Acad Sci U S A 104: 1289-1294.

42. Burgdorf S, Lukacs-Kornek V, Kurts C (2006) The mannose receptor mediates uptake of soluble but not of cell-associated antigen for cross-presentation. J Immunol 176: 6770-6776.

43. Burgdorf S, Kautz A, BÃ¶hnert V, Knolle PA, Kurts C (2007) Distinct pathways of antigen uptake and intracellular routing in CD4 and CD8 T cell activation. Science 316: 612-616.

44. Birkholz K, Schwenkert M, Kellner C, Gross S, Fey G, et al. (2010) Targeting of DEC-205 on human dendritic cells results in efficient MHC class II-restricted antigen presentation. Blood 116: 2277-2285.

45. Qian Z, LaRochelle JR, Jiang B, Lian W, Hard RL, et al. (2014). Early endosomal escape of a cyclic cell-penetrating peptide allows effective cytosolic cargo delivery. Biochemistry 53: 4034-4046.

46. Tewari MK, Sinnathamby G, Rajagopal D, Eisenlohr LC (2005) A cytosolic pathway for MHC class II-restricted antigen processing that is proteasome and TAP dependent. Nat Immunol 6: 287-294.

47. Comber JD, Robinson TM, Siciliano NA, Snook AE, Eisenlohr LC (2011) Functional macroautophagy induction by influenza A virus without a contribution to major histocompatibility complex class II-restricted presentation. J Virol 85: 6453-6463.

48. Eisenlohr LC, Hackett CJ (1989) Class II major histocompatibility complex-restricted T cells specific for a virion structural protein that do not recognize exogenous influenza virus. Evidence that presentation of labile T cell determinants is favored by endogenous antigen synthesis. J Exp Med 169: 921-931.

49. Centers for Disease Control and Prevention (CDC) (2010) Locally acquired Dengue-Key West, Florida, 2009-2010. MMWR Morb Mortal Wkly Rep 59: 577-581.

50. Gibney KB, Fischer M, Prince HE, Kramer LD, St George K, et al. (2011) Chikungunya fever in the United States: a fifteen year review of cases. Clin Infect Dis 52: e121-126.

51. Vega-RÃºa A, LourenÃ§o-de-Oliveira R, Mousson L, Vazeille M, Fuchs S, et al. (2015) Chikungunya virus transmission potential by local Aedes mosquitoes in the Americas and Europe. PLoS Negl Trop Dis 9: e0003780.

52. Hewlett AL, Varkey JB, Smith PW, Ribner BS (2015) Ebola virus disease: preparedness and infection control lessons learned from two biocontainment units. Curr Opin Infect Dis 28: 343-348.

53. Mora-Rillo M, Arsuaga M, Ramirez-Olivencia G, de la Calle F, Borobia AM (2015) Acute respiratory distress syndrome after convalescent plasma use: treatment of a patient with Ebola virus disease contracted in Madrid, Spain. Lancet Respir Med 3: 554-562.

54. Lee N, Hui D, Wu A, Chan P, Cameron P, et al. (2003) A major outbreak of severe acute respiratory syndrome in Hong Kong. N Engl J Med 348: 1986-1994.

55. Poutanen SM, Low DE, Henry B, Finkelstein S, Rose D, et al. (2003) Identification of severe acute respiratory syndrome in Canada. N Engl J Med 348: 1995-2005.

56. Drosten C, Preiser W, GÃ¼nther S, Schmitz H, Doerr HW (2003) Severe acute respiratory syndrome: identification of the etiological agent. Trends Mol Med 9: 325-327.

57. Guery B, Poissy J, el Mansouf L, Sejourne C, Ettahar N, et al. (2013) Clinical features and viral diagnosis of two cases of infection with Middle East Respiratory Syndrome coronavirus: a report of nosocomial transmission. Lance 381: 2265-2272.

58. Raj VS, Osterhaus AD, Fouchier RA, Haagmans BL2 (2014) MERS: emergence of a novel human coronavirus. Curr Opin Virol 5: 58-62.

59. de Wit E, Fouchier RA (2008) Emerging influenza. J Clin Virol 41: 1-6.

60. The Lancet ID (2014) Pandemic potential of emerging influenza. Lancet Infect Dis 14: 173.

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