Koch's time for COVID-19 and the other major epidemics of the 21st century

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Abstract

Koch's postulates are classical medical standard followed by researchers all over the world to identify a new pathogen for an emerging infectious disease (EID). In this article, we put forward a new concept of Koch's time for a new pathogen (KTNP), which stands for the time from the first case of attention of an EID to the first complete genome of the new pathogen in a public domain, to quantify the response of EIDs. Shortening KTNP expedites development of detection kits and/or devices world-wide. The KTNP of five EIDs occurred in this century are calculated respectively. The results demonstrated that KTNP is continuously shortening and Koch's time for COVID-19 is 15 days.

Introduction

Emerging infectious diseases (EIDs) are great challenges to human society and public health. EIDs can be caused by previously known pathogens such as new strains of influenza or unknown pathogens including newly identified novel coronaviruses. Once EID occurs, the pathogen should be identified as soon as possible to facilitate detection, diagnosis and treatment. Pathogen identification will allow physicians and epidemiologists to grasp the clinical and epidemiological characteristics of the disease and to control the outbreak.

Koch's postulates were proposed by German scientist Robert Koch in 1884 based on Jakob Henle's theory. It has been used to determine the correlation between certain diseases and pathogens (1). The main contents of original Koch postulates are: 1) the bacteria must be present in every case of the disease, 2) the bacteria must be isolated from the host with the disease and grown in pure culture, 3) the specific disease must be reproduced when a pure culture of the bacteria is inoculated into a healthy susceptible host, 4) the bacteria must be recoverable from the experimentally infected host. Koch's postulates have played an important role in pathogen determination, yet they have limitations, including the presence of non-symptomatic carriers, the population or individual-level variability in disease manifestation and EIDs caused non-bacterial pathogens.

In recent years, mega next-generation sequencing (mNGS) capacity has progressed enormously, providing an opportunity for obtaining pathogen's genome directly from clinical samples by

bypassing the isolation step in clinical microbiology laboratories. With complete pathogen genome sequences publicly available, medical research institutions world-wide may quickly establish an effective detection method and diagnosis and treatment scheme for the EID based on their epidemiological judgement and technological resources. Therefore, we believe that the time from emergence of the first EID case to deposition in public domain of the first complete genome of the new pathogen is the core indicator of the overall EID emergency management level. We defined this time as Koch's time for a new pathogen (KTNP). In this article, we calculated and analyzed Koch's time for five major epidemics: Severe Acute Respiratory Syndrome (SARS), Middle East Respiratory Syndrome (MERS), Coronavirus Disease -19 (COVID-19) and two recent influenza A outbreaks.

Method

The submission or release time of the corresponding virus genome sequence was acquired from NCBI GeneBank database. Cases and response information was provided by searching PubMed, World Health Organization (WHO), Chinese Center for Disease Control and Prevention (CCDC), US Center for Disease Control and Prevention (CDC) and other public data.

Result

Koch's time for COVID-19

Koch's time for COVID-19 is 15 days from December 21, 2019 to January 5, 2020. A cluster of pneumonia cases with an unknown cause occurred in Wuhan, China, starting on December 21, 2019 and some of them were linked to a seafood wholesale market (2). A previously unknown betacoronavirus showing 86.9% identity with a bat SARS-like CoV was discovered by sequencing three bronchoalveolar-lavage fluid samples collected on December 30 (3). The first complete genome sequence of SARS-CoV-2 virus was submitted to GeneBank on January 5, 2020 (accession no: MN908947). Another three complete genome sequences were submitted to GISAID on January 10 (accession ID: EPI_ISL_402119; EPI_ISL_402120; EPI_ISL_402121) (3). On January 11, Chinese Center for Disease Control and Prevention (CCDC) publicly shared the genetic sequence and further detailed information about the novel coronavirus outbreak with the WHO. On the same

day, the PCR test reagent was developed, produced, and distributed to hospitals and research institutions in Wuhan, enabling them to begin nucleic acid test of suspected cases. With the increase of confirmed cases, on January 23, the city of Wuhan began its lockdown. Later, several articles about clinical features and early transmission dynamics of COVID-19 in Wuhan were published online in eminent journals such as the Lancet and the New England Journal of Medicine (4, 5). On January 30, WHO declared the novel coronavirus outbreak a Public Health Emergency of International Concern (PHEIC). On March 11, the WHO announced that COVID-19 can be characterized as a pandemic (6).

Koch's time for SARS

Koch's time for SARS is 148 days from November 16, 2002 to April 13, 2003. Cases of severe acute respiratory syndrome, which was called atypical pneumonia then were first identified in Guangdong, China, on November 16, 2002 and the pathogen was unknown (7). Since February 2003, the epidemic began to spread gradually in countries and regions such as Hong Kong, Vietnam, and Singapore. On February 11, the Chinese Ministry of Health first reported 305 cases of acute respiratory syndrome of unknown cause to the WHO, with 5 deaths (8). On February 18, some Chinese experts concluded that the outbreak was caused by chlamydial organisms, while others believed a virus should be the cause. To identify the real pathogen, WHO gathered 11 laboratories in 9 countries and regions in a collaborative multi-center research project on SARS diagnosis (9). The US Center for Disease Control and Prevention (CDC) isolated virus strains, conducted tests and sequenced clinical specimens from patients in 7 countries and regions (10), and proposed on March 24 that a novel coronavirus may have an etiologic role in SARS. Canadian researchers analyzed clinical specimens and data of first 10 cases in Toronto (11), while British Columbia Cancer Agency (BCCA) sequenced Tor2 isolate from the patient and submitted the first complete genome sequence of SARS-CoV to GeneBank on April 13 (accession no: AY274119) (12). On April 16, the WHO announced that SARS-CoV is the cause of SARS (13). On April 17, seven other complete genome sequences were submitted to GeneBank, including the sequence of Urbani isolate by US CDC (accession no: AY278741) (14), the sequences of HKU-39849 isolate by Hong Kong University (accession no: AY278491) (15), CUHK-W1 isolate by the Chinese University of Hong

Kong (accession no: AY278554) (16), BJ01-03 and GD01 isolates by Academy of Military Medical Sciences/Beijing Genomics Institute (accession no: AY278487-90) (17).

Koch's time for other EIDs

Koch's time for MERS, another disease caused by a novel coronavirus in 2012, is 105 days. This previously unknown coronavirus was isolated from the sputum of a 60-year-old man who was hospitalized on June 13, 2012, with acute pneumonia and subsequent renal failure in Saudi Arabia (18). Researchers from Netherlands conducted sequencing of HCoV-EMC/2012 isolate and submitted to GeneBank on September 26 (accession no: NC_019843) (19), which was the first complete genome sequence of MERS-CoV.

The 21st century has also witnessed epidemics and pandemics of influenza A origins. Koch's time for influenza A (H1N1) pandemic in 2009 is 28 days and Koch's time for influenza A (H7N9) is 75 days. In 2009, a new strain of H1N1 occurred first in Mexico and USA and then spread to the world. On March 30, a 10-year-old boy in California had an onset of fever, cough, and vomiting. His sample was verified to be an influenza A strain that could not be subtyped in clinic and were subsequently sent to CDC. On April 14, the CDC received clinical specimens and determined that the virus was swine influenza A (H1N1) (20). Later they uploaded the complete genome sequence of A(H1N1)pdm09 virus to GISAID on April 20 (accession ID: EPI_ISL_29573, EPI_ISL_29575-7) and submitted it to GeneBank on April 27 (accession no: NC_026431-8). On April 25, the WHO declared influenza A(H1N1) a PHEIC (21). And later, on June 11, WHO announced the start of the 2009 influenza pandemic (22).

During February 25 and March 19, 2013, three patients of Shanghai and Anhui, China, were hospitalized with rapidly progressing lower respiratory tract infections of unknown cause. Further sequencing analysis showed that novel reassortant H7N9 viruses were associated with these cases (23). Complete genome sequences of the viruses from these patients were deposited in GISAID on March 31 (accession ID: EPI_ISL_138737-9) and sequence of A/Shanghai/02/2013 (H7N9) isolate was submitted on GeneBank on May 10 (accession no: NC_026422-9).

Fig 1. Koch's time for different pathogens of five EIDs in 21st century

Discussion

Koch's time for a new pathogen (KTNP) is the time from the first case of attention of an EID to the first complete genome of the new pathogen in a public domain. Although under some circumstances it might be difficult to trace the first case, when available, KTNP provides a way to estimate the global response reaction to EIDs. In twenty years since the beginning of this century, the KTNP of EID caused by unknown pathogens has continuously shortened. Among those caused by a coronavirus, the Koch's time for SARS is 148 days, Koch's time for MERS is 105 days and Koch's time for COVID-19 is 15 days. On the other hand, A(H1N1)pdm09 virus is a new strain of influenza A virus subtype H1N1 that has previously caused several epidemics since 20th century, while H7N9 is a new influenza A virus subtype first identified in 2013. Koch's time for 2013 influenza A(H7N9) is 74 days and Koch's time for 2009 influenza A(H1N1) is 28 days. In summary, the responding speed of COVID-19 is 9.87 times faster than SARS and 1.87 times that of 2009 influenza A(H1N1).

KTNP consists of three parts: clinical response time (sampling), laboratory response time (sequencing), and sequence disclosure time (disclosing). The clinical response time refers to the time from the first case of attention going to the clinic to the time when doctors finish collecting the samples and sending them to the laboratory. The laboratory response time is a period from sample to a report issued with new sequence information. The time when doctors get the report and decide to conduct whole genome sequencing of the pathogen until the result is obtained and released on a

public database (such as GeneBank) is called sequence disclosure time.

Traditional detection methods of pathogenic microorganisms mainly include morphological detection, culture separation, biochemical detection, immunology, and nucleic acid detection, which cannot quickly identify rare or unknown pathogenic microorganisms. Metagenomics next-generation sequencing (mNGS) has the ability to directly perform sequencing on clinical sample without culturing or separation and quickly & accurately determines pathogens by sequence alignment (24, 25), thus greatly shortening laboratory response time. At present, mNGS technology has been applied to the diagnosis of many clinically difficult infections (26-29). In 2019, China proposed an expert consensus on the application of metagenomic analysis and diagnostic technology in acute and critical infections, and introduced the indication of this new technology (30). As a result, clinical response time has significantly reduced in areas equipped with this new detecting method. On the other hand, in recent years much experience has been accumulated in complete genome sequencing, which effectively reduced the sequence disclosure time.

However, we must notice that EIDs could happen at any time in any region and obviously there are differences in pathogen detection and disease management among regions. The continuous shortening of KTNP in general aspect suggests that the identification ability of pathogens of EIDs has made great progress in recent years, providing solid foundation for controlling spread of diseases. EIDs are threats to global health systems, and all countries around the world should work collaboratively and cooperatively to face them.

References

1. Evans AS. Causation and disease: the Henle-Koch postulates revisited. *The Yale journal of biology and medicine*. 1976;49(2):175.
2. Wenjie T, Xiang Z, Xuejun M, Wenling W, Peihua N, Wenbo X, et al. A Novel Coronavirus Genome Identified in a Cluster of Pneumonia Cases Wuhan, China 2019-2020. *China CDC Weekly*. 2020;2(4):61-2.
3. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A novel coronavirus from patients with pneumonia in China, 2019. *N Engl J Med*. 2020.
4. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *The Lancet*. 2020;395(10223):497-506.
5. Li Q, Guan X, Wu P, Wang X, Zhou L, Tong Y, et al. Early transmission dynamics in Wuhan, China, of novel coronavirus – infected pneumonia. *N Engl J Med*. 2020.
6. WHO. WHO Director-General's opening remarks at the media briefing on COVID-19 - 11 March 2020. 2020-03-11 [cited 2020-05-01]; Available from: <https://www.who.int/dg/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing-on-covid-19---11-march-2020>
7. Control CfD, Prevention. Outbreak of severe acute respiratory syndrome--worldwide, 2003. *MMWR Morbidity and mortality weekly report*. 2003;52(11):226.
8. WHO. Severe Acute Respiratory Syndrome (SARS) multi-country outbreak - Update 6. 2003-03-21 [cited 2020-05-01]; Available from: https://www.who.int/csr/don/2003_03_21/en/
9. WHO. WHO collaborative multi-centre research project on Severe Acute Respiratory Syndrome (SARS) diagnosis. 2003-03-17 [cited 2020-05-01]; Available from: <https://www.who.int/csr/sars/project/en/>
10. Ksiazek TG, Erdman D, Goldsmith CS, Zaki SR, Peret T, Emery S, et al. A novel coronavirus associated with severe acute respiratory syndrome. *N Engl J Med*. 2003;348(20):1953-66.
11. Poutanen SM, Low DE, Henry B, Finkelstein S, Rose D, Green K, et al. Identification of severe acute respiratory syndrome in Canada. *N Engl J Med*. 2003;348(20):1995-2005.
12. Marra MA, Jones SJ, Astell CR, Holt RA, Brooks-Wilson A, Butterfield YS, et al. The genome sequence of the SARS-associated coronavirus. *Science*. 2003;300(5624):1399-404.
13. WHO. Update 31 - Coronavirus never before seen in humans is the cause of SARS. 2003-04-16 [cited 2020-05-01]; Available from: https://www.who.int/csr/sarsarchive/2003_04_16/en/
14. Rota PA, Oberste MS, Monroe SS, Nix WA, Campagnoli R, Icenogle JP, et al. Characterization of a novel coronavirus associated with severe acute respiratory syndrome. *Science*. 2003;300(5624):1394-9.
15. Peiris J, Lai S, Poon L, Guan Y, Yam L, Lim W, et al. Coronavirus as a possible cause of severe acute respiratory syndrome. *The Lancet*. 2003;361(9366):1319-25.
16. Tsui SK, Chim SS, Lo YD. Coronavirus genomic-sequence variations and the epidemiology of the severe acute respiratory syndrome. *N Engl J Med*. 2003;349(2):187-8.
17. Zhu Q, Yu M, Fan B, Chang G, Si B, Peng W, et al. A complete sequence and comparative analysis of a SARS-associated virus (Isolate BJ01). *Chin Sci Bull*. 2003;48(10):941-8.
18. Zaki AM, Van Boheemen S, Bestebroer TM, Osterhaus AD, Fouchier RA. Isolation of a novel coronavirus from a man with pneumonia in Saudi Arabia. *N Engl J Med*. 2012;367(19):1814-20.
19. van Boheemen S, de Graaf M, Lauber C, Bestebroer TM, Raj VS, Zaki AM, et al. Genomic characterization of a newly discovered coronavirus associated with acute respiratory distress syndrome

in humans. MBio. 2012;3(6):e00473-12.

20. CDC. Swine influenza A (H1N1) infection in two children-Southern California, March-April 2009. Morb Mortal Wkly Rep. 2009;58(15):400-2.

21. WHO. Swine influenza Statement by WHO Director-General, Dr Margaret Chan. 2009-04-25 [cited 2020-05-01]; Available from:

https://www.who.int/mediacentre/news/statements/2009/h1n1_20090425/en/

22. WHO. World now at the start of 2009 influenza pandemic-Statement to the press by WHO Director-General Dr Margaret Chan. 2009-06-11 [cited 2020-05-01]; Available from:

https://www.who.int/mediacentre/news/statements/2009/h1n1_pandemic_phase6_20090611/en/

23. Gao R, Cao B, Hu Y, Feng Z, Wang D, Hu W, et al. Human infection with a novel avian-origin influenza A (H7N9) virus. N Engl J Med. 2013;368(20):1888-97.

24. Westblade LF, van Belkum A, Grundhoff A, Weinstock GM, Pamer EG, Pallen MJ, et al. Role of clinicogenomics in infectious disease diagnostics and public health microbiology. J Clin Microbiol. 2016;54(7):1686-93.

25. Goldberg B, Sichtig H, Geyer C, Ledebuer N, Weinstock GM. Making the leap from research laboratory to clinic: challenges and opportunities for next-generation sequencing in infectious disease diagnostics. MBio. 2015;6(6):e01888-15.

26. Wilson MR, Naccache SN, Samayoa E, Biagtan M, Bashir H, Yu G, et al. Actionable diagnosis of neuroleptospirosis by next-generation sequencing. N Engl J Med. 2014;370(25):2408-17.

27. Ai J-W, Weng S-S, Cheng Q, Cui P, Li Y-J, Wu H-L, et al. Human endophthalmitis caused by pseudorabies virus infection, China, 2017. Emerg Infect Dis. 2018;24(6):1087.

28. Brown JR, Bharucha T, Breuer J. Encephalitis diagnosis using metagenomics: application of next generation sequencing for undiagnosed cases. J Infect. 2018;76(3):225-40.

29. Wang Q, Li J, Ji J, Yang L, Chen L, Zhou R, et al. A case of Naegleria fowleri related primary amoebic meningoencephalitis in China diagnosed by next-generation sequencing. BMC Infect Dis. 2018;18(1):349.

30. 宏基因组分析和诊断技术在急危重症感染应用的专家共识. 中华急诊医学杂志. 2019(02):151-5.