Chapter 24

Human gut virome and COVID-19: Modulating the extent of infection

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Introduction

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), accountable for the coronavirus disease pandemic in 2019 (COVID-19), led to more than two hundred million infections and four million fatalities as of September 2021. These numbers continued to swell up for a substantial length of time resulting in global healthcare crises and constrained health resources. Although COVID-19 initially manifests as pneumonia in the respiratory system, the virus, being more than just a "flu," can affect many other organs, especially the gastrointestinal (GI) system [1]. Reports claim that respiratory manifestations might lead to various GI symptoms, usually diarrhea, vomiting, digestive upset, etc., and COVID-19 is no different from that [2]. In this context, the role of gut-microbiota in respiratory tract infections is worth mentioning [3]. The "gut-microbiome" is a complex and dynamic pool of microorganisms in the human gut interfaces dominated by bacteria and viruses with a minor population of archaea, fungi, and protozoa. Being a significant component of the GI system, the gut-microbiome is well recognized for being the hub of enormous enzymes and the roles it plays for host metabolism, regulation of homeostasis, immunity development, and resistance against colonization of pathogens [4–7].

Evidences are being piled up showing the link between gut-microbiome and the severity of COVID-19 [8]. Dysbiosis in the gut-microbiome has become a noticeable feature in COVID patients and shifts in the composition of patient's intestinal microbiome as a result of treatment could be utilized as a potential biomarker [9]. In human GI tract, viruses are reported to be predominant among the assorted gut-microbiome [10]. Although viruses are usually considered as pathogens, recent studies highlight a relationship between the human and gut-viruses, as both beneficial and detrimental outcomes for the host could result in local and extra-intestinal immune variation [11]. Besides, SARS-CoV-2 has been reported to attack host intestinal epithelial cells that inputs stress on the microenvironment in the gut as well as, in turn, may proceed to alterations in the gut-virome (GV) [12]. According to several evidences, human GV is also crucial for immunity modulations, disease progression, and shifted homeostatic regulation, by both virus-microbiome and virus-host interactions [13–15], suggesting the possible reason of how changes in the GV can change in whole GI system as well as total health state. As COVID-19 still has many trackless links with the defense system of human bodies, insights into the GV may serve to deepen our sensing of the delicate interactions among host immune response, gut-microbiome and gut-viruses.

Gut-virome and its composition

The human GV comprises a diverse collection of viruses present in our GI tract. They can infect other enteric microbes, hence could directly impact on our well-being. Although virome dominate over the rest of the microbiota in gut, it remains least identified components of GI tract, as development of appropriate analytical toolkits is still in progress. The GV has become an object of study not long ago when viral sequence was first identified from a fecal sample from a healthy adult in 2003 [16]. It has been found that human feces contain around 10^8–10^9 virus-like particles (VLPs) per gram [17,18]. But till now below 1% of the virome has been identified, leaving the bulk of the virome yet to be characterized [19]. Among them, a great portion of these is composed of a staggering diversity of eukaryotic viruses and bacteriophages (prokaryotic viruses) able to cause infection in both host cells and gut-bacteriome, respectively. That being said, bacteriophages are almost 10-fold more prevalent in the gut than eukaryotic viruses in the gut [20,21]. A list of human gut-viruses is shown in Table 1.

Viral, Parasitic, Bacterial, and Fungal Infections. https://doi.org/10.1016/B978-0-323-85730-7.00002-3

Influences of eukaryotic viruses on immune responses

To date, little information is available regarding the connection between intestinal eukaryotic viruses and host immunity. However, the few studies performed so far suggest the possible degree of symbiosis established between enteric eukaryotic viruses and the host gut [9]. Reports about chronically residing viruses (such as Poliomaviruses, Papillomaviruses, Herpesviruses, Adenoviruses, and Hepatitis B and C viruses) in the healthy individuals propose that mutual GV can protect the host from further viral and bacterial infections by causing latent infections itself [33]. Supporting the statement, an interesting experiment on mice showed that chronic infection with its mutually symbiont gamma-herpes virus could increase resistance to both *Listeria monocytogenes* and *Yersinia pestis* pathogens in mice [33]. However, when cell-mediated immunity is jeopardized, selective enteric viruses start to damage intestinal epithelial cells, resulting in overall immune deficiency of the host. Subsequently, this would promote the shifting of enteric bacteria, commensal viruses, and other bacterial molecules from the intestinal surfaces to the circulatory system causing global inflammation [34]. Thereby, chronic viral infections are deemed to lessen the host immunity and promote susceptibility to infection at the same time.

Influences of bacteriophages on immune responses

Gut-bacteriophages are the "parasites" of enteric bacteria, which can cause bacteriome shifts in gut by predation mechanisms. In this framework, a displacement of commensal gut-bacteriome population might occur resulting in intestinal dysbiosis. Consequently, a number of diseases might take place in which microbial dysbiosis is prominent and contribute to the shift from healthy to diseased state in humans [32]. For decades, gut-bacteriophages were known for their indirect influences on human host through modulating the ecological network by infecting commensal bacteriome, but new findings suggests their direct stimulation on the immune system [35], outlined below.

Direct influences

- Enteric phages may directly modulate the immune response by passing through the intestinal epithelial cells once the cell layer is damaged due to any viral infection in gut and/or gut dysbiosis.
- They could promote systemic inflammation in the intestinal area as well as in the whole body via reacting with specific recognition sites of human immune cells that resemble bacterial receptors.
- On the contrary, these invading phages can interact with the intestinal immune system by inducing the production of phage-specific neutralizing antibodies upon nonspecific translocation from gut to other body compartments.

Indirect influences

Intestinal bacteriophages can induce synthesis of infectious phage particles via bacterial cell lysis (lytic cycle) or integrate with bacterial genetic material through lysogenic life cycle (prophage state) [36]. Recently, many reports have proposed some indirect mechanisms that express how commensal bacteriophages could play role in the transition from healthy to diseased state in the host by remaining in both lytic or prophage state [37–41] (Table 2).

SARS-CoV-2 affects gut through gut-lung axis

Recent investigations showed that SARS-CoV-2 caused gastrointestinal (GI) symptoms, for example, nausea, vomiting, and diarrhea in 3.34% to 11.40% of the COVID-19 patients [42]. Moreover, the presence of the viral RNA and live viruses in stool samples of COVID patients supports that lung infections caused by COVID-19 are associated with the host GI system [43,44]. Also, the detection of viral entry point, angiotensin converting enzyme 2 (ACE-2) into host

TABLE 2 Bacteria-mediated (Indirect) influence by gut-virome.

(1)	(2)	(3)
Intestinal phage-mediated bacterial cell rupture causes leakage of PAMPs, which can transit through the enteric epithelium and promote proinflammatory responses.	In the event of imbalanced phage communities in gut, bacteriophage infection may dramatically influence the commensal bacteriome community and lead to abundance of enteric pathogens.	Transmission of prophage-encoded genes can influence the pathogenicity of host bacteria and provide bacterial adaptation for immune evasion by inhibiting phagocytic cells. Also, bacteriophage-mediated genetic transfer can even occur between distinct species of bacteria that can convert friendly bacteria to virulent strains. Thus, enteric phage-mediated horizontal gene transfer might indirectly alter the immune responses to the gut microbial community.

cells of the GI tract provides a subtle suggestion about the capacity of SARS-CoV-2 to infect and replicate in the gut [45]. All these findings postulate that corona virus can modulate the composition of host's gut physiology and the stability of the microbiome in gut, as both depend on each other [46]. Previous studies report that lung-infected patients show characteristic gut issues, and patients with gut issues experience breathing problem contrarily [47]. This is because human gut remains in a two-way cross talk with the lungs, and the conversation happens between the microflora and immune cells of the organs [48]. This profound biological cross-talk is known as "gut-lung axis" that explains the relationship of the infection in lung and the forward imbalance occurred in the gut ecosystem and vice-versa [49]. According to the theory, once human lung is virally infected, it can cause intestinal inflammation by inducing massive release of proinflammatory cytokines (such as-IL-1, IL-2, IL-7, GCSF, IP-10, MCP-1, MIP-1A, and TNFα) leading to the phenomenon, termed as "cytokine storm" [50,51]. Afterward, the commensal gut-flora get affected resulting in the altered gut ecosystem devoid of health beneficial microbiome and supporting the emergence of opportunistic pathogens [52]. These flora release endotoxins and microbial metabolites that can affect the lung through circulatory system, and once further inflammation occurs in the lung, it can upset the gut micro-flora to a greater extent [49]. Such disruption in gut-microbiome balance might result in various metabolic disorders and enhanced susceptibility to secondary enteric infections [53]. Interestingly, the same consequences have been reported as common feature in severe COVID-19 disease [54] (Fig. 1).

SARS-CoV-2 and gut-virome

Following to the latest available SARS-CoV-2 research, a signature bacterial dysbiosis with poor diversity and alteration in viral abundance has been observed in COVID patients of different severities [9]. Then again, comparison of baseline viromes from COVID-19 patients with healthy individuals reported that people infected with SARS-CoV-2 showed an abundance of bacteriophages (including Inviridae and Microviridae) and other unclassified viruses in host gut [55]. The shift of viral composition in gut has long reaching effect on total gut-microbiome that not only influence viral infections in the gut system but also can promote pneumonia in the lung [56,57]. Owing to the "gut-lung axis" theory, corona viral infection is supposed to cause significant chaos by stimulating the "cytokine storm" in the immune system [46]. Once corona virus enters into human body via respiratory system, lung to gut signaling gets activated involving the translocation of immune cells and microbiome. Following that, corona virus enters into the intestine and infect the gut cells due to the presence of ACE-2 receptor in intestinal epithelium. As a result, gut-to-lung communication begins, and this bidirectional communication agitates the whole-body system for a long time. This might be the possible reason behind the prolonged post-COVID complications experienced by SARS-CoV-2 patients. Other associated factors like increment in opportunistic gut pathogens and reduced production of beneficial micro-metabolites are the common features of GI system noticeable in COVID patients [52]. In accordance with metabolic disorder, host intestinal barrier gets damaged due to massive inflammatory infection, leading to the translocation of viruses and other pathogens across the enteric epithelial barrier that exert further immune-modulatory effect on the host [58]. This could further paralyze the lung immunity and may affect the susceptibility and severity of various diseases [59] (Fig. 2).

Therapeutic treatment and gut virome

Enteric phageome in healthy adults comprises mostly of temperate phages that follows lysogenic lifestyle. However, under stressed conditions such as antibiotic treatment in diseased individual, temperate phages get influenced by its potential effects on gut community and the activation of lytic state occurs [60]. This phenomenon was supposed to happen due to the alteration of bacterial community in human body that lead to the shift of commensal phages from temperate to lytic state. Another study on gut bacteriome and phageome upon antibiotic treatment showed a dramatic reduction in bacterial abundance with an opposite trend of phage frequency in stool sample [61]. Furthermore, genomic analysis of overall phageome from that study showed the presence of antibiotic resistant genes suggesting a threat for any future treatment. Antibiotic therapy is well known to influence bacterial community in gut [62], but the dark side of this story signifies that if such bacterial hosts get damaged, it might activate their prophages to produce their gene encoded harmful products. For example, quinolone treatment in mice triggered the Shiga-toxin-encoding prophages residing in *Escherichia coli* to produce lethal toxin [63,64]. As for COVID-19 patients, no certain medication has been approved for their treatment yet. So, to protect them from subsequent bacterial infections, physicians usually prescribe some empirical antibiotics which in turn has been reported to show worse gut-dysbiosis and undesirable altered gut-flora [55]. Hence, random antibiotic therapies should be brought under careful consideration for treating COVID patients as drug use might induce the virome to act negatively on their host. Contrarily, the administration of probiotics as a prophylactic treatment for the management of COVID patients leads to the new insights of novel safeguard while we are still in the hunt for effective medication and vaccine [46].