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REVIEW

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The OM-85 bacterial lysate: a new tool against SARS-CoV-2?

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ABSTRACT

The emergence of SARS-CoV-2, a novel coronavirus, caused the global Coronavirus disease of 2019 (COVID-19) pandemic. Because SARS-CoV-2 mutates rapidly, vaccines that induce immune responses against viral components critical for target cell infection strongly mitigate but do not abrogate viral spread, and disease rates remain high worldwide. Complementary treatments are therefore needed to reduce the frequency and/or severity of SARS-CoV-2 infections. OM-85, a standardized lysate of 21 bacterial strains often found in the human airways, has immuno-modulatory properties and is widely used empirically in Europe, South America and Asia for the prophylaxis of recurrent upper airway infections in adults and children, with excellent safety profiles. *In vitro* studies from our laboratory recently demonstrated that OM-85 inhibits SARS-CoV-2 epithelial cell infection by downregulating SARS-CoV-2 receptor expression, raising the possibility that this bacterial extract might eventually complement the current COVID-19 therapeutic toolkit. Here we discuss how our results and those from other groups are fostering progress in this emerging field of research.

Key words: OM-85 bacterial lysate; SARS-CoV-2; epithelial cell infection; ACE2; TMPRSS2.

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Introduction

A novel coronavirus, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), emerged at the end of 2019 and rapidly spread across the world causing the Coronavirus disease of 2019 (COVID-19) global pandemic [1,2]. The initial steps of SARS-CoV-2 infection are mediated by the binding of viral spike (S) protein to its receptor angiotensin-converting enzyme 2 (ACE2) [3-5]. Cellular entry of the virus depends on S protein cleavage by cellular proteases (reviewed in Jackson *et al*. [6]), in particular, transmembrane protease serine 2 (TMPRSS2) [3]. The S1 proteolytic subunit of the S protein thus generated binds ACE2, while the S2 subunit mediates fusion of viral and cellular membranes [3,6]. Together, these two events are required for the efficient infection of target cells by SARS-CoV-2. The respiratory system is the main target of SARS-CoV-2 because multiple airway epithelial cell types express ACE2 [7,8] and the virus is preferentially spread by aerosol [9]. However, epithelial cells in other organs, such as the intestine and kidney, also express ACE2 and can become infected [10,11]. By April 2020, COVID-19 infections were detected in over 100 countries. To protect vulnerable populations and prevent overloading public health systems, entire countries went into lockdown and introduced harsh measures such as mandatory masking and social distancing [12-14]. Luckily, innovative technologies and massive investments by a number of governments allowed an unprecedentedly fast development of vaccines and drugs that could slow down the spread of the virus and decrease mortality rates [15-18]. Despite these advances, though, COVID-19 endures as a serious global threat to public health and well-being. Indeed, rapidly arising mutated variants of SARS-CoV-2 undermine the effectiveness of vaccines and drugs based on virus blocking-antibodies [19,20]. Immunity induced by previous infections and existing vaccines appears to wane over time [21]. Furthermore, vaccine hesitancy is widespread and many of the new drugs are prohibitively expensive. All of this implies that a significant proportion of the population remains susceptible to SARS-CoV-2 infection. Moreover, with the setting in of COVID-19 fatigue and the lifting of travel restrictions and mandatory masking, the risk of flareups in the number and severity of COVID-19 infections remains high. Clearly, the public health and social complexities of this unprecedented situation call for new drugs and treatments that not only mitigate the spread of SARS-CoV-2 but are also safe and widely affordable.

OM-85 might be one such drug. OM-85 is a standardized, low-endotoxin alkaline lysate of 21 bacterial strains from five genera (*Moraxella, Hemophilus, Klebsiella, Staphylococcus* and *Streptococcus*) found in the human airways [22]. The lysate was shown to have immunomodulatory [23] and anti-viral [24,25] properties, and is widely used to prevent recurrent upper respiratory infections in adults and children [26,27], with an impeccable safety profile [26,28]. Moreover, OM-85 reduced the rate and duration of wheezing attacks in pre-school children with acute respiratory infections [29] and increased the time to severe lower respiratory illnesses in at-risk infants [30]. Because wheezing-inducing lower respiratory tract infections are common harbingers of childhood asthma [31,32], an NIH-sponsored trial (NCT02148796) is currently testing whether OM-85 given to high-risk, 6-18 months old infants for 10 days, monthly, for two consecutive years can prevent or delay the development of wheezing or asthma-like symptoms during a three-year observation period off therapy in these young children.

We recently demonstrated that airway administration of OM-85 prevents cardinal manifestations of asthma in mouse models even when allergens, rather than viruses, induce those manifestations [33]. These effects depend primarily on profound reprogramming of innate immune responses in the lung [33]. It was from these mechanistic asthma studies that the first, unexpected suggestion of a protective effect of OM-85 against SARS-CoV-2 was provided. Indeed, while analyzing the impact of OM-85 on the mouse lung transcriptome, we found that the lysate downregulates the expression of *Ace2,* the primary SARS-CoV-2 receptor [34]. Notably, OM-85 downregulated *Ace2* (as well as *Tmprss2*) in wild-type mice from two different strains (BALB/c and C57BL6), but not in *MyD88/Trif*-deficient mice, pointing to a central role of innate immunity in these effects of OM-85 [34]. Needless to say, these results (initially obtained in the early spring of 2020, right at the time when COVID-19 was becoming a true pandemic), warranted our attention. However, further developing this project in mice would have been problematic because mice are not naturally susceptible to SARS-CoV-2(35). Therefore, the potential role of OM-85 in protecting against COVID-19 was evaluated *in vitro* using human epithelial cells [34].

Impact of OM-85 on discrete events in SARS-CoV-2 epithelial cell infection

Effects on S protein-mediated epithelial cell binding and entry

As we discussed, the first step in SARS-CoV-2 infection is the binding of the S protein to epithelial cells [3-5]. Therefore, we designed a specific assay to evaluate the effect of OM-85 on the binding of recombinant His-tagged S1 protein to the epithelial cell membrane using flow cytometry with fluorescently labeled anti-His antibodies [34]. Experiments were conducted on two epithelial cell lines: Vero E6, which are highly susceptible to infection with SARS-CoV-2 and derive from the kidney of an African green monkey, and Calu-3, adenocarcinoma from human lung. OM-85 dose-dependently inhibited binding of SARS-CoV-2 S1 protein to both cell lines by up to 70% [34].

Cell surface binding of S1 protein is only the first step in the viral infection process. Then, viral particles enter the cell – a step which, in the case of SARS-CoV-2, is mediated by host proteases that cleave the viral S protein (reviewed in Jackson *et al*. [6]). The effect of OM-85 on S protein-mediated cell entry was tested using recombinant lentiviral particles that are pseudotyped with SARS-CoV-2 S protein and carry GFP reporter [34]. Pretreatment of Vero E6 cells with OM-85 reduced the number of green fluorescent cells transduced with SARS-CoV-2 S protein-pseudotyped lentivirus by over 50% [34]. Using a similar approach, another group found that OM-85 strongly and dose-dependently inhibited SARS-CoV-2 S protein-mediated entry of pseudotyped lentiviral particles into human BEAS-2B and Nuli bronchial epithelial cell lines [36]. Furthermore, we demonstrated that OM-85 specifically affected SARS-CoV-2-dependent events because numbers of cells transduced with a control VSV-G-pseudotyped lentivirus were unchanged after treatment with the lysate [34].

Effects on ACE2 and TMPRSS2 expression

Although a number of mechanisms might be responsible for the OM-85-induced inhibition of SARS-CoV-2 S protein-mediated epithelial cell binding and entry [6], we focused on ACE2, which serves as the main receptor for this virus [3-5]. First, we studied whether OM-85 affects ACE2 protein expression on the epithelial cell surface. Flow cytometry with two different fluorochrome-conjugated anti-ACE2 antibodies showed that OM-85 dose-dependently decreased ACE2 surface expression on both Vero E6 and Calu-3 cells [34]. Another study that relied on Western blotting to measure ACE2 in BEAS-2B and Nuli cells as well as primary

bronchial epithelial cells from four healthy donors found significantly reduced ACE2 protein levels in cells treated daily with OM-85 (36). Both studies showed that OM-85 downregulates ACE2 protein expression by inhibiting *ACE2* transcription [34,36]. We found that the lysate significantly decreased *ACE2* mRNA levels in epithelial cell lines from lung (Calu-3), kidney (Vero E6) and colon (Caco-2), as well as primary bronchial epithelial cells from healthy donors [34]. Fang *et al*. on the other hand reported combined results from a different set of primary and immortalized human bronchial epithelial cells [36].

While binding of SARS-CoV-2 to target cells is mediated by ACE2, viral entry into these cells requires proteolytic cleavage of the viral S protein by the endogenous transmembrane protease serine 2 (TMPRSS2) [3]. To understand whether this step in the viral life cycle is also targeted by OM-85, we used quantitative RT-PCR to measure *TMPRSS2* mRNA levels in Vero E6, Calu-3, and Caco-2 cells, as well as in normal primary bronchial epithelial cells. *TMPRSS2* transcription was strongly and significantly inhibited by OM-85 in all cells tested, albeit to a varying degree and with a different kinetics [34]. Strong and significant OM-85-induced inhibition of *TMPRSS2* transcription in human bronchial epithelial cells was also independently reported by others, who also used Western blotting to demonstrate decreased levels of TMPRSS2 protein in OM-85-treated cells [36].

These data from two independent studies indicate that downregulation of *ACE2* and *TMPRSS2* expression is a/the major mechanism underlying OM-85-induced inhibition of SARS-CoV-2 S protein-mediated binding and cell entry. Furthermore, the decrease in *ACE2* and *TMPRSS2* mRNA levels strongly suggest that this downregulation is transcriptionally mediated. Functional evidence for the notion that transcriptional inhibition of the major SARS-CoV-2 S receptor is necessary for OM-85-dependent SARS-CoV-2 suppression in epithelial cells was obtained by comparing the effects of OM-85 on S1 protein binding in Vero E6 and Calu-3 epithelial cells, which naturally express *ACE2,* with those seen in transfected HEK293T cells, in which stable human *ACE2* expression is driven by a heterologous CMV promoter. Unlike Vero E6 and Calu-3 cells, ACE2/HEK293T cells incubated with the same dose of OM-85 showed neither *ACE2* downregulation nor a decrease in S1 binding [34]. Further experiments demonstrated that OM-85 was also unable to affect the entry of S protein-pseudotyped lentivirus into ACE2/HEK293T cells, thereby confirming the dependence of the lysate's effects on OM-85-responsive *ACE2* transcriptional regulation [34].

Inhibition of SARS-CoV-2 epithelial cell infection

The ability of OM-85 to inhibit S protein-mediated epithelial cell binding and virus cell entry suggested that the lysate might protect against COVID-19. To directly assess whether OM-85 suppresses epithelial cell infection with live SARS-CoV-2, we pretreated Vero or Calu-3 cells with different concentrations of OM-85 for 72 or 96 hours and then infected the cells with SARS-CoV-2 (isolate USA-WA1/ 2020). OM-85 dose-dependently inhibited SARS-CoV-2 infection in both cell lines. The inhibition was significant even at the lowest lysate concentration at all time points tested [34]. Of note, OM-85-induced suppression of SARS-CoV-2 infection *in vitro* was observed in epithelial cells from different organs.

Where we are…

SARS-CoV-2 infection is a complex process that starts with the binding of viral spike protein to ACE2 on the surface of target cells. Cleavage of viral S protein by cellular proteases initiates fusion of viral and cellular membranes and leads to viral entry into target cells [6]. Immediately after entry, viral genomic RNA is released, translated, and proteolytically processed into proteins that form the viral replication and transcription complex. This complex is responsible for replication of viral genomic RNA and transcription of viral structural proteins. New viral particles are assembled in the endoplasmic reticulum and Golgi compartments of infected cells and released by exocytosis (reviewed in V'Kovski *et al*. [37]).

Potentially, every step of the SARS-CoV-2 life cycle can be targeted for therapeutic purposes. Most of the vaccines currently in use aim to induce antibody response against the viral S protein to prevent it from binding ACE2. Therapeutic monoclonal antibodies against the S protein, such as bamlanivimab/etesevimab and casirivimab/imdevimab [38,39] as well as convalescent plasma have similar mechanism of action. Some protease inhibitors can block SARS-CoV-2 entry into the target cell, while others (such as nirmatrelvir/ritonavir) interfere with the production of viral proteins responsible for its replication. Another class of therapeutics (e.g., molnupiravir) inhibit replication of viral RNA by RNA-dependent RNA polymerase. Our work suggests that like vaccines, OM-85 may inhibit SARS-CoV-2 binding to its receptor. However, unlike vaccines, OM-85 targets not the virus but the receptor itself. Our study [34] demonstrated that OM-85 efficiently inhibit S1-mediated binding of SARS-CoV-2 to epithelial target cells by downregulating ACE2 expression on their surface. S protein-mediated viral cell entry was also strongly suppressed [34]. In addition to decreased ACE2 expression, OM-85-induced inhibition of *TMPRSS2,* which both we and others observed [34,36], was also likely to contribute to these effects.

In addition to ACE2 and TMPRSS2, other host cell factors that have been suggested to mediate or modulate SARS-CoV-2 infection [6] appear to be targeted by OM-85. For instance, heparan sulfate (HS) was shown to promote the interaction between S protein and ACE2 [40,41]. Incubation of epithelial cells with OM-85 for 3 or more days significantly and dose-dependently decreased the concentration of cellular heparan sulfate while soluble heparan sulfate was increased [36]. These effects have been hypothesized to play a protective role in SARS-CoV-2 infection by inhibiting viral binding to ACE2 or sequestering the virus. The increase in soluble heparan sulfate also correlated with increased production of heparanase after OM-85 treatment, but the significance of this observation is unclear because the role of heparanase in SARS-CoV-2 infection remains controversial. Fang et al also reported significant increases in soluble (s)ACE2 concentrations in cultures of human bronchial epithelial cells treated with OM-85 for 4-5 days [36]. Interestingly, OM-85 also upregulated expression of a disintegrin and metalloprotease 17 (ADAM17)] which was implicated in sACE2 shedding [42,43]. While the role of sACE2 in the pathophysiology of SARS-CoV-2 infection is unclear, increased ACE2 shedding may contribute to OM-85-mediated inhibition of SARS-CoV-2 infection by decreasing the availability of membrane ACE2. It is also worth mentioning that besides its specific inhibitory effects on SARS-CoV-2 infection mediated by ACE2 and TMPRSS2 downregulation, OM-85 has general anti-viral properties that are well-documented in humans [25,27,44] and mice [24,45,46]. OM-85 might inhibit viral infections by mobilizing monocytic cells and upregulating production of interferon [45,46], which can inhibit SARS-CoV-2 [47].

…and what is next

The scenario highlighted by the findings discussed above, and especially the ability of OM-85 to target early stages of SARS-CoV-2 infection, raises the possibility that the lysate might help prevent infection and/or decrease disease severity by limiting viral spread *in vivo*. Moreover, while the emergence of new SARS-CoV-2 variants with mutated S proteins is decreasing the effectiveness of existing vaccines, OM-85 would be expected to retain its activity against these variants because it targets the receptor, not the virus. Indeed, OM-85 might even prevent or limit infection by other coronaviruses, as long as they use ACE2 as their receptor.

Needless to say, the therapeutic potential and indications of OM-85 will remain speculative until its effects on SARS-CoV-2 receptor expression and infection are assessed directly *in vivo,* and optimal therapeutic regimens are experimentally identified. Moreover, besides affecting ACE2 and TMPRSS2 expression, OM-85 has well-documented anti-viral properties [24,25,27,44-46] that have been proposed to rely on monocyte mobilization and interferon upregulation [45,46]. The extent to which these properties might contribute to SARS-CoV-2 inhibition should also be determined. Finally, it will be critical to assess whether OM-85 administration effectively suppresses SARS-CoV-2 infection in cell types other than epithelial cells – first and foremost, endothelial cells, which are emerging as viral targets critical for COVID-19 pathogenesis [48,49]**.** Despite these important lingering issues, the robust safety record of OM-85 [23,26,28] and its approval for clinical use in many countries, combined with an impressive body of mechanistic evidence, imply that this lysate deserves serious consideration as a potential treatment for COVID-19.

Abbreviations

ACE2: angiotensin-converting enzyme 2;
ADAM17: a disintegrin and metalloprotease 17;
COVID-19: coronavirus disease of 2019;
CMV: cytomegalovirus;
GFP: green fluorescent protein;
RT-PCR: reverse transcription polymerase chain reaction;
SARS-CoV-2: severe acute respiratory syndrome coronavirus-2;
S: spike;
TMPRSS2: transmembrane protease serine 2;
VSV: vesicular stomatitis virus.

References

1. Coronaviridae Study Group of the International Committee on Taxonomy of Viruses. The species severe acute respiratory syndrome-related coronavirus: classifying 2019-nCoV and naming it SARS-CoV-2. Nat Microbiol 2020;5:536-44.
2. 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;382:727-33.
3. Hoffmann M, Kleine-Weber H, Schroeder S, Kruger N, Herrler T, Erichsen S, et al. SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor. Cell 2020;181:271-80.e8.
4. Walls AC, Park YJ, Tortorici MA, Wall A, McGuire AT, Veesler D. Structure, function, and antigenicity of the SARS-CoV-2 spike glycoprotein. Cell 2020;181:281-92.e6.
5. Wrapp D, Wang N, Corbett KS, Goldsmith JA, Hsieh CL, Abiona O, et al. Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation. Science 2020;367:1260-3.
6. Jackson CB, Farzan M, Chen B, Choe H. Mechanisms of SARS-CoV-2 entry into cells. Nat Rev Mol Cell Bio. 2022;23:3-20.
7. Kimura H, Francisco D, Conway M, Martinez FD, Vercelli D, Polverino F, et al. Type 2 inflammation modulates ACE2 and TMPRSS2 in airway epithelial cells. J Allergy Clin Immunol 2020;146:80-8.e8.
8. Sungnak W, Huang N, Becavin C, Berg M, Queen R, Litvinukova M, et al. SARS-CoV-2 entry factors are highly expressed in nasal epithelial cells together with innate immune genes. Nat Med 2020;26:681-7.
9. van Doremalen N, Bushmaker T, Morris DH, Holbrook MG, Gamble A, Williamson BN, et al. Aerosol and surface stability of SARS-CoV-2 as compared with SARS-CoV-1. N Engl J Med 2020;382:1564-7.
10. Cheung KS, Hung IFN, Chan PPY, Lung KC, Tso E, Liu R, et al. Gastrointestinal manifestations of SARS-CoV-2 infection and virus load in fecal samples from a Hong Kong cohort: Systematic Review and meta-analysis. Gastroenterology 2020;159:81-95.
11. Puelles VG, Lutgehetmann M, Lindenmeyer MT, Sperhake JP, Wong MN, Allweiss L, et al. Multiorgan and renal tropism of SARS-CoV-2. N Engl J Med 2020;383:590-2.
12. Chinazzi M, Davis JT, Ajelli M, Gioannini C, Litvinova M, Merler S, et al. The effect of travel restrictions on the spread of the 2019 novel coronavirus (COVID-19) outbreak. Science 2020;368:395-400.
13. Perra N. Non-pharmaceutical interventions during the COVID-19 pandemic: A review. Phys Rep 2021;913:1-52.
14. Sharma M, Mindermann S, Rogers-Smith C, Leech G, Snodin B, Ahuja J, et al. Understanding the effectiveness of government interventions against the resurgence of COVID-19 in Europe. Nat Commun 2021;12:5820.
15. Bok K, Sitar S, Graham BS, Mascola JR. Accelerated COVID-19 vaccine development: milestones, lessons, and prospects. Immunity 2021;54:1636-51.
16. Hurt AC, Wheatley AK. Neutralizing antibody therapeutics for COVID-19. Viruses 2021;13:628.
17. Li YD, Chi WY, Su JH, Ferrall L, Hung CF, Wu TC. Coronavirus vaccine development: from SARS and MERS to COVID-19. J Biomed Sc. 2020;27:104.
18. Golob JL, Lugogo N, Lauring AS, Lok AS. SARS-CoV-2 vaccines: a triumph of science and collaboration. JCI Insight 2021;6:e149187.
19. Tregoning JS, Flight KE, Higham SL, Wang Z, Pierce BF. Progress of the COVID-19 vaccine effort: viruses, vaccines and variants versus efficacy, effectiveness and escape. Nat Rev Immunol 2021;21:626-36.
20. Tao K, Tzou PL, Nouhin J, Gupta RK, de Oliveira T, Kosakovsky Pond SL, et al. The biological and clinical significance of emerging SARS-CoV-2 variants. Nat Rev Genet 2021;22:757-73.
21. Levin EG, Lustig Y, Cohen C, Fluss R, Indenbaum V, Amit S, et al. Waning immune humoral response to BNT162b2 Covid-19 vaccine over 6 months. N Engl J Med 2021;385:e84.
22. Bauer Jacques A, Salvagni M, Vigroux Jean-Pierre L, Chalvet L, Chiavaroli C. inventors. Bacterial extract for respiratory disorders and proces for its preparation. WO patent WO2008/109669 A3. 2008/03/05.
23. Esposito S, Soto-Martinez ME, Feleszko W, Jones MH, Shen KL, Schaad UB. Nonspecific immunomodulators for recurrent respiratory tract infections, wheezing and asthma in children: a systematic review of mechanistic and clinical evidence. Curr Opin Allergy Clin Immunol 2018;18:198-209.

24. Pasquali C, Salami O, Taneja M, Gollwitzer ES, Trompette A, Pattaroni C, et al. Enhanced mucosal antibody production and protection against respiratory infections following an orally administered bacterial extract. Front Med (Lausanne) 2014;1:41.
25. Roth M, Pasquali C, Stolz D, Tamm M. Broncho Vaxom (OM-85) modulates rhinovirus docking proteins on human airway epithelial cells via Erk1/2 mitogen activated protein kinase and cAMP. PLoS One 2017;12:e0188010.
26. Gutierrez-Tarango MD, Berber A. Safety and efficacy of two courses of OM-85 BV in the prevention of respiratory tract infections in children during 12 months. Chest 2001;119:1742-8.
27. Schaad UB, Mutterlein R, Goffin H, Group BV-CS. Immunostimulation with OM-85 in children with recurrent infections of the upper respiratory tract: a double-blind, placebo-controlled multicenter study. Chest 2002;122:2042-9.
28. Cao C, Wang J, Li Y, Li Y, Ma L, Abdelrahim MEA, et al. Efficacy and safety of OM-85 in paediatric recurrent respiratory tract infections which could have a possible protective effect on COVID-19 pandemic: A meta-analysis. Int J Clin Pract 2021;75:e13981.
29. Razi CH, Harmanci K, Abaci A, Ozdemir O, Hizli S, Renda R, et al. The immunostimulant OM-85 BV prevents wheezing attacks in preschool children. J Allergy Clin Immunol 2010;126:763-9.
30. Sly PD, Galbraith S, Islam Z, Holt B, Troy N, Holt PG. Primary prevention of severe lower respiratory illnesses in at-risk infants using the immunomodulator OM-85. J Allergy Clin Immunol 2019;144:870-2.e11.
31. Holt PG, Strickland DH, Custovic A. Targeting maternal immune function during pregnancy for asthma prevention in offspring: Harnessing the "farm effect"? J Allergy Clin Immunol 2020;146:270-2.
32. Martinez FD. Childhood Asthma inception and progression: Role of microbial exposures, susceptibility to viruses and early allergic sensitization. Immunol Allergy Clin North Am 2019;39:141-50.
33. Pivniouk V, Gimenes-Junior JA, Ezeh P, Michael A, Pivniouk O, Hahn S, et al. Airway administration of OM-85, a bacterial lysate, blocks experimental asthma by targeting dendritic cells and the epithelium/IL-33/ILC2 axis. J Allergy Clin Immunol 2022;149:943-56.
34. Pivniouk V, Pivniouk O, DeVries A, Uhrlaub JL, Michael A, Pivniouk D, et al. The OM-85 bacterial lysate inhibits SARS-CoV-2 infection of epithelial cells by downregulating SARS-CoV-2 receptor expression. J Allergy Clin Immunol 2022;149:923-33.e6.
35. Johansen MD, Irving A, Montagutelli X, Tate MD, Rudloff I, Nold MF, et al. Animal and translational models of SARS-CoV-2 infection and COVID-19. Mucosal Immunol 2020;13:877-91.
36. Fang L, Zhou L, Tamm M, Roth M. OM-85 Broncho-Vaxom®, a bacterial lysate, reduces SARS-CoV-2 binding proteins on human bronchial epithelial cells. Biomedicines 2021;9:1544.
37. V'Kovski P, Kratzel A, Steiner S, Stalder H, Thiel V. Coronavirus biology and replication: implications for SARS-CoV-2. Nat Rev Microbiol 2021;19:155-70.
38. Cohen MS. Monoclonal antibodies to disrupt progression of early Covid-19 infection. N Engl J Med 2021;384:289-91.
39. Dougan M, Nirula A, Azizad M, Mocherla B, Gottlieb RL, Chen P, et al. Bamlanivimab plus etesevimab in mild or moderate Covid-19. N Engl J Med 2021;385:1382-92.
40. Clausen TM, Sandoval DR, Spliid CB, Pihl J, Perrett HR, Painter CD, et al. SARS-CoV-2 infection depends on cellular heparan sulfate and ACE2. Cell 2020;183:1043-57.e15.
41. Zhang Q, Chen CZ, Swaroop M, Xu M, Wang L, Lee J, et al. Heparan sulfate assists SARS-CoV-2 in cell entry and can be targeted by approved drugs in vitro. Cell Discov 2020;6:80.
42. Lambert DW, Yarski M, Warner FJ, Thornhill P, Parkin ET, Smith AI, et al. Tumor necrosis factor-alpha convertase (ADAM17) mediates regulated ectodomain shedding of the severe-acute respiratory syndrome-coronavirus (SARS-CoV) receptor, angiotensin-converting enzyme-2 (ACE2). J Biol Chem 2005;280:30113-9.
43. Niehues RV, Wozniak J, Wiersch F, Lilienthal E, Tacken N, Schumertl T, et al. The collectrin-like part of the SARS-CoV-1 and -2 receptor ACE2 is shed by the metalloproteinases ADAM10 and ADAM17. FASEB J 2022;36e22234.
44. Aymard M, Chomel JJ, Allard JP, Thouvenot D, Honegger D, Floret D, et al. Epidemiology of viral infections and evaluation of the potential benefit of OM-85 BV on the virologic status of children attending day-care centers. Respiration 1994;61:s24-31.
45. Antunes KH, Cassao G, Santos LD, Borges SG, Poppe J, Goncalves JB, et al. Airway administration of bacterial lysate OM-85 protects mice against respiratory syncytial virus infection. Front Immunol 2022;13:867022.
46. Salzmann M, Haider P, Kaun C, Brekalo M, Hartmann B, Lengheimer T, et al. Innate Immune training with bacterial extracts enhances lung macrophage recruitment to protect from betacoronavirus infection. J Innate Immun 2022;14:293-305.
47. Felgenhauer U, Schoen A, Gad HH, Hartmann R, Schaubmar AR, Failing K, et al. Inhibition of SARS-CoV-2 by type I and type III interferons. J Biol Chem 2020;295:13958-64.
48. Gupta A, Madhavan MV, Sehgal K, Nair N, Mahajan S, Sehrawat TS, et al. Extrapulmonary manifestations of COVID-19. Nat Med 2020;26:1017-32.
49. Varga Z, Flammer AJ, Steiger P, Haberecker M, Andermatt R, Zinkernagel AS, et al. Endothelial cell infection and endotheliitis in COVID-19. Lancet 2020;395:1417-8.

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