Lessons from COVID-19 in children: Key hypotheses to guide preventative and therapeutic strategies

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Key points: Children experience milder COVID-19 disease compared to adults. Differences between pediatric and adult disease mitigating host factors and features of protective immunity can guide development of vaccines and therapeutics for all age groups.

Abstract

The current pandemic of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the causative agent of coronavirus disease 2019 (COVID-19), reveals a peculiar trend of milder disease and lower case fatality in children compared to adults. Consistent epidemiologic evidence of reduced severity of infection in children across different populations and countries suggests there are underlying biologic differences between children and adults that mediate differential disease pathogenesis. This presents a unique opportunity to learn about disease modifying host factors from pediatric populations. Our review summarizes the current knowledge of pediatric clinical disease, role in transmission, risks for severe disease, protective immunity, as well as novel therapies and vaccine trials for children. We then define key hypotheses and areas for future research that can use the pediatric model of disease, transmission, and immunity to develop preventive and therapeutic strategies for people of all age groups.

Keywords: COVID-19, SARS-CoV-2, Children, Pediatrics, Vaccines

Introduction

Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) originated in Wuhan, China in December 2019, and was deemed a public health emergency of international concern by the World Health Organization.[1] SARS-CoV-2 is the etiological agent of the disease known as coronavirus disease 2019 (COVID-19), which is characterized by fever, cough, dyspnea, and progression to acute respiratory distress syndrome (ARDS). In the four months since its identification, SARS-CoV-2 has led to more than 3 million cases and 228,000 deaths globally.[2] Sustained community-based spread is constraining healthcare resources, shutting down economies, and leading to unprecedented governmental recommendations for quarantining and social distancing to limit transmission. While these measures are necessary to slow the rate of new infections, they have been highly disruptive to society and other preventative and therapeutic approaches are urgently needed.

Surprisingly, epidemiological evidence across countries consistently reveals that children experience less severe disease and lower case fatality from COVID-19 than adults.[1,3,4] This trend suggests that there are underlying biological differences between children and adults that could inform the development of therapeutics, and preventative measures. Recent cohort studies indicate that only up to 6% of infected children experience severe disease, whereas up to 26% of adult cases progress to severe illness requiring ICU admission.[5,6] Notably, a similar trend of mild disease and low mortality rate in children was observed during the severe acute respiratory syndrome (SARS-CoV-1) outbreak in 2003 and Middle East respiratory syndrome (MERS)-CoV outbreak in 2012, indicating that this pattern is driven by common virologic features across coronaviruses (CoV).[7,8] Also, varicella disease is similarly known to be milder in young children compared to infants and adults.[9] In contrast, most other respiratory viruses, such as influenza and respiratory syncytial virus, cause more severe disease in young children compared to middle-aged adults[10]. This presents a unique opportunity to learn about disease-

modifying host factors to inform our understanding of CoV pathogenesis across age groups. Understanding differences in children's immunity, host cellular factors required for virus replication, and physiology can provide insights into the correlates of protection from SARS-CoV-2 and other CoVs. In this review, we summarize current pediatric-specific knowledge on clinical disease, transmission, risks for severe disease, protective immunity, and novel therapies and vaccines in trial. Importantly, we identify key unanswered questions in translating this evidence towards the development of preventive and therapeutic interventions for all ages (Table 1).

Epidemiology and Clinical Disease

Currently available clinical descriptions of COVID-19 consistently describe milder symptoms in children than that of adults. While children constitute 22% of the United States population, they only represent 1.7% of SARS-CoV-2 infections identified to date, consistent with estimates from China.[11] Yet, as more pediatric studies have become available, it is clear that children from birth to 18 years can be infected with SARS-CoV-2.[6,12] Infected children appear to be less symptomatic, and thus less likely to be tested for the virus in the setting of limited diagnostic capacity.

While definitions of clinical severity vary among studies, there are consistently fewer severe or critical cases among children than adults. In a retrospective review of over 2,000 pediatric cases in China, only 6% of cases were severe (112 cases) or critical (13 cases).[6] Moreover, in a case series of 36 children in China from whom SARS-CoV-2 was detected, 28% were asymptomatic.[13] In contrast, severe and critical cases represent up to 19% and 26% of adult cases reported in China and Italy, respectively, and occur mostly in people >60 years of age.[1,4]

The most common symptoms of COVID-19 include fever and cough, with fewer patients experiencing shortness of breath, upper respiratory symptoms, vomiting, diarrhea, myalgias, and fatigue.[11,12] Interestingly, only 56% of symptomatic children had fever and 54% had cough, while fever and cough were identified in 71% and 80% of adults, respectively.[11] Laboratory and radiographic abnormalities are also less common in children. While lymphopenia, elevated C-reactive protein (CRP), and abnormal coagulation tests are common in adults and correlate with disease severity, there are no consistent laboratory abnormalities across pediatric studies.[14] However, laboratory abnormalities that more closely reflect those of adults have been reported in children >5 years of age and adolescents.[13,15] In both adults and children, ground glass opacities and "patchy shadows" were the most common abnormalities on chest computed tomography.[16] Altogether, the differences in symptoms and disease severity between children and adults with COVID-19 imply that there are potential immunological or host factors that modulate disease in children.

Transmission

Children less than 15 years of age are primarily exposed to SARS-CoV-2 through close contact with a sick family or household member, although exposure may also occur with travel to an endemic area or contact with other infected individuals.[11] While transmission primarily occurs through aerosolized droplets and fomite contact, there is concern that fecal-oral transmission may also occur, particularly in children. In epidemiological investigations, viral RNA was detected in the stool of 8 of 10 children who tested positive for the virus via nasopharyngeal swab.[17] Moreover, virus was detected in stool up to 27 days after admission, compared with up to 15 days via nasopharyngeal swab and at higher magnitude of viral RNA detected in stool as compared to nasopharyngeal samples, however more studies are needed to determine if detection of viral RNA correlates to infectious virus in stool.[17] Notably, recent reports identified viable virus in fecal samples from adult patients.[18] Given the large proportions of asymptomatic

pediatric infections, lower severity of disease, and potential risk of fecal-oral transmission, it is highly likely that children have a distinct role in population transmission. Development of reliable and specific serological tests for SARS-CoV-2, such as those based on binding of serum antibodies to the viral spike protein, are important for accurate detection of rates of infection in children[19].

The possibility of vertical transmission remains of concern for maternal and neonatal health. In a case series of 33 neonates born to mothers with COVID-19 pneumonia, 3 presented with early onset of neonatal infection identified by detection of the virus by PCR in nasopharyngeal samples, and are suspected cases of perinatal transmission.[20] Also, amongst other cohorts, 17 infants born to SARS-CoV-2 positive mothers did not demonstrate evidence for vertical transmission[21,22]. However, elevated SARS-CoV-2 IgM antibodies detected in serum taken within two hours of birth from three newborns, despite negative testing of nasopharyngeal samples by PCR[21,23,24] is also suggestive of *in utero* SARS-CoV-2 exposure.[21,23,24] Nevertheless, these cases could represent false-positive IgM testing, as has been reported frequently with serological testing for other viruses.[25] Thus far, there is no report of detection of SARS-CoV-2 in amniotic fluid or breast milk, and it is unclear if vertical transmission occurs when pregnant women become infected during the first or second trimester of gestation.[21] Maternal infection can also lead to severe symptoms in the mother, which can result in birth asphyxia or premature birth.[21] In SARS-CoV-1, there was a higher case fatality among pregnant women and reported cases of miscarriage, spontaneous abortion, preterm birth and intrauterine growth restriction.[26] Further research is needed to understand the impact of SARS-CoV-2 infection on maternal and fetal health.

Risk factors for severe disease

While children represent a minority of severe COVID-19 cases, a third of the reported severe cases and more than half of the critical cases were among children less than one year of

age.[6] Children less than 1 year old also had the lowest percentage of asymptomatic cases as compared to older children.[6] An interesting observation in adults is that slightly higher rates of severe disease have been reported in men than women.[27] Similarly, of the >4000 pediatric cases reported in the US and China, 57% were male; however, there are currently no reports of sex differences related to disease severity in children[6,11] Further analysis is required to determine whether a sex bias exists in severe pediatric SARS-CoV-2 infections. Future studies will need to continue examining sex and age-related differences in COVID-19 severity as this might provide insights into host factors that mitigate severe disease outcomes. Moreover, studies should consider whether physiologic changes during puberty underlie age-dependent disease modifying factors in children[28].

The presence of medical comorbidities, such as hypertension, diabetes, chronic pulmonary disease, and cardiovascular disease is another risk factor for severe disease in adults[29], and the relative lack of comorbidities in children may contribute to the disparate COVID-19 severity between the age groups. Of the few reports of severe COVID-19 disease in children, all three critical cases had a significant underlying or concurrent medical condition, including acute lymphoblastic leukemia (ALL), hydronephrosis, and intussusception.[30] However, it should be noted that none of the five severe cases had significant comorbidities.[30] Given the low prevalence of severe and critical disease in children, it is difficult to determine the contribution of pre-existing comorbidities to COVID-19 severity. Specifically, underlying medical issues such as prematurity, chronic lung disease, congenital heart disease, asthma, and even lung injury from vaping and smoking, may result in an increase in the risk for severe COVID-19 disease.

Intriguingly, there are few reports of severe disease in immunocompromised patients with COVID-19 despite receipt of immunosuppressive agents and chemotherapies. While data are limited to small cohort studies, adult renal transplant recipients tended to have a typical

COVID-19 course, while adults with malignancy had more severe disease if they had recently received chemotherapy or underwent surgery.[31,32] Of immunocompromised SARS-CoV-2 infected children, the aforementioned child with ALL developed critical disease, but only mild to moderate disease has been observed in pediatric liver transplant recipients.[30,33] The surprisingly mild course of COVID-19 in immunocompromised patients could allude to the substantial role that the host immune system plays in the development of severe disease.

Disease mitigating characteristics of children

Protection from severe disease in children may be related to lower expression of host factors required for viral replication, and to differences in the magnitude and timing of innate or adaptive immune responses.

Host factors: SARS-CoV-2 uses the angiotensin-converting enzyme 2 (ACE2) as a cell entry receptor and the cellular transmembrane protease serine 2 (TMPRSS2) to activate the spike (S) viral protein for membrane fusion.[34] ACE2 modulates vasoconstriction to maintain homeostasis and is expressed in the oral mucosa, respiratory tract, and intestine.[35,36] Lower ACE2 expression in the lungs of children as compared to adults could contribute to the observed differences in disease pathogenesis across these groups.[37] However, given the large variability in human ACE2 expression profiles, further studies are required to confirm differences across age groups.[37] There are also age-dependent differences as lungs develop throughout childhood[38]. In particular, processes that impact the course of lung pathology and respiratory distress such as inflammation, apoptotic activation, surfactant secretion, alveolar fluid clearance, and tissue repair mechanisms differ in children compared to adults.[38] For example, a regulator of lung morphogenesis that is lower in childhood, nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κb), plays a pathologic role in inflammatory diseases and should be evaluated as a protective host factor in pediatric versus adult SARS-CoV-2 infections.[38,39] Indeed, even outside the context of SARS-CoV-2 infections, rates of ARDS are

lowest in children and increase with age, suggesting a role for protective host factors in the lungs of children.[40,41]

Innate and adaptive Immunity: Th1 responses are thought to be important for immune protection against SARS-CoV-1 since increased Th2 cytokines were identified in patients with fatal disease.[42] However, excess Th1 pro-inflammatory cytokine responses and circulating neutrophil levels are also associated with increased disease severity and delays in regulatory and repair responses.[43,44] In fact, over expression of serum IL-6 is associated with severe disease and mortality due to SARS-CoV-2 infection suggesting that aging-related inflammation may contribute to disease severity in elderly.[45] Whereas, children who recovered from SARS-CoV-1 infection demonstrated elevated plasma IL-1β but not TNF-α or IL-6 early in infection, suggesting a less destructive disease pathology.[46] Descriptions of lung pathology from SARS-CoV-1 and SARS-CoV-2 fatalities reveal that macrophages are the predominant leukocyte infiltrate in the alveoli[43,47]. Higher prevalence of macrophages in the alveoli may be due to prolonged IL-6 inflammation, in combination with monocyte chemoattractant protein-1 (MCP-1) expression, which induces a transition from neutrophil activation in early inflammation to monocyte accumulation in late inflammation[48]. Interestingly, lower levels of IL-6 and MCP-1 are observed in the lungs of children who survive ARDS compared to adults.[37] Although neutrophils are associated with lung pathology during ARDS, the role for lung neutrophils in COVID-19 severity remains unclear.[49] Neutrophil depletion in rodent models of respiratory viral infections such as SARS-CoV-1, influenza, and respiratory syncytial virus leads to worse clinical outcomes and higher levels of viral replication, suggesting that neutrophils may serve a protective function during these infections[38,50–53]. Thus, the role for neutrophils and macrophages in SARS-CoV-2 infections needs to be evaluated further and compared between children and adults. Effective immune responses to CoV require regulated Th1 immunity for viral control and infected cell killing, followed by regulatory signaling that mediate tissue repair[54]. Intriguingly, children

experience less leukopenia during SARS-CoV-2 infection than adults and have a relatively higher level of circulating lymphocytes compared to neutrophils, which may contribute to better viral control during acute infection.[5,12,55] Thus, milder SARS-CoV-2 infection in children may be driven by intrinsically lower levels of inflammation, higher lymphocyte to neutrophil ratio in blood, and less predominantly monocytic infiltration than adults.

Recent reports have demonstrated that neonates less than 1 year of age (<1 year) are more susceptible to severe COVID-19 disease compared to older children (1-18 years).[6] Dynamics of T cell-mediated immunity may contribute to the increased COVID-19 severity in adults and neonates (<1 year) compared to the milder disease observed in children (<18 years). For example, virus-specific CD8+ T cells play an important role in viral clearance by directly killing infected cells, but excess cytolytic activity can also mediate lung pathology[56]. The observed increase in lung pathology in both infants <1 year and older adults may be due to inappropriate levels of T cell activity. Indeed, in infants <1 year T cell activation is decreased and effector responses are characterized by Th2 cytokine secretion as infants transition from tolerogenic fetal immunity[57]. In contrast, higher inflammation associated with aging can lead to T cell exhaustion, which is linked with severe COVID-19 disease.[58] In comparison, children between 1-18 years may experience an intermediate level of T cell activation, leading to milder SARS-CoV-2 disease.[59] Also, an age-dependent increase in lung prostaglandin production may play a role in SARS-CoV-2 pathogenesis. For example, in mice lung prostaglandin concentrations correlated with decreased dendritic cell migration and T cell responses and greater SARS-CoV-1 induced lung pathology with age.[60] Further examination of children T cell immunity during SARS-CoV-2 infection compared to adult responses is required.

Our understanding of protective humoral responses to CoV infections comes from prior studies of SARS-CoV-1, and ongoing studies on the current SARS-CoV-2 pandemic. Typically, neutralizing antibody responses against the immunodominant S viral protein are elicited after

two weeks of infection and can protect from challenge in animal models.[61] Yet, high magnitude and early (< 2 week) peak neutralizing antibody responses were associated with more severe disease in SARS-CoV-1 infection, indicating antibody responses may also be related to disease pathology.[62] Moreover, the SARS-CoV-2 S protein contains neutralizing and non-neutralizing epitopes and is 76% identical to SARS-CoV-1 S at the amino acid level.[63,64] Further studies should evaluate whether this homology leads to cross-protective CoV antibody responses, that can be leveraged to design vaccines that target multiple CoVs for the development of a universal CoV vaccine. It is known that infants can elicit immune responses of greater breadth compared to adults and develop higher magnitude antibody responses to some protein vaccine antigens like Human Papillomavirus Virus-Like-Particle, HIV, and Hepatitis B.[65–69] Investigating the diversity and potency of pediatric antibody responses may help to define immune correlates of protection against COVID-19 and cross-protective epitopes on S to guide long-term CoV vaccine strategies.

Vaccine and therapeutic development

Globally, eight SARS-CoV-2 vaccine candidate platforms have entered into Phase 1 clinical trials: 1) a non-replicating lipid nanoparticle mRNA candidate encoding pre-fusion SARS-CoV-2 S protein (mRNA-1273; Moderna Inc and US NIH; Trial ID NCT04283461); 2) a replication-defective human adenovirus type-5 vectored SARS-CoV-2 candidate (Ad5-nCoV; CanSino); 3) a non-replicating chimpanzee adenoviral vectored SARS-CoV-2 S protein (ChAdOx1 nCoV-19, University of Oxford Jenner Institute); 4) A double stranded DNA plasma encoding spike protein that is delivered via electroporation (INO 4800; Inovio Pharmaceuticals, Trial ID: NCT04336410); 5) An inactivated SARS-CoV-2 candidate with multiple strains (PiCoVacc; Sinovac Biotech); 6) Probiotic with live *Bifidobacterium longum* that contain DNA plasmids for the SARS-CoV-2 spike protein (bacTRL-Spike, Symvivo; Trial ID: NCT04334980); 7) Four lipid nanoparticle-based mRNA candidates encoding the viral spike protein or receptor

binding domain nucleoside modifications (BNT162, BioNTech and Pfizer); 8) two lentiviral vector candidates expressing viral proteins and immunomodulatory genes (Shenzhen Geno-Immune Medical Institute; Trial IDs: NCT04299724 and NCT04276896).[70–75] Launching clinical trials within four months from SARS-CoV-2 discovery represents an incredible achievement for vaccine developers, with the mRNA vaccine being injected into an adult volunteer only 65 days after genomic elucidation.[70] This feat was made possible by advances in vaccine technology including the development of mRNA and vectored-based platforms that were tested in other emerging virus vaccines. Yet, all except the inactivated virus vaccine candidate, represent vaccine platforms that are not amongst licensed pediatric vaccines, with minimal data available for safety in children from clinical trials. In non-human primates and mice, delivery of double-inactivated SARS-CoV-1 vaccine candidates followed by challenge with homologous or heterologous strains, has led to eosinophilic lung immunopathology.[76,77] Intriguingly, young mice showed less immunopathology as compared to aged mice, indicating that age of vaccination may impact safety profile. Though it is unclear whether lung immunopathology reflects enhanced disease in humans, whole virus vaccine platforms must be carefully evaluated for safety. Lack of vaccine candidates with a proven safety and/or immunogenicity profile represents a gap in translating these technologies to pediatric populations during a pandemic. While rapidly testing candidates, it will be crucial to consider the earliest possible stage for inclusion of children in vaccine trials.

A key question for vaccine development in the current pandemic is the possibility of reinfection with SARS-CoV-2. Prior studies indicate that reinfection may be possible after several years, since SARS-CoV-1 neutralizing antibody titers reduced substantially 3 years after exposure and virus-specific memory B cells were undetectable 6 years after infection.[78,79] Further, virus-specific memory T cells were undetectable by 6 years post infection in 40% patients who recovered from SARS-CoV-1 infection.[79] Therefore, it will be important to assess if

SARS-CoV-2 immunity in children lasts longer than that of adults, which would indicate that childhood represents an opportune period for vaccination to elicit life-long protection. Also, differential waning of vaccine immunity in adults and children should be evaluated to optimize age of vaccination and develop boosting strategies to provide long term protective immune responses.

Two leading antivirals are currently being tested in patients with COVID19. Remdesivir is an intravenously delivered investigational antiviral that that is being tested in several randomized controlled clinical trials globally, largely in adults with moderate or severe COVID-19.[80] Remdesivir is a nucleoside analog that inhibits CoV replication by terminating the RNA genome transcription.[81] Assessments in children are underway to determine optimal pediatric dosing. Another option being tested is hydroxychloroquine, an approved oral antimalarial drug that is also used for rheumatoid arthritis and systemic lupus. While hydroxychloroquine demonstrates high antiviral activity in-vitro, underpowered clinical trials have indicated virologic control but no significant effect on clinical outcomes in patients with severe disease.[82]

In addition to antivirals, passive immunization strategies using convalescent plasma and purified immunoglobulins to limit virus replication and abrogate disease progression are under investigation. Reports of successfully treating critically ill COVID-19 patients with convalescent plasma from recovered individuals has enabled approval for emergency use in the US for cases of serious and life threatening COVID-19.[83] Meta-analysis of this therapy for SARS-CoV-1 suggests that this intervention appears safe and reduces mortality.[84] However, since antiviral potency of plasma may vary by donor, it is important to determine the characteristics of plasma that support efficacy and optimal prognosis. For example, poor treatment outcomes for SARS-CoV-1 patients were observed when convalescent plasma intervention was administered during PCR-positivity and before the 14th day of illness.[85] These observations allude to the relationship

between viral dynamics and IgG-mediated pathology that may differ between adults and children.

Conclusion

The current COVID-19 pandemic has resulted in more than 3 million cases worldwide, and the lack of protective vaccines and specific antiviral therapies to prevent severe disease has resulted in more than 228,000 deaths[2]. A pattern of milder COVID-19 in children in compared to adults offers a unique opportunity to identify protective host and immunologic factors within pediatric populations and apply findings to the design of interventions for all ages. In this review, we evaluated recent reports on the pathology and immunity to SARS-CoV-2 infection and offered several hypotheses for how these features may differ in children versus adults, and how they may differentially modulate disease in these populations. Further understanding of the pathogenesis of SARS-CoV-2 infection in children may provide important insights and guide development of therapeutic strategies and vaccines as we collectively strive to generate approaches to reduce the public health burden of SARS-CoV-2 pandemic.

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Table 1 - Characteristics of pediatric COVID-19 infections and key questions to inform prevention and treatment across all age groups.

	What We Know	Key Questions
Clinical presentation	• Less severe disease in children compared to adults • Substantial asymptomatic infections in children • All ages susceptible to infection; positive testing in ages 1 day-18 years	• What immunological or host factors predispose children to a milder illness compared to adults? • What, if any, differences in disease course exist for immunocompromised children?
Transmission	• Aerosolized droplets, contact with contaminated surfaces and potentially fecal-oral • Most pediatric infections acquired through familial clusters, some through community-based spread • Asymptomatic infections in children may facilitate spread • Suspected perinatal transmission	• Role of children's mild and asymptomatic infections in community transmission • Magnitude and duration of viral shedding in children compared to adults: super spreaders? • Does viral shedding in stool indicate potential fecal-oral route of transmission? • Are there pediatric-specific differences in reproductive number and incubation period as compared to adults? • Can SARS CoV-2 be vertically transmitted? • Is there differential risk of acquiring infection in infants versus adults, despite lower severity?
Risk factors for severe disease	• Among children, there is more severe and critical disease among neonates and infants <1 year • Severe presentation may have male preponderance in adults and children • Pediatric patients with severe/critical disease tend to have other underlying medical problems. There are few reports of severe or critical disease in infected immunocompromised patients	• What mechanisms lead to more severe disease in neonates and infants? • How does sex impose risk for severe disease? • What is the role of vaping, asthma, hypertension, prematurity, and cardiopulmonary disease on risk of severe disease in children? • What is the risk of severe disease in immunocompromised children?

Protective immunity	• Infants mount higher magnitude immune responses to protein vaccines like HPV and HepB as compared to adults, indicating the potential for early life immunization for lifelong protection
	• Several hypotheses about why children may have milder disease than adults:
	  ○ Host factors (e.g. cell entry enzymes such as ACE-2) are differentially expressed in infants as compared to adults
	  ○ Different magnitudes of aberrant immune responses in infant lung tissues as compared to adult
	  ○ Children are more likely to develop cross-protective immune responses as compared to adults
	• Phenotype is recapitulated in the monkey model of SARS-CoV1 infection, where older animals are more susceptible to disease than younger animals
		• What are immune correlates of protection for milder disease in children?
		• Does viral inoculum or load correlate with disease pathogenesis in infants as compared to adults?
		• To what extent does inflammation mediate protection versus pathology in infants?
		• Does immunity to other, more common circulating respiratory viruses or vaccines (such as BCG) offer [partial] immunity to SARS-CoV-2?
		• Does passive maternal immunity protect neonates and infants from severe disease?
		• What are the immunologic and virologic biomarkers that predict severe disease in infants?
		• How do infant respiratory tract cells respond differently to SARS-Cov2 infection as compared to adult cells?
		• Which host factors and underlying diseases modulate disease severity?

Therapies and vaccines	• No FDA-approved vaccines or antivirals • No therapy above supportive care has been shown to provide clinical benefit • Remdesivir and hydroxychloroquine demonstrate potent antiviral activity *in vitro*, though clinical trials are necessary to assess efficacy • Remdesivir in phase 3 clinical trials enrolling patients ≥12 years • mRNA vaccine encoding spike protein in Phase I clinical trial • Novel vaccine platform using measles live attenuated vector with SARS-CoV-2 antigens shows protection in mice	• Will antivirals suppress viral load in vivo as well as lower clinical pathology? • In which target population will the vaccine be most effective and durable? • What will be the effect of pre-existing immunity and maternal antibody on the vaccine? • Can children respond most effectively to this vaccine, and will infancy be the optimal timing to achieve lifelong protection? • How soon can we include vulnerable populations including pregnant women, neonates, and children in the vaccine development process to optimally tailor vaccine design to these populations? • Can we leverage understanding of protective pediatric immunity and pathophysiology to guide design of therapeutic targets and vaccines?