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Antigen epitopes of animal coronaviruses: a mini-review

Mingjun Su[1†], Guanghui Zheng[1†], Xiangwen Xu[1] and Houhui Song[1*]

Abstract

Coronaviruses are widespread in nature and can infect mammals and poultry, making them a public health concern. Globally, prevention and control of emerging and re-emerging animal coronaviruses is a great challenge. The mechanisms of virus-mediated immune responses have important implications for research on virus prevention and control. The antigenic epitope is a chemical group capable of stimulating the production of antibodies or sensitized lymphocytes, playing an important role in antiviral immune responses. Thus, it can shed light on the development of diagnostic methods and novel vaccines. Here, we have reviewed advances in animal coronavirus antigenic epitope research, aiming to provide a reference for the prevention and control of animal and human coronaviruses.

Keywords Animal coronavirus, Antigen epitope, B-cell epitope, T-cell epitope, Immune responses, Vaccines

Introduction

Coronaviruses (CoVs) are positive-sense, single-stranded RNA viruses belonging to the *Coronaviridae* family of the Nidovirales order. Based on their genome sequences, they fall into four genera: *alphacoronavirus* (α-CoV), *betacoronavirus* (β-CoV), *gammacoronavirus* (γ-CoV), and *deltacoronavirus* (δ-CoV) (Li 2016; Zhang et al. 2021a). Coronaviruses have a genome size of approximately 25,000–30,000 nucleotides (nt) and consist of at least six open reading frames (ORFs) in the following order: ORF1a, ORF1b, spike (S), envelope (E), membrane (M), and nucleocapsid (N). Some coronaviruses encode hemagglutinin esterase (HE) (Brian and Baric 2005).

Animal coronaviruses (animal CoVs) have a wide range of host tropisms and represent a health risk to livestock production. Since the discovery of infectious bronchitis virus (IBV) in the 1930s, numerous animal CoVs have been identified in pigs, rats, cats, dogs, cattle and horses, belonging to four subgenera of the family *Coronaviridae* (Fig. 1) (Hudson and Beaudette 1932; Woo et al. 2009; Zhang et al. 2021a). It is noteworthy that the prevention and control of emerging and re-emerging CoVs, such as porcine deltacoronavirus (PDCoV), swine acute diarrhea syndrome coronavirus (SADS-CoV), and porcine epidemic diarrhea virus (PEDV), represents a major global challenge (Wang et al. 2019). Antigen epitopes are chemical moieties located on the surface of an antigen molecule that possess a unique structure and antigenic activity. They represent a bioactive region on the antigen molecule that can stimulate the host immune system to produce antibodies or immunogenic lymphocytes and can be recognized by these immune cells. Therefore, the investigation of antigenic epitopes helps our understanding aboutvirus-mediated immune response and provides a basis for the design of antiviral strategies, which is an intense area of virology research. Although the high

†Mingjun Su and Guanghui Zheng contributed equally to this work.

*Correspondence:
Houhui Song
songhh@zafu.edu.cn
[1] Key Laboratory of Applied Technology On Green-Eco-Healthy Animal Husbandry of Zhejiang Province, Zhejiang Provincial Engineering Research Center for Animal Health Diagnostics & Advanced Technology, Zhejiang International Science and Technology Cooperation Base for Veterinary Medicine and Health Management, China-Australia Joint Laboratory for Animal Health Big Data Analytics, College of Animal Science and Technology & College of Veterinary Medicine of Zhejiang, A&F University, 666 Wusu Street, Lin'an District, Hangzhou 311300, Zhejiang Province, China

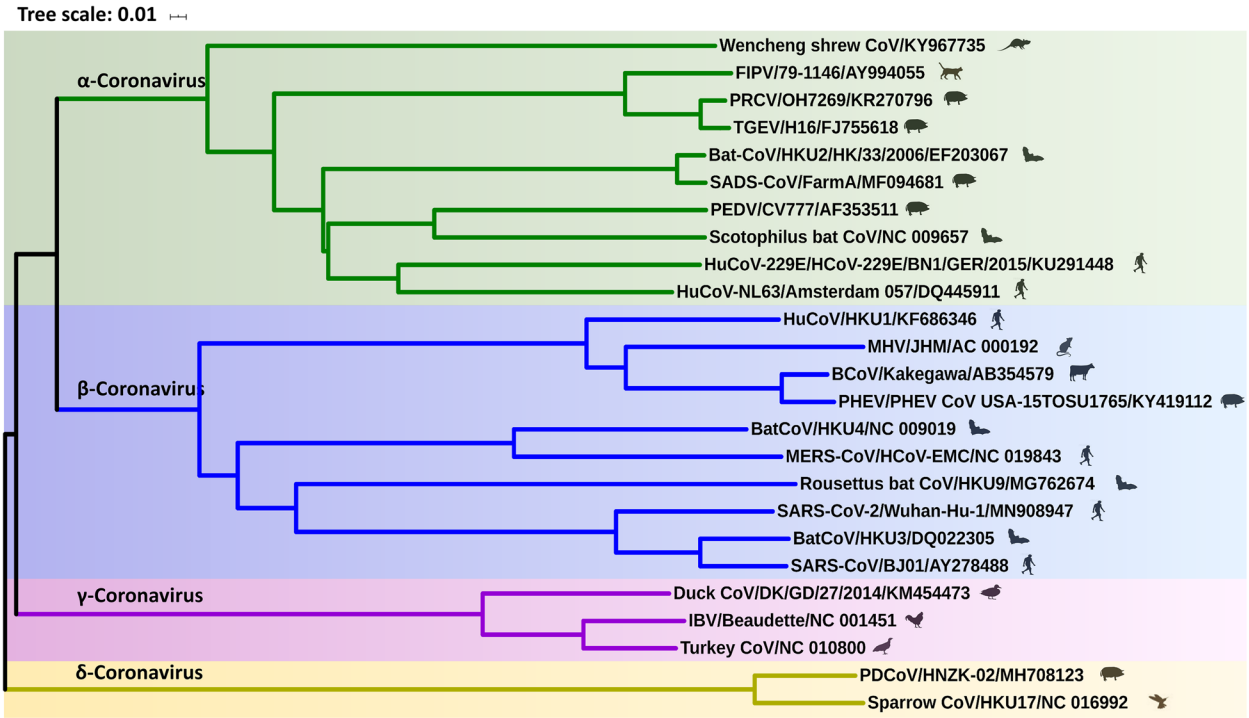

Fig. 1 Phylogenetic analysis based on the genome of coronavirus. A neighbor-joining phylogenetic tree was built using the p-distance model and 1,000 bootstrap replicates

output of research on animal CoV antigenic epitopes has promoted the study of antigenic epitope diagnostic methods and vaccines, there are still challenges in the identification and application of animal CoV antigen epitopes.

Distribution of antigen epitopes in animal CoVs

Based on the interaction with antigen receptors on cells, epitopes are classified as B-cell or T-cell epitopes, which can induce humoral or specific cellular immunity, respectively. Continuous antigenic epitopes, also known as linear epitopes, are characterized by a continuous stretch of amino acids in the peptide chain. They are typically found on T-cell epitopes and some B-cell epitopes. Conformational antigenic epitopes, on the other hand, are formed by amino acids that are adjacent in space but not continuous in sequence and are only present on B-cell epitopes (Barlow et al. 1986). According to the immune epitope database (IEDB; www.iedb.org) (Vita et al. 2010), multiple B- and T-cell epitopes have been identified in several animal CoVs, including TGEV, PEDV, SADS-CoV, FIPV, MHV, BCoV, IBV and PDCoV (Fig. 2A) (Table S1).

B-cell epitopes have been reported in all eight animal CoVs mentioned above. The main epitopes are on S, M and N proteins (Fig. 2A, Blue), and two B-cell epitopes have been found on NS7a protein of SADS-CoV (Qin et al. 2022). Additionally, four and six B-cell epitopes have been reported on the RNA-dependent RNA polymerase (RdRp) of TGEV and on the RNA polymerase of MHV, respectively (Mathieu et al. 2004; Nogales et al. 2011). Conformational epitopes are epitopes that bind specifically to antibodies that block the cellular receptors used by viruses to bind to cells. This is a critical step in the development of diagnostic reagents. However, only one potential epitope has been detected in the PEDV N protein (residues 18–133) (Wang et al. 2016). T-cell epitopes have been reported in four animal CoVs, including FIPV, MHV, IBV and TGEV (Fig. 2A, Orange). Similar to B-cell epitopes, T-cell epitopes are predominantly located on the S protein, followed by N and M proteins, and two T-cell epitopes have been reported on replicase polyprotein 1ab (pp1ab) of MHV (Croxford et al. 2006; Ercolini et al. 2007).

Coronavirus S protein is incorporated into the viral envelope and facilitates viral entry into target cells. In this process, the surface unit S1 binds to a cellular receptor, while the transmembrane unit S2 enables fusion of the viral membrane to the host cell membrane (Hulswit et al. 2016; Millet and Whittaker 2018). Membrane fusion requires S protein cleavage by host cell proteases at the S1/S2 site, resulting in S protein activation (Hoffmann et al. 2020; Hulswit et al. 2016; Millet and Whittaker

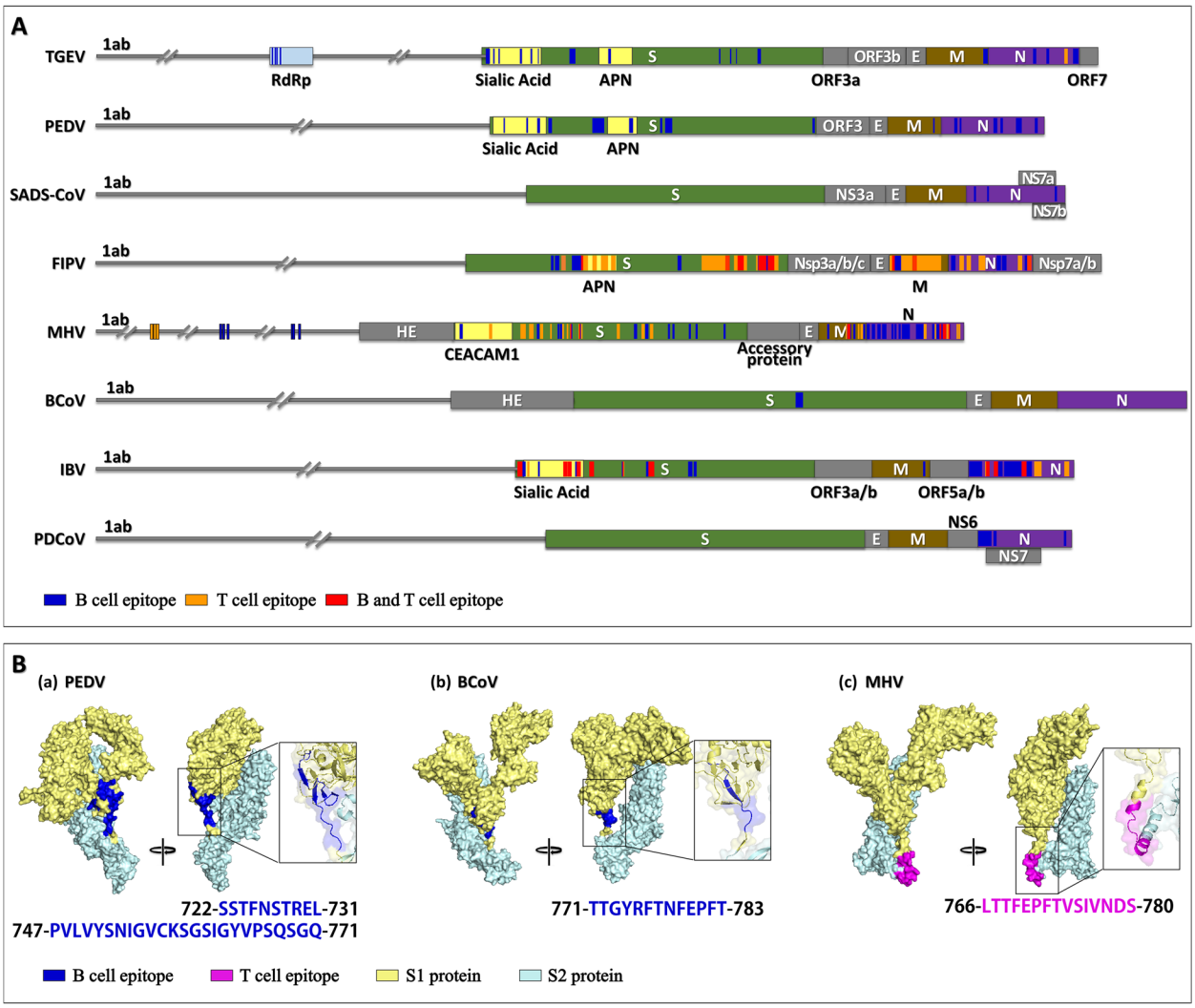

Fig. 2 **A** Location of antigen epitopes in animal CoV genomes. **B** Distribution of antigen epitopes in the S1/S2 junction of animal CoV S proteins. The predicted tertiary structures of the S region of PEDV (PDB ID: 6VV5), BCoV (PDB ID: 6NZK), and MHV (PDB ID: 3JCL) were modeled using the open-source modeling server SWISS-MODEL (https://swissmodel.expasy.org/) from the Swiss Institute of Bioinformatics (Biasini et al. 2014). Illustrations of these modeled tertiary structures were obtained using the python-based molecular viewer PyMOL (The PyMOL Molecular Graphics System, V. 1.7.4 Schrödinger, LLC)

2018). The 3D structure model of S protein showed that the antigen epitopes of PEDV (722–731, 747–771), MHV (766–780), and BCoV (711–783) were identified at the S1/S2 junction (Gillam and Zhang 2018; Khanolkar et al. 2010; Kong et al. 2020; Okda et al. 2017; Sun et al. 2008; Vautherot et al. 1992) (Fig. 2B).

Applications of animal CoV antigen epitopes

Application of antigen epitopes in vaccines

Antigenic epitopes can induce humoral immunity or specific cellular immunity that is crucial for inducing antiviral immune responses. They are commonly deployed in the development of safe and effective vaccines. Currently, the known animal CoV antigen epitopes offer a basis for the development of epitope vaccines (Table 1).

S proteins, especially the S1 of PEDV, are a key target for virus neutralization and vaccine development (Hain et al. 2016; Makadiya et al. 2016; Oh et al. 2014; Subramaniam et al. 2017). The B-cell epitope Y^{748}SNIGVCK^{755} on S1 protein of PEDV has been validated for potential use in vaccine development (Gillam et al. 2018; Okda et al. 2017). Gillam reported that a Y^{748}SNIGVCK^{755}-based epitope vaccine that used hepatitis B virus core antigen (HBcAg) as a vector efficiently elicited the production of anti-PEDV neutralizing antibodies in mice (Gillam

Table 1 Application of animal CoV antigen epitopes

Coronaviruses (protein)	Sequence	Application	References
PEDV (S-COE)	Y^{748}SNIGVCK^{755}	Vaccine: production of neutralizing antibody (used HBcAg as vector)	Gillam and Zhang 2018
PEDV (S)	T^{592}SLLASACTIDLFGYP^{607}	Vaccine: production of neutralizing antibody	Sun et al. 2019
FIPV (S)	N^{621}NYLTFNKFCLSLSPVGANC^{640}	Vaccine: induce Th1 activity	Takano et al. 2014
FIPV (M)	V^{81}YGIKMLIMWLLWPIVLALT^{100}	Vaccine: induce Th1 activity	Takano et al. 2014
FIPV (N)	G^{81}QRKELPERWFFYFLGTGPH^{100}	Vaccine: induce Th1 activity	Satoh et al. 2011
IBV (S)	S^{413}RIQTATDP^{421}, ^{517}RNATGSQP^{525}, G^{45}AYAVVNV^{52}, S^{413}RIQTATQP^{421}	Vaccine: stimulate CD8$^+$T-cell proliferation and IFN-γ secretion	Tan et al. 2016
TGEV (S)	TGEV S-A site (533–705)	Vaccine: increase the Th1 and Th2 cytokine levels	Gebauer et al. 1991
MHV (S: S1/S2 junction)	L^{766}TTFEPFTVSIVNDS^{780}	Vaccine: induces CD4 T-cell response in mice	Khanolkar et al. 2010
PEDV (S-COE)	T^{592}SLLASACTIDLFGYP^{607}	Diagnosis: mABs 4D8F10 and 6F3E3 recognize the COE, and highly conserved	Sun et al. 2019
TGEV (S)	T^{592}SLLASACTIDLFGYP^{607}	Diagnosis: highly conserved	Gebauer et al. 1991
MHV (N)	I^{24}LKKTTWADQTERGL^{38} R^{357}FDSTLPGFETIMKVL^{372}	Diagnosis (ELISA): more sensitive than the commercial tests	Asano et al. 2011
IBV (S)	TGEV (S1: 166–247, S1: 501–515, S2: 8–30)	Diagnosis (ELISA): more sensitive and specific than the commercial tests	Ding et al. 2015

and Zhang 2018). PEDV collagenase equivalent domain (COE), a crucial antigenic region within the viral S protein, has been widely used in the development of subunit vaccines (Ge et al. 2012; Ma et al. 2018). Sun reported a B-cell epitope, T^{592}SLLASACTIDLFGYP^{607}, within the COE that was conserved among G1 and G2 PEDV strains and may mediate the production of neutralizing antibodies (Sun et al. 2019).

Developing an FIP-preventive vaccine requires an antigen that does not induce antibody-dependent enhancement (ADE), and T helper (Th) 1 activity plays an important role in protecting against FIPV infection (Gelain et al. 2006; Kiss et al. 2004; Pedersen 2009; Weiss and Cox 1989). Satoh and Takano identified Th1 and linear immunodominant antibody-binding epitopes in the S1 domain, M protein and N protein of FIPV (Satoh et al. 2011; Takano et al. 2014). It has been shown that the T-cell epitopes N^{621}NYLTFNKFCLSLSPVGANC^{640} (II-S1-24), V^{81}YGIKMLIMWLLWPIVLALT^{100} (I-M-9), and G^{81}QRKELPERWFFYFLGTGPH^{100} (NP-7) strongly induce Th1 activity. This knowledge may guide the development of epitope vaccines against FIPV infection.

The broad cytotoxic T lymphocyte (CTL) response against IBV is a crucial factor in viral replication control (Cavanagh 2007; Collisson et al. 2000). Tan reported that four CTL epitopes of IBV (S^{413}RIQTATDP^{421}, S^{517}RNATGSQP^{525}, G^{45}AYAVVNV^{52} and S^{413}RIQTATQP^{421}) can stimulate CD8$^+$ T-cell proliferation and IFN-γ secretion (Tan et al. 2016). In vivo studies revealed that a poly-CTL epitope-based vaccine (pV-S1T) constructed by inserting the nucleotide sequences of the above four CTL epitopes into the pVAX1 vector provided 90% protection against an avirulent IBV strain.

The N-terminus of TGEV S protein contains four antigenic sites, A, B, C and D, which are involved in the stimulation of neutralizing antibodies (Delmas et al. 1990). Past studies have shown that the A site is predominantly responsible for stimulating neutralizing antibodies (Correa et al. 1990; Delmas et al. 1990; Laviada et al. 1990; Meng et al. 2013, 2011; Zhao et al. 2013). Yuan constructed a recombinant swinepox virus (rSPV-SA) expressing the TGEV S-A site (533–705) (Yuan et al. 2015). The results from passive immunity protection test of newborn piglets revealed that the recombinant live-vector vaccine rSPV-SA 100% protected piglets from SPV infection, and no significant clinical symptoms were observed in the rSPV-SA treatment group during this experiment. The antigen epitope M^{537}KSGYGQPIA^{547}, which is located in the TGEV S-A site, has been identified as a B-cell epitope of TGEV (Gebauer et al. 1991). However, whether this epitope can produce neutralizing antibodies remains unclear.

Antigen epitopes on the S1/S2 junction of animal CoVs have been shown to stimulate neutralizing antibody production. Okda found that the S1/S2 junction of PEDV is an immunodominant region of S protein with strong neutralizing activity (Okda et al. 2017; 2007). Previous studies have shown that the B-cell epitope Y^{748}SNIGVCK^{755} on the S1/S2 junction of PEDV has potential use in vaccine development

(Gillam and Zhang 2018). The T-cell epitope $L^{766}TTFEPFTVSIVNDS^{780}$ on the S1/S2 junction of MHV efficiently induces CD4 T-cell response in mice (Khanolkar et al. 2010). Vautherot found that the B-cell epitope $T^{771}TGYRFTNFEPFT^{783}$, which is on the S1/S2 junction of BCoV, is a potential immunodominant region (Vautherot et al. 1992). The above studies suggest that these antigen epitopes located on the S1/S2 junction might be used for developing epitope vaccines.

Application of antigen epitopes in diagnosis

Approaches based on antigen epitopes, which permit a high epitope density and careful choice of unique specific epitopes, have been used in the detection of antibodies against viruses and have achieved both high sensitivity and greater specificity in results (Anandarao et al. 2006; Gómara et al. 2010; He et al. 2011). Antigen epitope-based diagnosis relies on two strategies: one uses a highly conserved dominant antigen epitope, and the other combines multiple epitopes (Table 1).

The COE epitope $T^{592}SLLASACTIDLFGYP^{607}$, belonging to the B-cell epitope of PEDV, is highly conserved between G1 and G2 PEDV strains. The mAbs 4D8F10 and 6F3E3 that detect the COE epitope are suitable for PEDV by binding to the conserved epitope (Sun et al. 2019). Similarly, the TGEV B-cell epitope $T^{592}SLLASACTIDLFGYP^{607}$, which is highly conserved in coronaviruses, can be used as an antigenic peptide for diagnosis (Gebauer et al. 1991). Studies on other coronaviruses indicate that the N protein is highly conserved in different strains. Thus, it is widely used as a diagnostic antigen for the development of serologic diagnostic tools (Abdelwahab et al. 2015; Hou et al. 2007; Pradhan et al. 2014; Su et al. 2016). Asano established two indirect enzyme-linked immunosorbent assays (ELISAs) based on the B-cell epitopes $I^{24}LKKTTWADQTERGL^{38}$ and $R^{357}FDSTLPGFETIMKVL^{372}$, which are located in MHV N protein. ELISAs that rely on these peptides are more sensitive than the commercial tests used to screen laboratory mouse serum for unintended MHV infection (Asano et al. 2011). Diagnostic methods based on multiple antigens or synthetic peptides exhibit improved sensitivity and specificity (Chimeno Zoth and Taboga 2006; Hadifar et al. 2014; Shehata et al. 2012; Shenyang et al. 2011). Ding developed a multiepitope antigen of S protein (166–247 aa, S1 gene; 501–515 aa, S1 gene; 8–30 aa, S2 gene) for use in a highly sensitive and specific ELISA for detecting IBV-specific antibodies in chicken serum samples (Ding et al. 2015).

Prospect

Studies on the antigen epitopes on animal CoV have a high research output. However, they are mainly focused on IBV, FIPV, MHV, TGEV and PEDV. The relatively less harmful or newly emerged BCoV, PDCoV and SADS-CoV have not been studied extensively. Currently, technology for identification B-cell epitopes is well established. However, it is difficult to obtain high-quality monoclonal antibodies, especially those with neutralizing activity that are capable of recognizing conformational epitopes. For conformational B-cell antigen epitope identification, monoclonal antibodies recognized by whole viruses are more efficient than monoclonal antibodies from recombinant proteins. Identification of T-cell epitopes is crucial in the investigation of cellular immune mechanisms and the development of subunit peptide vaccines. Except for FIPV, MHV and IBV, few studies on T-cell antigenic epitopes of other animal CoVs have been reported. Since T cells only recognize antigenic polypeptides presented by major histocompatibility complex (MHC) molecules on the surface of antigen presenting cells (APCs), identification of T-cell epitopes is more challenging. Given that some coronaviruses, such as FIPV, have ADE characteristics, immune responses based on humoral immunity may not be suitable for preventing animal CoVs. Thus, further research on animal CoV T-cell epitopes is needed.

Relative to traditional vaccines, epitope vaccines are safer, nontoxic and stable and can more directly elicit immune responses against pathogenic microorganisms. However, in coronaviruses, applications of antigenic epitopes are poorly studied. Although several B- and T-cell epitopes that can induce antiviral immune responses have been identified, studies on the application of antigenic epitope-based diagnosis and vaccines are inadequate, with most having been done in vitro or in nonhost animals without evaluation of immune response and protection in susceptible animal hosts. The identification of different antigenic epitopes on various strains of the same virus limits the application of antigenic epitopes. Generally, antigenic epitopes are located on the surface of viral proteins with hydrophilicity and surface accessibility and are prone to certain mutations due to frequent contact with the external environment. In particular, coronavirus S proteins are located in the outermost layer of the virus and are most susceptible to mutation, resulting in poor conservation of S protein antigenic epitope. For example, Zhang identified that the residues of S protein at position 55–64 were specific for the recognition of PEDV classical G1 strains, whereas the residues at position 157–164 showed specificity to PEDV emerging G2 strains (Zhang et al. 2023). Thus, it is crucial to identify the conserved antigen epitopes, especially neutralizing epitopes, between different strains of the same coronaviruses.

Epitope vaccines based on multiepitope peptides can overcome the problem of low conservation between epitopes from different strains and trigger stronger immune responses. However, for animal CoV, the development of diagnoses and vaccines based on multiple epitopes is inadequate. Since multiepitope vaccines are based on different viral epitopes in tandem, they are suited for the development of universal vaccines against multiple animal CoVs that infect the same host, such as TGEV, PEDV, SADS-CoV and PDCoV. For example, multiple studies have shown that some people who have not been exposed to severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) have preexisting reactivity to SARS-CoV-2 sequences and that preexisting reactivity to SARS-CoV-2 is mediated by memory T cells (Braun et al. 2020; Grifoni et al. 2020; Le Bert et al. 2020). Cross-reactive T cells have been shown to specifically recognize a SARS-CoV-2 epitope as well as the homologous epitope from a common cold coronavirus (e.g., human coronavirus-OC43/229E/HL63/HKU1) (Mateus et al. 2020). Additionally, epitope vaccines based on B- and T-cell epitopes can elicit humoral and cellular immune responses, resulting in a stronger antiviral immune response, which deserves intensive investigation.

During the virus–host game, the host produces neutralizing antibodies to defend against viral invasion by recognizing viral antigenic epitopes. Meanwhile, the virus achieves immune escape by continuously mutating to reduce the neutralizing ability of host antibodies. When a virus enters a cell, the host immune system triggers the production of neutralizing antibodies that can effectively block the virus's ability to interact with susceptible cell receptors, interfere with the fusion of the virus with the cell membrane, and form immune complexes that are efficiently cleared by the immune system (Murin et al. 2019). S protein of SARS-CoV-2 mediates virus entry into cells and is the main recognition target of neutralizing antibodies. Mutations in individual amino acid sites on S protein can cause coronavirus to escape neutralizing antibodies. For example, the delta variant of SARS-CoV-2 has mutations in S protein that result in a sixfold reduction in the neutralizing ability of serum neutralizing antibodies compared to wild-type SARS-CoV-2 (Zhang et al. 2021b). Additionally, the clinically approved monoclonal antibodies bamlavinimab and imdevimab for the treatment of SARS-CoV-2-associated disease have shown a 1,000-fold and 50-fold reduction in neutralizing ability against the delta variant, respectively (Mlcochova et al. 2021). Therefore, identifying key antigenic epitope conserved regions of coronaviruses, such as the conserved receptor binding domain and the S1/S2 junction conserved region, is crucial for the development of broad-spectrum neutralizing antibodies against coronaviruses.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s44149-023-00080-0

Additional file 1: Table S1. IEDB inventory of B-cell and T-cell epitopes.

Authors' contributions

Conceptualization: Honghui Song, Mingjun Su; investigation: Mingjun Su, Guanghui Zheng, Xiangwen Xu; writing: Mingjun Su, Guanghui Zheng, Xiangwen Xu; funding acquisition: Houhui Song, Mingjun Su. All authors have read and approved the manuscript.

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Availability of data and materials

Not applicable.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The author declares that he/she has no competing interests.

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References

Abdelwahab, M., C.C. Loa, C.C. Wu, and T.L. Lin. 2015. Recombinant nucleocapsid protein-based enzyme-linked immunosorbent assay for detection of antibody to turkey coronavirus. *Journal of Virological Methods* 217: 36–41. https://doi.org/10.1016/j.jviromet.2015.02.024

Anandarao, R., S. Swaminathan, S. Fernando, A.M. Jana, and N. Khanna. 2006. Recombinant multiepitope protein for early detection of dengue infections. *Clinical and Vaccine Immunology* 13 (1): 59–67. https://doi.org/10.1128/cvi.13.1.59-67.2006

Asano, A., D. Torigoe, N. Sasaki, and T. Agui. 2011. Identification of antigenic peptides derived from B-cell epitopes of nucleocapsid protein of mouse hepatitis virus for serological diagnosis. *Journal of Virological Methods* 177 (1): 107–111. https://doi.org/10.1016/j.jviromet.2011.07.006

Barlow, D.J., M.S. Edwards, and J.M. Thornton. 1986. Continuous and discontinuous protein antigenic determinants. *Nature* 322 (6081): 747–748. https://doi.org/10.1038/322747a0

Biasini, M., S. Bienert, A. Waterhouse, K. Arnold, G. Studer, T. Schmidt, F. Kiefer, T. Gallo Cassarino, M. Bertoni, L. Bordoli, and T. Schwede. 2014. SWISS-MODEL: modeling protein tertiary and quaternary structure using evolutionary information. *Nucleic Acids Research* 42 (Web Server issue): W252-258. https://doi.org/10.1093/nar/gku340

Braun, J., L. Loyal, M. Frentsch, D. Wendisch, P. Georg, F. Kurth, S. Hippenstiel, M. Dingeldey, B. Kruse, F. Fauchere, et al. 2020. SARS-CoV-2-reactive T cells in healthy donors and patients with COVID-19. *Nature*. https://doi.org/10.1038/s41586-020-2598-9

Brian, D.A., and R.S. Baric. 2005. Coronavirus genome structure and replication. *Current Topics in Microbiology and Immunology* 287: 1–30. https://doi.org/10.1007/3-540-26765-4_1

Cavanagh, D. 2007. Coronavirus avian infectious bronchitis virus. *Veterinary Research* 38 (2): 281–297. https://doi.org/10.1051/vetres:2006055

ChimenoZoth, S., and O. Taboga. 2006. Multiple recombinant ELISA for the detection of bovine viral diarrhea virus antibodies in cattle sera. *Journal of*

Virological Methods 138 (1–2): 99–108. https://doi.org/10.1016/j.jviromet.2006.07.025
Collisson, E.W., J. Pei, J. Dzielawa, and S.H. Seo. 2000. Cytotoxic T lymphocytes are critical in the control of infectious bronchitis virus in poultry. *Developmental and Comparative Immunology* 24 (2–3): 187–200. https://doi.org/10.1016/s0145-305x(99)00072-5
Correa, I., F. Gebauer, M.J. Bullido, C. Suñé, M.F. Baay, K.A. Zwaagstra, W.P. Posthumus, J.A. Lenstra, and L. Enjuanes. 1990. Localization of antigenic sites of the E2 glycoprotein of transmissible gastroenteritis coronavirus. *The Journal of General Virology* 71 (Pt 2): 271–279. https://doi.org/10.1099/0022-1317-71-2-271
Croxford, J.L., A.M. Ercolini, M. Degutes, and S.D. Miller. 2006. Structural requirements for initiation of cross-reactivity and CNS autoimmunity with a PLP139-151 mimic peptide derived from murine hepatitis virus. *European Journal of Immunology* 36 (10): 2671–2680. https://doi.org/10.1002/eji.200635876
Delmas, B., D. Rasschaert, M. Godet, J. Gelfi, and H. Laude. 1990. Four major antigenic sites of the coronavirus transmissible gastroenteritis virus are located on the amino-terminal half of spike glycoprotein S. *The Journal of General Virology* 71 (Pt 6): 1313–1323. https://doi.org/10.1099/0022-1317-71-6-1313
Ding, M.D., H.N. Wang, H.P. Cao, W.Q. Fan, B.C. Ma, P.W. Xu, A.Y. Zhang, and X. Yang. 2015. Development of a multiepitope antigen of S protein-based ELISA for antibodies detection against infectious bronchitis virus. *Bioscience, Biotechnology, and Biochemistry* 79 (8): 1287–1295. https://doi.org/10.1080/09168451.2015.1025692
Ercolini, A.M., J. LudovicCroxford, M. Degutes, and S.D. Miller. 2007. Cross-reactivity between peptide mimics of the immunodominant myelin proteolipid protein epitope PLP139–151: comparison of peptide priming in CFA vs. viral delivery. *Journal of neuroimmunology* 186 (1–2): 5–18. https://doi.org/10.1016/j.jneuroim.2007.02.002
Ge, J.W., D.Q. Liu, and Y.J. Li. 2012. Construction of recombinant lactobacilli expressing the core neutralizing epitope (COE) of porcine epidemic diarrhea virus and a fusion protein consisting of COE and Escherichia coli heat-labile enterotoxin B, and comparison of the immune responses by orogastric immunization. *Canadian Journal of Microbiology* 58 (11): 1258–1267. https://doi.org/10.1139/w2012-098
Gebauer, F., W.P. Posthumus, I. Correa, C. Suñé, C. Smerdou, C.M. Sánchez, J.A. Lenstra, R.H. Meloen, and L. Enjuanes. 1991. Residues involved in the antigenic sites of transmissible gastroenteritis coronavirus S glycoprotein. *Virology* 183 (1): 225–238. https://doi.org/10.1016/0042-6822(91)90135-x
Gelain, M.E., M. Meli, and S. Paltrinieri. 2006. Whole blood cytokine profiles in cats infected by feline coronavirus and healthy non-FCoV infected specific pathogen-free cats. *Journal of Feline Medicine and Surgery* 8 (6): 389–399. https://doi.org/10.1016/j.jfms.2006.05.002
Gillam, F., and C. Zhang. 2018. Epitope selection and their placement for increased virus neutralization in a novel vaccination strategy for porcine epidemic diarrhea virus utilizing the Hepatitis B virus core antigen. *Vaccine* 36 (30): 4507–4516. https://doi.org/10.1016/j.vaccine.2018.06.015
Gillam, F., J. Zhang, and C. Zhang. 2018. Hepatitis B core antigen based novel vaccine against porcine epidemic diarrhea virus. *Journal of Virological Methods* 253: 61–69. https://doi.org/10.1016/j.jviromet.2017.11.003
Gómara, M.J., L. Fernández, T. Pérez, G. Ercilla, and I. Haro. 2010. Assessment of synthetic chimeric multiple antigenic peptides for diagnosis of GB virus C infection. *Analytical Biochemistry* 396 (1): 51–58. https://doi.org/10.1016/j.ab.2009.09.011
Grifoni, A., D. Weiskopf, S.I. Ramirez, J. Mateus, J.M. Dan, C.R. Moderbacher, S.A. Rawlings, A. Sutherland, L. Premkumar, R.S. Jadi, et al. 2020. Targets of T-Cell responses to SARS-CoV-2 Coronavirus in Humans with COVID-19 disease and unexposed individuals. *Cell* 181 (7): 1489–1501.e1415. https://doi.org/10.1016/j.cell.2020.05.015
Hadifar, F., J. Ignjatovic, S. Tarigan, R. Indriani, E. Ebrahimie, N.H. Hasan, A. Mcwhorter, S. Putland, A. Ownagh, and F. Hemmatzadeh. 2014. Multimeric recombinant M2e protein-based ELISA: a significant improvement in differentiating avian influenza infected chickens from vaccinated ones. *PloS one* 9 (10): e108420. https://doi.org/10.1371/journal.pone.0108420
Hain, K.S., L.R. Joshi, F. Okda, J. Nelson, A. Singray, S. Lawson, M. Martins, A. Pillatzki, G.F. Kutish, E.A. Nelson, E.F. Flores, and D.G. Diel. 2016. Immunogenicity of a recombinant parapoxvirus expressing the spike protein of Porcine epidemic diarrhea virus. *The Journal of General Virology* 97 (10): 2719–2731. https://doi.org/10.1099/jgv.0.000586
He, J., B. Xiu, G. Wang, K. Chen, X. Feng, X. Song, C. Zhu, S. Ling, and H. Zhang. 2011. Double-antigen sandwich ELISA for the detection of anti-hepatitis C virus antibodies. *Journal of Virological Methods* 171 (1): 163–168. https://doi.org/10.1016/j.jviromet.2010.10.019
Hoffmann, M., H. Kleine-Weber, and S. Pöhlmann. 2020. A multibasic cleavage site in the spike protein of SARS-CoV-2 Is essential for infection of human lung cells. *Molecular Cell* 78 (4): 779-784.e775. https://doi.org/10.1016/j.molcel.2020.04.022
Hou, X.L., L.Y. Yu, and J. Liu. 2007. Development and evaluation of enzyme-linked immunosorbent assay based on recombinant nucleocapsid protein for detection of porcine epidemic diarrhea (PEDV) antibodies. *Veterinary Microbiology* 123 (1–3): 86–92. https://doi.org/10.1016/j.vetmic.2007.02.014
Hudson, C.B., and F.R. Beaudette. 1932. Infection of the cloaca with the virus of infectious bronchitis. *Science (New York, N.Y.)* 76 (1958): 34. https://doi.org/10.1126/science.76.1958.34-a
Hulswit, R.J., C.A. De Haan, and B.J. Bosch. 2016. Coronavirus spike protein and tropism changes. *Advances in Virus Research* 96: 29–57. https://doi.org/10.1016/bs.aivir.2016.08.004
Khanolkar, A., R.B. Fulton, L.L. Epping, N.L. Pham, D. Tifrea, S.M. Varga, and J.T. Harty. 2010. T-cell epitope specificity and pathogenesis of mouse hepatitis virus-1-induced disease in susceptible and resistant hosts. *Journal of immunology (Baltimore, Md. :1950)* 185 (2): 1132–1141. https://doi.org/10.4049/jimmunol.0902749
Kiss, I., A.M. Poland, and N.C. Pedersen. 2004. Disease outcome and cytokine responses in cats immunized with an avirulent feline infectious peritonitis virus (FIPV)-UCD1 and challenge-exposed with virulent FIPV-UCD8. *Journal of Feline Medicine and Surgery* 6 (2): 89–97. https://doi.org/10.1016/j.jfms.2003.08.009
Kong, N., Q. Meng, Y. Jiao, Y. Wu, Y. Zuo, H. Wang, D. Sun, S. Dong, H. Zhai, W. Tong, H. Zheng, H. Yu, G. Tong, Y. Xu, and T. Shan. 2020. Identification of a novel B-cell epitope in the spike protein of porcine epidemic diarrhea virus. *Virology Journal* 17 (1): 46. https://doi.org/10.1186/s12985-020-01305-1
Laviada, M.D., S.P. Videgain, L. Moreno, F. Alonso, L. Enjuanes, and J.M. Escribano. 1990. Expression of swine transmissible gastroenteritis virus envelope antigens on the surface of infected cells: Epitopes externally exposed. *Virus Research* 16 (3): 247–254. https://doi.org/10.1016/0168-1702(90)90051-c
Le Bert, N., A.T. Tan, K. Kunasegaran, C.Y.L. Tham, M. Hafezi, A. Chia, M.H.Y. Chng, M. Lin, N. Tan, M. Linster, et al. 2020. SARS-CoV-2-specific T-cell immunity in cases of COVID-19 and SARS, and uninfected controls. *Nature* 584 (7821): 457–462. https://doi.org/10.1038/s41586-020-2550-z
Li, F. 2016. Structure, function, and evolution of coronavirus spike proteins. *Annual Review of Virology* 3 (1): 237–261. https://doi.org/10.1146/annurev-virology-110615-042301
Ma, S., L. Wang, X. Huang, X. Wang, S. Chen, W. Shi, X. Qiao, Y. Jiang, L. Tang, Y. Xu, and Y. Li. 2018. Oral recombinant *Lactobacillus* vaccine targeting the intestinal microfold cells and dendritic cells for delivering the core neutralizing epitope of porcine epidemic diarrhea virus. *Microbial Cell Factories* 17 (1): 20. https://doi.org/10.1186/s12934-018-0861-7
Makadiya, N., R. Brownlie, J. Van Den Hurk, N. Berube, B. Allan, V. Gerdts, and A. Zakhartchouk. 2016. S1 domain of the porcine epidemic diarrhea virus spike protein as a vaccine antigen. *Virology Journal* 13: 57. https://doi.org/10.1186/s12985-016-0512-8
Mateus, J., A. Grifoni, A. Tarke, J. Sidney, S.I. Ramirez, J.M. Dan, Z.C. Burger, S.A. Rawlings, D.M. Smith, E. Phillips, et al. 2020. Selective and cross-reactive SARS-CoV-2 T-cell epitopes in unexposed humans. *Science (New York, N.Y.)* 370 (6512): 89–94. https://doi.org/10.1126/science.abd3871
Mathieu, P.A., K.A. Gómez, J.P. Coutelier, and L.A. Retegui. 2004. Sequence similarity and structural homologies are involved in the autoimmune response elicited by mouse hepatitis virus A59. *Journal of Autoimmunity* 23 (2): 117–126. https://doi.org/10.1016/j.jaut.2004.05.006
Meng, F., Z. Zhao, G. Li, S. Suo, N. Shi, J. Yin, D. Zarlenga, and X. Ren. 2011. Bacterial expression of antigenic sites A and D in the spike protein of transmissible gastroenteritis virus and evaluation of their inhibitory effects on viral infection. *Virus Genes* 43 (3): 335–341. https://doi.org/10.1007/s11262-011-0637-1
Meng, F., Y. Ren, S. Suo, X. Sun, X. Li, P. Li, W. Yang, G. Li, L. Li, C. Schwegmann-Wessels, G. Herrler, and X. Ren. 2013. Evaluation on the efficacy and immunogenicity of recombinant DNA plasmids expressing spike genes from porcine transmissible gastroenteritis virus and porcine epidemic diarrhea virus. *PloS one* 8 (3): e57468. https://doi.org/10.1371/journal.pone.0057468

Millet, J.K., and G.R. Whittaker. 2018. Physiological and molecular triggers for SARS-CoV membrane fusion and entry into host cells. *Virology* 517: 3–8. https://doi.org/10.1016/j.virol.2017.12.015

Mlcochova, P., S.A. Kemp, M.S. Dhar, G. Papa, B. Meng, I. Ferreira, R. Datir, D.A. Collier, A. Albecka, S. Singh, et al. 2021. SARS-CoV-2 B.1.617.2 Delta variant replication and immune evasion. *Nature* 599 (7883): 114–119. https://doi.org/10.1038/s41586-021-03944-y

Murin, C.D., I.A. Wilson, and A.B. Ward. 2019. Antibody responses to viral infections: a structural perspective across three different enveloped viruses. *Nature Microbiology* 4 (5): 734–747. https://doi.org/10.1038/s41564-019-0392-y

Nogales, A., C. Galán, S. Márquez-Jurado, M. García-Gallo, L. Kremer, L. Enjuanes, and F. Almazán. 2011. Immunogenic characterization and epitope mapping of transmissible gastroenteritis virus RNA dependent RNA polymerase. *Journal of Virological Methods* 175 (1): 7–13. https://doi.org/10.1016/j.jviromet.2011.04.007

Oh, J., K.W. Lee, H.W. Choi, and C. Lee. 2014. Immunogenicity and protective efficacy of recombinant S1 domain of the porcine epidemic diarrhea virus spike protein. *Archives of Virology* 159 (11): 2977–2987. https://doi.org/10.1007/s00705-014-2163-7

Okda, F.A., S. Lawson, A. Singray, J. Nelson, K.S. Hain, L.R. Joshi, J. Christopher-Hennings, E.A. Nelson, and D.G. Diel. 2017. The S2 glycoprotein subunit of porcine epidemic diarrhea virus contains immunodominant neutralizing epitopes. *Virology* 509: 185–194. https://doi.org/10.1016/j.virol.2017.06.013

Pedersen, N.C. 2009. A review of feline infectious peritonitis virus infection: 1963–2008. *Journal of Feline Medicine and Surgery* 11 (4): 225–258. https://doi.org/10.1016/j.jfms.2008.09.008

Pradhan, S.K., N.M. Kamble, A.S. Pillai, S.S. Gaikwad, S.A. Khulape, M.R. Reddy, C.M. Mohan, J.M. Kataria, and S. Dey. 2014. Recombinant nucleocapsid protein based single serum dilution ELISA for the detection of antibodies to infectious bronchitis virus in poultry. *Journal of Virological Methods* 209: 1–6. https://doi.org/10.1016/j.jviromet.2014.08.015

Qin, Y., T. Feng, H. Shi, J. Zhang, L. Zhang, S. Feng, J. Chen, Y. He, X. Zhang, Z. Chen, J. Liu, D. Liu, D. Shi, and L. Feng. 2022. Identification and epitope mapping of swine acute diarrhea syndrome coronavirus accessory protein NS7a via monoclonal antibodies. *Virus research* 313: 198742. https://doi.org/10.1016/j.virusres.2022.198742

Satoh, R., T. Furukawa, M. Kotake, T. Takano, K. Motokawa, T. Gemma, R. Watanabe, S. Arai, and T. Hohdatsu. 2011. Screening and identification of T helper 1 and linear immunodominant antibody-binding epitopes in the spike 2 domain and the nucleocapsid protein of feline infectious peritonitis virus. *Vaccine* 29 (9): 1791–1800. https://doi.org/10.1016/j.vaccine.2010.12.106

Shehata, A.A., P. Fiebig, H. Sultan, M. Hafez, and U.G. Liebert. 2012. Development of a recombinant ELISA using yeast (Pichia pastoris)-expressed polypeptides for detection of antibodies against avian influenza A subtype H5. *Journal of Virological Methods* 180 (1–2): 18–25. https://doi.org/10.1016/j.jviromet.2011.12.004

Shenyang, G., L. Dandan, F. Chen, W. Shuliang, and Z. Tiezhong. 2011. Design and evaluation of a recombinant multiepitope-based ELISA for the serological surveillance of HEV infection in northern China. *Archives of Virology* 156 (9): 1621–1626. https://doi.org/10.1007/s00705-011-1007-y

Su, M., C. Li, D. Guo, S. Wei, X. Wang, Y. Geng, S. Yao, J. Gao, E. Wang, X. Zhao, Z. Wang, J. Wang, R. Wu, L. Feng, and D. Sun. 2016. A recombinant nucleocapsid protein-based indirect enzyme-linked immunosorbent assay to detect antibodies against porcine deltacoronavirus. *The Journal of Veterinary Medical Science* 78 (4): 601–606. https://doi.org/10.1292/jvms.15-0533

Subramaniam, S., D. Cao, D. Tian, Q.M. Cao, C. Overend, D.M. Yugo, S.R. Matzinger, A.J. Rogers, C.L. Heffron, N. Catanzaro, et al. 2017. Efficient priming of CD4 T cells by Langerin-expressing dendritic cells targeted with porcine epidemic diarrhea virus spike protein domains in pigs. *Virus Research* 227: 212–219. https://doi.org/10.1016/j.virusres.2016.10.007

Sun, D.B., L. Feng, H.Y. Shi, J.F. Chen, S.W. Liu, H.Y. Chen, and Y.F. Wang. 2007. Spike protein region (aa 636789) of porcine epidemic diarrhea virus is essential for induction of neutralizing antibodies. *Acta Virologica* 51 (3): 149–156.

Sun, D., L. Feng, H. Shi, J. Chen, X. Cui, H. Chen, S. Liu, Y. Tong, Y. Wang, and G. Tong. 2008. Identification of two novel B-cell epitopes on porcine epidemic diarrhea virus spike protein. *Veterinary Microbiology* 131 (1–2): 73–81. https://doi.org/10.1016/j.vetmic.2008.02.022

Sun, Y.G., R. Li, S. Xie, S. Qiao, Q. Li, X.X. Chen, R. Deng, and G. Zhang. 2019. Identification of a novel linear B-cell epitope within the collagenase equivalent domain of porcine epidemic diarrhea virus spike glycoprotein. *Virus Research* 266: 34–42. https://doi.org/10.1016/j.virusres.2019.04.003

Takano, T., H. Morioka, K. Gomi, K. Tomizawa, T. Doki, and T. Hohdatsu. 2014. Screening and identification of T helper 1 and linear immunodominant antibody-binding epitopes in spike 1 domain and membrane protein of feline infectious peritonitis virus. *Vaccine* 32 (16): 1834–1840. https://doi.org/10.1016/j.vaccine.2014.01.074

Tan, L., Y. Liao, J. Fan, Y. Zhang, X. Mao, Y. Sun, C. Song, X. Qiu, C. Meng, and C. Ding. 2016. Prediction and identification of novel IBV S1 protein derived CTL epitopes in chicken. *Vaccine* 34 (3): 380–386. https://doi.org/10.1016/j.vaccine.2015.11.042

Vautherot, J.F., J. Laporte, and P. Boireau. 1992. Bovine coronavirus spike glycoprotein: Localization of an immunodominant region at the amino-terminal end of S2. *The Journal of General Virology* 73 (Pt 12): 3289–3294. https://doi.org/10.1099/0022-1317-73-12-3289

Vita, R., L. Zarebski, J.A. Greenbaum, H. Emami, I. Hoof, N. Salimi, R. Damle, A. Sette, and B. Peters. 2010. The immune epitope database 2.0. *Nucleic Acids Research* 38 (Database issue): D854-862. https://doi.org/10.1093/nar/gkp1004

Wang, K., C. Xie, J. Zhang, W. Zhang, D. Yang, L. Yu, Y. Jiang, S. Yang, F. Gao, Z. Yang, Y. Zhou, and G. Tong. 2016. The Identification and characterization of two novel epitopes on the nucleocapsid protein of the porcine epidemic diarrhea virus. *Scientific Reports* 6: 39010. https://doi.org/10.1038/srep39010

Wang, Q., A.N. Vlasova, S.P. Kenney, and L.J. Saif. 2019. Emerging and re-emerging coronaviruses in pigs. *Current Opinion in Virology* 34: 39–49. https://doi.org/10.1016/j.coviro.2018.12.001

Weiss, R.C., and N.R. Cox. 1989. Evaluation of immunity to feline infectious peritonitis in cats with cutaneous viral-induced delayed hypersensitivity. *Veterinary Immunology and Immunopathology* 21 (3–4): 293–309. https://doi.org/10.1016/0165-2427(89)90038-x

Woo, P.C., S.K. Lau, Y. Huang, and K.Y. Yuen. 2009. Coronavirus diversity, phylogeny and interspecies jumping. *Experimental biology and medicine (Maywood, N.J.)* 234 (10): 1117–1127. https://doi.org/10.3181/0903-mr-94

Yuan, X., H. Lin, and H. Fan. 2015. Efficacy and immunogenicity of recombinant swinepox virus expressing the A epitope of the TGEV S protein. *Vaccine* 33 (32): 3900–3906. https://doi.org/10.1016/j.vaccine.2015.06.057

Zhang, G., B. Li, D. Yoo, T. Qin, X. Zhang, Y. Jia, and S. Cui. 2021a. Animal coronaviruses and SARS-CoV-2. *Transboundary and Emerging Diseases* 68 (3): 1097–1110. https://doi.org/10.1111/tbed.13791

Zhang, H., S. Deng, L. Ren, P. Zheng, X. Hu, T. Jin, and X. Tan. 2021. Profiling CD8(+) T cell epitopes of COVID-19 convalescents reveals reduced cellular immune responses to SARS-CoV-2 variants. *Cell reports* 36 (11): 109708. https://doi.org/10.1016/j.celrep.2021.109708

Zhang, H., Zou, C., Peng, O., Ashraf, U., Xu, Q., Gong, L., Fan, B., Zhang, Y., Xu, Z., Xue, C., Wei, X., Zhou, Q., Tian, X., Shen, H., Li, B., Zhang, X., Cao, Y., 2023. Global dynamics of porcine enteric coronavirus PEDV epidemiology, evolution, and transmission. *Molecular Biology and Evolution* 40 (3). https://doi.org/10.1093/molbev/msad052

Zhao, Q., J. Zhu, W. Zhu, X. Li, Y. Tao, X. Lv, X. Wang, J. Yin, C. He, and X. Ren. 2013. A monoclonal antibody against transmissible gastroenteritis virus generated via immunization of a DNA plasmid bearing TGEV S1 gene. *Monoclonal Antibodies in Immunodiagnosis and Immunotherapy* 32 (1): 50–54.https://doi.org/10.1089/mab.2012.0067