Journal of the Formosan Medical Association (2014) 113, 143–147

Available online at www.sciencedirect.com

ScienceDirect

journal homepage: www.jfma-online.com

REVIEW ARTICLE

# The receptor binding domain of MERS-CoV: The dawn of vaccine and treatment development

Nan Zhou [a], Yun Zhang [a], Jin-Chun Zhang, Ling Feng, Jin-Ku Bao*

*School of Life Sciences & Key Laboratory of Bio-resources, Ministry of Education, Sichuan University, Chengdu 610064, China*

Received 15 September 2013; received in revised form 20 October 2013; accepted 13 November 2013

**KEYWORDS**
coronavirus;
drug design;
Middle East;
vaccines

The newly emerged Middle East respiratory syndrome coronavirus (MERS-CoV) is becoming another "SARS-like" threat to the world. It has an extremely high death rate (~50%) as there is no vaccine or efficient therapeutics. The identification of the structures of both the MERS-CoV receptor binding domain (RBD) and its complex with dipeptidyl peptidase 4 (DPP4), raises the hope of alleviating this currently severe situation. In this review, we examined the molecular basis of the RBD-receptor interaction to outline why/how could we use MERS-CoV RBD to develop vaccines and antiviral drugs.


## Introduction

On 20 September 2012, a novel coronavirus isolated from a 60-year-old Saudi man with acute pneumonia and acute renal failure was reported.[1] In May 2013, the WHO adopted the virus name Middle East respiratory syndrome (MERS) coronavirus (MERS-CoV), which was defined by the Coronavirus Study Group of the International Committee on Taxonomy of Viruses.[2] As of 7 September 2013, the WHO has been notified of 114 laboratory-confirmed cases in the Middle East [Jordan, Saudi Arabia (KSA), the United Arab Emirates (UAE), and Qatar], Europe [France, Germany, United Kingdom (UK) and Italy], and North Africa (Tunisia), with 54 deaths.[3]

The world is facing a new challenge posed by a severe acute respiratory syndrome (SARS)-like infections in humans caused by MERS-CoV. Its main clinical manifestations in patients are pneumonia (or respiratory) failure and acute renal failure.[4] The short-lived but alarming epidemic SARS-CoV killed nearly 10% of approximately 8000 cases in the 2002–2003 outbreak.[5] The data so far indicate that

Conflicts of interest: The authors have no conflicts of interest relevant to this article.

* Corresponding author. No. 29, Wangjiang Road, School of Life Sciences & Key Laboratory of Bio-resources, Ministry of Education, Sichuan University, Chengdu 610064, China.
*E-mail address:* baojinku@scu.edu.cn (J.-K. Bao).
[a] These authors contributed equally to this work.

0929-6646/$ - see front matter 
http://dx.doi.org/10.1016/j.jfma.2013.11.006

MERS-CoV possesses an unusually high crude mortality rate of approximately 50%, implying a big threat to people who are infected. Clusters of cases including a UK family, and hospitals in KSA, France and UAE show epidemiological evidence of human-to-human transmission. Indeed, existing reports indicate person-to-person transmission occurs, raising significant concern on the possible emergence of a global epidemic in the near future.[6–8] It is therefore of utmost importance to pay worldwide attention to find antiviral drugs and effective vaccines to control its high death rate and spread.

Coronaviruses are a large family of enveloped, single-stranded RNA viruses that infect a number of different host species, including humans. In people, coronaviruses can cause illnesses ranging in severity from the common cold to SARS. Coronaviruses can be categorized into four genera, *Alphacoronavirus*, *Betacoronavirus*, *Gammacoronavirus*, and *Deltacoronavirus*.[9,10] The genus *Betacoronavirus* has four lineages: A, B, C, and D.[11] MERS-CoV belongs to lineage C and is the first lineage C *Betacoronavirus* known to infect humans.[12]

Coronaviruses infect animals and humans. The spike (S) entry protein binds to a cell surface receptor which primarily determines their tropism. Similar to other coronaviruses, MERS-CoV utilizes its large surface S protein to interact with and enter the target cell.[13,14] Raj et al[13] have identified that dipeptidyl peptidase 4 (DPP4; also known as CD26) is its functional receptor. MERS-CoV binds to DPP4 via the S protein and releases the RNA genome into the target cell.[2] The MERS-CoV S protein is a type-I membrane glycoprotein and contains S1 and S2 subunits.[15] The S1 domain determines cellular tropism and interacts with the target cell, while the S2 domain mediates membrane fusion.[13,16] According to recent studies, the MERS-CoV receptor binding domain (RBD) is mapped to the S1 region.[17,18] MERS-CoV RBD binds to the receptor and induces significant neutralizing antibody responses, indicating that it can be used as a candidate target for the development of vaccines and antiviral drugs.[19] Herein, we review recent studies on MERS-CoV to make suggestions on antiviral vaccine and drug development.

## Structure of MERS-CoV RBD

Lu et al[15] and other research teams have identified the crystal structure of MERS-CoV RBD after the discovery of its receptor.[17,20] Structural topology considers a sequence of secondary structure elements making protein structure easy to interpret by laying out the 3D structural information in two dimensions in a manner that makes the structure clear. Therefore we used Pro-origami to automatically generate the schematic representation of the MERS-CoV RBD topology.[21] The MERS-CoV RBD has a core and an external subdomain (Fig. 1A and B). The core subdomain is a five-stranded antiparallel β sheet (β1, β3, β4, β5, and β10) decorated by the connecting helices (α1–4 and η1, 2) and two small β-strands (β2 and β11) (Fig. 1B). Three disulfide bonds stabilize the fold by connecting C383 to C407, C425 to C478, and C437 to C585 (Fig. 1B). The external receptor binding subdomain reveals a four-stranded antiparallel β sheet with three large strands (β6, β8, and β9) and one small strand (β7) between strands β5 and β10 of the core domain (Fig. 1B). The β5/6, β7/8 and β9/10 intervening loops touch the core subdomain and anchor the external to the core (Fig. 1B). There is a long loop containing η3 crosses perpendicular to the β sheet connecting β7 and β8 strands, and a disulfide bond between C503 and C526 links the η3 helix to strand β6 (Fig. 1B).

## Mechanism of MERS-CoV binding to DPP4

Multifunctional DPP4 plays a major role in glucose metabolism, T-cell activation, chemotaxis modulation, cell adhesion, and apoptosis.[22,23] In humans, it is primarily expressed on the epithelial cells in the lungs, liver, small intestine, kidney, and prostate, and on activated leukocytes, while it also occurs in a soluble form in the circulation.[23,24] The structure of DPP4, as shown in previous studies, is composed of an N-terminal eight-bladed β-propeller domain (S39 to D496) and a C-terminal α/β-hydrolase domain (N497 to P766).[25,26] The β-propeller domain contains eight blades, each made up of four antiparallel β-strands. The receptor-binding subdomain of MERS-CoV RBD binds to the DPP4 β-propeller, contacting blades four and five and a small bulged helix in the blade-linker.[15,20] Structural analysis and mutational analysis by Lu et al[15] and Wang et al[20] have identified Y499, L506, W533, and E513 in the RBD to be critical for receptor binding and viral entry, and mutations of these significantly abrogate its interaction with DPP4.

## MERS-CoV RBD-based vaccine design

One reason for the exceptionally high crude mortality rate of nearly 50% in MERS-CoV infection is the lack of vaccines. Therefore, it is necessary to develop efficient and safe vaccines to control MERS quickly. MERS-CoV infects a wide variety of host species whereas coronaviruses generally tend to have a narrow host tropism.[1] DPP4 sequence alignment demonstrates that its orthologs are highly conserved for MERS-CoV acquiring cross-species transmissibility by binding to an evolutionarily conserved receptor.[27] Minor mutations within the RBD domain can disturb the lock-and-key interaction of the RBD-receptor binding interface and then places a barrier for cross-species transmission.

The roles of MERS-CoV RBD in receptor binding indicate that vaccines based on it could induce antibodies to block virus binding or infection. Among structural proteins of MERS-CoV, S protein is known to be the main antigenic component to induce significant neutralizing antibody responses up to now.[19] Du et al[19] found that MERS-CoV RBD binds to the receptor and induces significant neutralizing antibody responses. Mou et al[18] revealed that MERS-CoV RBD can efficiently elicit neutralizing antibodies. Besides, previous studies have shown that the related RBD of SARS-CoV strongly reacts with antisera from patients with SARS and the depletion of the RBD-specific antibodies results in significant elimination of the neutralizing activity.[28] Lu et al[2] also proposed that SARS vaccines based on SARS-CoV RBD are more effective and safer than vaccine candidates based on the inactivated virus, DNA or viral vectors. In