Is the ChAdOx1 Vaccine Safe and Immunogenic as Prophylactic Measure Against the Lethal Human-Coronaviruses? A Systematic Review

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Abstract- Knowledge of other Coronaviruses has contributed to the development of a vaccine for the Severe Acute Respiratory Syndrome-Coronavirus-2 (SARS-CoV-2). As soon as the genetic sequence of SARS-CoV-2 was released, intense global activity around different vaccine platform technologies started. Among these platforms, the viral vectored chimpanzee adenovirus Oxford1 (ChAdOx1)-previously studied for various indications, including for the Middle East Respiratory Syndrome Coronavirus (MERS-CoV) vaccine, and currently is adapted for the ChAdOx1 nCoV-19 (novel Coronavirus-19). Therefore, this systematic review aimed to investigate the potential of the ChAdOx1 platform for the development of a vaccine for SARS-CoV and MERS-CoV, the Lethal Human-Coronaviruses (Lh-CoVs). For this purpose, a highly sensitive literary search was conducted through electronic databases that reached 1,445 related articles, of which, eight articles were elected according to previous eligibility criteria. The gathering of the articles demonstrated that the previous approaches, referring to the ChAdox1 platform, have contributed to the development of vaccines against Lh-CoVs and, that thus far, ChAdOx1 (nCoV-19 and MERS) vaccines are safe and immunogenic. However, it is important to emphasize that further studies are needed to ensure the effectiveness of vaccines in humans.

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Introduction

A vaccine against many strains of Coronavirus (CoVs), is the effective alternative to contain the emerging pandemics (1). Coronavirus is a large family of viruses that cause illnesses ranging from the common cold to severe disease (2). Thus far, among the seven zoonotic CoVs identified, four are related to minimal symptoms, while three-Severe Acute Respiratory Syndrome-Coronavirus (SARS-CoV), Middle East Respiratory Syndrome-Coronavirus (MERS-CoV), and Severe Acute Respiratory Syndrome-Coronavirus-2 (SARS-CoV-2)-have been leading with about 9.6%, 35.5%, and 6.76% mortality rates, respectively (2-3).

Several resemblances in the genome and in the pathogenesis of the Lethal Human-Coronaviruses (Lh-CoVs) have been identified. To demonstrate, the SARS-CoV-2 genome has 79% similarity to SARS-CoV and 50% to MERS-CoV (4). Moreover, regardless of whether coronaviruses are lethal or not, these viruses are enveloped, single-stranded Ribonucleic Acid (RNA), with a distinctive spike (S) protein responsible for mediating the attachment of the virus to the host cell receptor.

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SARS-CoV S-protein attaches primarily to the human Angiotensin Converting Enzyme-2 (ACE-2), whereas MERS-CoV S-protein attaches to human Dipeptidyl Peptidase-4 (DPP4) (4-6). Therefore, the similarities among the coronaviruses and the role of the S-protein, have been target factors for the development of vaccines (7).

Current approaches to vaccines against Lh-CoVs include Deoxyribonucleic Acid (DNA) vaccine; subunit vaccine; virus-like particles (VLPs) vaccine; Inactivated Whole-Virus (IWV) vaccine; live attenuated vaccine; and viral vector vaccines, including from replication-deficient chimpanzee adenovirus (ChAd) (8). Chad platform boasts a very good safety and immunogenicity profile in humans as demonstrated in clinical trials against a wide range of indications such as malaria, Human Immunodeficiency Virus (HIV), tuberculosis, influenza, hepatitis C, Ebola, and recently in trials with the ChAdOx1 vaccine candidates against MERS and SARS-CoV-2 (9-11).

The ChAdOx1 platform emerged from the vector based on a Chad Y25, which has undergone genetic modifications made through a bacterial artificial chromosome system, resulting in a molecular clone of the virus, ChAdY25-E, named ChAdOX1, and currently renamed AZD1222 (11-12). The ChAdOx1 vaccines against MERS and SARS-CoV-2 are ChAdOx1 MERS, which contain MERS-CoV S-protein, and ChAdOx1 nCoV-19, which contain SARS-CoV S-protein (11-13). The Chad virus cannot be reproduced in the human body, but the genetic coronavirus material, that is inserted in the ChAdOx1 vaccine transmits instructions for the production of coronavirus S-protein by the adenovirus (14-15). As a result, the human Immune System (IS) induces the formation of antibodies (Abs) against the Coronavirus S-protein, protecting the individual from a possible future infection (16).

Thus far, pre-clinical and clinical trials with the ChAdOx1 platform against Lh-CoVs, have demonstrated rapid induction of both humoral and cellular Immune Responses (IR), by increasing rate after a second dose, in addition to an acceptable safety profile (11). These results, according to Doremalen *et al*., (1), render the ChAdOx1 platform ideal for the development of vaccines against emerging CoVs, such as SARS-CoV-2 (1).

Therefore, due to the threat of coronaviruses to public health, relevant studies regarding vaccines and Lh-CoVs can be important tools for the medical-scientific community. Then, the present systematic review aimed to investigate the benefits of the ChAdOx1 platform as a prophylactic measure for Lh-CoVs, mainly regarding SARS-CoV-2.

Materials and Methods

Guidelines

We conduct this systematic review according to the PRISMA-P (Preferred Reporting Items for Systematic Reviews and Meta-Analyses-Protocols) statement.

Criteria

The eligible articles for the research were those related to pre-clinical and clinical trials from ChAdOx1 as a vaccine for Lh-CoVs, without restriction about the date of publication or language.

The ineligible studies were review articles, editorials, letters, presentations, case reports, small communications, preprints, comments, book chapters, and original studies with other vaccine platforms. However, original articles about the ChAdOx1 and other approaches were selected to be used in the introduction and discussion, aiming to add value to the review.

Literary search

We performed a highly sensitive literary search to answer the leading question: Is the ChAdOx1 safe and immunogenic to be administered as a prophylactic measure for the lethal human Coronaviruses?

We searched the electronic databases from PubMed Central, CAPES periodicals, and ResearchGate, from December 2019 up to September 30, 2020, with the terms: ChAdOx1 and coronavirus; ChAdOx1 and MERS; ChAdOx1 and SARS; ChAdOx1 and SARS-CoV-2. Then, we tabulated the articles and removed the duplicates; next, we analyzed the titles and the abstracts; and finally, we thoroughly analyzed the remained articles. The strategy is demonstrated in the PRISMA flow diagram (Figure 1).

Figure 1. PRISMA flow diagram for the current Systematic Review process, indicating the study selection and analyzed results.

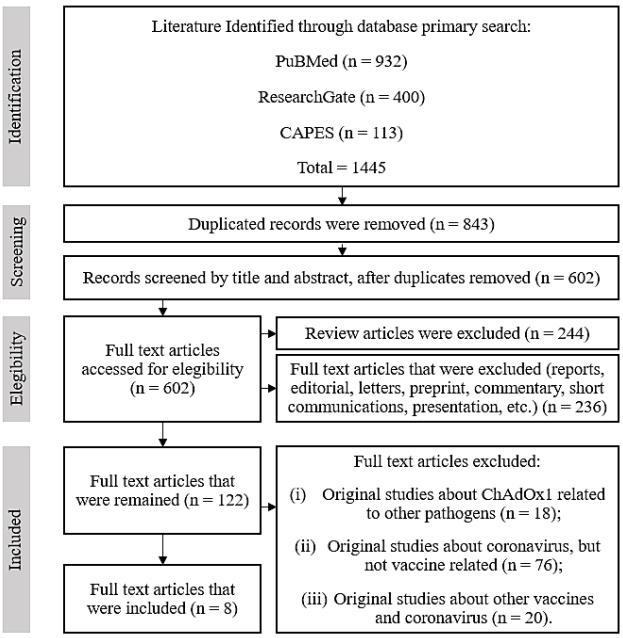

Figure 1. Prisma flow chart for the systematic review

To facilitate the data collection and to determine the inclusion or exclusion of the studies with greater reliability, the clinical question was structured according to the acronym PICOT. P-participant: animals or human; I-intervention: ChAdOx1 vaccine (regardless of dosage or regimen); C-Control: placebo, another vaccine, or ChAdOx1 in different applications; O-Outcome: adverse events and clinical aspects after vaccination, cellular and humoral responses (regardless of the viral challenge or animal sacrifice); T-study design (17).

To appraise the methodological and evidence quality of the studies, we access the Critical Appraisal Skills Program (18), focusing on the following issues: the aim of the research; methodology; assertiveness of the results; rigor in data analysis; confounding factors, and research value (Table 1). Beyond that, to minimize the risk of bias, any ambiguity during the study was resolved by authors' mutual discussion and consensus.

Table 1. Methodological quality assessment and evidence quality assessment from the articles.

Article	Was there a clear statement of the aims of the research?	Was the methodology appropriate for addressing the research goal?	Was the data analysis sufficiently rigorous?	Was there a clear statement of findings?	Which confusing variables did the authors elucidate?	Did the research add value to the medical/scientific community?
N. van Doremalen *et al*., 2020b	Yes	Yes	Yes	*Partially	Case-control	Yes
Simon P. Graham *et al*., 2020	Yes	Yes	Yes	Yes	Case-control	Yes
N. van Doremalen *et al*., 2020a	Yes	Yes	Yes	*Partially	Case-control	Yes
Naif Khalaf Alharbi *et al*., 2019	Yes	Yes	Yes	Yes	Case-control	Yes
Naif Khalaf Alharbi *et al*., 2017	Yes	Yes	Yes	Yes	Case-control	Yes
Vincent J Munster *et al*., 2017	Yes	Yes	Yes	Yes	Case-control	Yes
Pedro M. Folegatti *et al*., 2020a	Yes	Yes	Yes	Yes	Case-control	Yes
Pedro M. Folegatti *et al*., 2020b	Yes	Yes	Yes	Yes	Case-control	Yes

Table 1. * The tests referring to Rhesus macaques were very clear. However, the data regarding to the number of the mouse in the experiment group were not completely evident.

Results

We identified from PubMed Central, CAPES periodicals, and ResearchGate, 932, 113, and 400 articles respectively, totaling 1,445 articles. Of the 602 articles screened, only eight (8) articles contained eligible trials about the ChAdOx1 vaccine and the Lh-CoVs. Five (5) articles addressed MERS-CoV, three (3) articles approached SARS-CoV-2, and zero (0) articles referred to SARS-CoV. The selected articles were written in English and published between 2017-2020. The remaining 594 articles did not meet the eligibility criteria and were excluded (Figure 1).

Referring to elected articles for ChAdOx1-MERS, four (4) presented pre-clinical tests using mice, camels, and Rhesus macaques (about 6 animals on average per experiment group) and 1 article presented a clinical trial with 19 participants of the experiment group. Regarding ChAdOX1 nCoV-19, of the three (3) elected articles, 2 presented pre-clinical trials using mice, pigs, and Rhesus macaques (about 6.5 animals on average per experiment group) and one (1) article presented one ongoing clinical trial with 543 participants of the experiment group.

Pre-Clinical Trials

Pre-clinical trials: two (2) articles presented data for the analysis of the immunological response after ChAdOx1 nCoV-19 vaccination

The studies were based on the administration of ChAdOx1 nCoV-19 containing the full-length spike protein of SARS-CoV-2 (GenBank YP_009724390.1) or SARS-CoV-2 (GenBank MN908947), with or without human tissue Plasminogen Activator (tPA) gene - a serine protease that cleaves peptide bonds in proteins, and with or without Tissue Culture Infectious Dose $(TCID)_{50}$ of SARS-CoV-2 (GenBank MN985325.1) as a challenge.

Doremalen *et al*., (19), to test the protective efficacy of the ChAdOx1 nCoV-19 and the absence of enhanced disease upon infection, conducted two trials: with mice and with Rhesus macaques.

1) In mouse test, the author administered $6x10^9$ ChAdOx1 nCoV-19 virus particle (vp) containing tPA. As a result, 100% of vaccinated mice produced specific Imunoglobulina-G (IgG) against S-protein and virus-specific neutralizing antibodies (nAbs) as early as 14 days post-vaccination.

2) In Rhesus macaques, the author administered $2.5x10^{10}$ vp/tPA ChAdOx1 nCoV-19, prime-only regimen (28 days before challenge) or prime-boost regimen (56 and 28 days before challenge). As a result, 100% of the macaques developed S-specific Abs as early as 14 days post-vaccination, and after challenge with $2.6x10^6$ $(TCID)_{50}$-SARS-CoV-2, 100% prime-boost and 33 % prime-only animals produced Imunoglobulina-M (IgM).

Graham *et al.*, (20), to compare the immunogenicity of either one or two doses of ChAdOx1 nCoV-19, performed two trials: with mice and with pigs.

1) in the mouse test, the author administered 6.02×10^9 vp of ChAdOx1 nCoV-19 on day 0 and on day 28, whereas prime-only mouses received a single dose on day 28. As a result, after 49 days, was observed a significant increase in Flavonol synthase/flavanone 3-hydroxylase Abs (FL-S) binding titers in prime-boost animals compared to their prime-only counterparts.

2) in pigs, the author administered 5.12×10^{10} vp ChAdOx1 nCoV-19 on day 0 (prime-only) and on day 0 and day 28 (prime-boost) and, tested the animal's blood for up to 42 days after vaccination. As a result, after the prime immunization, SARS-CoV-2 nAbs titers were detected in 2/3 prime-boost and 2/3 prime-only pigs, with increased titlers in all prime-boost pigs, which were significantly greater than the earlier time points and the titlers measured in the prime-only group. In addition, S-specific T-cell responses were significantly greater in the prime-boost pigs compared to prime-only animals.

Pre-clinical trials: three (3) articles presented data for the analysis of the immunological response after ChAdOx1 MERS vaccination

The studies were based on the administration of ChAdOX1 MERS-CoV containing the full-length spike protein (Genbank KJ650098.1), with or without a challenge of viral injection, or with a challenge based on the natural exposure.

Doremalen *et al*., (19), conducted two trials, to investigate the ChAdOx1 MERS efficacy after a single dose: with Rhesus macaques and with mice.

1) in the Rhesus macaques test, the author administered 3.9×10^8 vp ChAdOx1 MERS either via prime-boost or prime-only regimen. Then, they challenged the macaques with 7×10^6 $TCID_{50}$-HCoV. As a result, 100% of animals developed S-protein-specific IgG, as early as 14 days post-vaccination, and more than 90% of animals developed nAbs after a second dose.

2) in mice, the authors analyzed the vaccine cross-

protection for six different MERS-CoV strains. They administered 10^8vp ChAdOx1 MERS at day 0 and, 28 days later, they challenged the animals with 10^4 $TCID_{50}$ of one of six diverse MERS-CoV strains (Table 2). As a result, 100% of the animals survived regardless of the challenge virus used.

Alharbi *et al*., (21) also accomplished two simultaneous trials: 1) Evaluation of the immunogenicity of ChAdOx1 MERS in dromedary camels, either seropositive or seronegative for MERS-CoV; 2) Evaluation of the age effect on vaccine immunogenicity in seronegative dromedary calves (less than 1 year and between 1 year and two years).

1) In the study with camels, they administered 10^9 vp of the ChAdOx1 MERS, either prime-boost or prime-only regimen. Then, they placed vaccinated camels in the same location as the infected ones, and as a result: in the seronegative group, before the challenge, 100% of camels that have received boost vaccination had the anti-S1-Abs detectable and, after the challenge, 100% of camels had Abs levels increased; in seropositive group, before the challenge, 100% of camels enhanced anti-S1-Abs titer and, after challenge, the Abs levels increased (21).

2) In the study with calves, 50% of them (under 1 year old), received one (1) dose of the ChAdOx1 MERS, while 50% of calves (between 1 and 2 years old) received two (2) doses of the vaccine. As a result, no anti-S1-Abs were detected in the calves (under 1-year-old), except for one calve with a very low level of Abs. Whereas Abs were detected in the older calves (between 1 and 2 years) (21).

In a mouse model study that lasted 42 days, Alharbi *et al*., (22) investigated the functionality of tPA in the vaccine, from the administration of 10^8 vp ChAdOx1 MERS with or without tPA, at prime-only and prime-boost regimens. As a result, ChAdOx1 MERS/tPA induced significantly higher S-specific Abs than ChAdOx1 MERS without tPA, as well as serum Abs induced by ChAdOx1 MERS/tPA showed higher neutralization activity than without tPA (22).

Pre-clinical trials: adverse events and clinical aspects of ChAdOx1 nCoV-19 and of ChAdOx1 MERS

The analysis regarding adverse events and clinical aspects after ChAdOx1 vaccination was conducted based on one study about ChAdOx1 nCoV-19 and three studies about ChAdOx1-MERS.

Doremalen *et al*., (1,19), accomplished two studies: ChAdOx1 nCoV-19 and ChAdOx1 MERS. Both studies were based on standard nonhuman primate scoring sheets (sNHPs), focusing on areas such as general appearance, nasal discharge, and food intake. Regarding the ChAdOx1 nCoV-19, 100% of the vaccinated Rhesus macaques had no adverse events, or symptoms of lung disease, and all lungs were histologically normal without evidence of viral pneumonia or immune-enhanced inflammatory disease. Likewise, ChAdOx1 MERS-vaccinated animals had no lung lesions, consolidation, or pulmonary congestion, and lower clinical sNHPs than the control group (1,19).

Alharbi *et al*., (21) evaluated the clinical aspects after ChAdOx1 MERS vaccination from the camel's nasal discharge's abundance, scored from 0 (normal) to 3 (severe) during 14 days after challenge. As a result, vaccinated seropositive and seronegative camels obtained a significant reduction in nasal discharge over time as compared to control camels.

In like manner, Munster, *et al.*, (23) in a study that lasted 56 days, administered 10^8 vp ChAdOx1 MERS in mice. Then, after 28 days, they injected a lethal dose of 10^4 $TCID_{50}$ MERS-CoV into the animals. As a result, no signs of disease were found, no significant loss of body weight, no ruffled fur, and lethargy in any vaccinated mice (23).

Pre-clinical trials: Viral load of ChAdOx1 nCoV-19 and of ChAdOx1 MERS.

The analysis of viral load after vaccination was conducted based on one study about ChAdOx1 nCoV-19 and three studies about ChAdOx1-MERS.

Referring to ChAdOx1 nCoV-19, Doremalen *et al*., (19), demonstrated that only 17% of vaccinated Rhesus macaques had viral guide RNA (gRNA) and single guide RNA (sgRNA), even as the viral load was significantly lower in vaccinated macaques compared to the control group.

Doremalen *et al*., (1) detected infectious viruses in 50% and 25% of Rhesus macaques vaccinated with ChAdOx1 MERS prime-only and prime-boost regimens, respectively. Moreover, viral messenger RNA (mRNA) of animals in the prime-boost group was only detected at 1-Day Post Infection (DPI) in 41.6% of animals, and 3 DPI in 8.3% of the animals. Besides that, Doremalen *et al.*, (1) detected a reduced amount of viral mRNA in the mice that received prime-only ChAdOx1 MERS.

Alharbi *et al*., (21), reported in the study referring to ChAdOX1 MERS, that 100% of vaccinated animals had a decrease in viral RNA to undetectable levels by 14 post-challenge. These results added value to the study by Munster, *et al*., (23), a trial on a mouse model, in which even after the administration of a lethal MERS-CoV dose, no viral RNA was detected in any of the ChAdOx1 MERS vaccinated mice.

Clinical Trials

Clinical trials: immunological and adverse events of ChAdOX1 nCoV-19 and ChAdOx1 MERS

Currently, there is one (1) complete human study with ChAdOx1 MERS and one (1) ongoing human study with ChAdOx1 nCoV-19. Both were conducted by Folegatti *et al*., (11,13).

1) ChAdOx1 MERS study began in 2018, aiming to access: a) safety and tolerability; b) cellular and humoral immunogenicity. A total of 24 enrolled participants received a single intramuscular injection of ChAdOx1 MERS at (low, medium, and high) doses, with or without tPA. All participants were available for follow-up at 6 months, but five were lost to follow-up at 12 months. a) Referring to safety and tolerability: 92 (74%) of 124 solicited adverse events reported were mild, 31 (25%) were moderate, and all were self-limiting; possible unsolicited adverse events reported were predominantly mild and resolved with a period of 12 months; the proportion of moderate to severe adverse events was significantly higher in the high-dose group. b) Referring to the cellular and humoral immunogenicity: a significant increase from baseline in T-cell and IgG responses to the MERS-CoV S-protein was observed at all doses, and nAbs against live MERS-CoV were observed in (44%) of the high-dose group.

2) The study of ChAdOx1 nCoV-19 established to access: a) safety; b) reactogenicity; c) immunogenicity. The trial is still ongoing, and so far there are 1077 adult participants, of whom 543 were vaccinated with 5×10^{10} vp ChAdOx1 nCoV-19, (533 by prime-only, and 10 by boost regimen at day 0 and 28). As a result: a) Local and systemic reactions such as pain, feeling feverish, chills, muscle ache, headache, and malaise were common; b) So far, no serious adverse events were proved to be vaccine-related; c) S-specific T-cell responses peaked on day 14 at 7,91% of participants, as well as Anti-S-IgG response rose by day 28 at 23,38% and were boosted following a second dose for the 10 participants. The summary of results is demonstrated in (Table 2,3).

Table 2. Summary of results

Reference article	Participants	Sample size	Duration of experiment (days)	ChAdOX1 nCoV-19 vaccine dosage/vp	Study Design	End point
N. van Doremalen et al., 2020b	Rhesus macaques BALB/c and outbred CD1 mice	12 *---	63 14	$2.5x10^{10}$ $6x10^{9}$ / tPA	Case-control, randomized and blind	Prevented pneumonia Promoted nAbs
Simon P. Graham et al., 2020	BALB/c) and Outbred (CD1) mice Pigs	13 6	49 42	$6.02x10^{9}$ $5.12x10^{10}$	Case-control, randomized and blind	Increased Abs/FLS Boost regimen Promoted Higher immunogenic response
Reference	**Participant**	**Sample size**	**Total period (days)**	**ChAdOX1 MERS Dosage vp**	**Study Design**	**Endpoint**
N. van Doremalen et al., 2020a	Rhesus macaques Balb/c mice transgenic for hDPP4	12 *---	62 28	3.9×10^{8} 10^{8}	Case-control, randomized, and blind	Reduced severe illness Promoted limitation of mRNA, besides the absence of viral RNA in boost regimen
Naif Khalaf Alharbi et al., 2019	Dromedaries camels Dromedaries calves	10 05	365 196	10^{9} 10^{9}	Caso-control	A single dose protected 100% of the mice against six MERS-CoV strains A single dose boosted Abs in old seropositive camels and decreased viral title to undetectable At least two doses of vaccine were required to induce Abs in young seronegative calves
Naif Khalaf Alharbi et al., 2017	BALB/c mice	06	42	10^{8} / tPA	Case-control	A single dose with and without tPA elicited cellular immunity and nAbs

Cont. Table 2

Vincent J Munster et al., 2017	BALB/c mice expressing the human dipeptidyl peptidase hDPP4	10	65	10^8	Case-control, randomized, and blind.	Protected mice from infection even after a lethal dose of the MERS-CoV
Reference	**Participant**	**Sample size**	**Total period (days)**	**ChAdOx1 MERS or nCoV-19 dosage/vp**	**Study Design**	**End point**
Pedro M. Folegatti et al., 2020a	Human 18-50 years old	19	365	ChAdOx1 MERS 5×10^9 2.5×10^{10} 5×10^{10}	Randomized and uncontrolled	Was safe at all doses Elicited both humoral and cellular responses
Pedro M. Folegatti et al., 2020b	Human 18-55 years old	533 10	ongoing	ChAdOx1 nCoV-19 5×10^{10} prime-only 5×10^{10} prime-boost	Single-blind, randomized, and controlled	Elicited both humoral and cellular responses Booster immunization augmented Abs titers

Table 3. Summary of vaccines and challenges

Reference	Participant	Vaccine	Genbank vaccine	Challenge	Genbank Challenge
N. van Doremalen et al., 2020b	Rhesus macaques	SARS-CoV-2	YP_009724390.1	2.6×10^6 $TCID_{50}$ SARS-CoV-2 strain nCoV-WA1–2020	MN985325.1
N. van Doremalen et al., 2020a	Rhesus macaques Balb/c mice transgenic for hDPP4	MERS-CoV	KJ650098.1	7×10^6 $TCID_{50}$ HCoV-EMC/2012	JX869059
				10^4 $TCID_{50}$ Hu/HCoV/EMC/ 2012	JX869059
				Hu/Saudi Arabia/Rs924/2015	KY688119
				Hu/Korea- /Seoul/SNU1- 035/2015	KU308549
				Riyadh/KSA18013832/2018	MN723544
				Camel/Saudi Arabia/KFUHKU1/2013	KJ650297
				Camel/Burkina Faso/CIRADHKU785/2015	MG923471
Vincent J Munster et al., 2017	BALB/c mice expressing the human dipeptidyl peptidase hDPP4	MERS-CoV	KJ650098.1	10^4 $TCID_{50}$ HCoV-EMC2012	JX869059

Table 2. Reference article, participants, sample size, duration of experiment, type of vaccine and dosage/vp, study design, and end point. * The tests referring to Rhesus macaques were very clear. However, the data regarding the number of the mouse in the experiment group were not completely evident.

Table 3. Challenges reference: GenBank ® -The National Institutes of Health (NIH).

Discussion

To the best of our knowledge, herein we reported the first systematic review summarizing the benefits of the ChAdOX1 platform as a prophylactic measure for Lh-CoVs. The articles approaches agreed that both ChAdOx1-MERS and ChAdOx1 nCoV-19 vaccine candidates elicit nAbs, and reduce the severity of the disease, in addition to demonstrating an acceptable safety profile.

Certainly, there were some limiting factors in the research. For instance, the heterogeneity of the study designs, the sample sizes, the different vaccine dosages, and unfinished trials. Nevertheless, the previous approaches with the ChAdOx1 MERS contributed to elucidating the search target, the benefits of the ChAdOx1 platform as a prophylactic measure against Lh-CoVs, mainly regarding the SARS-CoV-2 (3,24-25).

According to WHO, an ideal vaccine for Coronaviruses must fulfill requirements inherent to those expected for other vaccines: be safe in the short term and

in the long term, be safe at different groups of age, ideally prime-only regimen, and provide immunity for a long as possible (8).

In this context, Doremalen *et al*., (11) added: "A unique vaccine against many lethal Coronavirus is the alternative to control the current pandemic". They demonstrated that the ChAdOx1 MERS Qatar/2/2014, not only protected animals from severe disease but also provided cross-immunity for six different Coronavirus strains.

Thus far, there is no study regarding ChAdOx1 nCoV-19 cross-immunity. In fact, there are few studies with attention to ChAdOx1 nCoV-19 (11,19-20). However, previous tests using the ChAdOx1 MERS platform from animal models, vaccination regimens, safety and immunogenicity (1,21-23), as well as clinical trials, have been facilitating the development of the ChAdOx1 nCoV-19 vaccine (13). To demonstrate, as soon as the SARS-CoV-2 genome was sequenced, Doremalen *et al*., (1) immediately adapted the ChAdOx1 MERS platform for the ChAdOx1 nCoV-19, thus enabling pre-clinical and clinical trials from the new ChAdOx1 platform (22).

Equally under those circumstances, Graham *et al.*, (20), demonstrated that ChAdOx1 nCoV-19 prime-boost regimen has the potential to induce higher T cell response, as well as enhance Abs response. These results imply that the combination of nAb and antigen-specific T cells would act in synergy to prevent and control infection, and notably suggest that boost regimen has better reliability.

Comparatively, Alharbi *et al*., (21) demonstrated that at least two doses of ChAdOx1 MERS are required to induce Abs in young seronegative calves and conversely, in older seronegative camels, a single dose of the ChAdOx1 MERS elicits systemic and local nAbs. These events suggest that seronegative calves were truly immune and naive to MERS-CoV, possibly without any cross-protection, and that greater vaccine responsiveness occurs in older animals because they have more mature IS.

Since articles (20,21) elucidated the advantages of boost vaccination, and article (21) connects IR to age in large animals, it is important to emphasize that the trials have not yet reached at least two criteria of those established by WHO: be effective with just one shot, as well as for the extremes of ages.

Another important observation is that, whether we compare human trials conducted by Folegatti *et al*., (11 and 13) and WHO standards for an ideal vaccine, we are about to infer that ChAdOx1 MERS fulfills several WHO criteria. As an illustration, Folegatti *et al*., (11) demonstrated that the vaccine-induced seroconversion in the majority of participants, T-cell responses in all of them, in addition to protection last up to 1 year, without severe adverse events.

As noted before, there are no human studies related to ChAdOx1 MERS with participants of extreme ages (children and elderly). As a matter of fact, not even to ChAdOx1 nCoV-19. However, ongoing trials with ChAdOx1 nCoV-19 are going to fill these and other gaps regarding the ChAdOx1 platform and the standards established by WHO. Thus far, according to Folegatti *et al*., (11), the ChAdOx1 nCoV-19 has an acceptable safety profile, and homologous boosting increased Abs responses, besides the induction of humoral and cellular immune responses.

As the final analysis, the gathering of articles relating the benefits of the ChAdOx1 platform vaccine against Lethal Human Coronaviruses demonstrated that previous approaches with the ChAdOx1 MERS have been contributing to the development of the ChAdOx1 nCoV-19. Although more time and more studies are needed, thus far, the ChAdOx1 platform has been shown to be safe and immunogenic as a prophylactic measure against Lh-CoVs, including for SARS-CoV-2.

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