Le Infezioni in Medicina, n. 4, 421-433, 2024
doi: 10.53854/liim-3204-2

Crimean-Congo haemorrhagic fever (CCHF): present and future therapeutic armamentarium

Stefano Di Bella[1], Stella Babich[2], Roberto Luzzati[1], Rosario Alessandro Cavasio[3], Barbara Massa[3], Neva Braccialarghe[3], Verena Zerbato[2], Marco Iannetta[3,4]

[1]Clinical Department of Medical, Surgical and Health Sciences, Trieste University, Trieste, Italy;
[2]Infectious Diseases Unit, Trieste University Hospital, Trieste, Italy;
[3]Department of Systems Medicine, Tor Vergata University, Rome, Italy;
[4]Infectious Disease Clinic, Policlinico Tor Vergata, Rome, Italy

Article received 29 July 2024 and accepted 23 October 2024

SUMMARY

Crimean-Congo haemorrhagic fever (CCHF) is an emerging severe tick-borne illness. The expanding habitat of *Hyalomma* ticks, coupled with migratory birds harbouring CCHF-infected ticks, contributes to an increasing number of potential hosts. The seroprevalence of anti-CCHF virus antibodies in livestock is approximately one-quarter, with a noticeable upward trend in recent years. The management of CCHF patients predominantly relies on supportive therapy, although a potential arsenal of antivirals, convalescent and hyperimmune plasma, monoclonal antibodies, and vaccines exists, both currently and in the future. This review aims to critically examine the current therapeutic approaches to managing CCHF, highlighting both the potential and limitations of existing treatments, and identifying future directions for improving patient outcomes.

Keywords: Crimean-Congo haemorrhagic fever, CCHFV, tick-borne illness, *Hyalomma* tick vaccines.

INTRODUCTION

Crimean-Congo haemorrhagic fever (CCHF) is a severe tick-borne illness with a wide geographical distribution. The etiological agent is the CCHF virus (CCHFV - *Bunyavirales* order). The virus was initially identified in the 1940s when Soviet soldiers fell ill with a hemorrhagic disease in Crimea, and later, in the 1960s, a virus with similar clinical symptoms was found in the Belgian Congo (now Democratic Republic of Congo), confirming its antigenic similarity and resulting in the naming of the Crimean-Congo Hemorrhagic Fever Virus (CCHFV) [1]. It is transmitted to humans by *Hyalomma* ticks. Available evidence demonstrates that CCHFV can productively infect a wide variety of domestic and wild animal species, but only humans develop severe symptomatic disease [2-4]. Approximately 10,000-15,000 cases of human CCHF occur annually worldwide [5]. From an epidemiological perspective, interest in this disease is growing. The expanding range of *Hyalomma* ticks, and migratory birds harbouring CCHF-infected ticks, contribute to an increasing number of potential hosts [6, 7]. A meta-analysis assessing the seroprevalence of anti-CCHFV antibodies in animals, primarily livestock, demonstrated a seroprevalence of 25% (with an upward trend in recent years) [8]. Historically, CCHF has affected areas in Africa, the Middle East, Asia, and southern and eastern Europe. However, the re-

Corresponding author
Stefano Di Bella
E-mail: stefano932@gmail.com

ports of cases of infections occurring outside these areas have shown that the possibility of potential outbreaks is not uncommon. For example, since 2013, 15 cases of CCHF have been identified in Spain, of which eight were retrospectively identified [9]. Diagnostic challenges have likely contributed to underdiagnosis, both due to the nonspecific nature of symptoms and because only a few laboratories in non-endemic areas have diagnostic kits for CCHF. The mortality rate in CCHF ranges from 10% to 40% [10]. To date, no strong evidence of the effectiveness of specific treatment against CCHF has been identified.

CCHFV STRUCTURE AND REPLICATION CYCLE

CCHFV is a single-strand, negative-sense RNA virus of the *Orthonairovirus* genus, *Nairoviridae* family, *Bunyavirales* order [11]. Its genetic material is characterized by three segments, namely large (L), medium (M) and small (S). The L segment encodes the RNA-dependent RNA-polymerase (RdRp) and an ovarian tumor-like protease (OTU). The M segment encodes for the glycoprotein precursor (GPC), the structural glycoproteins of the envelope, Gc and Gn and the medium non-structural protein (NSm). This segment exhibits up to 31% sequence divergence, which accounts for the existence of seven virus subclasses with varying virulence [12]. In the S segment one open reading frame (ORF) encodes for the nucleoprotein (NP), while the small non-structural protein (NSs) is encoded in an opposite-sense open reading frame [4]. CCHFV enters the host cell through pH-dependent and clathrin-dependent endocytosis; once in the cytoplasm, the viral RdRp produces complementary positive-strand RNA intermediates, which are used for the synthesis of viral proteins. These viral proteins coordinate the production of negative-strands RNA molecules, using positive-strand RNA intermediates as templates [13]. The GPC is proteolytically processed to produce mature glycoproteins along with the accessory proteins through the endoplasmic reticulum and Golgi apparatus. Newly synthesised negative-strands RNA molecules serve as viral genomes and are enveloped in new viral particles, which are released via the secretory pathway and are ready to infect other cells [4].

CLINICAL COURSE AND PATHOGENESIS OF CCH IN HUMANS

The pathogenesis of CCHF is multifaceted, involving complex mechanisms that contribute to disease severity and development (Figure 1).
The CCHFV primarily spreads to humans through contact with infected animal blood, bodily fluids, or bites from infected ticks. Humans can also become infected through contact with a patient during the acute phase of CCHF [14]. Once inside the human body, the virus replicates in target cells such as endothelial cells, macrophages, and epithelial cells, triggering both innate and adaptive immune responses. Upon exposure to the virus, the incubation period varies depending on the mode of transmission: after a tick bite, it is generally between 1 and 3 days (up to a maximum of 9 days), while after contact with infected blood or tissues, it is between 5 and 6 days (up to a maximum of 13 days) [5]. During this incubation peri-

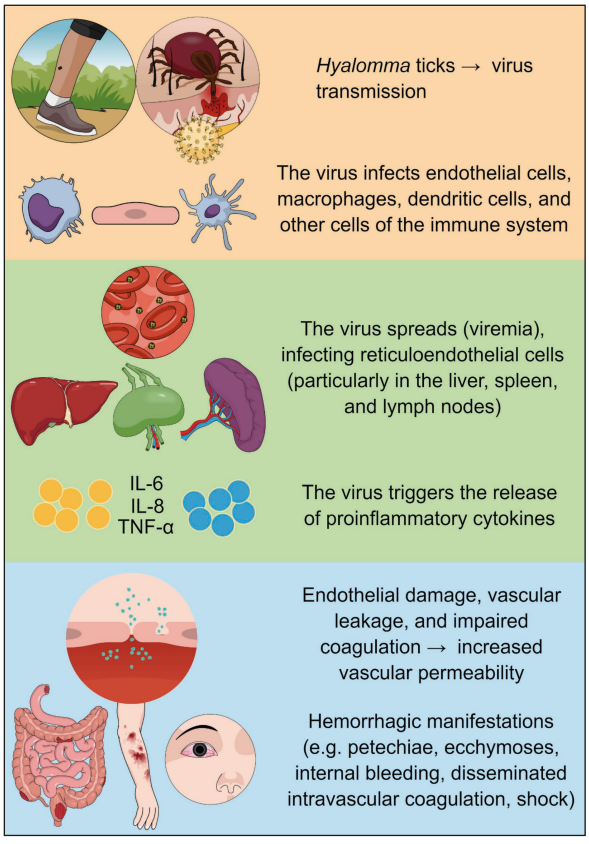

Figure 1 - CCHF pathogenesis.

od, individuals may not exhibit any symptoms. At the early stages of the disease, the virus disseminates through the bloodstream and lymphatic vessels, leading to systemic symptoms including fever, severe headache, photophobia, muscle aches, and malaise, backache and conjunctivitis. Viral replication and immune responses can damage blood vessel walls, causing increased vascular permeability and inflammation. These early symptoms often mimic those of common viral infections, making diagnosis challenging in the early stages. As the disease progresses, patients may develop more severe symptoms such as nausea, vomiting, abdominal pain, and diarrhoea. The hemorrhagic phase of CCHF, typically begins 3-5 days after symptom onset, and usually lasts 1-3 days, is marked by severe haemostatic impairment, resulting in widespread bleeding and coagulopathy [13]. This phase is characterised by petechiae, ecchymosis, and mucosal bleeding, occurring as a consequence of the virus directly damaging endothelial cells within blood vessels. Epistaxis is present in less than 50% of patients in the hemorrhagic phase, hematemesis in less than 35% of patients, haematuria, melena and haematochezia in 10%-20% of patients, and intra-abdominal or intracerebral bleeding in 1%-2% of cases [5]. Additionally, the host's immune response may exacerbate coagulopathy by producing inflammatory cytokines storms and causing haemostatic dysfunction as disseminated intravascular coagulation (DIC). Concurrently, patients may experience hepatomegaly and splenomegaly due to liver and spleen involvement. The disease can progress rapidly, with patients deteriorating into a severe and potentially fatal stage within a few days. This stage is marked by the development of DIC, severe haemorrhage, and multi-organ failure [15].

The mortality rate in CCHF ranges from 10% to 40%, influenced by factors such as healthcare infrastructure availability and timely antiviral therapy. Despite medical management advances, mortality remains high, with death typically occurring within two weeks of symptom onset, primarily due to shock and multi-organ failure [10, 16].

When obtainable, CCHF viremia levels have been correlated with disease severity, and viral loads equal to or exceeding 10^8 copies/mL and 10^9 copies/mL are significantly associated with high CCHF mortality [17]. Estimating the infection fatality rate for CCHF is complicated by the presence of asymptomatic and often undiagnosed infections, making further research and advanced surveillance methods necessary to achieve a more precise assessment [18].

The convalescent phase of CCHF typically sets in between the 10th and 20th day of illness, lasting up to 1 year. Most patients recover without complications or long-term effects. Among those experiencing symptomatic convalescence, common symptoms include fatigue, malaise, hair loss, anorexia, and polyneuritis. Tachycardia and dyspnoea have also been reported, along with memory, visual, and auditory impairments [4, 19]. A study from Turkey found that 48.4% of patients exhibited symptoms of post-traumatic stress disorder (PTSD), with 18.5% experiencing PTSD after recovery [20].

Currently, there have been no documented cases of relapses or reinfections of CCHF, especially among individuals re-exposed in endemic areas. Nevertheless, the duration of protective immunity against the virus has not yet been fully understood [21].

GENERAL OVERVIEW OF CURRENT THERAPEUTIC APPROACHES

The care and management of CCHF patients require careful monitoring and a multifaceted therapeutic approach. Laboratory monitoring is crucial, including blood count, liver enzymes, total bilirubin, serum creatinine, coagulation parameters such as the International Normalised Ratio (INR) and activated partial thromboplastin time, creatine kinase, and lactate dehydrogenase. Timely testing is recommended, especially in cases of low platelet counts or suspected DIC [22].

Supportive therapy is the mainstay of treatment, involving the administration of blood products including thrombocytes, fresh frozen plasma, and erythrocyte preparations [23]. Replacement therapy should be guided by daily complete blood count checks. Platelet transfusion is effective in preventing or resolving bleeding due to thrombocytopenia or platelet dysfunction. Considering the incidence of DIC in CCHF, some platelet destruction is expected, and an immediate increase in platelet levels after transfusion may not be observed [24]. Maintaining proper electrolyte balance and adjusting fluid therapy is also fundamental. Hypotonic solutions should be avoided and in cases of kidney failure requiring renal re-

placement therapy, haemodialysis is preferred over peritoneal dialysis. Consideration should be given to potential sites of bleeding, and precautionary measures should be implemented. This may include the use of antacids for individuals with peptic ulcers, avoidance of intramuscular injections (to prevent haematomas and local bleeding at puncture sites), and refraining from the use of aspirin or other medications that affect the coagulation system [25]. Antivirals such as ribavirin have shown poor efficacy in treating CCHF patients [26-29].

In severe CCHF cases characterised by a dysregulated inflammatory response, anti-inflammatory drugs like high-dose methylprednisolone, when combined with ribavirin, have shown improved outcomes [30]. A clinical trial, conducted in Iran from 2010 to 2011, aimed to assess the treatment outcomes for patients experiencing severe thrombocytopenia (less than 50,000/mm^3) due to CCHF. Thirteen patients in the intervention group were administered ribavirin and high-dose methylprednisolone, while 22 patients in the control group received ribavirin alone. Results indicated that patients who received high-dose methylprednisolone exhibited a significant increase in platelet and white blood cell counts within 36 hours ($p < 0.001$) [30].

In regions with limited healthcare, combating CCHFV demands urgent public health education and vaccination campaigns [31]. The strategy gives special importance to proper protective measures, including personal equipment in healthcare and abattoir settings. Agricultural workers are advised to wear protective clothing to minimise tick exposure. Control measures targeting the tick vector, such as acaricides and animal quarantine, are decisive, and vaccination is essential for preventing CCHFV infections in both humans and livestock. Public health education aims to reduce exposure activities and prompt symptom recognition, forming a combined approach in areas with restricted healthcare access [4]. An infographic on present and future potential options against CCHFV is reported in Figure 2.

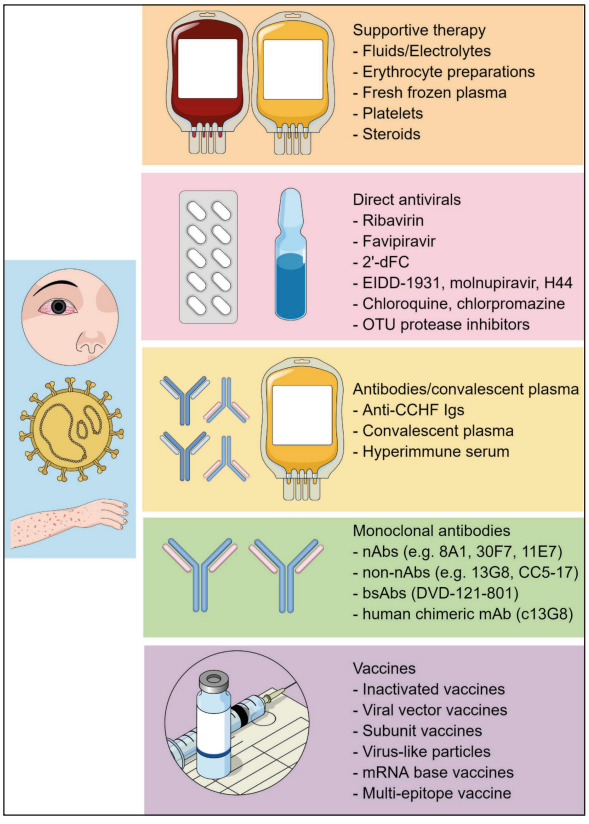

Figure 2 - Present and future potential options against CCHFV.

Ribavirin

Different antiviral therapies against CCHFV have been investigated. Ribavirin is the only one that has been widely used. According to the World Health Organization (WHO) and the Centers for Disease Control and Prevention (CDC) the advised dose is: 30 mg/kg *per os* as an initial loading dose, then 15 mg/kg *per os* every 6 hours for 4 days, then 7.5 mg/kg *per os* every 8 hours for 6 days [32, 33]. Ribavirin (1-beta-D-ribo-furanosyl-1,2,4-triazole-3-carboxamide) is a synthetic purine nucleoside analogue with known broad-spectrum antiviral activity. It was first tested in 1972 against both positive- and negative-sense DNA and RNA viruses [34]. Later it demonstrated activity against viruses responsible for several hemorrhagic diseases, such as Lassa fever but also hepatitis C and some respiratory viruses (respiratory syncytial virus, influenza A and B, measles) [35, 36]. However, the use of ribavirin, both oral and intravenous, as treatment for CCHFV is still controversial and few prospective randomised studies are available [37]. In 1985 it was employed for the first time as post-exposure prophylaxis in a nosocomial CCHF outbreak in South Africa. Six CCHFV contacts were treated

with ribavirin and only one developed a mild clinical disease, the others developed neither clinical CCHF nor antibodies to the virus [38]. In 1995 three CCHF patients in Pakistan were treated with ribavirin with reported rapid benefit [39]. Subsequently, ribavirin was successfully employed as a treatment protocol for CCHF in Iran demonstrating its efficacy even when orally administered [40]. Oral ribavirin efficacy was determined to be 34% (RR 0.66, 95% CI, 0.45-0.98) among suspected CCHF patients and 80% (RR 0.20, 95%CI 0.09-0.45) among patients with confirmed CCHF disease [40]. Ergonül *et al.* reported that ribavirin significantly reduced the mortality rate when administered within 48 hours from symptom onset (OR 0.03, 95%CI 0-0.58) [41]. However, in 2011, a systematic review and meta-analysis stated that the evidence for the effectiveness of ribavirin was limited [26]. The low quality of evidence has been reported also by Johnson S. *et al.* in 2018 [27]. In 2018, Ergonül *et al.* showed ribavirin efficacy as post-exposure prophylaxis reducing significantly the risk for CCHFV infection (OR 0.01, 95%CI 0.003-0.03; p<0.001 at univariate and OR 0.009, 95%CI 0.001-0.039; p<0.001 at multivariable analysis) [28]. Arab-Bafrani *et al*. in 2019 conducted a systematic review and meta-analysis including 24 studies with 1287 confirmed CCHF patients and, differently from previous meta-analyses, excluding suspected cases and overlapping retrospective studies. They showed that in the early phases of CCHF disease (within four days from symptoms onset), ribavirin has a crucial role in treatment and co-administration of corticosteroids could benefit in the hemorrhagic phases because the cytokine storm (overproduction of proinflammatory cytokines) is the major factor in causing severe tissue damage, dysfunction and organ failure [29]. In 2022 D'Addiego *et al.* conducted a non-randomized controlled trial evaluating the effect of ribavirin on the mutation rate of the CCHFV genome in CCHF patients compared to ones who received supportive care only. They observed a similar rate of decrease in virus titers for both groups, suggesting that ribavirin treatment did not contribute to a reduction of the viral load and therefore other factors must be involved in CCHFV clearance [42]. Currently there is an ongoing trial (NCT02483260) on the intravenous use of ribavirin in people with CCHF or Lassa fever [43].

Favipiravir

Favipiravir (T-705) is an antiviral drug that selectively inhibits the RNA-dependent RNA polymerase of influenza virus. Favipiravir blocks the replication of many other RNA viruses, including flaviviruses (Yellow Fever and West Nile), paramyxovirus, respiratory syncytial virus and noroviruses [44]. Both Oestereich *et al.* and Hawman *et al.* showed the efficacy of favipiravir in inhibiting viral replication and preventing a lethal outcome even when administered in the late phases of CCHFV infection *in vivo* [45, 46]. Furthermore, it seems that favipiravir and ribavirin exert synergistic effects *in vitro* suggesting that a combined treatment may provide benefits rather than adverse effects [45]. There is an ongoing Phase 1/2 trial (NCT05940545) investigating the combination of ribavirin and favipiravir in the treatment of CCHF patients [47].

OTU Protease Inhibitors

Deubiquitinating (DUB) enzymes encoded by many viruses have an important role in viral replication and innate immune evasion. CCHFV L protein contains an OTU-domain with DUB activity that binds to and removes ubiquitin and interferon-stimulated gene15 from their target to circumvent host antiviral innate immune responses [48]. Kocabs and Aslan developed a fluorescent reporter assay of CCHFV OTU protease to screen CCHFV OTU inhibitors that might possess potential antiviral activity against CCHFV. In 2015 they showed *in vitro* inhibitory activity of two molecules, especially the compound phenanthrenequinone [49].

UbV-CC4 is another screened OTU protease inhibitor that showed *in vitro* strong inhibition of viral replication either inhibiting the IFN antagonist function of OTU and blocking the formation of ribonucleoprotein complexes, an assembly of molecules containing both protein and RNA necessary for virus survival [50, 51].

Other new antivirals

2′-deoxy-2′-fluorocytidine (2′-dFC) is a molecule investigated in 2017 by Welch *et al.* through a high-throughput screening assay for its antiviral effect against CCHFV, showing potentially superior inhibitory activity compared to favipiravir or ribavirin *in vitro* [52]. The authors also determined that 2′-dFC acts synergistically with favipiravir to inhib-

it CCHFV replication without adverse cytotoxic effects. 2′-dFC efficacy against non-encephalitic bunyaviruses was also shown by Smee *et al.* [53].
In 2022 Wang *et al.* evaluated several nucleoside analogues with activity against CCHFV, including remdesivir (GS-5734), EIDD-1931 and its pro-drug molnupiravir (EIDD-2801) and H44, a favipiravir-derived compound. While remdesivir did not show any inhibitory effect, the other three molecules inhibited CCHFV infection *in vitro* at the "post-entry" stage of viral infection but only H44 showed activity *in vivo*, even with delayed administration [54].
New horizons in antiviral therapy of CCHF consist in compounds that target host cell pathways supporting viral replication. CCHFV enters the cells via a clathrin- and early endosome-dependent pathway, which can be potential targets of treatments [55].
Among these, chloroquine and chlorpromazine - drugs already used as antimalarial and antipsychotic, respectively - showed antiviral activity and synergistic effects with ribavirin, suggesting that a combined therapy may represent a better treatment strategy for CCHFV infection [50].

Passive immunisation and convalescent plasma
Passive immunisation and convalescent plasma therapy have emerged as potential treatment options for CCHF. Although large-scale trials have not been conducted, limited evidence, as discussed below, suggests that the administration of plasma or antibodies from survivors of CCHF can confer a benefit in seriously ill patients.
The idea of treating CCHF patients with specific immunoglobulin was first proposed by Chumakov in 1944 [56]. Antibodies to CCHFV are typically not present within 5-9 days after the onset of illness, and patients who die from the disease do not usually develop a measurable antibody response [57].
A similar lack of antibody development among fatal cases is seen in other viral hemorrhagic fevers, such as Argentine haemorrhagic fever and Lassa fever [58]. This finding encouraged the therapeutic use of antibodies derived from recovered CCHF patients or from animals. In fact, there was early recognition of the potential benefits of treatments using serum prepared from the blood of recovered CCHF patients or gamma-globulin obtained by immunising horses [59].
Regarding the use of intramuscular or intravenous anti-CCHF immunoglobulin, Bulgarian investigators described the rapid improvement of seven severely ill patients treated simultaneously with intramuscular and intravenous anti-CCHF immunoglobulin, prepared from the plasma of CCHF survivor donors, boosted with one dose of CCHF vaccine, but its efficacy has still not been assessed in a randomised clinical trial [60]. Lazarev *et al.* in 1969, suggested that the use of convalescent sera in the first 2-3 days of disease was better than treatment after 4 days [61]. Moreover, Van Eeden *et al.* described an outbreak of CCHF in South Africa in 1985. A total of 5 patients were enrolled. Two patients received 1, and 3 patients received 2 injections of 250 mL of serum intravenously. All the patients who received serum survived. Four patients demonstrated symptom improvement after administration of the first dose. In contrast, two untreated patients died and showed no antibody response at the time of death, suggesting the importance of immune serum therapy for survival [62]. Although the results were not statistically significant, the authors suggested that a continuous infusion of hyperimmune serum over 48-72 hours might more effectively influence the disease outcome [63].
Moreover, intravenous immunoglobulin may be effective in patients with CCHF associated with secondary hemophagocytic lymphohistiocytosis (HLH), during the hemorrhagic period of the disease [64]. Kubar *et al*. reported interesting results in CCHF patients (n=15) with a high viral load after administration of hyperimmune globulins prepared from the serum of 22 convalescent patients. A total of 26 CCHF patients were enrolled in this study. A single dose of 400 KU of hyperimmunoglobulin was given to each patient before viral load was detected. Between 15 patients with a viral load of 108 copies/mL (a high-risk group) the survival rate was found to be 86.6% and 2 patients died despite CCHFV hyperimmunoglobulin administration [65]. The lack of a control group made it difficult to analyse this rate.
The European Centre for Disease Prevention and Control 2008 recommendations reported the experience of specific intramuscular human immunoglobulin (CCHF-Bulin) used in Bulgaria [66]. They have been derived from the plasma of convalescent patients and have been used since 1975 for the post-exposure prophylaxis of people who have

been in contact with suspected CCHF cases or treatment of CCHF patients and for prevention of bioterrorism. Depending on the case, different doses of immunoglobulins were recommended: for prophylaxis, 3 mL; for suspected cases, 6 mL; and for confirmed cases, 6-9 mL on days 1-5, or until a therapeutic effect is achieved [63].

Monoclonal antibodies

Monoclonal antibodies (mAbs) have gained attention in recent years due to their use against COVID-19, but several molecules are in different stages of development for other viral infections such as hepatitis C, hepatitis B, Ebola Virus and arboviruses, like CCHFV [67].

Studies on serum of infected patients showed that low levels of antibodies are associated with worse outcomes, while the development of specific IgG within 7-9 days from symptom onset is protective from the fatal disease [68, 69]. Therefore, the administration of mAbs against CCHFV could theoretically be helpful.

We can categorize mAbs into neutralizing antibodies (nAbs) and non-neutralizing antibodies (non-nAbs). nAbs target the CCHFV Gc or Gn antigen and can block the virus from entering the host's immune system cells. In contrast, non-nAbs are directed to the secreted glycoprotein GP38 of CCHFV and can induce activation of complement, natural killer cells and phagocytosis, resulting in the elimination of infected cells [70, 71].

Among isolated nAbs, 8A1, 30F7 and 11E7 bind the Gc antigen and exhibit broad cross-neutralization of CCHFV strains analysed between the ones isolated throughout Africa, Asia, Middle East and southern Europe. Notably, 8A1 displayed a higher neutralising activity at lower concentrations *in vitro*, while combining two or all three molecules did not enhance their potency, even if they bind different epitopes [72]. Studies conducted on suckling mice showed that nAbs inoculated before the exposure to the virus were protective against the lethal infection; when they were inoculated after the exposure the protection was only partial. Moreover, the protection was higher when suckling mice received anti-Gn nAbs such as 6B12, 11F6, 7F5 or 8F10, even though they did not prevent cells' infection *in vitro* [73].

Fels *et al*. were able to isolate nAbs against distinct epitopes of Gc antigen with various degrees of neutralisation potency from CCHF human survivors. Then, the group engineered bispecific antibodies (bsAbs) selecting the molecules that displayed a higher synergism. Efficacy was assessed *in vitro* and two bsAbs were identified to test their therapeutic effect *in vivo* on 4-8 weeks old mice. DVD-121-801 was the only bsAbs that ensured protection from the lethal infection [73]. Golden *et al*. evaluated the efficacy of non-nAb 13G8. Prophylactic administration to adult mice later exposed to CCHFV resulted in 60% of survival compared to the negative control mice group, which succumbed to the infection. The association of pre- and post-exposure administration of 13G8 led to an increased survival of >90%, while post-exposure treatment determined 60% of survival if administered on day 1 and on day 4. Delayed treatment to day 2 and day 5 lowered the chances of survival to 20%. Moreover, it was observed that 13G8 prevents the virus from spreading to the liver and the spleen of mice creating hemorrhagic lesions. Due to the high diversity of the M segment, the cross-protective efficacy against a different viral strain was examined. Despite the initial promising results, it was observed a reduction in efficacy down to 20% [71].

Further studies confirmed that GP38 could be an antigen of interest for new therapies, even if its role in viral replication is still unclear. A human chimeric mAb, c13G8, and a non-nAb isolated from serum of human survivor of CCHF, CC5-17, have been compared as they compete for the same antigenic site. Their difference in binding is 22 degrees and enables CC5-17 to engage GP38 more stably. Therefore, it was investigated if this characteristic increased the protective action *in vivo* using infected mouse models, testing two dosing regimens. Surprisingly, the higher dosage of CC5-17 showed a 50% rate of survival, while the higher dosage of c13G8 reached a 65-80% rate of survival [74].

The efficacy of mAbs in humans is yet to be tested and this represents a great challenge for researchers in the future.

Table 1 presents an overview of various therapies being investigated for their efficacy against viral infections. Each therapy is categorised based on its class, mechanism of action, and available efficacy data. The efficacy data presented reflect the current status of clinical and preclinical research, highlighting the need for further investigation and validation.

Table 1 - Summary of therapies in use and under in study.

Therapy	*Class*	*Mechanism of action*	*Efficacy data*
Corticosteroids	Anti-inflammatory	*Affecting genes and transcription factors involved in the inflammatory pathway to reduce inflammation*	More preclinical and clinical data need [30]
Convalescent plasma	Antibodies	*Neutralization of viral particles. Signalling inhibition of inflammatory pathway*	More preclinical and clinical data needed [60]
Ribavirin	Nucleoside analogue	*Action on RdRp*	Poor efficacy; more data needed on combination therapy [26-29]
Favipiravir	Nucleoside analogue	*Action on RdRp*	Good efficacy in late treatment in rodents; more clinical data needed [45, 46, 54]
2'-deoxy-2'-fluorocytidine	Nucleoside analogue	*Action on RdRp*	More preclinical and clinical data needed [52, 53]
Molnupiravir	Nucleoside analogue	*Action on RdRp*	No efficacy demonstrated [54]
Remdesivir	Nucleoside analogue	*Action on RdRp*	No efficacy demonstrated [54]
UbV-CC4	OTU protease inhibitors	*Inhibition of the IFN antagonist function of OTU blocking the formation of ribonucleoprotein complexes*	More preclinical and clinical data needed [50, 51]
8A1, 30F7, 11E7, 6B12, 11F6, 7F5, 8F10, DVD-121-801	Neutralizing monoclonal antibodies	*Binding Gc and Gn antigen to block the virus from entering the host's cell*	More preclinical and clinical data needed [50, 72, 73]
13G8	Non neutralizing monoclonal antibodies	*Directed to GP38 of CCHFV inducing activation of immune system*	More preclinical and clinical data needed [71]

CCHFV: Crimean-Congo haemorrhagic fever virus; IFN: interferon; OUT: ovarian tumor-like protease; RdRp: RNA-dependent RNA-polymerase.

Vaccines

Preventive strategies for CCHFV are necessary due to the lack of available and adequate medical treatments. Currently, different strategies for the development of an effective and safe CCHFV vaccine are under evaluation, including inactivated, viral vector-based, subunit and transgenic plant-based, virus-like particles, and nucleic acid-based vaccines [75].

Some aspects hinder the development of an effective and safe vaccine, such as the genetic variability of CCHFV, the lack of naturally susceptible animal models, incomplete understanding of protective epitopes and the dynamics of protective immune responses, the paucity of heterologous challenge studies and the lack of a world-wide interdisciplinary research consortium [75, 76].

Inactivated vaccines are produced by chemical or physical inactivation of infectious agents. A mouse brain-derived vaccine was in use in Bulgaria (1974-1996) in humans [77]. This vaccine is not a valid option for widespread use due to safety concerns (potential induction of autoimmune and allergic responses) and lack of efficacy trials [78]. For these reasons, the Bulgarian vaccine has never been approved by the Food and Drug Administration or the European Medicines Agency. New attempts have been made to develop a cell culture-derived vaccine capable of triggering a robust immune response, avoiding CCHFV culture on suckling mouse brain, thus improving the safety profile of these formulations compared to the Bulgarian vaccine [79, 80]. These approaches are generally based on the Turkey-Kelkit06 strain of CCHFV cultured in Vero-E6 cells [75].

Viral vectored vaccines consist of modified viral particles which contain one or more foreign genes encoding antigens of the target pathogen. They trigger both innate and adaptive immune responses and can be replication-competent or either replication-deficient. One possible limitation is represented by the immunisation to the antigenic com-

ponents of the vector that can weaken the immune response to the targeted antigen [81]. Genes encoding either nucleocapsid proteins (NP) or glycoprotein precursors (GPC) of CCHFV have been inserted in different types of viral vectors, including Modified Vaccinia virus Ankara, Adenoviruses, Bovine Herpesvirus 4, Vesicular Stomatitis Virus [75, 82-86].

Subunit vaccines contain CCHFV antigens, such as the structural Gn and Gc, or NP. After production, viral antigens are purified and can be associated with adjuvants [75, 76]. CCHFV specific antigens can be produced using the baculovirus expression system in insect-cells [87]. Alternatively, viral antigens can be produced by transgenic plants [88]. New attempts are under evaluation to increase immunogenicity of CCHFV subunit vaccines, exploiting nanoparticles technology [89].

Virus-like particles represent a specific form of subunit vaccines, in which structural proteins assemble into viral constructions that resemble the actual pathogen [84]. In the specific case of CCHFV vaccine, virus replicon particles have been realised, including the complete S and L (not M) genome segments in the viral particle, allowing for a single round of replication. Virus Replicon Particles (VRP)-based vaccines have been used in animal models to induce specific humoral and cell-mediated immunity against CCHFV [90, 91].

In the last three decades DNA vaccines have been extensively investigated and attempts to obtain a DNA-based CCHFV vaccine started in 2006 [92]. DNA vaccines encoding GPC, Glycoprotein Gn, Gc, non-structurally secreted protein GP38 and NP have been studied in mouse models [93-97]. Recently, a DNA-based vaccine, containing NP and GPC genes of the CCHFV strain Hoti, was tested in a cynomolgus macaque model for CCHFV. The vaccine was well tolerated and induced CCHFV-specific antibody and T cell responses. This is the first vaccine to show efficacy in a non-human primate model of CCHF and supports advancement into human clinical trials [98, 99].

After the approval of the first two mRNA-based vaccines for COVID-19, the application of this platform to different pathogens was rapidly implemented. Although first attempts to develop a mRNA based vaccine for CCHFV started in 2019 (S segment of the CCHFV Ank-2 strain) [82], the development of a carrier system for mRNA with lipid nanoparticles improved the immunogenicity and efficacy of mRNA vaccines, which were based on mRNA encoding GPC, Gc, Gn and NP [100, 101].

Current and future directions for CCHFV vaccine development rely on Immunoinformatics and reverse vaccinology methods to design a multi-epitope vaccine for CCHFV that has the potential to elicit a protective humoral and cellular immune response [102-105]. One phase 1 study on a CCHFV vaccine in humans was initiated in 2017 (NCT03020771); although enrollment was completed, data are still unavailable [106].

Another phase 1 study is currently ongoing in healthy adult volunteers in the United Kingdom (ISRCTN 12351734), investigating the safety and immunogenicity of a vectored-vaccine against CCHFV called "ChAdOx2 CCHF". This is a replication-deficient adenoviral-vectored vaccine expressing the CCHFV GPC [84], developed by the University of Oxford.

CONCLUSIONS

CCHF is a severe tick-borne disease that has expanded geographically, driven mainly by ticks migration. Despite its high fatality rate, no definitive treatment exists. Ribavirin shows poor efficacy. New antivirals like favipiravir and monoclonal antibodies are promising but require more clinical data. Vaccines are under development. Prevention and early intervention remain critical until effective vaccines and treatments will be available.

Author contribution

S.D.B., R. L., V.Z., M.I.: formal analysis, methodology, supervision, writing – original draft, writing – review & editing; S.B., R.A.C., B.M., N.B.: data curation; formal analysis, writing – original draft, writing – review & editing. All authors approved the submitted version and have agreed both to be personally accountable for the author's own contribution and to ensure that questions related to the accuracy or integrity of any part of the work, even ones in which the author was not personally involved, are appropriately investigated, resolved, and the resolution documented in the literature.

Conflict of interest

Authors have no conflict of interest to disclose.

Funding

Authors received no funding for this study.

REFERENCES

[1] Kuehnert PA, Stefan CP, Badger CV, Ricks KM. Crimean-Congo Hemorrhagic Fever Virus (CCHFV): A Silent but Widespread Threat. *Curr Trop Med Rep*. 2021; 8(2): 141-147.
[2] Spengler JR, Bergeron É, Rollin PE. Seroepidemiological Studies of Crimean-Congo Hemorrhagic Fever Virus in Domestic and Wild Animals. *PLoS Negl Trop Dis*. 2016; 10(1): e0004210.
[3] Kar S, Rodriguez SE, Akyildiz G, Cajimat MNB, Bircan R, Mears MC, et al. Crimean-Congo hemorrhagic fever virus in tortoises and Hyalomma aegyptium ticks in East Thrace, Turkey: potential of a cryptic transmission cycle. *Parasit Vectors*. 2020; 13(1): 201.
[4] Hawman DW, Feldmann H. Crimean-Congo haemorrhagic fever virus. *Nat Rev Microbiol*. 2023; 21(7): 463-477.
[5] Frank MG, Weaver G, Raabe V, State of the Clinical Science Working Group of the National Emerging Pathogens Training and Education Center's Special Pathogens Research Network. Crimean Congo Hemorrhagic Fever Virus for Clinicians-Virology, Pathogenesis, and Pathology. *Emerg Infect Dis*. 2024; 30(5): 847-853.
[6] Valcárcel F, Elhachimi L, Vilá M, et al. Emerging Hyalomma lusitanicum: From identification to vectorial role and integrated control. *Med Vet Entomol*. 2023; 37(3): 425-459.
[7] Mancuso E, Toma L, Pascucci I, et al. Direct and Indirect Role of Migratory Birds in Spreading CCHFV and WNV: A Multidisciplinary Study on Three Stop-Over Islands in Italy. *Pathogens*. 2022; 11(9): 1056.
[8] Nasirian H. Crimean-Congo hemorrhagic fever (CCHF) seroprevalence: A systematic review and meta-analysis. *Acta Trop*. 2019; 196: 102-120.
[9] Negredo A, Sánchez-Ledesma M, Llorente F, et al. Retrospective Identification of Early Autochthonous Case of Crimean-Congo Hemorrhagic Fever, Spain, 2013. *Emerg Infect Dis*. 2021; 27(6): 1754-1756.
[10] Who Crimean-congo haemorrhagic fever fact sheets [Internet]. Available at: https://www.who.int/en/news-room/fact-sheets/detail/crimean-congo-haemorrhagic-fever. [accessed 29th Sept 2024].
[11] Serretiello E, Astorri R, Chianese A, et al. The emerging tick-borne Crimean-Congo haemorrhagic fever virus: A narrative review. *Travel Med Infect Dis*. 2020; 37: 101871.
[12] Freitas N, Enguehard M, Denolly S, et al. The interplays between Crimean-Congo hemorrhagic fever virus (CCHFV) M segment-encoded accessory proteins and structural proteins promote virus assembly and infectivity. *PLoS Pathog*. 2020; 16(9): e1008850.
[13] Bente DA, Forrester NL, Watts DM, McAuley AJ, Whitehouse CA, Bray M. Crimean-Congo hemorrhagic fever: history, epidemiology, pathogenesis, clinical syndrome and genetic diversity. *Antiviral Res*. 2013; 100(1): 159-189.
[14] Ergonul O, Whitehouse CA. Crimean-Congo Hemorrhagic Fever: A Global Perspective. *Springer Science & Business Media*. 2007; 334.
[15] Saksida A, Duh D, Wraber B, Dedushaj I, Ahmeti S, Avsic-Zupanc T. Interacting roles of immune mechanisms and viral load in the pathogenesis of crimean-congo hemorrhagic fever. *Clin Vaccine Immunol*. 2010; 17(7): 1086-1093.
[16] Raabe VN. Diagnostic Testing for Crimean-Congo Hemorrhagic Fever. *J Clin Microbiol*. 2020; 58(4): e01580-19.
[17] Cevik MA, Erbay A, Bodur H, et al. Viral load as a predictor of outcome in Crimean-Congo hemorrhagic fever. *Clin Infect Dis*. 2007; 45(7): e96-100.
[18] Riccò M, Baldassarre A, Corrado S, Bottazzoli M, Marchesi F. Seroprevalence of Crimean Congo Hemorrhagic Fever Virus in Occupational Settings: Systematic Review and Meta-Analysis. *Trop Med Infect Dis*. 2023; 8(9): 452.
[19] Shahhosseini N, Wong G, Babuadze G, et al. Crimean-Congo Hemorrhagic Fever Virus in Asia, Africa and Europe. *Microorganisms*. 2021; 9(9): 1907.
[20] Gul S, Gul EU, Yesilyurt M, Ozturk B, Kuscu F, Ergonul O. Health-related quality of life and the prevalence of post-traumatic stress disorder among Crimean-Congo hemorrhagic fever survivors. *Jpn J Infect Dis*. 2012; 65(5): 392-395.
[21] Mendoza EJ, Warner B, Safronetz D, Ranadheera C. Crimean-Congo haemorrhagic fever virus: Past, present and future insights for animal modelling and medical countermeasures. *Zoonoses Public Health*. 2018; 65(5): 465-480.
[22] de la Calle-Prieto F, Martín-Quirós A, Trigo E, et al. Therapeutic management of Crimean-Congo haemorrhagic fever. *Enferm Infecc Microbiol Clin*. 2018; 36(8): 517-522.
[23] Erduran E, Bahadir A, Palanci N, Gedik Y. The treatment of crimean-congo hemorrhagic fever with high-dose methylprednisolone, intravenous immunoglobulin, and fresh frozen plasma. *J Pediatr Hematol Oncol*. 2013; 35(1): e19-24.
[24] Ergonul O. Treatment of Crimean-Congo hemorrhagic fever. *Antiviral Res*. 2008; 78(1): 125-131.
[25] Ergönül O. Crimean-Congo haemorrhagic fever. *Lancet Infect Dis*. 2006; 6(4): 203-14.
[26] Ascioglu S, Leblebicioglu H, Vahaboglu H, Chan KA. Ribavirin for patients with Crimean-Congo haemorrhagic fever: a systematic review and meta-analysis. *J Antimicrob Chemother*. 2011; 66(6): 1215-1222.
[27] Johnson S, Henschke N, Maayan N, Mills I, Buckley BS, Kakourou A, et al. Ribavirin for treating Crimean Congo haemorrhagic fever. *Cochrane Database Syst Rev*. 2018; 6(6): CD012713.
[28] Ergönül Ö, Keske Ş, Çeldir MG, et al. Systematic Review and Meta-analysis of Postexposure Prophylaxis for Crimean-Congo Hemorrhagic Fever Virus among Healthcare Workers. *Emerg Infect Dis*. 2018; 24(9): 1642-1648.

[29] Arab-Bafrani Z, Jabbari A, Mostakhdem Hashemi M, Arabzadeh AM, Gilanipour A, Mousavi E. Identification of the crucial parameters regarding the efficacy of ribavirin therapy in Crimean-Congo haemorrhagic fever (CCHF) patients: a systematic review and meta-analysis. *J Antimicrob Chemother*. 2019; 74(12): 3432-3439.
[30] Sharifi-Mood B, Alavi-Naini R, Metanat M, Mohammadi M, Shakeri A, Amjadi A. Efficacy of high-dose methylprednisolone in patients with Crimean-Congo haemorrhagic fever and severe thrombocytopenia. *Trop Doct*. 2013; 43(2): 49-53.
[31] Milano A, Robbiati C, Declich S, et al. Assessing the Adoption of One Health Approaches in National Plans to Combat Health Threats: The Pilot of a One Health Conceptual Framework in Armenia. *Trop Med Infect Dis*. 2024; 9(1): 22.
[32] Crimean-Congo Hemorrhagic Fever Virus for Clinicians-Diagnosis, Clinical Management, and Therapeutics [Internet]. Available at: https://wwwnc.cdc.gov/eid/syn/en/article/30/5/23-1648.htm [accessed 29th Sept 2024].
[33] Who Clinical Management of patients with viral haemorragic fever [Internet]. Available at: https://iris.who.int/bitstream/handle/10665/205570/9789241549608_eng.pdf?sequence=1 [accessed 29th Sept 2024].
[34] Sidwell RW, Huffman JH, Khare GP, Allen LB, Witkowski JT, Robins RK. Broad-spectrum antiviral activity of Virazole: 1-beta-D-ribofuranosyl-1,2,4-triazole-3-carboxamide. *Science*. 1972; 177(4050): 705-706.
[35] Fernandez H, Banks G, Smith R. Ribavirin: a clinical overview. *Eur J Epidemiol.* 1986; 2(1): 1-14.
[36] Dietz J, Schelhorn SE, Fitting D, et al. Deep sequencing reveals mutagenic effects of ribavirin during monotherapy of hepatitis C virus genotype 1-infected patients. *J Virol*. 2013; 87(11): 6172-6181.
[37] Koksal I, Yilmaz G, Aksoy F, et al. The efficacy of ribavirin in the treatment of Crimean-Congo hemorrhagic fever in Eastern Black Sea region in Turkey. *J Clin Virol*. 2010; 47(1): 65-68.
[38] van de Wal BW, Joubert JR, van Eeden PJ, King JB. A nosocomial outbreak of Crimean-Congo haemorrhagic fever at Tygerberg Hospital. Part IV. Preventive and prophylactic measures. *S Afr Med J*. 1985; 68(10): 729-7232.
[39] Fisher-Hoch SP, McCormick JB, Swanepoel R, Van Middlekoop A, Harvey S, Kustner HG. Risk of human infections with Crimean-Congo hemorrhagic fever virus in a South African rural community. *Am J Trop Med Hyg*. 1992; 47(3): 337-345.
[40] Mardani M, Jahromi MK, Naieni KH, Zeinali M. The efficacy of oral ribavirin in the treatment of crimean-congo hemorrhagic fever in Iran. *Clin Infect Dis*. 2003; 36(12): 1613-8.
[41] Ergönül O, Celikbaş A, Dokuzoguz B, Eren S, Baykam N, Esener H. Characteristics of patients with Crimean-Congo hemorrhagic fever in a recent outbreak in Turkey and impact of oral ribavirin therapy. *Clin Infect Dis*. 2004; 39(2): 284-287.
[42] D'Addiego J, Elaldi N, Wand N, et al. Investigating the effect of ribavirin treatment on genetic mutations in Crimean-Congo haemorrhagic fever virus (CCHFV) through next-generation sequencing. *J Med Virol*. 2023; 95(2): e28548.
[43] ClinicalTrials.gov. Intravenous Ribavirin Protocol to Treat Individuals With Viral Hemorrhagic Fever. Available at: https://clinicaltrials.gov/study/NCT02483260?cond=Crimean-Congo%20Hemorrhagic%20Fever&intr=Ribavirin&rank=2 [accessed 29th Sept 2024].
[44] Furuta Y, Gowen BB, Takahashi K, Shiraki K, Smee DF, Barnard DL. Favipiravir (T-705), a novel viral RNA polymerase inhibitor. *Antiviral Res*. 2013; 100(2): 446-454.
[45] Oestereich L, Rieger T, Neumann M, et al. Evaluation of antiviral efficacy of ribavirin, arbidol, and T-705 (favipiravir) in a mouse model for Crimean-Congo hemorrhagic fever. *PLoS Negl Trop Dis*. 2014; 8(5): e2804.
[46] Hawman DW, Haddock E, Meade-White K, et al. Favipiravir (T-705) but not ribavirin is effective against two distinct strains of Crimean-Congo hemorrhagic fever virus in mice. *Antiviral Res*. 2018; 157: 18-26.
[47] ClinicalTrials.gov. UMIT-1 Trial Favipiravir & Ribavirin for the Treatment of CCHF (UMIT-1) Available at: https://clinicaltrials.gov/study/NCT05940545?tab=table [accessed 29th Sept 2024].
[48] Scholte FEM, Zivcec M, Dzimianski JV, et al. Crimean-Congo Hemorrhagic Fever Virus Suppresses Innate Immune Responses via a Ubiquitin and ISG15 Specific Protease. *Cell Rep*. 2017; 20(10): 2396-2407.
[49] Kocabas F, Aslan GS. Fluorometric CCHFV OTU protease assay with potent inhibitors. *Virus Genes*. 2015; 51(2): 190-197.
[50] Dai S, Deng F, Wang H, Ning Y. Crimean-Congo Hemorrhagic Fever Virus: Current Advances and Future Prospects of Antiviral Strategies. *Viruses*. 2021; 13(7): 1195.
[51] Scholte FEM, Hua BL, Spengler JR, et al. Stable Occupancy of the Crimean-Congo Hemorrhagic Fever Virus-Encoded Deubiquitinase Blocks Viral Infection. *MBio*. 2019; 10(4): e01065-19.
[52] Welch SR, Scholte FEM, Flint M, et al. Identification of 2'-deoxy-2'-fluorocytidine as a potent inhibitor of Crimean-Congo hemorrhagic fever virus replication using a recombinant fluorescent reporter virus. *Antiviral Res*. 2017; 147: 91-99.
[53] Smee DF, Jung KH, Westover J, Gowen BB. 2'-Fluoro-2'-deoxycytidine is a broad-spectrum inhibitor of bunyaviruses in vitro and in phleboviral disease mouse models. *Antiviral Res*. 2018; 160: 48-54.
[54] Wang Q, Cao R, Li L, et al. In vitro and in vivo efficacy of a novel nucleoside analog H44 against Crimean-Congo hemorrhagic fever virus. *Antiviral Res*. 2022; 199: 105273.
[55] Garrison AR, Radoshitzky SR, Kota KP, et al. Crimean-Congo hemorrhagic fever virus utilizes a clathrin-

and early endosome-dependent entry pathway. *Virology*. 2013; 444(1-2): 45-54.
[56] Chumakov, M.P., 1945. A new tick-borne virus disease-Crimean hemorrhagic fever (acute infectious capillary toxicosis). In: Sokolov, A.E., Chumakov, M.P., Kolachev, A.A. (Eds.), Crimean Hemorrhagic Fever (Acute Infectious Capillary Toxicosis). Izdanie Otdel'noj Primorskoj Armii, Simferopol. USSR [Russian]. 13-43.
[57] Keshtkar-Jahromi M, Kuhn JH, Christova I, Bradfute SB, Jahrling PB, Bavari S. Crimean-Congo hemorrhagic fever: current and future prospects of vaccines and therapies. *Antiviral Res*. 2011; 90(2): 85-92.
[58] Enria DA, Briggiler AM, Fernandez NJ, Levis SC, Maiztegui JI. Importance of dose of neutralising antibodies in treatment of Argentine haemorrhagic fever with immune plasma. *Lancet*. 1984; 2(8397): 255-256.
[59] Hoogstraal H. The epidemiology of tick-borne Crimean-Congo hemorrhagic fever in Asia, Europe, and Africa. *J Med Entomol*. 1979; 15(4): 307-417.
[60] Ergonul O. Treatment of Crimean-Congo hemorrhagic fever. *Ant*iviral Res. 2008; 78(1): 125-131.
[61] Lazarev, V.N., 1969. Treatment of Crimean hemorrhagic fever patients with conva lescent sera. In: Chumakov, M.P. (Ed.), Arboviruses (Tick-borne and Japanese Encephalitides, Hemorrhagic Fevers, and Other Arboviral Infections). Materi als of the 16th Scientific Session of the Institute of Poliomyelitis and Viral Encephalitides, vol. 2. Academy of Medical Sciences, Moscow, USSR [Russian]. 142-143.
[62] van Eeden PJ, van Eeden SF, Joubert JR, King JB, van de Wal BW, Michell WL. A nosocomial outbreak of Crimean-Congo haemorrhagic fever at Tygerberg Hospital. Part II. Management of patients. *S Afr Med J*. 1985; 68(10): 718-721.
[63] Keshtkar-Jahromi M, Kuhn JH, Christova I, Bradfute SB, Jahrling PB, Bavari S. Crimean-Congo hemorrhagic fever: current and future prospects of vaccines and therapies. *Antiviral Res*. 2011; 90(2): 85-92.
[64] Erduran E, Bahadir A, Palanci N, Gedik Y. The treatment of crimean-congo hemorrhagic fever with high-dose methylprednisolone, intravenous immunoglobulin, and fresh frozen plasma. *J Pediatr Hematol Oncol*. 2013; 35(1): e19-24.
[65] Kubar A, Haciomeroglu M, Ozkul A, et al. Prompt administration of Crimean-Congo hemorrhagic fever (CCHF) virus hyperimmunoglobulin in patients diagnosed with CCHF and viral load monitorization by reverse transcriptase-PCR. *Jpn J Infect Dis*. 2011; 64(5): 439-43.
[66] Consultation on Crimean-Congo haemorragic fever prevention and control. Available at: https://www.ecdc.europa.eu/sites/default/files/media/en/publications/Publications/0809_MER_Crimean_Congo_Haemorragic_Fever_Prevention_and_Control.pdf [accessed 29th Sept 2024]
[67] Otsubo R, Yasui T. Monoclonal antibody therapeutics for infectious diseases: Beyond normal human immunoglobulin. *Pharmacol Ther*. 2022; 240: 108233.
[68] Ergonul O, Celikbas A, Baykam N, Eren S, Dokuzoguz B. Analysis of risk-factors among patients with Crimean-Congo haemorrhagic fever virus infection: severity criteria revisited. *Clin Microbiol Infect.* 2006; 12(6): 551-554.
[69] Kaya S, Elaldi N, Kubar A, et al. Sequential determination of serum viral titers, virus-specific IgG antibodies, and TNF-α, IL-6, IL-10, and IFN-γ levels in patients with Crimean-Congo hemorrhagic fever. *BMC Infect Dis*. 2014; 14: 416.
[70] Gholizadeh O, Jafari MM, Zoobinparan R, et al. Recent advances in treatment Crimean-Congo hemorrhagic fever virus: A concise overview. *Microb Pathog.* 2022; 169: 105657.
[71] Golden JW, Shoemaker CJ, Lindquist ME, et al. GP38-targeting monoclonal antibodies protect adult mice against lethal Crimean-Congo hemorrhagic fever virus infection. *Sci Adv*. 2019; 5(7): eaaw9535.
[72] Zivcec M, Guerrero LIW, Albariño CG, Bergeron É, Nichol ST, Spiropoulou CF. Identification of broadly neutralizing monoclonal antibodies against Crimean-Congo hemorrhagic fever virus. *Antiviral Res*. 2017; 146: 112-20.
[73] Fels JM, Maurer DP, Herbert AS, et al. Protective neutralizing antibodies from human survivors of Crimean-Congo hemorrhagic fever. *Cell*. 2021; 184(13): 3486-501.e21.
[74] Durie IA, Tehrani ZR, Karaaslan E, et al. Structural characterization of protective non-neutralizing antibodies targeting Crimean-Congo hemorrhagic fever virus. *Nat Commun*. 2022; 13(1): 1-12.
[75] Ahata B, Akçapınar GB. CCHFV vaccine development, current challenges, limitations, and future directions. *Front Immunol*. 2023; 14: 1238882.
[76] Ozdarendeli A. Crimean-Congo Hemorrhagic Fever Virus: Progress in Vaccine Development. *Diagnostics*. 2023; 13(16): 2708.
[77] Papa A, Papadimitriou E, Christova I. The Bulgarian vaccine Crimean-Congo haemorrhagic fever virus strain. *Scand J Infect Dis.* 2011; 43(3): 225-229.
[78] Tipih T, Burt FJ. Crimean-Congo Hemorrhagic Fever Virus: Advances in Vaccine Development. *Biores Open Access*. 2020; 9(1): 137-150.
[79] Pavel STI, Yetiskin H, Kalkan A, Ozdarendeli A. Evaluation of the cell culture based and the mouse brain derived inactivated vaccines against Crimean-Congo hemorrhagic fever virus in transiently immune-suppressed (IS) mouse model. *PLoS Negl Trop Dis*. 2020; 14(11): e0008834.
[80] Berber E, Çanakoğlu N, Tonbak Ş, Ozdarendeli A. Development of a protective inactivated vaccine against Crimean-Congo hemorrhagic fever infection. *Heliyon*. 2021; 7(10): e08161.
[81] Ewer KJ, Lambe T, Rollier CS, Spencer AJ, Hill AV, Dorrell L. Viral vectors as vaccine platforms: from immunogenicity to impact. *Curr Opin Immunol*. 2016; 41: 47-54.

[82] Aligholipour Farzani T, Földes K, Ergünay K, Gurdal H, Bastug A, Ozkul A. Immunological Analysis of a CCHFV mRNA Vaccine Candidate in Mouse Models. *Vaccines*. 2019; 7(3): 115.
[83] Buttigieg KR, Dowall SD, Findlay-Wilson S, et al. A novel vaccine against Crimean-Congo Haemorrhagic Fever protects 100% of animals against lethal challenge in a mouse model. *PLoS One*. 2014; 9(3): e91516.
[84] Saunders JE, Gilbride C, Dowall S, et al. Adenoviral vectored vaccination protects against Crimean-Congo Haemorrhagic Fever disease in a lethal challenge model. *EBioMedicine*. 2023; 90: 104523.
[85] Rodriguez SE, Cross RW, Fenton KA, Bente DA, Mire CE, Geisbert TW. Vesicular Stomatitis Virus-Based Vaccine Protects Mice against Crimean-Congo Hemorrhagic Fever. *Sci Rep*. 2019; 9(1): 7755.
[86] Dowall SD, Carroll MW, Hewson R. Development of vaccines against Crimean-Congo haemorrhagic fever virus. *Vaccine*. 2017; 35(44): 6015-6023.
[87] Kortekaas J, Vloet RPM, McAuley AJ, et al. Crimean-Congo Hemorrhagic Fever Virus Subunit Vaccines Induce High Levels of Neutralizing Antibodies But No Protection in STAT1 Knockout Mice. *Vector Borne Zoonotic Dis*. 2015; 15(12): 759-764.
[88] Ghiasi SM, Salmanian AH, Chinikar S, Zakeri S. Mice orally immunized with a transgenic plant expressing the glycoprotein of Crimean-Congo hemorrhagic fever virus. *Clin Vaccine Immunol*. 2011; 18(12): 2031-2037.
[89] Zhang G, Wang P, Jiang L, et al. Evaluation of the immunogenicity of a Crimean-Congo hemorrhagic fever virus vaccine candidate in mice developed based on a baculovirus Zera nanoparticle delivery system. *Front Vet Sci*. 2023; 10: 1126785.
[90] Scholte FEM, Karaaslan E, O'Neal TJ, et al. Vaccination with the Crimean-Congo hemorrhagic fever virus viral replicon vaccine induces NP-based T-cell activation and antibodies possessing Fc-mediated effector functions. *Front Cell Infect Microbiol*. 2023; 13: 1233148.
[91] Spengler JR, Welch SR, Scholte FEM, et al. Heterologous protection against Crimean-Congo hemorrhagic fever in mice after a single dose of replicon particle vaccine. *Antiviral Res*. 2019; 170: 104573.
[92] Spik K, Shurtleff A, McElroy AK, Guttieri MC, Hooper JW, SchmalJohn C. Immunogenicity of combination DNA vaccines for Rift Valley fever virus, tick-borne encephalitis virus, Hantaan virus, and Crimean Congo hemorrhagic fever virus. *Vaccine*. 2006; 24(21): 4657-4666.
[93] Suschak JJ, Golden JW, Fitzpatrick CJ, et al. A CCHFV DNA vaccine protects against heterologous challenge and establishes GP38 as immunorelevant in mice. *NPJ Vaccines*. 2021; 6(1): 31.
[94] Golden JW, Fitzpatrick CJ, Suschak JJ, et al. Induced protection from a CCHFV-M DNA vaccine requires CD8 T cells. *Virus Res*. 2023; 334: 199173.
[95] Hu YL, Zhang LQ, Liu XQ, et al. Construction and evaluation of DNA vaccine encoding Crimean Congo hemorrhagic fever virus nucleocapsid protein, glycoprotein N-terminal and C-terminal fused with LAMP1. *Front Cell Infect Microbiol*. 2023; 13: 1121163.
[96] Hinkula J, Devignot S, Åkerström S, et al. Immunization with DNA Plasmids Coding for Crimean-Congo Hemorrhagic Fever Virus Capsid and Envelope Proteins and/or Virus-Like Particles Induces Protection and Survival in Challenged Mice. *J Virol*. 2017; 91(10): e02076-16.
[97] Tipih T, Heise M, Burt FJ. Immunogenicity of a DNA-Based Sindbis Replicon Expressing Crimean-Congo Hemorrhagic Fever Virus Nucleoprotein. *Vaccines*. 2021; 9(12): 1491.
[98] Hawman DW, Ahlén G, Appelberg KS, et al. A DNA-based vaccine protects against Crimean-Congo haemorrhagic fever virus disease in a Cynomolgus macaque model. *Nat Microbiol*. 2021; 6(2): 187-195.
[99] Hawman DW, Meade-White K, Leventhal S, et al. Accelerated DNA vaccine regimen provides protection against Crimean-Congo hemorrhagic fever virus challenge in a macaque model. *Mol Ther*. 2023; 31(2): 387-397.
[100] Appelberg S, John L, Pardi N, et al. Nucleoside-Modified mRNA Vaccines Protect IFNAR Mice against Crimean-Congo Hemorrhagic Fever Virus Infection. *J Virol*. 2022; 96(3): e0156821.
[101] Leventhal SS, Meade-White K, Rao D, et al. Replicating RNA vaccination elicits an unexpected immune response that efficiently protects mice against lethal Crimean-Congo hemorrhagic fever virus challenge. *EBioMedicine*. 2022; 82: 104188.
[102] Omoniyi AA, Adebisi SS, Musa SA, et al. In silico design and analyses of a multi-epitope vaccine against Crimean-Congo hemorrhagic fever virus through reverse vaccinology and immunoinformatics approaches. *Sci Rep*. 2022; 12(1): 8736.
[103] Imran MA, Islam MR, Saha A, Ferdousee S, Mishu MA, Ghosh A. Development of Multi-epitope Based Subunit Vaccine Against Crimean-Congo Hemorrhagic Fever Virus Using Reverse Vaccinology Approach. *Int J Pept Res Ther*. 2022; 28(4): 124.
[104] Mears MC, Bente DA. Design of a Crimean-Congo Hemorrhagic Fever Virus Glycoprotein Multi-Epitope Antigen for Vaccine Development. *Zoonoses*. 2022; 2(1): 34.
[105] Shah SZ, Jabbar B, Mirza MU, et al. An Immunoinformatics Approach to Design a Potent Multi-Epitope Vaccine against Asia-1 Genotype of Crimean-Congo Haemorrhagic Fever Virus Using the Structural Glycoproteins as a Target. *Vaccines*. 2022; 11(1): 61.
[106] ClinicalTrials.gov. Phase I Study to Evaluate Basic Pharmacodynamic, Pharmacological and Toxicological Effects of the Newly Developed Crimean-Congo Hemorrhagic Fever Vaccine for Humans. Available from: https://clinicaltrials.gov/study/NCT03020771 [accessed 29th Sept 2024].