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EDITORIAL COMMENTARY

Experimental Therapies for Ebola Virus Disease: What Have We Learned?

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(See the brief report Dörnemann et al on pages 171–4.)

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The terrible mortality rate of Ebola virus (EBOV) disease (EVD) is most pronounced in the vulnerable groups of pregnant women and neonates. During the 2013–2016 West African outbreak, hundreds of EBOV-infected pregnant women were reported, with maternal mortality rate estimated at >70% and neonatal mortality rate nearly 100%. Thus, Dornemann et al's [1] interesting case report in this issue of *The Journal of Infectious Diseases*, describing an EBOV-infected neonate who not only survived but had no apparent sequelae at 8 months of age, represents a first. The surviving baby, one of the last cases of EVD seen in Guinea, gives hope that perhaps we are finally turning the corner in finding effective treatments for this disease. Of course, one case does not constitute scientific proof of effectiveness, and it remains possible that she is simply a very fortunate outlier. The report is not only instructive in itself, but also raises a number of points with respect to clinical management and assessment of investigational EVD therapeutics.

In addition to aggressive supportive care, the baby received 3 experimental therapies for EVD—ZMapp (a cocktail of 3 human-mouse chimeric anti-EBOV monoclonal antibodies), a buffy coat infusion, and GS-5734, the prodrug of a nucleoside viral RNA polymerase inhibitor. In addition, the fetus may have also been exposed in utero to the RNA polymerase inhibitor favipiravir, which the mother received for 3 days before delivery and her subsequent death. Thus, like most patients with EVD who received care in hospitals in the United States and Europe [2], this infant received several investigational therapeutic agents, making it very difficult to determine the impact of any particular one.

Mortality in EVD correlates directly with blood viral load [3–5], usually reflected in the field by the cycle threshold (Ct) noted on quantitative reverse-transcription polymerase chain reaction (RT-PCR), the most readily available diagnostic technique. The Ct varies inversely with the viral RNA load [5]. Initial review of the Ct profile of the child's case indicates that the 4 ZMapp infusions were insufficient to clear EBOV RNA from her blood, although it is possible that their administration controlled viral replication sufficiently to enable the child's immune system to respond and ultimately eliminate the virus. After an initial increase in Ct value following the first ZMapp dose, it fell again, reflecting an increase in viral load. One consideration is the emergence of ZMapp-resistant variants, as observed in EBOV-infected nonhuman primates (NHPs) given another antibody cocktail, MB-003 [6]. The eventual clearance of EBOV RNA from blood occurred well after the last ZMapp plus buffy coat infusions and likely before use of GS-5734. Unfortunately, the absence of serologic data from EBOV-specific enzyme-linked immunosorbent or neutralizing antibody assays during the patient's acute course or later during convalescence does not allow conclusions regarding her adaptive immune responses.

But just as it is impossible to attribute the child's survival to the experimental therapies, we should not discount their potential influence. As noted above, survival of an EBOV-infected neonate is extremely rare. We might be further encouraged by the 18.5% mortality rate seen in patients with EVD who received care in hospitals in the United States and Europe, 85% of whom also received 1 or more experimental therapies [2], compared with case-fatality rates ranging from 31% to 76% for patients treated in West Africa without access to these experimental therapies [7]. Of course, all of the aforementioned patients received a level of aggressive supportive care, including close attention to fluid and electrolyte balance, that was not available to the vast majority of patients with EVD in West Africa. But even if we conclude a causal benefit from these collective interventions, an 18.5% case-fatality rate is still unacceptably high. Antiviral agents with greater potency, likely achieved through use of combination regimens to rapidly control virus replication, together with host-response-modifying agents to mitigate the consequences of infection, will likely be needed.

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Although we need more conclusive efficacy data on the various experimental therapeutics for EVD, the progress made in their assessment during the outbreak in West Africa should not be underestimated. Excluding a handful of patients who received convalescent whole blood in 1995 [8], the West Africa outbreak—nearly 40 years after the discovery of EBOV in 1976—was the first time that any experimental therapy has been administered in EBOV-infected humans, either on a compassionate use basis or in the context of a clinical trial. Based primarily on industrialized countries' concerns regarding the use of EBOV as a bioweapon, a foundation of preclinical research on EVD therapeutics and vaccines has been built over the last few decades [9]. Nevertheless, faced with little economic incentive and the daunting logistics of sporadic EVD outbreaks in remote locations, even the most promising products did not proceed from preclinical testing to evaluation in clinical trials prior to the West African outbreak.

The magnitude and urgency of the outbreak finally provided both a moral imperative and potential opportunity for testing experimental therapies. In August 2014, as EVD case counts in West Africa skyrocketed, the World Health Organization (WHO) convened a meeting of medical ethicists to address the key question of whether use of experimental interventions, which had varied safety and preclinical efficacy profiles, was ethical given the extreme suffering in West Africa, to which the committee unanimously responded in the affirmative [10]. A September 2014 WHO meeting in Geneva brought together diverse stakeholders, including representatives from the ministries of health, pharmaceutical companies, drug regulatory agencies, nongovernmental organizations providing clinical care, and experts in virology, anthropology, and medical ethics, to consider options for studying vaccines and therapeutics [11]. WHO also created a Scientific and Technical Advisory Committee for Ebola Experimental Interventions to help guide the process. One of the first objectives of the Committee was to identify the most promising therapeutics among a long list of proposed candidates, including many of dubious plausibility. This process required consideration of not only the evidence for safety and efficacy, but also the anticipated feasibility of potential use and conducting a clinical trial under the conditions on the ground and the limited production capacities or intermittent drug availability for some candidates [12]. There was rigorous and sometimes contentious debate around acceptable study designs for EVD therapeutics, but eventually several novel approaches, including adaptive trials [13] and sequential, multistage trials [14], were successfully implemented (Table 1). These trials faced numerous challenges, including relatively long delays in their initiation, which meant that some only started late in the outbreak and were unable to include enough patients for adequate statistical power. An opportunity was also missed to enroll more patients in clinical trials in resource-rich settings. Many West African patients presented late at treatment centers, by which time high levels of viral replication and associated organ damage were present, likely reducing the therapeutic value of antiviral interventions.

There were also surprising empirical observations of apparent reduced mortality rates with so-called repurposed drugs, such as artesunate-amodiaquine given to treat possible malaria coinfection, although these findings still need to be confirmed through formal clinical trials [15]. Other agents, like GS-5734, which is effective in controlling viral replication and as salvage therapy in NHPs when initiated up to 3 days after EBOV challenge [16], were not available in time to initiate clinical trials. However, GS-5734 was administered on a compassionate use basis to both the neonate in Guinea [1] and to a nurse with late-onset EBOV meningoencephalitis [17], who also survived.

Several drug candidates progressed through early clinical trials at an unprecedented pace, and the recognition that some agents were ineffective and others are promising (Table 1) provides a starting point for prioritization of future human studies and assessing the predictability of available animal models. Yet there can be no ignoring that, despite enormous effort, we still lack a specific therapeutic option with proven antiviral efficacy or clear clinical benefit for EVD. Many difficult lessons were learned regarding the challenges of inconsistent reproducibility of in vitro experiments, inadequately predictive animal models, and the operational demands of conducting trials in Ebola treatment units during an outbreak centered in countries without preexisting research infrastructure.

Antivirals should be assessed carefully for various indications in EBOV infection. Virus persistence and recurrent disease at immune-privileged sites like the eye and central nervous system have been described in several survivors, each of whom has received investigational antiviral treatments [17, 18]. More importantly from a public health perspective is the persistence of virus in the semen of male survivors, one of whom was implicated in reigniting an EVD cluster 470 days after his acute infection [19]. Studies to test clearance of virus from semen are urgently needed, and one such trial, using GS-5734, has been launched in Liberia (clinical trials registration NCT02818582). Interventions like favipiravir [20] and high doses of the recombinant vesicular stomatitis virus–vectored EBOV glycoprotein (rVSV-GP) vaccine [21] were also used for postexposure prophylaxis in healthcare workers. However, it remains unclear whether these patients were actually exposed, so no conclusions on protective efficacy can be made; in addition, systemic side effects occurred with the latter intervention. Lower, better-tolerated doses of the rVSV-GP vaccine appeared to reduce the risk of EBOV infection in household contacts, with protection starting about 6 days after administration [22]. In contrast, an effective antiviral could potentially provide immediate protection and/or early treatment, as established for other viral infections, such as human immunodeficiency

Table 1. Registered Clinical Trials Reporting Results of Experimental Therapeutics for Ebola Virus Disease (EVD) During the 2013–2016 West African Outbreak

	Trial Characteristics			Dose Characteristics			
Agent	Name	Sponsor and/or Funder	Design	Route	Regimen	Outcomes/Primary End Point	Comment
ZMapp [23]	PREVAIL II	NIAID	Open-label RCT with adaptive design; comparison to optimized SOC alone (including favipiravir in Guinea)	Intravenous	50 mg/kg within 24 h of enrollment, followed by 2 more doses every third day	Enrollment not met (72 of 200 targeted). Overall mortality by day 28 after EVD onset: 13/35 (37%) in SOC group vs 8/36 (22%) in SOC + ZMapp group. Mortality among those with high virus levels (Ct ≤ 22) at entry: 9/15 (60%) in SOC group vs 7/15 (47%) in SOC + ZMapp group.	No statistically significant survival benefit in patients with EVD, but underpowered. Infusions require 2–12 h and may be associated sometimes with systemic reactions; these can be ameliorated by pretreatment with antihistamines and antipyretics. Specific for Zaire EBOV including Makona strain.
TKM 130803 [24]	RAPIDE-TKM	University of Oxford, UK	Open label, single arm with historical and concurrent controls, as part of a multistage approach	Intravenous	0.3 mg/kg once daily for up to 7 days	Halted after meeting prespecified futility end point (survival to day 14 of ≤55%). Survival at day 14 after EVD onset in 3/12 (25%), after excluding 2 who died <48 h after enrollment. Infusions generally well tolerated, except for 1 possible reaction.	No apparent survival benefit as compared to historical controls, but potential confounding by enrollment of patients with high viral loads and late-stage disease. Dose-limiting systemic infusion reactions (acute cytokine release syndrome) in healthy volunteers. Infused over a minimum of 2 h. EBOV Makona specific.
Favipiravir [5]	JIKI	Institut National de la Sante et de la Recherche Medicale, France	Open label, single arm with historical controls	Oral	6 g on day 1, followed by 2.4 g/day on days 2–10 in divided doses	Completed. Among 99 evaluable adults and adolescents, mortality rate was 20% (95% CI, 11.6%–32.4%) in those with a Ct ≥ 20 and 91% (95% CI, 78.8%–91.1%) in those with a Ct < 20. Viral RNA loads and mortality rates were not significantly different between 31 adults starting favipiravir within <72 h of symptom onset and 68 who started later. No grade 3 or 4 clinical AEs.	Mortality did not significantly differ from the predefined target values of 30% for patients with high Ct values and 85% for patients with low Ct values. Much less active against EBOV than influenza virus in preclinical models. Dose regimen was approximately 2 times higher than that tested in phase 3 trials of uncomplicated influenza but appeared to be generally well tolerated.
Convalescent plasma [25]	Ebola-Tx	Institute of Tropical Medicine, Belgium	Open label, single arm with historical controls	Intravenous	2 transfusions of 200–250 mL of ABO-compatible convalescent plasma, with each plasma unit obtained from a separate donor, given within 2 days of EVD diagnosis; those weighing <45 kg received 2 transfusions of 10 mL/kg body weight	Completed. Among 84 evaluable subjects, the mortality rate from day 3 to day 16 after diagnosis was 31% in the convalescent plasma group and 38% in the control group (odds ratio, 0.88 [95% CI, 0.51–1.51], adjusted for Ct values and age). No serious AEs related to the infusions.	No overall survival benefit as compared to historical controls, but levels of anti-EBOV antibodies were not determined in the plasma units. No survival or antiviral effects seen in EBOV-infected NHPs given convalescent blood with high titers of neutralizing antibodies [26], but hyperimmune globulin effective in NHPs [27]. Transfusion-associated acute lung injury reported in a separate patient with EVD given convalescent plasma [28].
Brincidofovir [29]	RAPIDE-BCV	University of Oxford, UK	Open label, single arm with historical controls, as part of a multistage approach	Oral	200 mg as a loading dose on day 1, followed by 100 mg on days 4, 8, 11, and 15; further adjusted for patients weighing <50 kg	Recruitment halted by manufacturer after 4 patients enrolled. Survival at day 14 after EVD onset in 0/4 patients. No serious or unexpected AEs.	Variable, assay-dependent antiviral activity and selectivity for EBOV in cell culture. Antiviral action linked to brincidofovir's lipid moiety [30]. No survival benefit in murine model studies at nontoxic doses. Unable to be studied in NHPs owing to pharmacokinetic profile.
rIFN-β1a[a]	. . .	Canadian Institutes of Health Research	Open label, single arm, single center with historical controls	Subcutaneous	30 μg (6×10^6 IU) rIFN-β1a daily for up to 10 days	Enrollment of 9 patients. Primary outcome of blood viral load reduction based on Ct values appeared faster than in controls. Mortality rates of 84% among 38 controls and in 33% among rIFN-β1a recipients.	Patients enrolled within 6 days of symptom onset. Analysis to address differences in baseline Ct values revealed that the probability of dying in the untreated group was 1.8 times that in the treated group.

More-detailed information on these and other investigational therapeutics is available from the World Health Organization [12].

Abbreviations: AE, adverse event; CI, confidence interval; Ct, cycle threshold; NHP, nonhuman primate; NIAID, National Institute of Allergy and Infectious Diseases; RCT, randomized controlled trial; rIFN-β1a, recombinant interferon β1a; SOC, standard of care.

[a] Data are from E. Fish, personal communication, 16 September 2016.

virus infection and influenza, and potentially could be combined with vaccine for this purpose.

Much work remains to capitalize on the lessons learned from West Africa and make the accelerated pace of clinical trials during future outbreaks the norm, including prioritizing drug candidates, completing phase 1 pharmacology and safety studies, working out trial designs and protocol details, addressing ethics committee and regulatory reviews, and setting logistical frameworks for rapid operationalization. The WHO R&D Blueprint for Action to Prevent Epidemics [31] is one activity that is currently trying to address these parameters for a range of priority diseases before outbreaks, so that the world will have a set of tools at its disposal to rapidly evaluate experimental interventions. Discussion continues regarding the scientific and ethical merits of the various clinical trial designs used in this outbreak. For acute infections with predictably high lethality, as in EVD in pregnant women and neonates, open-label case series and individual case experiences [17] can be highly informative. We also must not forget the importance of the upstream pipeline, recognizing that the implementation of clinical trials during the West Africa 2013–2016 outbreak was only possible because of years of preclinical research to provide viable candidates for field testing. Ongoing scrutiny of the existing therapeutic landscapes for other high-consequence pathogens, support for clinical research networks to conduct studies in the interevent period, and improved surveillance and diagnostic capacities should help to reduce response times for initiating clinical research in future outbreaks of EVD and other emerging threat pathogens.

Note

Potential conflicts of interest. F. G. H. reports receiving personal fees from the Wellcome Trust and fees from the World Health Organization related to Ebola research outside the submitted work and for being a nonpaid consultant to Medivector for studies on the treatment of influenza with favipiravir, which was also evaluated as treatment for Ebola, and to Gilead Sciences for non-EVD studies on GS-5734. All other authors report no potential conflicts of interest. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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