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EBOLA VIRUS DISEASE – A CLINICAL REVIEW

Muhas C.*, Fanish U., Mufeedha P., Shafna Hussain, Abhila E. P. and Saleena C.

Assist. Professor, Department of Pharmacy Practice, Jamia salafiya Pharmacy College, Pulikkal, Kerala.

***Corresponding Author: Muhas C.**

Assist. Professor, Department of Pharmacy Practice, Jamia salafiya Pharmacy College, Pulikkal, Kerala.

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ABSTRACT

Ebola virus is one of the most virulent pathogens, killing a very high proportion of patients within 5-7 days. Most patients developed high fever, head ache, diarrhea, vomiting and hemorrhagic manifestations. Since Ebola virus has a high mortality rate and is very infectious, it is public health imperative to investigate and improve upon ways to manage out breaks. This article mainly discusses about the etiological factors, pathophysiology, clinical manifestations, diagnostic techniques, management and control of infection. To reduce worldwide panic and possibly to save lives, its urgent for medical researches to propose some alternatives remedies which may have positive effect on this epidemic during in intervening period from now to when effective vaccines become available.

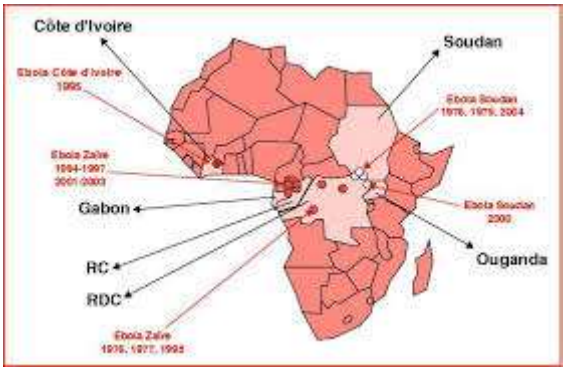
KEYWORDS: Ebola virus, Filoviridae outbreak, Transmission.

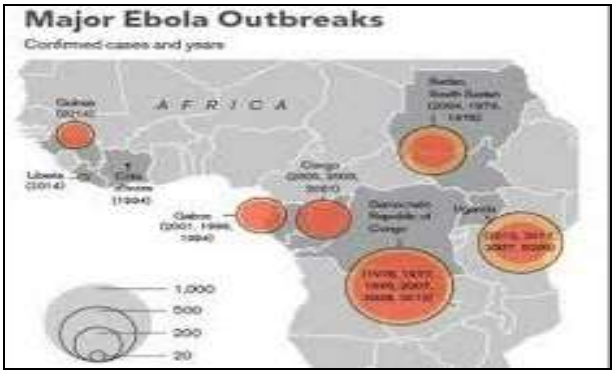
INTRODUCTION

Ebola virus disease is a severe often fatal zoonotic filovirus infection. Ebola virus belongs to the filoviridae family and is subdivided in to four subtypes: Zaire, sudan, Coted' Ivoire, and Reston. Ebola virus is one of the most virulent pathogens, killing a very high proportion of patients within 5-7 days.[1] Most patients developed high fever, head ache, diarrhea, vomiting and hemorrhagic manifestations.[2]

Transmission occurs by close contact with body fluids of infected patients. The incubation period after infection is usually 5-9 days, range of 1-21 days in 95% more of patients[3] and patients are not considered infectious until they develop symptoms. Ebola virus infections is part of a group of diseases know as viral hemorrhagic fevers.[4] Internal and external hemorrhages, the signature symptoms of Ebola, are only seen in the late stages of infection.[5] Because the early symptoms are known specific, Ebola must be diagnosed with specialized laboratory test. First, antigen-capture enzyme-linked immunosorbent assays (ELISA) and immunoglobulin M (IgM) testing use antibodies to detect virus in the blood, then polymerase chain reaction (PCR) amplify viral DNA in blood samples and finally, blood cultures isolate the virus. The presence of immunoglobulin G (IgG), an antibody produced by the body during recovery can show prior infection in survivors.[6]

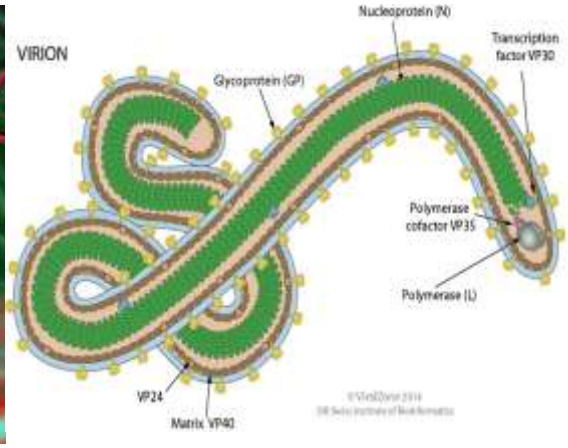
Since Ebola virus has a high mortality rate and is very infectious, it is public health imperative to investigate and improve upon ways to manage out breaks. The out breaks in Uganda and Republic of the Congo, which spanned 2000-2001 and 2001-2002 respectively, were selected for the study because of several specific similarities and differences that make them appropriate for comparison.





Both were the first known outbreak reported in each region, so there would be equally limited community knowledge about the disease and no previous containment policies in place. In contrast, two out breaks occurred in country with vastly different political and social climates. The major difference between the out breaks was the strain of the virus; in the republic of the Congo it was Zaire strain and in Uganda it was Sudan strain. Controls used for expected mortality and spread for each strain were designated by WHO based on previous laboratory and human out breaks that occurred without any official outbreak containment practices in place. This comparison was designed to elucidate the differences in the effects of the two viral strains, as well as effects of social and political environment on identification and containment of the disease, length of outbreak, numbers affected and mortality.[6]

Controlling an EBOV outbreak of this magnitude has proven to be a challenge and the outbreak is predicted to last at least several more months.[7] In the absence of licensed vaccines and therapeutics against EBOV, there is little that can be done for infected patients outside of supportive care, which includes fluid replenishment, administration of antiviral and management of secondary symptoms.[8]



Over past decade, several experimental strategies have shown promise in treating EBOV-challenged non-human primates (NHPs) after infections. These include recombinant human activated protein C(rhAPC),[10] recombinant nematode anticoagulant protein C_2(rNAPC$_2$),[11] small interfering RNA (SiRNA), positively charged-phophorodiamidate morpholino oligomers (PMOplus), vesicular stomatitis virus vaccine (VSV AG-EB OV GP),[12] as well as monoclonal antibody (mAb) cock tails MB-003 and ZMab. Of these, only the antibody- based candidates have demonstrated substantial benefits in NHPs when administered greater than 24 h past EBOV exposure.[13]

The natural host for Ebola virus is unknown, so it has not been possible to implement programs to control or eliminate viral reservoir of transmission to human populations. The rapid progression of Ebola virus infection has further complicated the control of this disease, affording little opportunity to develop acquired immunity.[11] Because of its highly contagious nature and extremely high death rate, WHO has declared this epidemic has an international public health emergency. There is no effective cure currently available for this disease and treatment is palliative. WHO has given a green light to an experimental antibody (Zmapp) and allowed this potential remedy which has not undergone clinical trials, to be used to treat a few select Ebola patients.

To reduce worldwide panic and possibly to save lives, its urgent for medical researches to propose some alternative remedies, which may have positive effects on this epidemic during in intervening period from now to when effective vaccines become available.[14]

Etiology

The virus is thought to initially acquired by exposure to body fluids or tissue from infected animals, such has bats and non-human primates; however, the natural reservoir and mode of transmission to humans has not be confirmed. Animal to human transmission may occur during hunting and consumption of the reservoir specious or infected non-human primates. The practice of butchering or eating bush meat or food contaminated with bat faeces (these specious of tree roosting bats, have been implicated has a reservoir) is also thought to contribute.

Human to human transmission occurs thorough contact with body fluids from infected patients.[15] In early epidemics, the reuse of non-sterile injections was responsible for many healthcare associated transmissions.[16] However, although this remains a risk, most cases result from close physical contact or contact with body fluids (such as sweat, blood, faeces, vomit, saliva, genital secretions, urine and breast milk) of infected patients.

In a study of viral shedding in various body fluids, Ebola virus was isolated from saliva, breast milk, stool, tears,

and semen up to 40 days after onset of illness,[17,18] confirming possibility of delayed sexual transmission. Infection through inhalation is possible in non-human primates but there is no evidence for air borne transmission in humans.[19]

It also transmitted from traveler from affected area and lab scientist. The virus occurs naturally in animal population and can be transmitted to and among human population through direct contact with the body, body fluids or contaminated clothes or inners of infected persons. Human infection has been associated with entering caves or mines or handling bush meat.[20] However, like influenza and SARS, there is some evidence of aerosol transmission of EVD.[21]

Large outbreaks of EHF are usually driven by person to-person transmission, with caregivers both at home and in hospital being at particular risk. Although direct contact with bodily fluids is considered to be the major risk factor, other than confirmation of EBOV in blood during acute illness, few data exist on which specific bodily fluids pose a risk and at what stages of infection. Furthermore, although extreme caution is recommended to prevent environmental contamination and exposure in isolation ward and detailed safety guidelines and protocols for decontamination have been developed, the role of fomites in the transmission of EBOV has not been explored. To better understand the precise modes of transmission, is sampled various clinical specimens from patients as well as from environmental surfaces in an isolation ward for EHF and analyzed them for the presence of EBOV.[22]

REBOV was originally reported in wild-caught cynomolgus monkeys (Macaca fascicularis) imported from the Philippines in to the United States, and has also be reported in pigs both in the Philippines and China as an infection concurrent with porcine reproductive and respiratory syndrome virus (PRRSV), so it was not clear if REBOV was causing any clinical signs.[20]

This infection runs its course within 14-21 days. The virus eventually infects micro vascular endothelial cells and compromises vascular integrity. The terminal stages of Ebola virus infection usually include diffuse bleeding and hypotensive shock.[23]

Signs and Symptoms

There are typically three faces of illness, starting with a few days of nonspecific fever, headache, and myalgia, followed by gastrointestinal phase in which diarrhea and vomiting, abdominal symptoms, and dehydration are prominent. In the second week, the patient may recover or deteriorate with third phase of illness including collapse, neurological manifestation, bleeding, diarrhea and fatigue.

Children presents with similar symptoms to adults: however younger children are reported to have more respiratory (such as cough and dyspnea) and gastrointestinal symptoms but less bleeding and neurological sign than adults.

- Viral hemorrhagic fever (conjunctival infection, purpuric rash, bleeding), hypotension, bradycardia, tachycardia in the later stages and tachypnea.
- Also maculopapular rash, bleeding, hiccups, hepatomegaly, lymphadenopathy, multi organ dysfunction include acute kidney injury, pancreatitis and liver damage, splenomegaly, confusion and seizures.

Pathophysiology

The virus genome consists of a single 19 kb strand of negative sense RNA with seven viral genes that are transcribed by viral RNA dependent RNA polymerase present in the virion. The single strand of RNA is covered by helically arranged viral nucleoproteins NP and VP30, which are linked by matrix proteins. NP24 andVP4 to the lipid bilayer that coats the virion.[25]

Tissue invasion occurs through infected fluid coming in to contact with breaks in the mucosa or skin. This can occur with animal to human or human to human transmission. Monocytes, macrophages, and dendritic cells are the preferred replication site for filoviruses on initial infection. Infected cells migrate to the regional lymph nodes, liver and spleen, there by disseminating the infection. Ebola virus has a wide cell tropism and can infect a variety of cell types. It also has remarkable ability to modulate the expression of genes involved in the host immune response, causing lymphocyte apoptosis and attenuation of protective effects of interferon.[26,27]

The host immune response is crucial and dictates the outcome of infection. Progression to sever diseases occurs when virus triggers expression of a host of pro inflammatory cytokines, including interferon, interleukins(ILs) such as IL-2, IL-6, IL-8 and IL-10; Interferon inducible protein and tumor necrosis factor α(TNF-α). This causes endothelial activation and reduced vascular integrity, release of tissue factor and increased nitric oxide levels.[27]

Thrombocytopenia is most commonly caused by loss of platelets from damaged tissue or more generalized virus induced disseminated intravascular coagulation, where coagulation factors are depleted.[2] Impairment, predisposes the patient to bleeding complications. Other complications of severe disease include acute kidney injury, hepatitis and pancreatitis.[25] An early antibody response, along with reduced lymphocyte depletion is associated with effective viral clearance and survival.[28]

The Ebola virus C_1P is synthesized in a secreted (sGP) or full length transmembrane form, and each gene product has distinct biochemical and biological properties.[29] Preferential binding of Ebola virus GP to the endothelium was demonstrated by use of two independent methodologies as follows: direct binding

was assessed by fluorescence-activated cell sorter analysis and pseudo typing experiment were performed in which virus titers, cell numbers, and confluence were carefully determined so that the multiplicity of infection was controlled and equal in all cell types.[30]

The receptors required for cell binding and infection are not completely understood. A folate related receptor can serve as a cofactor to facilitate infection.[31] but whether it serves as a receptor remains unclear. The cell surface lectn DC-sign can also facilitate GP binding to cells through viral carbohydrate determinants, but it does not appear to mediate entry by itself.[32,33] In contrast to GP, sGP gives rise to a dimeric protein that interact with neutrophils.[29] sGP mediates neutrophil binding, directly or indirectly, through CD 16 b, the neutrophil-specific form of the Fcλ receptor lll.[30] After the initial description of the neutrophil binding of sGP, it was shown that immunoglobulin G (IgG), but not an Fab fragment, against sGP was needed to detect neutrophil binding.

Neutrophils incubated with sGP showed a significant reduction in the CR3-Fcλ R lll B RET signal[34] demonstrating that sGP alters the physical and functional interaction between Fcλ R lll B and CR3. Through this interaction, sGP may contribute to immune evasion by inhibiting early steps in neutrophil activation.

Viral GP plays a key role in the manifestation of Ebola virus infection. The transmembrane form of GP targets the Ebola virus to cells that are relevant to its pathogenesis. Specifically GP allows the virus to introduce its contents into monocytes or macrophages, where cell damaged or exposure to viral particles may cause the release of cytokines[35] associated with inflammation and fever, and into endothelial cells, which damages vascular integrity.[30] Thus sGP may alter the immune response by inhibiting neutrophil activation, while the transmembrane GP may contribute to the hemorrhagic fever symptoms by targeting virus to cells of the reticuloendothelial network and the lining of blood vessels.[23]

Further in vitro analysis have begun to elucidate the molecular mechanisms underlying GP-induced cytotoxicity. Critical mediators of cell adhesion to the matrix and immune signaling (e.g.: - integrin and major histocompatibility complex class l cell surface proteins) are among the cell surface molecules that are dysregulated.[36,37] Transient expression of Ebola virus GP in human kidney 293 T cells caused a reduction of specific integrins (primary molecules responsible for cell adhesion to the extracellular matrix on the cell surface. GP mutants lacking the membrane-spanning region of the ectox domain did not cause this down-regulation, suggesting that anchorage of GP to the cell membrane is required for this effect. Disruption of MHC class l expression on the cell surface is a mechanism for evading host immune responses that is shared by several pathogens, including cytomegalo virus, human immuno deficiency virus (HIV), and herpes viruses.[39] It is not known whether GP effects integrin levels by altering intracellular trafficking or by modulation of protein synthesis or degradation, but suggest a role for cellular protein transport machinery in GP-mediated cytotoxicity. In any event, the biologic effects of GP alone may account largely for the features of Ebola virus infection that lead to fatal disease, including inflammatory dysregulation immune suppression, and loss of vascular integrity.[23]

The most important findings was that acutely ill patients are intensely viremic that ELISA determination of viral antigens in serum provides a sensitive and specific way to quickly screen large numbers of suspect human samples.[9] During acute diseases, there was m-RNA evidence of activation of multiple cytokines.[39] These cytokines have been implicated in the pathogenesis of several forms of shock and cause specific defects in vascular permeability in filovirus infections studied invitro.[40] Another interesting finding in acute- phase in infections of humans and non-human primates[41] was the presence of a circulating soluble glycoprotein, which shares ~300 amino acids with the viral glycoprotein that is produced through transcriptional editing of the same gene.[42] It has been speculated that this protein may serve as same form of immunologic decoy, preventing an effective immune response. There are several other possible immuno suppressive mechanisms including the extensive necrosis of spleen and lymph nodes from fatal human and non-human primate cases.[43]

Extensive infection and co-localized necrosis found in parenchymal cells, macrophages, and endothelial cells of many organs.[44,45] The virology and pathologic findings are important for the way we think about therapy of patients infected with EBO-Z. The pathogenetic hurdle is the extensive nature of infection with a cytopathic virus and the lack of an effective immune response. In fact, the infection and related necrotic lesions are so widespread in fatal cases that it seems, unlikely that supportive care will have much impact on survival unless some form of antiviral of immunologic therapy can be instituted relatively early in disease. The extensive cytokine activation explains some features of the diseases, and it may well be that disseminated intravascular coagulation occurs on the severely affected endothelial cell surfaces, as seen in some animal models, but these are not the driving forces behind the fatal diseases process. Treatment of this phenomena as well as traditional supportive care may be useful in some cases but should not destruct research energies from antiviral drugs, effective passive antibody, or other forms of therapy designed to modify the underlying problem.[9]

Diagnosis[1]

A full physical examination should be undertaken with precautionary isolation procedures and use of PPE. The aim of examination is to exclude a focus for sepsis while

looking for viral hemorrhagic fever (such as conjunctival infection, purpuric rash, or other signs of bleeding).

Vital signs should be taken

- **Fever (≥37.5°C)** – Fever is the presenting symptom in about 90% of patients. Wide variations in body temperature are seen during the course of illness, with normothermia or hypothermia occurring in the later stages of fatal infection.
- **Blood pressure** – Hypotension is the important feature of this disease.
- **Pulse rate** – Bradycardia may be present in the initial stages of illness, whereas tachycardia may be seen in the later stages of fatal infections.
- **Respiratory rate –** Tachypnea, along with tachycardia, correlates with a more severe or advanced infection.

Other possible findings include a maculopapular rash, bleeding, hiccups, hepatomegaly, lymphadenopathy. Hepatitis is common, with aspartate aminotransferase (AST) higher than alanine aminotransferase (ALT), although jaundice is rare and renal dysfunction is common in advanced disease.

Initial investigations: - The main confirmatory test for Ebola virus infection is a positive Ebola RT-PCR. This test should be ordered in all patients with suspected Ebola infection while the patient is isolated. A positive result implies that the patient is potentially infectious, particularly if there is active diarrhea, vomiting, or bleeding. If test result is negative, the test should be repeated within 48 hours because viral load can be low and undetectable early in the illness.

Other investigations: - Nowadays there are many additional investigations that may add valuable information to help further management is available. These include Antigen capture enzyme linked immunosorbent assay (ELISA) testing, full blood count, coagulation studies, renal function tests, arterial blood gases, liver function tests, blood cultures, Ebola specific IgM and IgG antibodies, chest radiography etc.

Management

The main stay of treatment is early recognition of infection, together with effective isolation and best available supportive care in a hospital setting.[46] Failures to provide full supportive care to those with suspected (not confirmed) infection may result in substandard care for these patients who may later be shown to have a treatable disease such as malaria.[47]

Isolation and infection control

Patients identified as being at risk of infection should immediately be isolated in room with private bathroom facilities. All attending health care personnel must wear PPE that conforms with published protocols. All contaminated materials (such as cloths & bed lines) should be treated as potentially infectious.[1]

Fluid and electrolyte replacement

Vomiting and diarrhea are common, so patients are often dehydrated & hypovolemic. In such cases oral rehydration solution can be used. The volume of intravenous fluids needed should be assessed on the basis of clinical examination (level of dehydration, signs of shock) & fluid loss (volume of diarrhea or vomitus or both). Large volume of fluid replacement (> 10L/day) may be needed in febrile patients with diarrhoea.[48] There should be perform electrolyte monitoring daily and high blood lactate values are a reliable measure of hypo perfusion and can help guide fluid resuscitation.[47]

Symptomatic management: The following strategies are recommended:

Fever & pain: - It should be treated with paracetamol first. Opioid analgesics (such as morphine) are preferable for more severe pain. NSAIDs (including aspirin) should be avoided because of the associated increase risk of bleeding & potential for nephrotoxicity.[49]

Nausea & vomiting: - Oral or intravenous antiemetic (such as ondansetron & metoclopramide) are recommended.[49]

Heart burn, dysphagia, and upper abdominal pain: - An antacid or proton pump inhibitor (such as omeprazole) may be beneficial.[49]

Seizures: - Although uncommon, seizures can be seen in advanced disease and a benzodiazepine can be used to abort the seizure and can be given intramuscularly or rectally if intravenous access is unavailable. An anticonvulsant (such as phenobarbital) can be given for repeated seizures.[49]

Agitation: - Although uncommon, agitation can occur in advanced disease. Judicious use of a sedative (such as haloperidol or a benzodiazepine) will help to keep the patient calm.[49]

Sepsis & septic shock: - It should be treated with broad spectrum antibiotics (such as ceftriaxone, piperacillin-tazobactum, or meropenem) in the first hour after sending blood cultures, rapid intravenous fluid resuscitation with assessment of response, appropriate airway management and oxygen administration, & monitoring of urine output preferably by urethral catheterization.[1]

Other emerging treatments

Although experimental treatments for Ebola virus infection are under development, they have not yet been fully tested for safety or effectiveness.[1]

Z Mapp: - The best known emerging treatment so far, Z Mapp, is a combination of 3 humanized monoclonal antibodies targeted at 3 Ebola virus glycoprotein epitopes & is engineered for expression in tobacco patients. It has not yet been tested in humans for safety and efficacy.

TKM – Ebola: - It consists of a combination of small interfering RNAs that target Ebola virus RNA polymerase L, formulated with lipid nanoparticle technology. It has been shown to be protective in non-human primates.[1]

Brincidofovir: - It is currently undergoing phase III trials for the treatment of cytomegalovirus & adenovirus. It also shows activity against Ebola in vitro. This drug has been used in patients with Ebola virus infection in the US under Emergency Investigational New Drug applications approved by the FDA.[1]

Favipiravir: - It selectively inhibits viral RNA dependent RNA polymerase. This drug is approved in Japan for influenza pandemics and is effective against Ebola virus in mouse models.[1]

BCX -4430: - It is an adenosine analogue that is active against Ebola virus in rodents. However, no human's studies have been performed.

Other agents: - interferons have been used in the past. Therapeutic agents used for other diseases, such as amiodarone, clomiphene, and chloroquine inhibit Ebola virus interaction with human cells in models.[1]

Melatonine is a derivative of an essential amino acid, tryptophan. The particular and most important reason for melatonin being potentially useful for EVD is that this molecule seems to directly target all the immuno-inflammatory responsive events associated with the Ebola virus infection.[14]

Vaccines: - Two experimental vaccines are currently undergoing trials.

cAd3-ZEBOV is a chimpanzee derived adenovirus vector with an Ebola virus gene inserted. rVSV- ZEBOV is an attenuated vesticular stomatitis virus without one of its genes replaced by an Ebola virus gene.

Infection controlled measures

Infection control measures for health care workers:

- Wear protective clothing.
- Practice proper infection control and sterilization measures.
- Isolate the suspected patient from each other (if possible) and patients with confirmed disease from those with suspected disease.
- Avoid direct with bodies of people who have died from Ebola, or suspected Ebola. During experiments, avoid direct contact with any dead body.
- Notify health officials if you have direct contact with the body fluids of an infected patients.[24]

Infection control measures for people in affected areas

- Practice care full hygiene (for example, wash hands with soap and water, alcohol based hand sanitizer or diluted chlorine).
- Avoid contact with body fluids.
- Do not handle items that have come in to contact with an infected persons body fluids (such as clothes, medical equipment's and needles).
- Avoid funeral or burial rituals that require handling of the body of someone who has died from proven or suspected Ebola.
- Avoid contact with non-human primates and bats including body fluids or raw meat prepared from these animals.
- If infection is suspected on the basis of the initial screening, immediate isolation is warranted before further investigation. This is crucial to reduce contact with other patients and health care workers while the patient is being investigated. Isolation measures should be continued until the patient has tested negative.[24]

CONCLUSION

The natural host for Ebola virus is unknown, so it has not been possible to implement programs to control or eliminate viral reservoir of transmission to human populations. Extensive infection and co-localized necrosis found in parenchymal cells, macrophages and endothelial cells of many organs. The main stay of treatment is early recognition of infection together with effective isolation and host available supportive care in a hospital setting.

To reduce worldwide panic and possibly to save lives, its urgent for medical researches to propose some alternatives remedies which may have positive effect on this epidemic during in intervening period from now to when effective vaccines become available.

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