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No. 28

医薬品 研究報告 調査報告書

識別番号・報告回数		報告日	第一報入手日	新医薬品等の区分	総合機構処理欄
			2009. 9. 16	該当なし	
一般的名称	解凍人赤血球濃厚液	研究報告の公表状況	Aguilar PV, Camargo W, Vargas J, Guevara C, Roca Y, Felices V, Laguna-Torres VA, Tesh R, Ksiazek TG, Kochel TJ. Emerg Infect Dis. 2009 Sep;15(9):1526-8.	公表国 ボリビア	
販売名(企業名)	解凍赤血球濃厚液「日赤」(日本赤十字社) 照射解凍赤血球濃厚液「日赤」(日本赤十字社) 解凍赤血球-LR「日赤」(日本赤十字社) 照射解凍赤血球-LR「日赤」(日本赤十字社)				

研究報告の概要

○ボリビア出血熱の再興(2007～2008年)
ボリビア出血熱(BHF)は、1959年に、ボリビア東部でのアウトブレイク発生時に初めて報告された。しかし、病原体のマチュポウイルスが死亡患者の脾臓から分離されたのは、1963年であった。1976～1993年は症例が報告されなかったが、1994年にアウトブレイクが起こり、以降、散発症例が観察されていた。
2007年の2月、3月に、BHF疑い症例20例以上(死亡3例)がボリビア北東部ベニの保健当局(SEDES)に報告されていた。2008年2月には、疑い症例200例以上(死亡12例)がSEDESに報告された。
疑い例患者から採取した血清19検体について、間接免疫蛍光法とPCRで検査を行った。アレナウイルスの分離株が5株得られ、遺伝子配列の解析からマチュポウイルスであることが確認された。

使用上の注意記載状況・その他参考事項等

解凍赤血球濃厚液「日赤」
照射解凍赤血球濃厚液「日赤」
解凍赤血球-LR「日赤」
照射解凍赤血球-LR「日赤」

血液を介するウイルス、
細菌、原虫等の感染
vCJD等の伝播のリスク

報告企業の意見	今後の対応
2007年の2月、3月に、ボリビア出血熱の疑い症例20例以上、2008年2月には200例以上が報告され、19例について検査したところ、5例でマチュポウイルスが確認されたとの報告である。	日本赤十字社では、輸血感染症対策として問診時に海外渡航歴の有無を確認し、帰国(入国)後4週間は献血不適としている。また、発熱などの体調不良者を献血不適としている。今後も引き続き、新興・再興感染症の発生状況等に関する情報の収集に努める。

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Table. Characteristics *Mycobacterium bovis* BCG complication cases, Taiwan, 2005–2007*

Patient no.	Sex/age at diagnosis, y	Year reported	Specimen	Diagnosis and site of involvement
1	F/2	2005	Biopsy sample	BCG osteitis/osteomyelitis, right ankle
2	M/1	2005	Bacterial isolate	Subcutaneous abscess, left anterior chest wall
3	M/2	2005	Bacterial isolate	Severe combined immunodeficiency, disseminated BCGitis
4	M/9	2005	Bacterial isolate	Suppurative lymphadenitis
5	F/1	2005	Bacterial isolate	Injection-site abscess
6	M/1	2005	Biopsy sample	Suppurative lymphadenitis
7	M/2	2006	Bacterial isolate	BCG osteitis/osteomyelitis, right distal femoris
8	M/2	2006	Bacterial isolate	BCG osteitis/osteomyelitis
9	F/1	2006	Bacterial isolate	BCG osteitis/osteomyelitis, left distal femoris
10	F/1	2006	Bacterial isolate	BCG osteitis/osteomyelitis, left distal radius
11	F/2	2007	Bacterial isolate	BCG osteitis/osteomyelitis, right knee
12	M/1	2007	Bacterial isolate	Subcutaneous abscess, left wrist
13	M/2	2007	Biopsy sample	BCG osteitis/osteomyelitis, right ankle
14	F/1	2007	Bacterial isolate	Suppurative lymphadenitis
15	M/2	2007	Bacterial isolate	BCG osteitis/osteomyelitis, left proximal tibia

*BCGitis, disseminated BCG infection.

age. In particular, suspected childhood TB patients without an identifiable TB contact and with normal immune status were subjected to further investigations. Multidisciplinary management, including enhanced laboratory diagnosis of atypical bony lesions in infants and children, is recommended for any suspected TB infection. Once BCG-related infection is confirmed, medical treatment has to be consistent.

Acknowledgments

We thank Steve H. S. Kuo, Toru Mori, and Jen Suo for comments and Chen-Che Chiu and Chien-Chung Huang for excellent technical assistance.

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Reemergence of Bolivian Hemorrhagic Fever, 2007–2008

To the Editor: Bolivian hemorrhagic fever (BHF) was first described in 1959 during outbreaks affecting isolated human communities in eastern Bolivia. However, it was not until 1963 that the etiologic agent, Machupo virus, was isolated from the spleen of a patient who died from this disease (*1*). Although no cases were reported between 1976 and 1993, an outbreak occurred in 1994 and sporadic cases have been observed since then.

In February and March 2007, at least 20 suspected BHF cases (3 fatal) were reported to the El Servicio Departamental de Salud (SEDES) in Beni, Bolivia. In February 2007, physicians at the Hospital Santa Maria Magdalena reported 3 male patients (23, 27, and 29 years of age), who worked at a ranch in Magdalena, Itenez Province (13°14′0″S, 64°12′0″W). The patients sought treatment for fever, gingivorrhagia, petechiae, nausea, hematemesis, melena and tremors; clinical laboratory examinations showed thrombocytopenia (<130,000 cells/mm³), leukopenia (<3,900 cells/mm³), and hematuria. Because physicians suspected BHF, patients received supportive therapy, including intravenous hydration, corticoids, antipyretic drugs, antimicrobial drugs, and blood transfusions from donors who had survived Machupo virus infection. Nonetheless, 2 of the patients died 3 and 4 days after admission.

In February 2008, at least 200 suspected new BHF cases (12 fatal) of BHF were reported to SEDES. A febrile hemorrhagic illness developed in a 19-year-old man from Huacaraje, Itenez Province (13°33′S, 63°45′W). On first examination at the Hospital Santa Maria Magdalena, the patient had fever, tremor, gingivorrhagia, petechiae, bruises, asthenia, and anorexia and was admitted with a presumptive diagnosis of BHF. Despite supportive treatment (including administration of plasma from a BHF survivor), his condition worsened; hematemesis, melena, hematochezia, hematuria, anuria, respiratory alkalosis, and metabolic acidosis developed in the patient, eventually resulting in death. A fifth case was detected in a 46-year-old man from San Ramon, Mamore Province (13°17′0″S, 64°43′0″W). A febrile hemorrhagic illness developed in the patient and he was admitted to the Hospital German Busch in Trinidad. The patient recently had been hired as a farm worker. When first seen by the attending physicians, he had fever, thrombocytopenia, leukopenia, petechias, tremors, gingivorrhagia, and dehydration, consistent with symptoms of BHF. The patient received hydration, corticoids, antipyretic therapy, and a plasma transfusion from a BHF survivor. The patient's condition improved and he was subsequently discharged from the hospital ≈10 days after admission.

Nineteen serum samples collected from suspected BHF patients, including the cases described above, were sent to Centro Nacional de Enfermedades Tropicales (Santa Cruz, Bolivia) and the US Naval Medical Research Center Detachment (Lima, Peru) for testing. Serum was injected into Vero and C6/36 cells; 10 days later, the cells were tested for flaviviruses, alphaviruses, and arenaviruses by indirect immunofluorescent assay and PCR. Five arenavirus isolates were obtained from the patients described in this report.

Viral RNA was extracted from the cell culture supernatant and the small (S) segment (≈3,200 bp) was amplified and sequenced. Phylogenetic analyses were conducted using the neighbor-joining and maximum likelihood program implemented in PAUP 4.0 software (Sinauer Associates, Inc., Sunderland, MA, USA). Sequence analyses confirmed the isolates as Machupo virus (Figure). Eight major Machupo phylogenetic lineages were described based on partial sequence of the nucleocapsid protein gene (*2*). We observed a similar tree topology based on the glycoprotein gene sequences (Figure). Two distinct lineages were distinguished among the isolates from the Itenez and Mamore provinces: V and VII and I and II, respectively. The recent isolates (2007–2008) from Magdalena and Huacaraje (Itenez Province) grouped within lineage V whereas the 2008 isolate from San

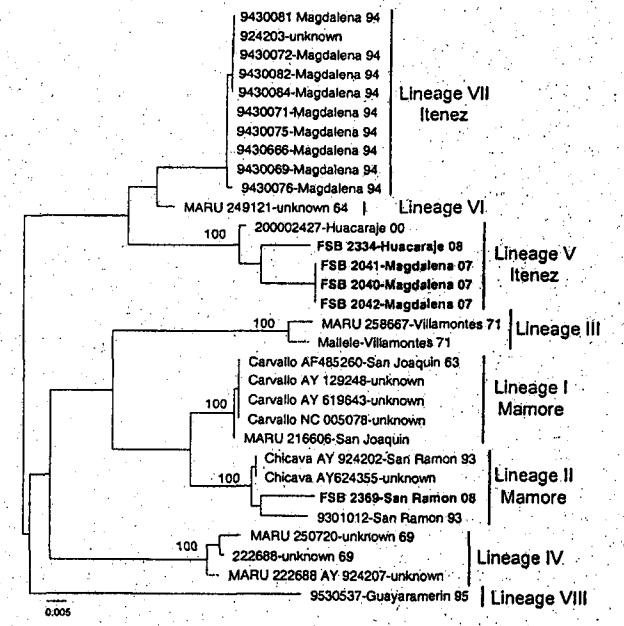

Figure. Neighbor-joining phylogenetic tree of Machupo virus derived from the glycoprotein precursor gene sequence. The neighbor-joining and maximum likelihood analyses yielded similar phylogenetic trees. **Boldface** indicates 2007–2008 isolates. Numbers indicate bootstrap values for 1,000 replicates. Scale bar indicates nucleotide substitutions per site.

Ramon (Mamore Province) belonged to lineage II. These isolates showed 10% nucleotide difference within the S segment and a 6% amino acid difference within the glycoprotein precursor gene. Similar genetic diversity has been described with Machupo virus and other arenaviruses (2–4). Sequences generated were deposited in GenBank (accession nos. FJ696411, FJ696412, FJ696413, FJ696414, and FJ696415).

It is not known whether lineage VII and I viruses continue to circulate or have been replaced by lineage V and II viruses, respectively. This study confirms the long-term maintenance of distinct phylogenetically forms of Machupo virus in a small area within Beni. Although the distribution of the Machupo virus rodent reservoir (*Calomys callosus*) extends beyond the geographic area of the Machupo cases described, factors that limit the endemic distribution of the virus remain unknown. However, population differences among *C. callosus* may account for the natural nidality of BHF (5). Studies are needed to fully identify and understand the ecology of the rodent reservoir and Machupo virus transmission.

Machupo virus continues to cause sporadic cases and focal outbreaks of BHF in Bolivia. We describe 5 confirmed human cases (3 fatal) of Machupo virus infection in Beni Department, Bolivia, an area in which BHF is endemic. That all 5 patients were farmers suggests their infections were probably acquired through occupational exposure. Although all the patients received plasma transfusion from patients who had survived BHF infection, 3 patients still died. An early diagnosis and the rapid administration of Machupo immune plasma before the hemorrhagic phase may increase the chance of survival, as has been observed with other arenavirus infections (6–8).

Acknowledgments

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Relapsing Fever Spirochete in Seabird Tick, Japan

To the Editor: Tick-borne relapsing fever (TBRF) is caused by infection with spirochetes belonging to the genus *Borrelia*. We previously reported a human case of febrile illness suspected to be TBRF on the basis of serologic examination results; the vector most likely was a genus *Carios* tick that had fed on a seabird colony (*1*). However, surveillance of ticks in the area did not identify *Borrelia* spp. in any of the *Carios* ticks sampled (*2*). In 2007 and 2008, a borreliosis investigation was conducted on Kutsujima Island (35.71°N, 135.44°E) because

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研究報告の概要

○慢性疲労症候群患者の血液細胞における感染性レトロウイルスXMRVの検出
慢性疲労症候群(CFS)は原因不明の疾患で、全世界に1700万人の患者がいると推定されている。CFS患者の末梢血単核細胞(PBMCs)を検討することにより、患者101名中68名(67%)、健常者の対照群218名中8名(3.7%)において、ヒトガンマレトロウイルスの一種であるxenotropic murine leukemia virus-related virus(XMRV)のDNAを同定した。細胞培養試験では、患者由来XMRVに感染性があり、細胞結合性感染、無細胞性感染のいずれも起こりうることが判明した。CFS患者由来の活性化PBMCs、B細胞、T細胞、血漿への暴露後に、非感染初代リンパ球と指標細胞株において二次感染が成立した。これらの知見はXMRVがCFSの病原因子である可能性を提起する。

使用上の注意記載状況・その他参考事項等

赤血球濃厚液-LR「日赤」
照射赤血球濃厚液-LR「日赤」

血液を介するウイルス、細菌、原虫等の感染
vCJD等の伝播のリスク

報告企業の意見	今後の対応
慢性疲労症候群(CFS)患者の血液細胞から感染性レトロウイルスXMRVのDNAが検出され、XMRVがCFSの病原因子である可能性が提起されたとの報告である。	今後も引き続き、新たなウイルス等に関する情報の収集に努める。

(12)

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the pathophysiology of chytridiomycosis appears to be disruption to the osmoregulatory functioning of the skin and consequent osmotic imbalance that leads to cardiac standstill.

To test whether treating electrolyte abnormalities would reduce the clinical signs of disease, we administered an oral electrolyte supplement to *L. caerulea* in the terminal stages of infection, when they lost the righting reflex and could no longer correct their body positions (*26*). Frogs under treatment recovered a normal posture and became more active; one individual recovered sufficiently to climb out of the water onto the container walls, and two individuals were able to jump to avoid capture. These signs of recovery were not observed in any untreated frogs. In addition, treated frogs lived >20 hours longer than untreated frogs [mean time after treatment ± SEM: treated frogs (N = 9), 32 ± 2.8 hours; control frogs (N = 6), 10.7 ± 2.2 hours; Student's t test, $P < 0.001$]. All treated frogs continued to shed skin and ultimately died from the infection, as expected. It is unlikely that electrolyte treatment could prevent death unless the epidermal damage caused by *Bd* is reversed. Although amphibians can generally tolerate greater electrolyte fluctuations than other terrestrial vertebrates (*18*), we suggest that depletion of electrolytes, especially potassium, is important in the pathophysiology of chytridiomycosis. Amphibian plasma potassium concentrations are maintained at constant levels across seasons (*27*), and even moderate hypokalemia is dangerous in humans (*28*).

Our results support the epidermal dysfunction hypothesis, which suggests that *Bd* disrupts cutaneous osmoregulatory function, leading to electrolyte imbalance and death. The ability of *Bd* to compromise the epidermis explains how a superficial skin fungus can be fatal to many species of amphibians; their existence depends on the physiological interactions of the skin with the external environment (*16*–*19*). Disease outbreaks capable of causing population declines require the alignment of multiple variables, including a life-compromising pathophysiology (*1*). Resolving the pathogenesis of chytridiomycosis is a key step in understanding this unparalleled pandemic.

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Supporting Online Material

www.sciencemag.org/cgi/content/full/326/5952/582/DC1
Materials and Methods
SOM Text
Figs. S1 and S2
Tables S1 and S2
References

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Detection of an Infectious Retrovirus, XMRV, in Blood Cells of Patients with Chronic Fatigue Syndrome

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Chronic fatigue syndrome (CFS) is a debilitating disease of unknown etiology that is estimated to affect 17 million people worldwide. Studying peripheral blood mononuclear cells (PBMCs) from CFS patients, we identified DNA from a human gammaretrovirus, xenotropic murine leukemia virus–related virus (XMRV), in 68 of 101 patients (67%) as compared to 8 of 218 (3.7%) healthy controls. Cell culture experiments revealed that patient-derived XMRV is infectious and that both cell-associated and cell-free transmission of the virus are possible. Secondary viral infections were established in uninfected primary lymphocytes and indicator cell lines after their exposure to activated PBMCs, B cells, T cells, or plasma derived from CFS patients. These findings raise the possibility that XMRV may be a contributing factor in the pathogenesis of CFS.

Chronic fatigue syndrome (CFS) is a disorder of unknown etiology that affects multiple organ systems in the body. Patients with CFS display abnormalities in immune system function, often including chronic activation of the innate immune system and a deficiency in natural killer cell activity (*1*, *2*). A number of viruses, including ubiquitous herpesviruses and enteroviruses, have been implicated as possible environmental triggers of CFS (*1*). Patients with CFS often have active β herpesvirus infections, suggesting an underlying immune deficiency.

The recent discovery of a gammaretrovirus, xenotropic murine leukemia virus–related virus (XMRV), in the tumor tissue of a subset of prostate cancer patients prompted us to test whether XMRV might be associated with CFS. Both of these disorders, XMRV-positive prostate cancer and CFS, have been linked to alterations in the antiviral enzyme RNase L (*3*–*5*). Using the Whittemore Peterson Institute's (WPI's) national tissue repository, which contains samples from well-characterized cohorts of CFS patients, we isolated nucleic acids from PBMCs and assayed the samples for XMRV *gag* sequences by nested polymerase chain reaction (PCR) (*5*, *6*). Of the 101 CFS samples analyzed, 68 (67%) contained XMRV *gag* sequence. Detection of XMRV was confirmed in 7 of 11 WPI CFS samples at the Cleveland Clinic by PCR-amplifying and sequencing segments of XMRV *env* [352 nucleotides (nt)] and *gag* (736 nt) in CFS PBMC DNA (Fig. 1A) (*6*). In contrast, XMRV *gag* sequences were detected in 8 of 218 (3.7%) PBMC DNA specimens from healthy individuals. Of the 11 healthy control DNA samples analyzed by PCR for both *env* and *gag*, only one sample was positive for *gag* and none for *env* (Fig. 1B). In all positive cases, the XMRV *gag* and *env* sequences were more than 99% similar to those previously reported for prostate tumor–associated strains of XMRV (VP62, VP35, and VP42) (fig. S1) (*5*).

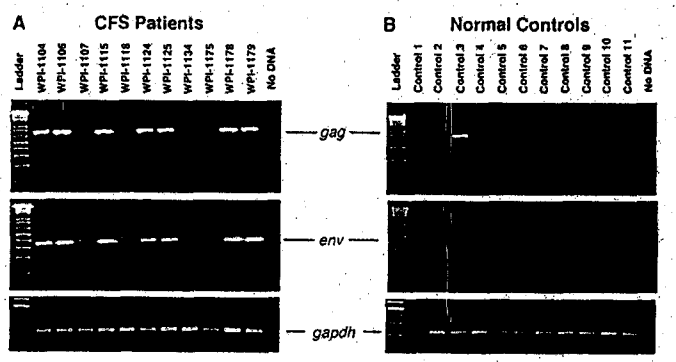

Fig. 1. XMRV sequences in PBMC DNA from CFS patients. Single-round PCR results for *gag*, *env*, and *gapdh* sequences in PBMCs of (**A**) CFS patients and (**B**) healthy controls are shown. The positions of the amplicons are indicated and DNA markers (ladder) are shown. These are representative results from one group of 20 patients.

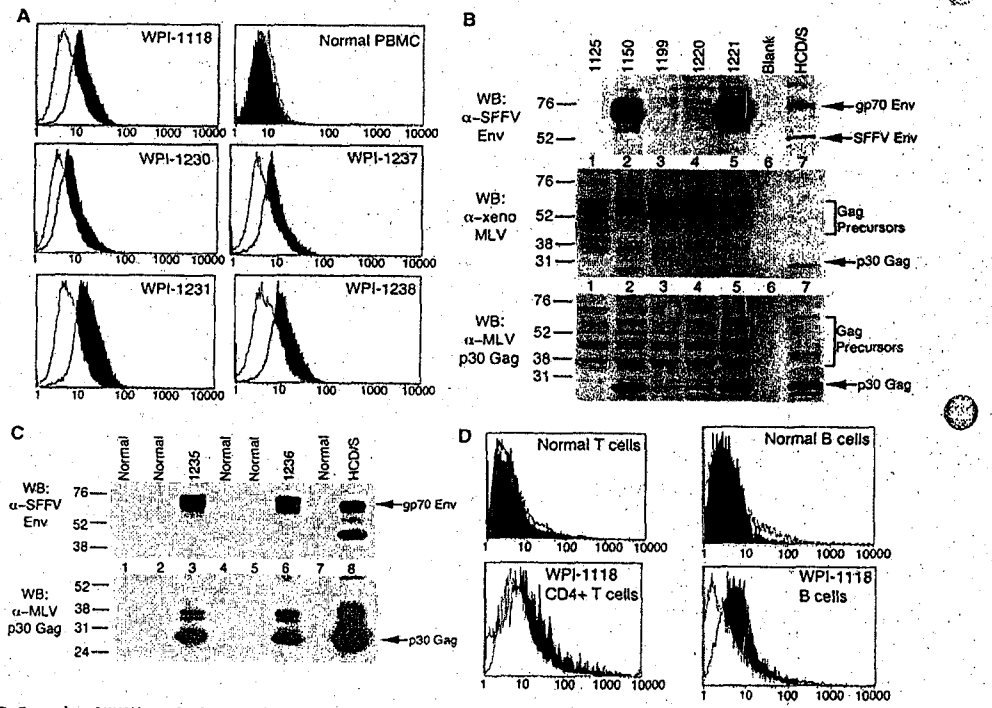

Fig. 2. Expression of XMRV proteins in PBMCs from CFS patients. (**A**) PBMCs were activated with phytohemagglutinin and interleukin-2, reacted with a mAb to MLV p30 Gag, and analyzed by IFC. (**B**) Lysates of activated PBMCs from CFS patients (lanes 1 to 5) were analyzed by Western blots with rat mAb to SFFV Env (top panel), goat antiserum to xenotropic MLV (middle panel), or goat antiserum to MLV p30 Gag (bottom panel). Lane 7, lysate from SFFV-infected HCD-57 cells. Molecular weight markers in kilodaltons are at left. (**C**) Lysates of activated PBMCs from healthy donors (lanes 1, 2, 4, 5, and 7) or from CFS patients (lanes 3 and 6) were analyzed by Western blots using rat mAb to SFFV Env (top panel) or goat antiserum to MLV p30 Gag (bottom panel). Lane 8, SFFV-infected HCD-57 cells. Molecular weight (MW) markers in kilodaltons are at left. (**D**) $CD4^+$ T cells (left) or $CD19^+$ B cells (right) were purified, activated, and examined by flow cytometry for XMRV Gag with a mAb to MLV p30 Gag.

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