



HAL
open science

Variola virus in a 300-year-old Siberian mummy.

Philippe Biagini, Catherine Thèves, Patricia Balaesque, Annie Geraut, Catherine Cannet, Christine Keyser, Dariya Nikolaeva, Patrice Gerard, Sylvie Duchesne, Ludovic Orlando, et al.

► **To cite this version:**

Philippe Biagini, Catherine Thèves, Patricia Balaesque, Annie Geraut, Catherine Cannet, et al.. Variola virus in a 300-year-old Siberian mummy.. *New England Journal of Medicine*, 2012, 367 (21), pp.2057-9. 10.1056/NEJMc1208124 . hal-00967584

HAL Id: hal-00967584

<https://hal.science/hal-00967584v1>

Submitted on 29 Mar 2014

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

different standards for patients on the basis of their socioeconomic status. Although this is a laudable long-term goal, it means that hospitals with a high proportion of Medicaid patients are much more likely to suffer a penalty for excessive readmissions than a hospital with a lower proportion of Medicaid patients. This is incredibly bad social policy, discouraging hospitals from admitting Medicaid patients. Objections to it are not merely theoretical — the published penalties show the results of this decision. The method used also makes it more likely that a large hospital will be hit with a penalty than a small hospital with the same readmission rate after adjustment for case mix. The data shown in Figure 1 support these arguments.

This issue will grow in importance in the next 2 years as the maximum allowable penalty increases from 1% of payments to 2% then 3%. An additional problem with the method is that patients cannot use the results to assess the probability of having a readmission at hospitals of different sizes or with different disproportionate share percentages.

J. Graham Atkinson, D.Phil.

1449 44th St., NW
Washington, DC
jgatkinton@aol.com

Disclosure forms provided by the author are available with the full text of this article at NEJM.org

1. Centers for Medicare and Medicaid Services. Hospital inpatient Prospective Payment Systems for acute care hospitals and

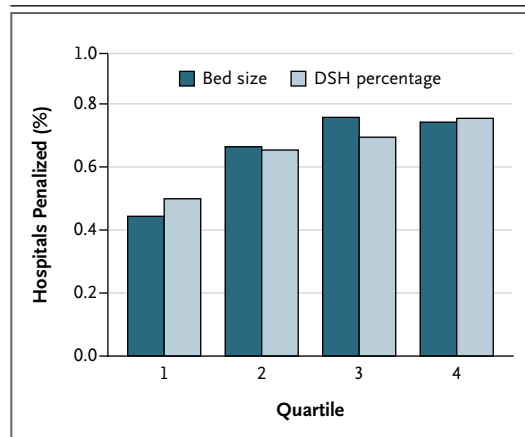


Figure 1. Percentage of Hospitals Penalized, According to Quartile of Bed Size and Disproportionate Share Percentage.

Data are from the Centers for Medicare and Medicaid Services.^{2,3} DSH denotes disproportionate share.

the long term care hospital Prospective Payment System and fiscal year 2013 rates: final rule (<http://cms.gov/Medicare/Medicare-Fee-for-Service-Payment/AcuteInpatientPPS/FY-2013-IPPS-Final-Rule-Home-Page-Items/CMS-1588-F-Text-Version.html>).

2. *Idem.* FY 2013 IPPS final rule: Hospital Readmissions Reduction Program — supplemental data (<http://cms.gov/Medicare/Medicare-Fee-for-Service-Payment/AcuteInpatientPPS/FY-2013-IPPS-Final-Rule-Home-Page-Items/FY2013-Final-Rule-Tables.html>).

3. *Idem.* FY 2013 final rule date files (<http://cms.gov/Medicare/Medicare-Fee-for-Service-Payment/AcuteInpatientPPS/FY-2013-IPPS-Final-Rule-Home-Page-Items/FY2013-Final-Rule-Data-Files.html>).

DOI: 10.1056/NEJMc1212281

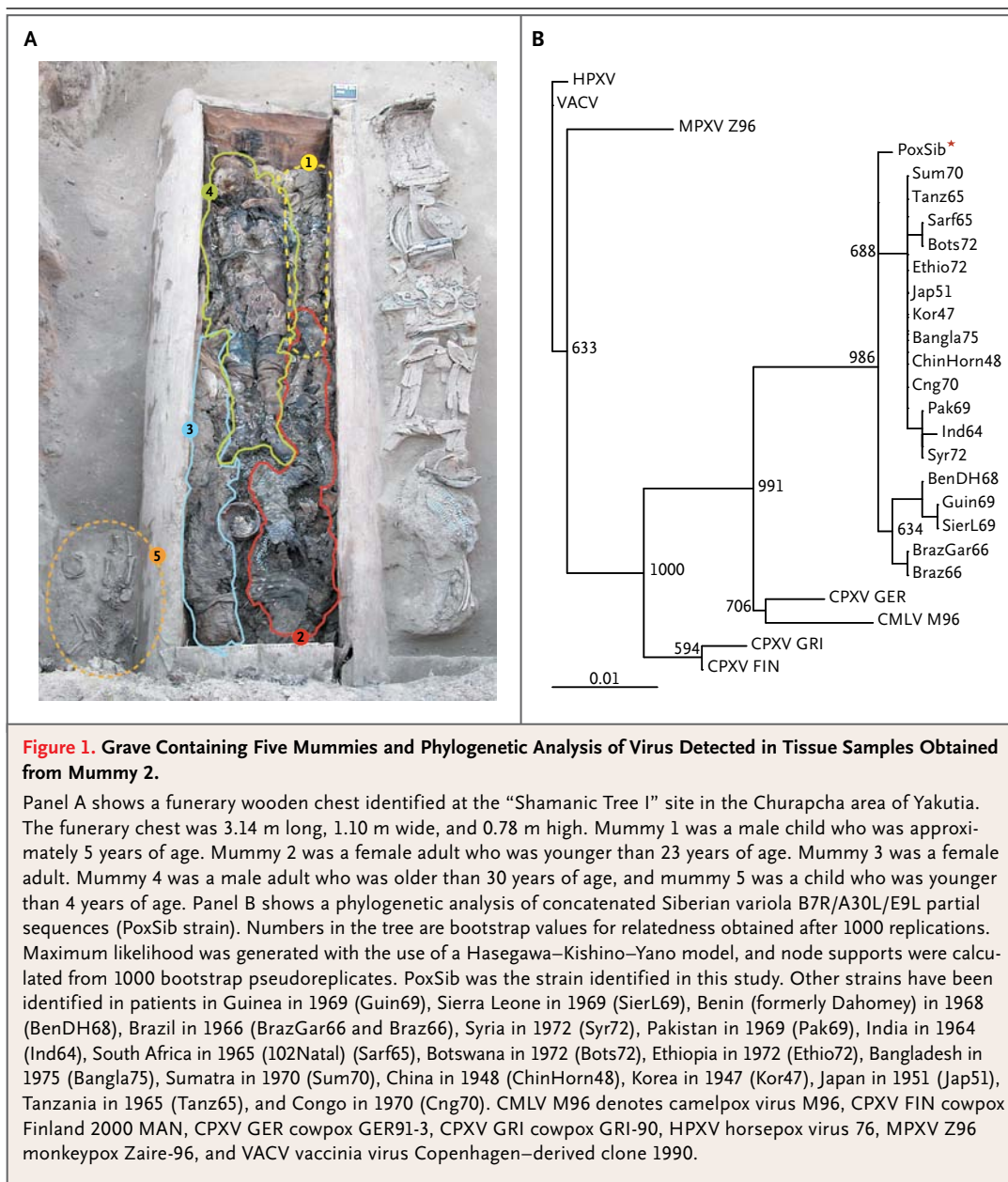
Variola Virus in a 300-Year-Old Siberian Mummy

TO THE EDITOR: Smallpox, which is caused by the variola virus of the Poxviridae family and the orthopoxvirus genus, is among the most devastating human diseases. It may have originated and spread from Egypt, the Near East, or the Indus Valley 3000 to 4000 years ago, and historical reports indicate epidemics in China as early as the first century A.D. and in Europe during the 6th century. By the mid-18th century, smallpox was a worldwide endemic disease. It was eradicated after vaccination campaigns began more than 200 years ago.¹

Variola DNA is about 186 kbp, with genes distributed across conserved (central) or variable

(termini) regions. Sequence analysis has revealed two main clusters: clade 1 includes variants of variola major, and clade 2 includes West Africa strains and variola minor (alastrim).² The oldest sequences that have been characterized originate from biologic samples obtained from patients during the past five to six decades.

In 2004, a French and Russian team identified several archeological sites in northeastern Siberia (in Sakha Republic [Yakutia], Russian Federation). Each site consisted of frozen wooden graves buried in the permafrost and dating from the late 17th to early 18th century.³ One of these graves contained five frozen mummies (Fig. 1A;



and see the Supplementary Appendix, available with the full text of this letter at NEJM.org). This discovery was very unusual, since burial of bodies individually was the standard practice in Yakutia at that time. Analysis of the grave also suggested that the corpses were buried shortly after death.⁴

Biologic samples from mummy 2 were obtained for histologic and molecular investigations. Microscopical examination of pulmonary tissue showed iron inclusions suggestive of the

presence of blood after a possible hemorrhagic episode (Fig. S1 in the Supplementary Appendix). On the basis of these observations, the hypothesis of a sudden and lethal infection was considered, one of which was variola infection.

We confirmed this hypothesis by performing successful polymerase-chain-reaction (PCR) amplification of three short fragments (B7R/hemagglutinin, A30L/14-kD protein, and E9L/DNA polymerase) of the variola genome (PoxSib strain, 718 bp of sequence information) (Fig. S2 in the

Supplementary Appendix). To rule out the persistence of intact viral particles, long-distance PCR analyses (E9L assay, approximately 2 kb) were performed. No positive results were obtained, suggesting an extensive fragmentation of the viral genome. Phylogenetic analyses confirmed that PoxSib was variola-related, clustering together with 18 representative variola human sequences, but distinct from contemporary clades 1 and 2 (Fig. 1B). Bayesian analysis that included PoxSib extended the origin of smallpox viral strains as far back as A.D. 120 (geometric mean, A.D. 928). Thus, PoxSib could be a direct progenitor of modern viral strains or a member of an ancient lineage that did not cause outbreaks in the 20th century. It could be linked to the epidemic of 1714, which was described in studies conducted during the 18th century. The disease may have been imported to Yakutia during Russian conquest.⁵

These data show that mummified bodies frozen in the Siberian permafrost are a reservoir of DNA fragments from ancient pathogens. This genetic information could provide clues to past epidemics.

Philippe Biagini, Ph.D.

Viral Emergence and Co-Evolution Unit
Marseille, France
philippe.biagini@efs.sante.fr

Catherine Thèves, Ph.D.

Patricia Balaesque, Ph.D.
Molecular Anthropology and Image Synthesis Laboratory
Toulouse, France

Annie Gérard, M.D.

Catherine Cannet, M.Sc.
Institute of Legal Medicine
Strasbourg, France

Christine Keyser, Ph.D.

Molecular Anthropology and Image Synthesis Laboratory
Strasbourg, France

Dariya Nikolaeva, M.Sc.

North-Eastern Federal University
Yakutsk, Russian Federation

Patrice Gérard, M.Sc.

Sylvie Duchesne, M.Sc.
Molecular Anthropology and Image Synthesis Laboratory
Toulouse, France

Ludovic Orlando, Ph.D.

Eske Willerslev, Ph.D.
Center for GeoGenetics
Copenhagen, Denmark

Anatoly N. Alekseev, Ph.D.

North-Eastern Federal University
Yakutsk, Russian Federation

Philippe de Micco, M.D., Ph.D.

Viral Emergence and Co-Evolution Unit
Marseille, France

Bertrand Ludes, M.D., Ph.D.

Eric Crubézy, M.D., Ph.D.

Molecular Anthropology and Image Synthesis Laboratory
Toulouse, France

Supported by the French Archaeological Mission in Oriental Siberia (Ministry of Foreign and European Affairs, France), North-Eastern Federal University (Yakutsk, Sakha Republic), and the Human Adaptation program of the French Polar Institute Paul Émile Victor.

Disclosure forms provided by the authors are available with the full text of this letter at NEJM.org.

1. Fenner F, Henderson DA, Arita I, Jezek Z, Ladnyi ID. Smallpox and its eradication. Geneva: World Health Organization, 1988.
2. Lefkowitz EJ, Wang C, Upton C. Poxviruses: past, present and future. *Virus Res* 2006;117:105-18.
3. Crubézy E, Amory S, Keyser C, et al. Human evolution in Siberia: from frozen bodies to ancient DNA. *BMC Evol Biol* 2010;10:25.
4. Crubézy E, Alexeev A, Chamane, Kyss, jeune fille des glaces. Paris: Editions Errance, 2007.
5. Zinner EP. Sibir v izvestiakh zapadnoevropeiskikh puteshestvennikov i outchenykh XVIII v. Vostoshno-Sibirskoe Kniznoe Izdatelstvo Ed, 1968.

DOI: 10.1056/NEJMc1208124

Correspondence Copyright © 2012 Massachusetts Medical Society.

INSTRUCTIONS FOR LETTERS TO THE EDITOR

Letters to the Editor are considered for publication, subject to editing and abridgment, provided they do not contain material that has been submitted or published elsewhere. Please note the following:

- Letters in reference to a *Journal* article must not exceed 175 words (excluding references) and must be received within 3 weeks after publication of the article.
- Letters not related to a *Journal* article must not exceed 400 words.
- A letter can have no more than five references and one figure or table.
- A letter can be signed by no more than three authors.
- Financial associations or other possible conflicts of interest must be disclosed. Disclosures will be published with the letters. (For authors of *Journal* articles who are responding to letters, we will only publish new relevant relationships that have developed since publication of the article.)
- Include your full mailing address, telephone number, fax number, and e-mail address with your letter.
- All letters must be submitted at authors.NEJM.org.

Letters that do not adhere to these instructions will not be considered. We will notify you when we have made a decision about possible publication. Letters regarding a recent *Journal* article may be shared with the authors of that article. We are unable to provide prepublication proofs. Submission of a letter constitutes permission for the Massachusetts Medical Society, its licensees, and its assignees to use it in the *Journal's* various print and electronic publications and in collections, revisions, and any other form or medium.