Next Generation Sequencing –
The Role of New Sequence Technologies in Shaping the Future of Veterinary Science

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Discovery

Alan Radford

Institute of Infection and Global Health, School of Veterinary Science
LE SRAS DANS MONDE
au 26 Mai 2003 on dénombre
au total : 8202 cas

Canada 148 cas
Belgique 1 cas
Espagne 7 cas
Irlande 1 cas
Italie 9 cas
Koweït 1 cas
Lettone 1 cas
Lituanie 1 cas
Laos 1 cas
Mongolie 9 cas
Malaisie 7 cas
Singapour 206 cas
Indonésie 2 cas
Australie 6 cas
Corée du sud 3 cas
Chine 5316 cas
France 7 cas
Hong Kong 1726 cas
Japon 2 cas
Koweït 1 cas
Macao 2 cas
Philippines 10 cas
Taiwan 585 cas
Thaïlande 8 cas
Vietnam 63 cas

Etats-unis 65 cas
Brésil 2 cas
Colombie 1 cas
Finlande 1 cas
Pologne 1 cas
Taiwan 585 cas

Afrique du sud 1 cas
Australie 6 cas
Corée du sud 3 cas

Pays touchés par le SRAS et recensés par l'OMS

travelsante.com

STOP
Coronavirus never before seen in humans is the cause of SARS

Unprecedented collaboration identifies new pathogen in record time

16 April 2003 | GENEVA -- Today, the World Health Organization announced that a new pathogen, a member of the coronavirus family never before seen in humans, is the cause of Severe Acute Respiratory Syndrome (SARS). The speed at which this virus was identified is the result of the close international collaboration of 13 laboratories from 10 countries. While many lines of evidence have found strong associations between this virus and the disease over the last weeks, final confirmation came today.

"The pace of SARS research has been astounding," said Dr. David Heymann, Executive Director, WHO Communicable Diseases programmes. "Because of an extraordinary collaboration among laboratories from countries around the world, we now know with certainty what causes SARS."

The successful identification of the coronavirus means that scientists can now confidently turn to other SARS challenges. For example, various laboratories continue to work to unravel the genetic information of the SARS virus and compare the sequences obtained from viruses in different parts of the world. Experts are gathering at WHO this week to map future work on SARS.

"Today, the collaboration continues as top laboratory researchers have come to WHO to design the next steps, a strategy for transforming these basic research discoveries into diagnostic tools which will help us to successfully control this disease," said Heymann.

This collaboration has brought together leading scientific expertise, and was established after WHO issued a global alert on SARS on 12 March 2003. The priority has been to find the cause and to develop diagnostic tests. Two laboratories in China recently joined this network of laboratories from Canada, France, Germany, Hong Kong Special Administrative Region of China, Japan, the Netherlands, Singapore, the United Kingdom, and the United States of America.
<table>
<thead>
<tr>
<th>Date</th>
<th>Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>16 November 2002</td>
<td>First known case of atypical pneumonia occurs in Foshan City, Guangdong Province, China, but is not identified until much later.</td>
</tr>
<tr>
<td>21 February 2003</td>
<td>64-year-old medical doctor Guangzhou (Guangdong Province) arrives in Hong Kong to attend a wedding.</td>
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<tr>
<td>12 March</td>
<td>WHO issues a global alert about cases of severe atypical pneumonia following mounting reports of spread among staff at hospitals in Hong Kong and Hanoi.</td>
</tr>
<tr>
<td>27 March</td>
<td>Scientists in the WHO lab network identify causative agent, with results from several labs consistently pointing to a new member of the coronavirus family.</td>
</tr>
<tr>
<td>30 May</td>
<td>Full SARS genome sequenced.</td>
</tr>
<tr>
<td>5 July</td>
<td>WHO declares that SARS outbreaks have been contained worldwide, but calls for continued vigilance.</td>
</tr>
</tbody>
</table>
In 2008 an outbreak of unexplained hemorrhagic fever was reported in South Africa. The index patient was admitted on September 12 to a clinic in South Africa. Over the following two weeks secondary and tertiary cases were reported, with four out of the five patients dying. Such outbreaks of haemorrhagic fever are highly emotive events, necessitating a rapid response both to control infection and to control public anxiety. RNA extracts from two post-mortem liver biopsies (cases 2 and 3) and one serum sample (case 2) were submitted to NGS on the Roche 454. Blast analysis of the resulting sequences identified contigs corresponding to about half of a novel arenaviruses ~10kb genome (Briese et al., 2009). The majority of sequences were obtained from serum rather than tissue, presumably a reflection of the higher levels of host DNA obtained from the highly cellular tissue samples.

Novel Orthobunyavirus in Cattle, Europe, 2011

Bernd Hoffmann,¹ Matthias Scheuch,¹ Dirk Höper, Ralf Jungblut, Mark Holsteg, Horst Schirrmieier, Michael Eschbaumer, Katja V. Goller, Kerstin Wernike, Melina Fischer, Angele Breithaupt, Thomas C. Mettenleiter, and Martin Beer

Emerging Infectious Diseases 2012. Vol 8 (3), 469-72

Seven orthobunyavirus sequences were detected in the library prepared from pooled RNA from 3 animals of 1 farm

<table>
<thead>
<tr>
<th>Table. Output of raw sequence data for the sequencing libraries in the analysis of a novel orthobunyavirus in cattle, Europe, 2011</th>
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<tbody>
<tr>
<td>Sample</td>
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<td>--------</td>
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<td>BH 80/11 RNA (3 pooled samples)</td>
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</table>
11 June – Schmallenberg virus: further update on GB testing results

There are 267 UK farms reporting SBV: 45 in cattle and 219 in sheep and 3 premises which reported sheep (earlier in the year) and are now also reporting cattle cases. There are no new reported cases since the 6 June 2012.

We have adjusted some of the current total numbers for the counties where some premises lie close to a county boundary. This has no bearing on the distribution of infection when it occurred last summer or on our assessment of the risk of incursion of potentially infected midges from Continental Europe. It is a consequence of more detailed ongoing work following up affected premises in order to assess impact.

Figures correct as of 11 June 2012

<table>
<thead>
<tr>
<th>County</th>
<th>Positive holdings (Sheep)</th>
<th>Positive holdings (Cattle)</th>
<th>Other species</th>
<th>Positive holdings (cattle and sheep)</th>
<th>Total</th>
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<tr>
<td><strong>Total</strong></td>
<td><strong>219</strong></td>
<td><strong>45</strong></td>
<td><strong>0</strong></td>
<td><strong>267</strong></td>
<td></td>
</tr>
</tbody>
</table>
A case–control study of pathogen and lifestyle risk factors for diarrhoea in dogs

Jenny Stavisky\textsuperscript{a, *}, Alan David Radford\textsuperscript{b}, Rosalind Gaskell\textsuperscript{b}, Susan Dawson\textsuperscript{b}, Alex German\textsuperscript{c}, Bryony Parsons\textsuperscript{b}, Simon Clegg\textsuperscript{b}, Jenny Newman\textsuperscript{b}, Gina Pinchbeck\textsuperscript{b}

In 80 cases of diarrhoea...

7 CECov
4 CPV
24 Campylobacter
1 Salmonella
5 Helminths
39 ??
AB034652 JAPAN A848/88 3CD Aichi virus...
AB040749 JAPAN A virus polyprotein A...
AB010145 JAPAN A846/88 Aichi virus g...
AB034651 JAPAN A844/88 3CD Aichi virus...
EF079158 BANGLADESH B370/05 3CD Aichi...
EF079161 VIETNAM VN636/03 3CD Aichi...
AB034655 INDONESIA N128/91 3CD Aichi...
AB092828 JAPAN 494/97(SIN) 3CD Aichi...
AY747174 GERMANY BAY/1/03/DEU Aichi...
DQ145760 FRANCE RN57 CD3 Aichi virus...
DQ145762 FRANCE R380 CD3 Aichi virus...
DQ145761 FRANCE R586 CD3 Aichi virus...
EU159246 FRANCE E1197/2006 3CD Aichi...
EF079160 THAILAND T132/02 3CD Aichi virus...

AB034662 PAKISTAN P880/91 3CD Aichi virus...
AB034661 PAKISTAN P840/91 3CD Aichi virus...
AB034658 PAKISTAN P766/90 3CD Aichi virus...
A. virus Chshc7 polyprotein+VP0+VP3+V...
DQ028632 BRAZIL Goiania/GO/03/01/Braz...
EF079157 BANGLADESH B171/05 3CD Aichi virus...
EF079155 BANGLADESH B5/05 3CD Aichi virus...
EF079156 BANGLADESH B101/05 3CD Aichi virus...
EF079159 BANGLADESH1055/05 3CD Aichi virus...

gi|345846142|gb|JN088541.1| Canine ko...
DQ145759 FRANCE/MALI RN48 CD3 Aichi virus...
EU787450 HUNGARY P. kobuvirus swine/S...

HUMAN Genotype A
HUMAN Genotype B
Canine
HUMAN Genotype C
Characterization of a canine homolog of hepatitis C virus

Amit Kapoor\textsuperscript{a,1}, Peter Simmonds\textsuperscript{b}, Gisa Gerold\textsuperscript{c}, Natasha Qaisar\textsuperscript{a}, Komal Jain\textsuperscript{a}, Jose A. Henriquez\textsuperscript{a}, Cadhla Firth\textsuperscript{a}, David L. Hirschberg\textsuperscript{a}, Charles M. Rice\textsuperscript{c}, Shelly Shields\textsuperscript{d}, and W. Ian Lipkin\textsuperscript{a}

\textsuperscript{a}Center for Infection and Immunity, Columbia University, New York, NY 10032; \textsuperscript{b}Centre for Immunology, Infection and Evolution, Ashworth Laboratories, University of Edinburgh, Edinburgh EH9 3JT, United Kingdom; \textsuperscript{c}Center for the Study of Hepatitis C, Laboratory of Virology and Infectious Disease, The Rockefeller University, New York, NY 10065; and \textsuperscript{d}Pfizer Veterinary Medicine Research and Development, New York, NY 10017

Edited by Robert H. Purcell, National Institutes of Health, Bethesda, MD, and approved April 28, 2011 (received for review February 1, 2011)

Helicase

Flavivirus

Hepacivirus

2 of 5 respiratory outbreaks
0 of 60 healthy dogs
5 of 19 livers
General principles

- Samples low in host material
  - Serum, diarrhoea, CSF
- Enrichment
  - Centrifugation, DNase / RNase digestion, filtration
- Extraction
  - RNA +/- DNA
- Amplification
  - Random
- Informatics
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