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RODENTS AS CARRIERS OF DISEASE: PRELIMINARY STUDIES IN THE UNITED KINGDOM

COLIN V PRESCOTT AND ALEX STUART
School of Biological Sciences, University of Reading,
Whiteknights, Reading, Berkshire RG6 6AS, UK

Abstract The University of Reading has conducted some preliminary work on the prevalence of *Campylobacter spp*., *Salmonella spp*. and *Arenavirus* in Norway rats trapped from farms and semi-urban areas in central southern England. *Campylobacter* is the cause of a notificable disease in the UK, with 57,772 cases reported for England and Wales in 2009. Transmission to humans is believed to be primarily through undercooked meat, from contaminated water, and through contact with pets; and symptoms include a high temperature, severe diarrhoea, vomiting and abdominal pain. Ninety-seven per-cent of sporadic cases have been attributed to farm animals, and in particular the meat and poultry industry. There are eighteen species of *Campylobacter*, eleven of which can be pathogenic to humans; although the principal species that cause gastrointestinal disease in humans are *C. jejuni* and *C. coli*; although *C. lari*, *C. helveticus* and *C. upsaliensis* are also involved. *Salmonella* species also causes a gastrointestinal disease, and in the UK, is common in chicken and has been linked to egg production. Species are typed using antigen specific agglutination tests, or by their susceptibility to specific bacteriophage. Some strains are known to be linked with human disease (eg. *S. enteritidis* PT4).
Key Words Disease, *Campylobacter*, Lymphocytic Choriomeningitis, *Salmonella*

INTRODUCTION

The"Rodentia" is the most diverse order of living mammals, comprising some 2,024 species and 88 infraspecific taxa, in 27 families that include 444 genera, making up over 41% of living mammal species (Orrell, 2011). In many places, rodents live in close association with man, his farm animals and pets. Some rodent species are referred to as 'commensal' species (Norway rat, roof rat, and house mouse) because of their close association with humans. Macdonald and Fenn (1994) considered 'kleptoparasitic' to be a more appropriate descriptor, because the term 'commensal' implies no damage to man, which is clearly not the case. Rodents often can provide the link with wildlife communities, exposing humans to some zoonoses circulating in natural ecosystems. Fear of wild rats as carriers of disease is likely linked to their frequent occurrence in areas put aside for human waste, in particular sewers and refuge areas.

Rodents are known to be reservoirs of a large number of infectious organisms, many of which are transmitted to humans and his domesticated animals, where they cause disease outbreaks, often with high morbidity and some mortality (Gratz, 1994). From a global perspective, Meerburg et al. (2009) tabulated twenty viral diseases, nineteen bacterial diseases, eight protozoan diseases and fourteen helminth diseases that are linked to rodent infestations. In the UK, Webster and Macdonald (1995) presented prevalence levels for four Bacteria, two Protozoa and seven Helminths in Norway rats trapped from UK farms, and Battersby et al. (2002) presented prevalence data for three Bacteria, three Protozoa and six Helminths in Norway rats trapped from urban areas. So clearly, the concerns over public and veterinary health are well founded.

The University of Reading has conducted some preliminary work on the prevalence of *Campylobacter spp*., *Salmonella spp*. and *Arenavirus* in Norway rats trapped from farms and semi-urban areas in central southern England. Work on the bacterial species is being conducted in collaboration with Dr Sheila MacIntyre, and work on the *Arenavirus* is being conducted in collaboration with Dr Ben Neuman.

Campylobacter is the cause of a notificable disease in the UK, with 57,772 cases reported for England and Wales in 2009. Transmission to humans is believed to be primarily through undercooked meat, from contaminated water, and through contact with pets; and symptoms include a high temperature, severe diarrhoea, vomiting and abdominal pain. Ninety-seven per-cent of sporadic cases have been attributed to farm animals, and in particular

the meat and poultry industry. There are eighteen species of *Campylobacter*, eleven of which can be pathogenic to humans; although the principal species that cause gastrointestinal disease in humans are *C. jejuni* and *C. coli*; although occassionally, *C. lari*, *C. helveticus* and *C. upsaliensis* are also involved.

Salmonella species also causes a gastrointestinal disease, and in the UK, is common in chicken and has been linked to egg production. Species are typed using antigen specific agglutination tests, or by their susceptibility to specific bacteriophage. Some strains are known to be linked with human disease (eg. *S. enteritidis* PT4).

MATERIALS AND METHODS

For both *Salmonella* and *Campylobacter* species, samples were initially obtained by extracting intestinal content of freshly killed wild Norway rats into saline water; and subsequently, samples were obtained from fresh Norway rat droppings collected into peptone water. For *Campylobacter* species, the extracts were cultured within 24 h of collection, in an enrichment broth at 42^{o}C within a microaerophilic environment. A positive result was obtained from one of two sites where samples were collected from intestinal content, and positive results were obtained from three of four sites where samples were collected as fresh droppings.

For the *Campylobacter* work, collaboration has been established with Professor Martin Maiden and Dr Noel McCarthy from the department of Zoology at the University of Oxford. The Maiden group have established a molecular methodology to identify the strain of *Campylobacter*, using a 'Multi-Locus Sequence Typing (MLST), which targets seven house-keeping genes (Hastings et al. 2011; Nielsen et al., 2010). By amplifying and sequencing short fragments of nucleic acid it has been possible to confirm the species of *Campylobacter*, and identify the strain. This work has produced some very interesting and novel results, and is being prepared for publication.

For *Salmonella* species, the extracts were cultured within 24 h of collection, in a non-selective broth, and then plated out on a selective media (XLD agar) at 42^{o}C to isolate individual colonies. Results to date are preliminary and still require verification. However, positive results were obtained from one of two sites where samples were collected from intestinal content, and a site where the sample was collected as fresh droppings.

RESULTS AND DISCUSSION

Arenaviruses are rodent-borne viruses that contain an RNA genome. The genome consists of only four genes, two of which are very similar in all known arenaviruses. When arenaviruses are transmitted to humans, severe disease, including haemorrhagic fever can develop (eg. 'Lassa Fever' from both east and west Africa, 'South American Arenavirus' and 'North American Arenavirus'). In addition there is Lymphocytic Choriomeningitis Virus (LCMV), which is distributed worldwide, and carried by *Mus spp*. and *Rattus spp*. It is carried as a life-long persistent infection (passing from mother to offspring), and is asymptomatic in persistently infected rodents. This virus has been reported in the UK, and human infections do not result in haemorrhagic fever.

Identification is achieved using molecular techniques. Short fragments of nucleic acid are amplified and sequenced to confirm the species. Using this technique, positives indicate that the animal had an on-going infection at the time of sampling. Many of the rats in this study also had enlarged spleens, another common sign of persistent LCMV infection. This method is not able to detect whether a rat was infected in the past, and thus provides a conservative assessment of the infected population. In total, 35 samples were collected from four sites. The virus was extracted from liver or spleen tissue that was collected into Trizol reagent from rats that had previously been stored at -21^{o}C. Ten samples taken from each of the first three sites, and five samples taken from the fourth site. Nine positive samples were obtained (3/10, 3/10, 2/10, 1/5), giving an overall incidence of 25% across all four sites.

With such a high incidence of LCMV in the Norway rat population, the chance of human infection must be significant. But what are the symptoms? Some infected individuals simply do not become ill ! But for those that do symptoms usually develop 8-13 days after exposure to the virus. Initial Phase: fever, malaise; lack of appetite; muscular aches; headache; nausea; vomiting; and paratoid (salivary gland) pain following a few days of recovery. The Second Phase: symptoms of meningitis (eg. fever, headache and a stiff neck) or characteristics of encephalitis (eg. Drowsiness, confusion, sensory disturbance, motor abnormalities (eg paralysis). LCMV has also been known to cause acute hydrocephalus (fluid on the brain), and in rare instances myelitis (inflammation of the spinal cord)

LCMV has been reported in the Americas, Europe, Australia and Japan; and is believed to be contracted by humans through breathing air that is contaminated with rodent excrements. Person to person transmission has not been reported. Infections may therefore occur wherever infected rodent hosts of the virus are found. The disease has historically been under-reported. Several serological studies in urban areas have shown that the prevalence of LCMV infection among humans ranges from 2% to 5%

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