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CASE REPORT

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Characteristics analysis of Orf virus from goats to human

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Abstract

Contagious pustular dermatitis (orf) is an exanthemous disease affecting sheep and goats primarily. As a zoonotic infectious disease caused by parapoxvirus, orf should be managed not only in animals but also in humans. In this study, the typical orf clinical symptoms in goat and humans were observed. Human and goat samples were drawn. The Orf virus (orfv) was identified using an electron microscope, and PCR was used to amplify the target for B2L gene sequence. Molecular analysis of other B2L gene sequences downloaded from GenBank was performed by Mega4 soft. Results indicated that a 21-year old girl who worked in a goat farmcontacted orfv infection from infected goats directly. Eleven amino acid (AA) mutations were detected in the goat orfv transmitted to human orfv. Phylogenetic analysis showed that human orfv in this study was closely related genetically to FJ-SJ2 (KC568397), which was isolated from Fujian province 2012. The results facilitate the development of programs to control Orf virus infections not only in goats but also in humans.

Keywords: *Orfv, human, goat, zoonosis, public health*

Introduction

As a zoonotic infectious disease, contagious pustular dermatitis (orf) is a serious threat to sheep industry and human health [1]. Orf not only has an economic impact on farmers but also generates substantial challenges for animal welfare. The severity of orf depends on the location of lesions and the age of sheep and goats, and mortality is often higher in young ruminants [2], especially in the presence of bacterial or fungal secondary infections [3,4]. Many wild mammals are infected by orf virus (orfv) [5,6] and orfv from deer infects humans [7]. Human orf was first described in 1879 and confirmed in 1934. It was first isolated in 1961 using human amnion cell sheet [8]. Since then, nearly 107 human cases were diagnosed and reported, including one from China. In China, the first orf case was reported in sheep in 1955. Since then, most studies investigating orf in China were focused on sheep and goats, and rarely in other animals or humans. Orfv-infected human was first described in 2006 and the diagnostic analysis was reported in 2012. Comparison and phylogenetic analysis based on the B2L gene of orfv was conducted in goats and sheep in China, during 2009-2011 [9]. The observation of Orf virus-like particle with electron microscopy was rapid using the classical diagnostic method [10-12]. Polymerase chain reaction (PCR) or real-time PCR were used for orf diagnosis [13-15]. PCR and phylogenetic studiestargeted the B2L gene in orf diagnosis, and orfv molecular analysis [16-18].

Surprisingly, orfv was transmitted between humans [19,20]. Therefore, attention should be paid to human orf, for prevention, control and cure. In this study, we diagnosed and analyzed a human orf case. The findings may be used to prevent and control orf infection worldwide.

Case presentation

About 400 black goats were located in a boer goat farm located in Sichuan Province (103.45°E, 30.42°N). Approximately, 20 goats manifested nodular lesions on the lips, tongue and around the mouth. A 21-year-old girl feeder developed fever and apparently exhibited suspected orf symptoms on the borders of her palm. Tissue samples were collected from goats on the farm. Human tissue scrapings from the hand were collected and anticoagulant blood was collected in hospital by the physician. The goats were handled according to the International Guiding Principles for Biomedical Research Involving Animals, as issued by the Council for the International Organizations of Medical Sciences. Ethics related to human studies were compliant with the regulations issued by the World Health Organization.

Identification of Orfv infection

The tissue scrapings from goats and human were triturated in PBS (1:200 V/V), respectively, and freeze-thawed twice between -80ºC and 37ºC. After centrifugation at 3000 rpm for 10 min at 4ºC, 400 μL supernatant and human anticoagulant blood, were used to isolate the total DNA with a genomic DNA purification kit (Promega). The remainder of the supernatant was centrifuged at 35000 rpm for 3 h at 4ºC. The precipitatewas observed with electron microscope. Primers targeting the orfv B2L gene were designed and synthesized (Takara Dalian Co., Ltd). Amplification of the full-length B2L gene was performed using PCR [16]. Sequencing of B2L gene was conducted after PCR.

Sequence analysis

Complete B2L sequences were downloaded from GenBank database. The sequence homologies of nucleotides and amino acids were analyzed by ClustalW method [21]. A phylogenetic tree based on B2L amino acid sequences was constructed by the neighbor-joining method with 1000 bootstrap replicates using MEGA version 4.0 [22].

Results and discussion

Typical orf symptoms were observed in goat and humans clinically. The orfv and its unique criss-cross arrangement of tubular surface were found in goat and human specimens (Figure 1). Orfv was confirmed in goat and human anticoagulant blood by PCR and blast analysis. Sequence analysis showed that B2L

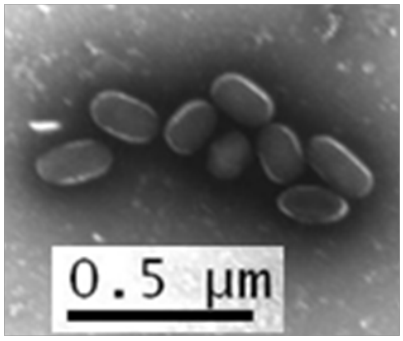

Figure 1. *Electron microscopic study of orfv. Electronmicrograph showing the characteristic tubular surface of orf virions from the lip skin lesions (ba=200 nm).*

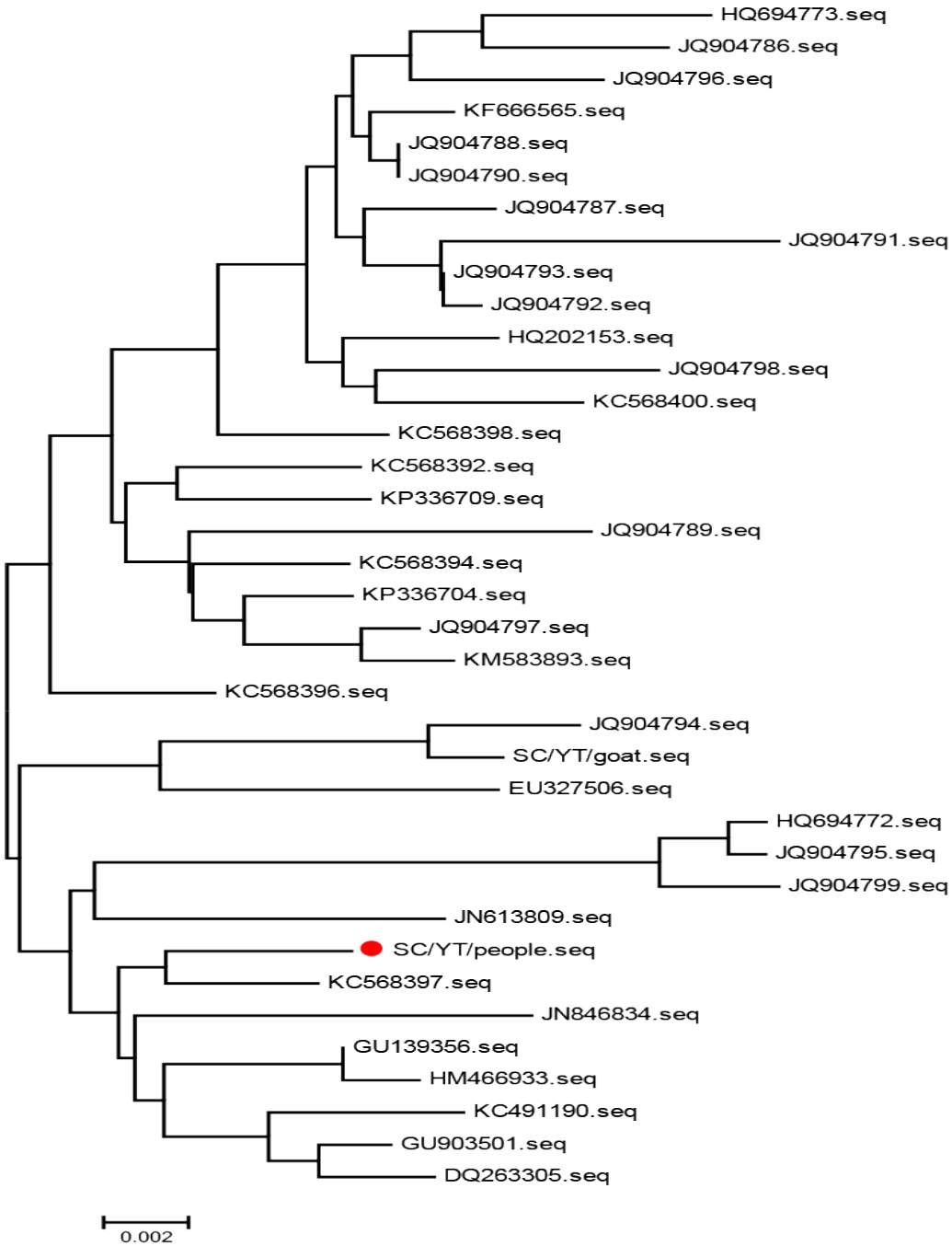

Figure 2. *Phylogenetic analysis based on deduced amino acid sequence of the complete B2L gene. The phylogenetic tree was constructed by the neighbor-joiningalgorithm using MEGA 4.0,and bootstrap analysis was conducted with 1000 trials. Allthe sequences were collected from GenBank. The red spot indicates SC/YT/individuals identified in this study.*

sequences from human and goats were identical (named SC/YT/people). We reported the diagnostic results to the doctor. The girl was rehabilitated after anti-viral and antibiotic treatment administered intravenously for three days in the hospital.

Phylogenetic analysis based on B2L gene indicated that SC/YT/2015 was most closely related phylogenetically to FJ-SJ2(KC568397), which was isolated from goats in FuJian province of China in 2012 (Figure 2), with 99.2% and 98.9% homology at the nucleotide and amino acid level. The study revealed 11 amino acid changes (L9→V, P43→S, L57→P, N79→D, K111→R, D196→N, F204→S, R256→Q, T258→I, D267→N, K354→E) from KC568397 to ORFV/SC/2015.Significance of these amino acid alterations with virus spread from goat to human will be focus of our next study.

Until now, most studies investigating orf were focused on goats and sheep, with few investigations related to humans. Rapid diagnosis and effective interventions are imperative for the control of zoonotic diseases such as orf to prevent human spread. Direct contact with infected animals is one of the primary transmission routes from animals to humans [23]. This case report prompts the need for measures to prevent orfv animal-to-human transmission, especial in individuals handling the infected animals.

People who contact orfv-infected animals or meat are susceptible to infection. Orfv enters the body via skin through scratches, cuts, burns or blisters. Preventive mesasures include wearing plastic gloves, protection of wounds exposed to possibile orfv, hand wash with soap and flowing water, washing clothes that may be contaminated with orfv, and wearing protective equipment. Individuals infected with orfv should not open the sore with a needle, or rub the sore on the face. The sore must be left dry and reported to a physician as soon as possible. Anti-virals and antibiotics are indicated for severe and persistent orf infection.

Conflict of interest statement

We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work; there is no professional or other personal interest of any nature or kind in any product, service, and/or company that could be construed as influencing the position presented in, or the review of, the manuscript entitled.

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Animal rights statement

The goats were handled according to the International Guiding Principles for Biomedical Research Involving Animals, as issued by the Council for the International Organizations of Medical Sciences. Ethics related to human studies were compliant with the regulations issued by the World Health Organization.

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References

1. Maganga GD, Relmy A, Bakkali-Kassimi L, Ngoubangoye B, Tsoumbou T, et al. Molecular characterization of Orf virus in goats in Gabon, Central Africa.*Virol J.* 2016; 13: 79.
2. de la Concha-Bermejillo A, Guo J, Zhang Z, Waldron D. Severe persistent orf in young goats. *J Vet Diagn Invest.* 2003; 15:423-431.
3. Meynink SE, Jackson PG, Platt D. Treatment of intraoral orf lesions in lambs using diathermy and cryosurgery.*Vet Rec.* 1987; 121:594.
4. Chan KW, Lin JW, Lee SH, Liao CJ, Tsai MC, et al. Identification and phylogenetic analysis of orf virus from goats in Taiwan.Virus *Genes.* 2007; 35:705-712.
5. Pedersen AB, Jones KE, Nunn CL, Altizer S. Infectious diseases and extinction risk in wild mammals. *Conserv Biol.* (2007); 21:1269-1279.
6. Khalafalla AI, El-Sabagh IM, Al-Busada KA, Al-Mubarak AI, Ali YH. Phylogenetic analysis of eight sudanese camel contagious ecthyma viruses based on B2L gene sequence.*Virol J.* 2015; 12:124.
7. Kuhl JT, Huerter CJ, Hashish H. A case of human orf contracted from a deer. *Cutis.* 2003; 71:288-290.
8. Nagington J, Whittle CH. Human orf. Isolation of the virus by tissue culture. *Br Med J.*1961; 2:1324-1327.
9. Zhang K, Liu Y, Kong H, Shang Y, Liu X. Comparison and phylogenetic analysis based on the B2L gene of orf virus from goats and sheep in China during 2009-2011. *Arch Virol.* 2014; 159:1475-1479.
10. Harkness JW, Scott AC, Hebert CN. Electron microscopy in the rapid diagnosis of ORF. *Br Vet J.* 1977; 133:81-87.
11. Peng F, Chen Z, Zheng SY, Li HM, Du J, Zhang JZ. A Case of Orf Identified by Transmission Electron Microscopy. *Chin Med J (Engl).* 2016; 129:108-109.
12. Guenova E, Schaller M. Residents' corner March 2012. (Carpe) DIEM--dermatological indications for electron microscopy: orf infection. *Eur J Dermatol.* 2012; 22:293.
13. Gallina L, Dal Pozzo F, Mc Innes CJ, Cardeti G, Guercio A, et al. A real time PCR assay for the detection and quantification of orf virus. *J Virol Methods.* 2006; 134:140-145.
14. Du H, Li W, Hao W, Liao X, Li M, et al. Taqman real-time PCR assay based on ORFV024 gene for rapid detection of orf infection. *Toxicol Mech Methods.* 2013; 23:308-314.
15. Venkatesan G, Balamurugan V, Bhanuprakash V. TaqMan based real-time duplex PCR for simultaneous detection and quantitation of capripox and orf virus genomes in clinical samples. *J Virol Methods*. 2014; 201:44-50.
16. El-Tholoth M, Elnaker YF, Shiha G. Phylogenetic analysis of B2L gene of Egyptian orf virus from naturally infected sheep. *Virus disease.* 2015; 26:147-150.
17. Karakas A, Oguzoglu TC, Coskun O, Artuk C, Mert G, et al. First molecular characterization of a Turkish orf virus strain from a human based on a partial B2L sequence. *Arch Virol*. 2013; 158:1105-1108.
18. Hosamani M, Bhanuprakash V, Scagliarini A, Singh RK. Comparative sequence analysis of major envelope protein gene (B2L) of Indian orf viruses isolated from sheep and goats. *Vet Microbiol.* 2006; 116:317-324.
19. Rajkomar V, Hannah M, Coulson IH, Owen CM. A case of human to human transmission of orf between mother and child. *Clin Exp Dermatol.* 2016; 41:60-63.
20. Turk BG, Senturk B, Dereli T, Yaman B. A rare human-to-human transmission of orf. *Int J Dermatol.* 2014; 53:e63-e65.
21. Thompson JD, Higgins DG, Gibson TJ. CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Res.* 1994; 22:4673-4680.
22. Tamura K, Dudley J, Nei M, Kumar S. MEGA4: Molecular Evolutionary Genetics Analysis (MEGA) software version 4.0. *Mol Biol Evol*. 2007; 24:1596-1599.
23. Zimmerman JL. Orf. *JAMA.* 1991; 266:476.