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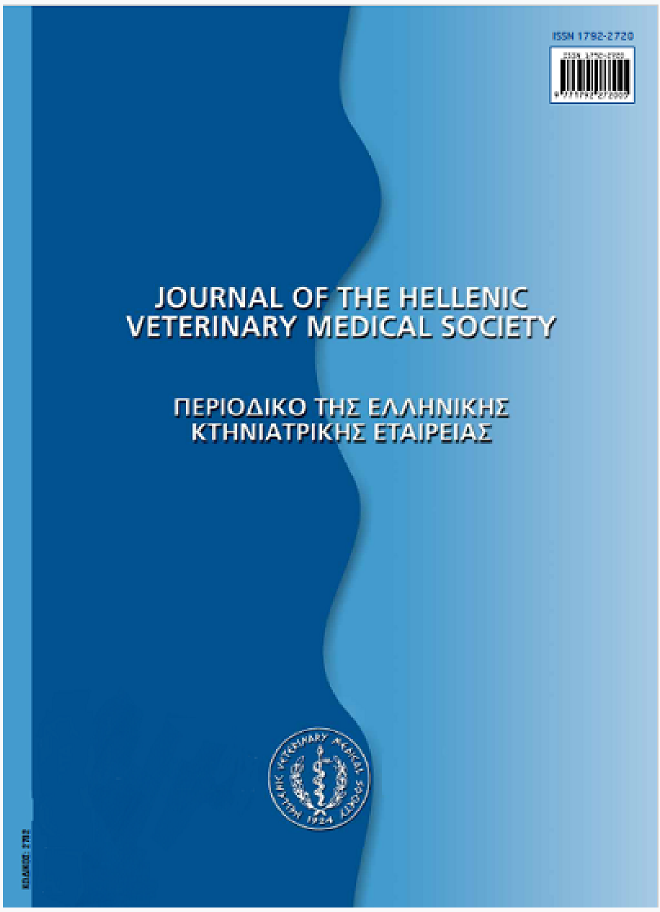

Interspecies transmission of Rotaviruses among ruminants, dogs and humans: Current facts and remarks

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Review article
Ανασκόπηση

Interspecies transmission of Rotaviruses among ruminants, dogs and humans: Current facts and remarks.

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Μετάδοση των Ροταϊών ανάμεσα σε μηρυκαστικά, σκύλους και ανθρώπους: Τωρινά δεδομένα και παρατηρήσεις.

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ABSTRACT . *Rotaviruses* are considered to be a major cause of diarrhoea to humans as well as a wide variety of animals and may cause serious economic losses in livestock animals, especially swine and ruminants. This fact, along with the genetic diversity that characterizes members of the *Rotavirus* group, raised concerns regarding the potential of virus interspecies transmission among various species of animals and humans. Regarding the presence and the epidemiology of *Rotaviruses* in ruminants in association with closely related humans and dogs, research is limited and few data have been presented in recent years. In this review we present all the latest information regarding the distribution of genotypes of *Rotavirus* strains in ruminants, dogs and humans.

Keywords: bovines, dogs, goats, humans, *Rotaviruses*, sheep, transmission

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Περίληψη. Οι ροταϊοί θεωρούνται ένας σημαντικός αιτιολογικός παράγοντας διάρροιας σε ανθρώπους και σε διάφορα είδη ζώων, προκαλώντας σοβαρές οικονομικές απώλειες σε εκτρεφόμενα ζώα, ιδιαίτερα χοίρους και μηρυκαστικά. Το γεγονός αυτό σε συνδυασμό με τη γενετική ποικιλομορφία που χαρακτηρίζει τα μέλη της ομάδας των ροταϊών, δημιουργεί ανησυχίες σχετικά με τη δυνατότητα μετάδοσης των στελεχών του ιού, μεταξύ των διαφόρων ειδών ζώων και ανθρώπων. Οι μελέτες και τα δεδομένα που έχουν παρουσιαστεί τα τελευταία χρόνια σχετικά με την παρουσία των ροταϊών στα μηρυκαστικά και την επιδημιολογία τους σε σχέση με τους ανθρώπους και τους σκύλους με τους οποίους έρχονται σε στενή επαφή, είναι περιορισμένα. Σε αυτή την επισκόπηση παρουσιάζουμε όλες τις τελευταίες πληροφορίες σχετικά με την κατανομή των γενοτύπων των ροταϊών σε μηρυκαστικά, σκύλους και ανθρώπους.
Λέξεις-κλειδιά: βοοειδή, σκύλοι, αίγες, άνθρωποι, ροταϊοί, πρόβατα, μετάδοση

INTRODUCTION

Rotavirus is considered to be one of the most important pathogens of gastrointestinal tract, associated with acute diarrhoea cases mainly in young animals and children (Parashar et al., 2006; Chatzopoulos et al., 2013). The severity of disease decreases as the age progress, probably because of the secretion of gastric acids into the stomach, causing viruses inactivation, as well as the development of adaptive immunity due to previous exposures (Gentsch et al., 2005; Estes and Kapikian, 2007).

The *Rotavirus* genus belongs to the Reoviridae family and is divided into 8 genetic groups (A–H). *Rotaviruses* of group A (RVA) are of the highest importance causing disease in humans and various animal species. *Rotavirus* genome consists of 11 segments of double-stranded RNA and encodes six structural (VP1–VP4, VP6 and VP7) and five or six nonstructural proteins (NSP1–NSP6). The virion consists of a non-enveloped trip-layered icosahedral capsid with the external layer looking like a sponge because of the VP4 spikes extending on the surface (Settembre et al., 2011). A binary classification system is used nowadays to characterize the two outer capsid proteins, VP4 and VP7, which independently elicit neutralizing antibodies. RVA strains are classified into P types (for protease-sensitive) and G types (for glycoprotein) respectively (Estes and Kapikian, 2007). At least 27 G types and 35 P types have been described so far (Matthijnssens et al., 2011a; Banyai et al., 2012; Tam et al., 2014). The *Rotavirus* Classification Working Group has now established a naming manual including (a) the segment's serotype group, (b) the type of the host (c) the country of detection of the strain, (d) the name that was given to the segment, (e) the year of detection and (f) its genotype combination of the GxP[x] format i.e. RVA/sheep-wt/CHN/CC0812-2/2—8/G10P[15] (Matthijnssens et al., 2011a). An improved and more detailed classification scheme has also been adopted by the *Rotavirus* Classification Working Group. The new scheme identifies each of the 11 RNA segments, setting a letter code for each viral protein; thus, VP7-VP4-VP6-VP1-VP2-VP3-NSP1-NSP2-NSP3-NSP4-NSP5/6 are represented by Gx-Px-Ix-Rx-Cx-Mx-Ax-Nx-Tx-Ex-Hx, respectively, with 'x' representing number of corresponding genotype. The classification in genotypes is performed based on previously established cut off phylogenetic identity values, which vary for each of viral genes. The widespread use of full genome analysis has been already proved to be the cornerstone in identifying genomic reassortments and interspecies transmission events, highlighting the potential of a zoonotic viral transmission (Matthijnssens et al., 2011a). According to data collected from whole genome analysis, human *Rotaviruses* can be organized in two major and one minor group. The so-called Wa genogroup in which strains share their backbone gene constellation with porcine strains, the DS1 genogroup with strains sharing genes with bovine-origin *Rotaviruses* and AU1-like strains which relate to feline strains (Doro et al., 2015).

Surveillance of *Rotaviruses* and strain molecular characterization has been largely extended in recent years, in various domestic and wild susceptible animal species, considered earlier to not be of the utmost importance. This has to do mainly with the introduction of vaccines in human and animal populations and the need to monitor their impact on the pathogen epidemiology (Matthijnssens et al., 2012;

2014). According to various results, it is obvious that *Rotavirus* strains cross species barriers either causing a direct interspecies transmission, or exploiting genetic reassortments with other strains. One microenvironment that has become really interesting to investigate these phenomena is the livestock farm where farm keepers, shepherd dogs and ruminants co-exist and thus, during an acute diarrhoea incidence caused by *Rotaviruses*, strains could theoretically cross the species barrier.

This review aims to summarize the updated available data concerning genetic diversity of *Rotavirus* strains in ruminants, as well as to evaluate the genetic relatedness and evolutionary course of *Rotaviruses* among strains detected to young dogs, ruminants and children.

ROTAVIRUSES IN SHEEP

Neonatal diarrhoeic syndrome of lambs is a considerable cause of lamb mortality associated with approximately the half of lamb losses. *Escherichia coli, Rotaviruses, Salmonella spp, Cryptosporidium spp*. and *Clostridium perfigens* are the most frequently detected pathogens, involved in onset and course of diarrhoea. (Schoenian, 2007). Ovine *Rotaviruses* belong to groups A and B, while the epidemiology of infection and the burden of disease remain unclear. Older studies showed that group B *Rotaviruses* could be the cause of severe outbreaks of neonatal diarrhoea with remarkable mortality rates (Chasey and Banks, 1984; Theil et al., 1996). Concerning group A strains, many studies have reported them as the causative agents of lamb diarrhoea in various countries (Wani et al., 2004; Khafagi et al., 2010; Galindo-Cardiel et al., 2011; Gazal et al., 2012). Regarding ovine strains molecular characterization, very limited data has been reported so far. In Spain, two *Rotavirus* strains isolated from diarrhoeic lambs have been molecularly recognized as G8P[14] and G8P[1] (Matthijnssens et al., 2009; Galindo-Cardiel et al., 2011). In India, as the result of a large scale surveillance study, a total of 52 *Rotavirus* strains were detected and characterized. G6 was the predominant G genotype (25/52; 48.07%), followed by G10 (19/52; 36.54%) whereas, the predominant P genotype was P[11] (46/52; 88.46%) (Gazal et al., 2012). In China, all three cases of *Rotaviruses* associated diarrhoea in lambs which have been reported so far, all characterized as G10P[15]-I10-R2-C2-M2-A11-N2-T6-E2-H3. (Shen et al., 1993; Chen et al., 2009; Zhang et al 2011). In the UK, four lamb strains were characterized, showing different specificities, (G3P[1]; G6P[11], G9P[8], G10P[14]) (Fitzgerald et al. 1995). In Greece, during a recent study a G10P[8] strain derived from sheep has been detected and characterized (Chatzopoulos et al., 2015).

ROTAVIRUSES IN GOATS

Similarly to sheep farms, diarrhoeic syndrome of newborn kids is a significant pathological condition reported in goat farms. Group A, B and C *Rotaviruses* have been implicated in diarrhoea cases of a few days old kids, either in pure infections or in combination with E. coli., Cryptosporidium spp and other opportunistic pathogens. Identification of strains has been reported from various countries, and characterization has shown a high similarity to sheep strains. The presence of *Rotaviruses* in goat diarrheic samples has been reported to range from 8% (Munoz et al., 1996; Khafagi et al., 2010) to as high as 40% to 70% (Legrottaglie et al., 1993; Munoz et al., 1995; Alkan et al., 2012). Regarding infective strains characterization, the following G/P combinations have been reported so far: several G6P[1] strains were characterized in Turkey, Italy and Bangladesh, a G8P[1] strain in Turkey, a G6P[14] strain from South Africa, a G3P[3] strain from South Korea, and one G10P[15] from China (Pratelli et al., 1999; de Beer et al., 2002; Lee et al., 2003; Ghosh et al., 2010; Alkan et al., 2012; Liu et al., 2012; Timurkan and Alkan, 2012). Recently, a G8-P[1]-I2-R5-C2-M2-A3-N2-T6-E12-H3 caprine strain was detected for first time in Argentina. Full genomic analyses of this strain resembles similar strains of Bovidae and Camelidae origin, indicating possible interspecies transmission (Louge Uriarte et al., 2014).

ROTAVIRUSES IN CATTLE

In cattle, various RVA strains have been detected, classified into 12 G types (G1–G3, G5, G6, G8, G10, G11, G15, G17, G21, and G24) and 11 P types (P[1], P[3], P[5,6,7], P[11], P[14], P[17], P[21], P[29], and P[33]). The most common worldwide bovine RVA genotypes are considered to be G6 (range, 39.8–78.3%), followed by G10 (21%) in Americas, Europe, Asia and Australia, and G8 (3%) in Africa. Regarding

P typing, P[5] strains (range, 37.1–50.0%) are the most prevalent in Europe, the Americas, Asia, and Australia followed by P[11] (range, 15.4–34.8%) and P[1] (2%) ; strains belonging to G1–G3, G5, and G11 and P[3], P[6], P[7], and P[14] have been also sporadically reported. A total of 20 individual G and P combinations have been described so far and three combinations, G6P[5], G6P[11], and G10P[11] are predominant (combined prevalence, 40%) in many areas worldwide (Papp et al., 2013; Doro et al., 2015). An unusual human G6P[14] *Rotavirus* strain was recently isolated from a child with diarrhoea in Thailand and the whole genome analysis revealed evidence for a bovine-to-human interspecies transmission and reassortment events (Tacharoenmuang et al., 2015). The potential of zoonotic transmission has also recently been proven by the structural basis of glycan specificity in neonate-specific bovine-human reassortant *Rotavirus* (Hu et al., 2015).

ROTAVIRUSES IN DOGS

There have been only a few studies that have reported results regarding the characterization of *Rotaviruses* strains detected in faecal samples derived from dogs. Noteworthy is that all reported strains have been classified in the G3P[3] genotype (Matthijnssens et al., 2011b). Although canine to human *Rotavirus* transmission has not been described yet, several *Rotavirus* strains isolated from children have been shown to possess characteristics of canine origin. In 1997 De Grazia et al. reported the detection of a G3P[3] canine *Rotavirus* strain (PA260/97) in a child suffering from severe gastroenteritis in Palermo, Italy (De Grazia et al., 2007). Additionally, a G3P[3] *Rotavirus* strain was identified in a 2-year-old child who was attended in a hospital's emergency ward in Taiwan in February 2005 (Wu et al., 2012).). A *Rotavirus* C strain classified as G10P[8] was also reported in Hungary, providing further evidence for the genetic diversity of Group C strains (Marton et al., 2015). Moreover, an unusual bovine-like *Rotavirus* A strain G8P[1] multiple reassortant *Rotavirus* strain was isolated from an asymptomatically infected dog (Sieg et al., 2015). Finally, two unusual *Rotavirus* strains were detected in faecal specimens from sheltered dogs in Hungary by viral metagenomics, tentatively named *Rotavirus* serogroup I (Mihalov-Kovacs et al., 2015).

ROTAVIRUSES IN HUMANS

A great variety of group A *Rotaviruses* genotypes have been identified in humans. A total of 14 G types (G1–G6, G8–G12, G14, G20 and G26) and 17 P types (P[1] to P[11], P[14], P[15], P[19], P[24], P[25] and P[28]) and nearly 90 G/P combinations have been recognized in humans (Banyai et al., 2012; Doro et al. 2015). The most commonly isolated from humans *Rotavirus* strains are G1P[8] (31–53%), G2P[4] (10–13%), G3P[8] (5–11%), G4P[8] (5– 14%), G9P[8] (10–11%) and G12P[8] (1–3%). Interestingly, the last two combinations have been being detected in the last 25 years with a dramatically increasing incidence. In many cases, unusual genotypes have been detected as a consequence of reassortments among predominant human strains e.g. G1P[4] and G2P[8] combinations have been detected in areas where typical G1P[8] and G2P[4] strains circulate. Furthermore, a genotype may be highly prevalent in an area along with the six major strains e.g. G6P[6] and G8P[6] in African regions (Cunliffe et al., 1999; Ndze et al., 2014). There are indications that the introduction of vaccines contributed in changing the strains prevalence or even in the appearance of unusual strains derived from reassortment mechanisms. G2P[4] strains appear to predominate in areas where the G1P[8] Rotarix (GlaxoSmithKline Biologicals, Belgium) vaccine is used probably because of the pressure applied by the vaccination on the G1P[8] wild type strains. genotypes like G1P[5], G2P[5] or G6P[8] share either the G6 or the P[5] gene with the parental bovine strain which is contained in the RotaTeq (Merck, Whitehouse Station, NJ, USA) vaccine, indicating possible reassortments with the vaccine strain (Doro et al., 2014). Some rare *Rotavirus* strains are thought to have evolved from animal reservoirs enhancing the conviction of interspecies transmission events. Although these strains have been detected in humans, they are commonly detected in various animal species. The most representative human strains of animal origin are: G4P[6] and G5P[7] strains of swine, G10P[11] and G6P[11] of bovine origin, G3P[3] strains of canine origin, G3P[9] strains of feline origin (Doro et al., 2015). However, there are also various reports of other unusual strains detected in humans and considered to be of animal origin, through direct transmission or reassortment mechanisms: Recently, a Vietnamese G2P[4] *Rotavirus* strain possessing the NSP2 gene sharing an ancestral sequence with Chinese sheep and goat *Rotavirus* strains was reported (Do et al., 2015). Earlier, a study strongly indicated that human P[14] *Rotavirus* strains may have been the result of interspecies transmissions from sheep or other ungulates (Matthijnssens et al., 2009). An Australian G3P[14] strain was detected that had a mixture of bat, feline/canine and bovine *Rotavirus* genes. A G4P[14] strain from Barbados may be a reassortant of human, porcine and bovine *Rotaviruses* (Donato et al., 2014; Tam et al., 2014). In Greece, genotypes G4P[8], G9P[8] and G12P[8] have been detected in samples collected during 2008-2010 (Trimis et al, 2011; Kokkinos et al., 2013; Konstantopoulos et al., 2013). A large surveillance study conducted in the paediatric department of a University hospital between 2009-2013, in a total of 126 *Rotavirus* positive cases, the genotypes found were the following: G4P[8] (58.7%), G1P[8] (14.7%), G12P[8] (9.3%), G3P[8] (9.3%), G12P[6] (5.3%), G9P[8] (1.3%) and G2P[4] (1.3%) (Koukou et al., 2015). A high percentage of genotypes detected (approximately 85%) were similar with the 5 most frequent genotypes circulating around the world and in the community; G1P[8], G2P[4], G3P[8], G4P[8] and G9P[8]. The remaining genotypes (15%) were combination of the possibly animal originated G12 genotype with the common P[8] or the animal originated P[6] genotype.

CONCLUDING REMARKS

Current knowledge regarding *Rotavirus* strains strongly supports the zoonotic potential of the infection. However, the relative reports are scarce and the majority of surveillance studies indicate a rather low prevalence of human infection by animal origin *Rotaviruses*. Nevertheless, reassortments seem to be very likely to occur, as many human strains carry genomic fragments derived from animal strains and these novel strains are readily transmitted from human to human following an adequate adaptation to the new host, as shown by full genome analysis results. Approaching sequencing data in an epidemiological perspective, many scenarios of interspecies reassortments have been reported worldwide in various cases of unusual strains detection in humans. Given that strain surveillance mainly focuses on hospitalized cases, which are only a small proportion of diarrheic cases caused by *Rotaviruses* in total, it is safe to hypothesize that implication of animals in transmission and reassortments events is underestimated.

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CONFLICT OF INTEREST STATEMENT

The authors report no conflict of interest.

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