



Letter

## Postpartum Transmission as a Major Route of Mother-to-Child *Helicobacter felis* Infection

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In this study we investigated maternal *Helicobacter felis* (*H. felis*) infection status to determine the potential of maternal transmission. Pregnant Beagle dogs were infected experimentally with *H. felis*. Following the experimental design, the stools of the mother and litters were isolated and assessed for transmission of *H. felis* at parturition day, 1-week old age and 6-week old age respectively. Polymerase chain reaction (PCR) was used to examine the presence of transmitted *H. felis*. All litters showed no transmission of *H. felis* at parturition day. However, they revealed 14.3 % and 100 % at 1-week old age and 6-week old age respectively by PCR. These results suggested that vertical infection during prenatal period or delivery procedure is unlikely as a route of mother-to-child *H. felis* infection. It might be acquired *H. felis* through breast-feeding, contaminating saliva and fecal-oral during co-habitat.

**Key words:** *Helicobacter*, *H. felis*, dog, transmission, route, postpartum

Received 27 August 2010; Revised version received 11 September 2010; Accepted 14 September 2010

In-depth knowledge of the transmission patterns of *Helicobacter* species may constitute important information for future intervention strategies. In the absence of consistent and verified environmental reservoirs, a predominantly person-to-person transmission has been postulated. *Helicobacter pylori* (*H. pylori*) infection is associated with poor living conditions, and possible transmission routes are fecal-oral, oral-oral, or gastro-oral, but firm evidence is lacking (Torres et al., 2000; Kim et al., 2006). Young children are particularly vulnerable to infection by transmission of *H. pylori* from their infected parents, especially infected mothers (Rothenbacher et al., 1999), and it is generally believed that such transmission is influenced by socio-economic status. However, little is known about how and when maternal transmission occurs

during perinatal period, especially whether this occurs before or after parturition. Also, *H. felis* infection is suspected to acquire mainly in early aged animal but the exact modes of transmission remain elusive (Lee et al., 2007; Oh et al., 2009). In this study, we investigated maternal *H. felis* infection status to determine the potential of maternal transmission.

*H. felis* (ATCC 49179; American Type Culture Collection, USA) was incubated in a brain-heart infusion broth containing 10% fetal bovine serum at 37°C overnight under a microaerophilic atmosphere and allowed to grow to a density of  $\sim 2.0 \times 10^9$  colony-forming units (CFU) per 1 mL of culture broth. Two pregnant Beagle dog were inoculated twice at 3-day intervals by oral administration of  $1.0 \times 10^{10}$  CFU of *H. felis* suspended in 10 mL of broth. All studies were performed in accordance with the Guide for Animal Experimentation by Wonkwang University and approved by the Institutional Animal Care and Use Committee of Wonkwang University (Iksan, Korea). All efforts were made to minimize pain or discomfort of animals used. The challenged animals were confirmed to be *H. felis*-positive by polymerase chain reaction (PCR) of their fecal samples as described previously (Baele et al., 2004). The pregnant dogs were cared until delivery and the mother and her litters were housed in one cage per family. The stools of the mother and litters

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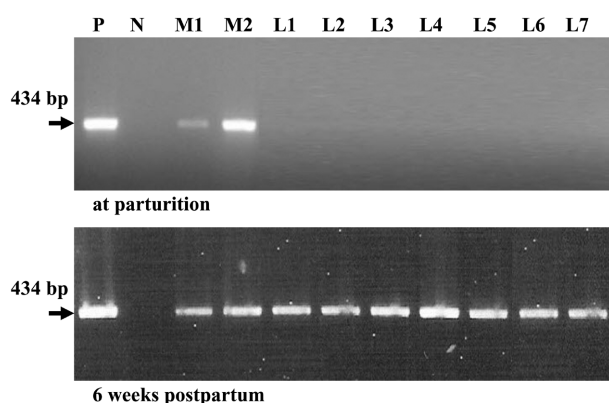
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**Table 1.** Results of polymerase chain reaction for assessment of transmission of *Helicobacter felis*

Evaluated time	Subject	% Detection rate of <i>H. felis</i> (No. of positive/No. of animal)
Delivery	Mothers	100 (2/2)
	Litters	0 (0/7)
1-week postpartum	Mothers	100 (2/2)
	Litters	14.3 (1/7)
6-week postpartum	Mothers	100 (2/2)
	Litters	100 (7/7)

**Figure 1.** Result of *Helicobacter felis* specific polymerase chain reaction with the fecal samples. P, positive control; N, negative control; M1, mother 1; M2, mother 2; L1~L7, litter 1~litter 7. On parturition day, there was no positive reaction in litters, while 6 weeks postpartum, all litters showed positive reaction.

were sampled and assessed for transmission of *H. felis* at parturition day, 1 week and 6 weeks after delivery respectively. Bacterial DNAs were extracted from the stools by bead beater-phenol extraction method (Kim and Kim, 2004; Oh *et al.*, 2009). PCR was performed as previously described method (Baele *et al.*, 2004), which predicted PCR products of 434 base pairs for the final round of amplification. The PCR products were electrophoresed on a 1.2% (weight/volume) agarose gel.

Vertical transmission was examined at parturition day. The mothers were identified *H. felis* infection by PCR (Table 1). However, their fetuses were not infected with *H. felis* (Figure 1). Maternal transmission was examined at 1 week and 6 weeks postpartum (corresponding to transitional milk and weaning stage, respectively). Each stage group was composed of 7 litters. At 1 week postpartum, the mothers were revealed *H. felis* infected status by PCR and one of their litters was infected with *H. felis* (Table 1). At 6 weeks postpartum, the mothers and all their litters were infected with *H. felis* (Table 1 and Figure 1). The frequency of maternal transmission was increased during the nursing period. The transmission rate at 6 weeks postpartum (weaning stage) was significantly higher than at 1 week postpartum (lactating stage) (Table 1).

The results indicated that vertical transmission of *H. felis* was not occurred at pregnant and delivery period. However, they revealed 14.3 % and 100 % at lactating and weaning stage respectively. Recent epidemiological studies in humans suggest that the acquisition of *H. pylori* occur during childhood. For example, Rothenbacher *et al.* (2000) reported that *H. pylori* acquisition seems to occur mainly between the first and second year of life: that is, after the age of weaning. Our results are in agreement with those reports. Also, Rothenbacher *et al.* (2002) reported that infected parents, especially infected mothers, play a key role in the transmission of *H. pylori* within families. Maternal contact behavior during the breastfeeding period may be responsible for the high frequency of maternal transmission. Our results also showed that the maternal-transmission of *H. felis* was not observed during pregnancy and delivery stage, but detected at lactating and weaning stage. The virulence mechanisms of non-*H. pylori* gastric *Helicobacters* like as *H. felis* are hitherto largely unknown nor is it clear whether the same differences in terms of virulence are present among these organisms (De Bock *et al.*, 2006). On the basis of these findings, vertical infection during pregnancy or at delivery is unlikely as a route of mother-to-child *H. felis* infection. We suggested that *H. felis* infection of transplacental route during pregnancy might not be occurred and that *H. felis* transmission by discharges of uterine or vagina, obstetric delivery tract, during parturition might not be occurred. It might be acquired *H. felis* through breast-feeding, contaminating saliva and fecal-oral during co-habitat.

In conclusion, the present study provides new and important information on maternal transmission of *H. felis*. This study implied that maternal transmission of *H. felis* might be developed during latency or later postpartum stage.

## Acknowledgements

This study was supported by the research fund of Wonkwang University in 2009, Iksan, Korea. The authors thank Prof. Kook Y. for invaluable discussions during the preparation of the manuscript.

## References

- Baele, M., Van den Bulck, K., Decostere, A., Vandamme, P., Hänninen, M.L., Ducatelle, R. and Haesebrouck, F. (2004) Multiplex PCR assay for differentiation of *Helicobacter felis*, *H. bizzozeronii*, and *H. salomonis*. *J. Clin. Microbiol.* 42(3), 1115-1122.
- De Bock, M., Decostere, A., Hellemans, A., Haesebrouck, F. and Ducatelle, R. (2006) *Helicobacter felis* and *Helicobacter bizzozeronii* induce gastric parietal cell loss in Mongolian

- gerbils. *Microbes Infect.* 8(2), 503-510.
- Kim, S.H. and Kim, O. (2004) Application of Consensus Polymerase Chain Reaction for Monitoring of *Helicobacter* Species. *Lab. Anim. Res.* 20(4), 316-320.
- Kim, S.K., Cho, S.J. and Kim, O. (2006) Detection and identification of secreting *Helicobacter* species from cats. *Lab. Anim. Res.* 22(3), 243-247.
- Lee, H.A., Park, Y.S. and Kim, O. (2007) Prevalence of *Helicobacter* Species in Feces of Dogs. *Lab. Anim. Res.* 23(3), 339-344.
- Oh, H.G., Kim, Y., Lee, H.A. and Kim, O. (2009) Detection of *Helicobacter heilmannii* infection in a cat. *Lab. Anim. Res.* 25(4), 385-391.
- Rothenbacher, D., Inceoglu, J. and Bode, G.. (1999) *Helicobacter pylori* among preschool children and their parents: evidence of parent-child transmission. *J. Infect. Dis.* 179(2), 398-402.
- Rothenbacher, D., Inceoglu, J. and Bode, G. (2000) Acquisition of *Helicobacter pylori* infection in a high-risk population occurs within the first 2 years of life. *J. Pediatr.* 136(6), 744-748.
- Rothenbacher, D., Winkler, M. and Gonser, T. (2002) Role of infected parents in transmission of *Helicobacter pylori* to their children. *Pediatr. Infect. Dis. J.* 21(7), 674-679.
- Torres, J., Perez-Perez, G. and Goodman, K.J. (2000) A comprehensive review of the natural history of *Helicobacter pylori* infection in children. *Arch. Med. Res.* 31(5), 431-469.