---Review---

**Tupaia Belangeri** as an Experimental Animal Model for Viral Infection

Kyoko TSUKIYAMA-KOHARA1,2) and Michinori KOHARA3)

1) Transboundary Animal Diseases Center, Joint Faculty of Veterinary Medicine, Kagoshima University, 1-2-24 Korimoto, Kagoshima 890-0065, Japan
2) Laboratory of Animal Hygiene, Joint Faculty of Veterinary Medicine, Kagoshima University, 1-2-24 Korimoto, Kagoshima 890-0065, Japan
3) Department of Microbiology and Cell Biology, Tokyo Metropolitan Institute of Medical Science, Tokyo 113-8613, Japan

**Abstract:** Tupaias, or tree shrews, are small mammals that are similar in appearance to squirrels. The morphological and behavioral characteristics of the group have been extensively characterized, and despite previously being classified as primates, recent studies have placed the group in its own family, the Tupaiidae. Genomic analysis has revealed that the genus **Tupaia** is closer to humans than it is to rodents. In addition, tupaias are susceptible to hepatitis B virus and hepatitis C virus. The only other experimental animal that has been demonstrated to be sensitive to both of these viruses is the chimpanzee, but restrictions on animal testing have meant that experiments using chimpanzees have become almost impossible. Consequently, the development of the tupaia for use as an animal infection model could become a powerful tool for hepatitis virus research and in preclinical studies on drug development.

**Key words:** genome, HBV, HCV, *Tupaia*, virus

---

**Taxonomic Classification**

*Tupaia belangeri* belongs to the family Tupaiidae, which consists of four genera and 19 extant species (Table 1) [13, 19]. The members of *Tupaia*, which are colloquially referred to as tree shrews, were first recorded in a sketch by William Ellis on a voyage with Captain Cook in 1780 [7]. With a body weight ranging between 45–350 g (Table 1), members of the genus *Tupaia* are similar in appearance to squirrels (Fig. 1). The natural habitat of *Tupaia* spp. consists of the tropical rainforest in South East Asia where they feed on fruits, insects and small vertebrates [7].

Similarities between *Tupaia* spp. and primates were first reported in the 1920s; for example, Le Gros Clark proposed that tree shrews and primates were closely related based on brain anatomy [20]. However, recent molecular studies have separated tupaias from the primates and placed them in the order Scandentia and within the grandorder Euarchonta, which also contains the Primates and Dermoptera [17].

---

**Handling of Tupaia**

Tupaia is active during daytime, and animal rooms are illuminated from 7:00 am to 9:00 pm with a relative humidity of 50–60%, and temperature at 26°C. Their foods are CMS-1M (CREA, Japan) 20 g, apple, banana and boiled egg, everyday. They usually slip into the boxes as soon as somebody enters the room, then we can catch them by net. We can bleed approximately 0.5 ml from the tail or leg vein once in 2 weeks. Tupaias can be
breeding after 6–9 months age and easily to give average 4 babies after approximately 45 days of pregnancy. Tu-

paia usually possesses few health problems, but some-
times shows diarrhea by Escherichia coli, Klebsiella
pneumonia or protozoa, which can be checked by quar-
tantine. The inbred tupaias has not been established yet.

Table 1. Composition of family Tupaiidae [1]

| Taxa | Morphological characteristics | Reproductive characteristics | Weaning and longevity | Distribution |
|------|-------------------------------|-----------------------------|-----------------------|--------------|
| Family: Tupaiidae | | | | |
| Genus: Tupaia | | | | |
| *Species: belangeri | BW: 50–270 g | GP: 41–55 d | W: ca. 30 d | Tropical forests in Southeast Asia |
| Subspecies: belangeri chinensis | HBL: 12–21 cm | L: 1–5 | L: 9–12 yr |
| *Species: chrysogaster, dorsalis, glis, gracilis, javanica, longipes, minor, moellendorffi, montana, nicobarica, palawanensis, picta, splendidula, tana | NN: 1–3 pairs | NBW: 6–10 g | |
| Genus: Anathana ellioti | BW: 180 g | UK | UK |
| Genus: Dendrogale melanura, murina | BW: 60 g | GP: 41–55 d | W: ca. 30 d |
| | HBL: 13 cm | L: 1–5 | L: 9–12 yr |
| | NN: 1 pair | NBW: 6–10 g | |
| Genus: Urogale everetti | BW: 220–359 g | GP: 30 d | W: ca. 30 d |
| | HBL: 20 cm | L: 1–4 | L: 6 yr |
| | NN: 2 pairs | NBW: 10 g | |

*BW: body weight; HBL: head-body length; NN: number of nipples; GP: gestation period; L: litter size; NBW: Newborn body weight; W: weaning; L: life span; UK: unknown.

![Fig. 1. Adult female tupaias](image)

Evolutionary characterization of 7S RNA-derived short interspersed elements (SINEs) revealed that 7S RNA is a component of the cytoplasmic signal recognition particle [33] in primates [5], tupaias [25] and rodents [18], i.e. all of the members of the placental mammalian order Supraprimates and the superorder Euarchontogli-

res. The fossil Alu monomer was previously considered to be the oldest common ancestor of all 7S RNA-derived SINEs [27], and was thought to be restricted to primates [17]. Tupaia possesses specific, chimeric, Tu-type II SINEs, which may share a common ancestor with rodent B1 SINEs [27]. Phylogenetic analysis of 7S L RNA-

derived SINEs has shown that tupaias can be grouped with primates and Dermoptera in the Euarchonta, while the Rodentia and Lagomorpha can be grouped with the Glires [17].

Whole-genome analysis by several groups ([8], Tsu-

kiyama-Kohara et al., in preparation) revealed a ge-

netic relationship between tupaias and humans. Simi-

larly, phylogenetic analysis based on whole genome sequences showed that humans are closer to tupaias than they are to mice (Fig. 2). Further, several of the same highly conserved and variable genes have been identified
in both tupaia and humans. For example, relatively high homology has been observed between human and Tupaia hepatitis C virus (HCV) viral receptor CD81 (Fig. 3A), scavenger receptor class B member I (SR-BI), the tight junction proteins claudin I and occludin I [16], as well as the hepatitis B virus (HBV) receptor, sodium-taurocholate cotransporting polypeptide (NTCP) (Fig. 3B) [38], particularly in the receptor and virus envelope surface glycoprotein regions that interact with the transmembrane proteins. It is possible that these highly conserved molecules could be a missing link during the evolution of tupaia, and detailed analysis of this hypothesis is currently underway.

**Tupaia as an Experimental Animal Model**

The high degree of genetic homology between several neuromodulator receptor proteins in tree shrews and primates has meant that Tupaia has been extensively utilized in preclinical research, particularly in the areas of toxicology and virology [10]. Although adult male tupaia exhibit strong territoriality in their natural habitat, the coexistence of two males in visual and olfactory contact in the laboratory leads to the establishment of a stable dominant-subordinate relationship, with subordinates showing distinct stress-induced alterations to behavior, physiology and central nervous activity [9]. These alterations exhibited by the subordinate male tupaia are similar to those observed in depressed human patients, and could be applicable to preclinical research of antidepressant drugs [11]. Various aspects of human behavior, infant development, communication and social structure could also potentially be studied in tupaia [22, 23].

**Tupaia as Viral Hepatitis Model**

Tupaia have also been employed in studies of viral infection, especially on hepatitis B and C viruses (HBV and HCV) [12]. For these viruses, the only existing natural-infection animal model is the chimpanzee. However, because chimpanzees are long-lived (>50 years), very expensive, and subject to stringent animal welfare regulations, several groups have attempted to develop Tupaia for use as an animal infection model. Pathogenesis of HCV was characterized using various transgenic mouse animal models and they can develop chronic hepatitis, liver cirrhosis and hepatocellular carcinoma [30], however natural infection is difficult to be established in these mice. HCV can successfully establish infection in the humanized chimeric mice liver [15, 24], but they do not have immune response, therefore, patho-
genicity of HCV could not be characterized.

We previously conducted infection experiments using HCV in Tupaia and characterized the pathogenesis in this animal [2]. Chronic HCV infection, which manifests as liver cirrhosis and hepatocellular carcinoma, is easily established [1]. Currently, approximately 170 million people around the world may be infected with HCV [35].

The current standard therapy for chronic hepatitis C is a combination of pegylated interferon (IFN) alpha-2a and nucleoside analog ribavirin. Recently, IFN-free combinations of direct-acting antiviral agents have been tested for clinical use and can achieve significant antiviral activity [29]. However, no vaccines against HCV infection have been developed to date, mainly because of the lack of suitable animal experimental systems.

We injected tupaias with serum from a chronic hepatitis C patient (HCR6; \(3.7 \times 10^4\) 50% chimpanzee infectious dose/ml) or reconstituted virus (RCV; genotype 1b). Inoculation with patient serum caused marked fluctuations in the serum alanine aminotransferase (ALT) concentrations − from 2–5 fold in both tupaias − suggesting acute hepatitis (Figs. 4 and 5). Quantitation of viral RNA by reverse transcription PCR revealed HCV viremia in Tupaia (Tup. 5 and 6, Fig. 5A). Inoculation with RCV showed sustained viremia for up to 10 weeks (Tup. 4 and 8, Fig. 5B). Histological examination re-
Monitoring

Long-term follow up
- Serum ALT values
- Serum HCV RNA (Quantification by RTD-RT-PCR)

Fig. 4. Experimental design of HCV infection and re-infection of tupaia.

Fig. 5. Course of HCV infection in tupaia. (A) Tupaias No. 5 and 6 were inoculated with patient serum HCR6. Serum ALT (IU/ml) and viral loads, measured as amount of HCV RNA (copies/ml), were measured for over 120 weeks. Set point for serum ALT in untreated tupaia was 22.3 IL/ml (n=23). Negative control animals showed no significant ALT fluctuations for more than 2 years (n=3). No HCV RNA was detected in the negative controls after more than 2 years (n=3). (B) Tupaias No. 4 and 8 were inoculated with RCV as for the HCR6 inoculated animals.
K. TsuKiyama-Kohara and M. Kohara

revealed that HCV caused chronic hepatitis, fibrosis and cirrhosis (Fig. 6), with progressive lipid degeneration observed in tupaias over the course of infection. Macroscopic observations also indicated that liver cirrhosis worsened and large surface nodules were observed (Fig. 6). Transmission of viral RNA-positive serum to naïve animals reproduced acute hepatitis and viremia, indicating that hCV infection could reproduce the pathogenesis typically associated with acute and chronic hepatitis in tupaia. However, sustained seroconversion was not observed in tupaias and production of HCV antibody only occurred at specific time points. To increase the susceptibility of tupaia to hCV infection and to develop a sensitive hCV infection model, these differences between hCV infection in tupaia and humans should be examined in future. hCV infection studies in tupaia have been examined using x-rays [41] and metabolic analysis [31], and the efficacy of natural products for treating hCV-infected tupaia has also been evaluated [39].

Several groups have successfully infected tupaia with HBV, as follows. In culture medium, infection by HBV has been shown to produce HBs antigen (Ag) and HBeAg. HBV infection in newborn and adult tupaia induced the production of HBsAg, HBsAb, HBCab and HBeAb; all of the adults were successfully infected [34]. Experimental infection of tupaia with HBV was successful in approximately 55% of the animals inoculated [38]. HBV infection and aflatoxin B1 exhibited a synergistic effect in hepatocarcinogenesis [21]. To establish chronic infection by HBV, newborn tree shrews were infected with HBV [36]. Six of 46 newborn babies were found to be susceptible to HBV infection at 48 weeks post inoculation. Histological analysis of liver tissues from infected tupaia revealed chronic hepatitis symptoms, such as hydropic, fatty and eosinophilic degeneration of hepatocytes, lymphocytic infiltration, and hyperplasia of small bile ducts in the portal area [28]. One tupaia infected with HBV for more than 6 years showed multiple necrotic areas [28]. These findings show that although the efficacy of infection needs to be improved in future, tupaia are potentially well suited for use as a model for HBV infection.

Tupaia have also been reported to be infected by
specific viruses, such as tupaia herpes virus, which induces tumorigenicity [4], and potentially with non-pathogenic tupaia paramyxovirus [32]. Tupaias have also been infected with TTV [26], tupaia adenovirus [3], and influenza virus [40].

Conclusion

Tupaia shares considerable genetic homology with both humans and primates, and is considered to be well suited for use as a model for studies on viral infection and preclinical drug development. At present, difficulties associated with maintaining and handling tupaia are major factors limiting the widespread adoption of this animal for use in infection studies. However, optimizing these issues will facilitate the use of tupaia as an experimental animal. In addition, development of genetic methods for modifying the tupaia genome would also increase the potential value of tupaia as a model animal, as this would facilitate detailed studies of virus pathogenesis and drug evaluation.

Acknowledgments

We thank the staff of the Department of Animal Hygiene in the Joint Faculty of Veterinary Medicine at Kagoshima University, and at the Department of Microbiology and Cell Biology of The Tokyo Metropolitan Institute of Medical Science for their assistance with animal care, especially Dr. Yutaka Amako. This work was supported by grants from the Ministry of Education, Culture, Sports, Science and Technology of Japan and the Ministry of Health, Labour and Welfare of Japan.

References

1. Alter, M.J., Kruszon-Moran, D., Nainan, O.V., McQuillan, G.M., Gao, F., Moyer, L.A., Kaslow, R.A., and Margolis, H.S. 1999. The prevalence of hepatitis C virus infection in the United States, 1988 through 1994. N. Engl. J. Med. 341: 556–562. [Medline] [CrossRef]
2. Amako, Y., Tsukiyama-Kohara, K., Katsume, A., Hirata, Y., Sekiguchi, S., Tobita, Y., Hayashi, Y., Hishima, T., Funata, N., Yonekawa, H., and Kohara, M. 2010. Pathogenesis of hepatitis C virus infection in Tupaia belangeri. J. Virol. 84: 303–311. [Medline] [CrossRef]
3. Brinckmann, U., Darai, G., and Flügel, R.M. 1983. Tupaia (tree shrew) adenovirus DNA: sequence of the left-hand fragment corresponding to the transforming early region of human adenoviruses. EMBO J. 2: 2185–2188. [Medline]
4. Darai, G., Koch, H.G., Flügel, R.M., and Gelderblom, H. 1982. Tree shrew (Tupaia) herpesviruses. Dev. Biol. Stand. 52: 39–51. [Medline]
5. Deininger, P.L., Jolly, D.J., Rubin, C.M., Friedmann, T., and Schmid, C.W. 1981. Base sequence studies of 300 nucleotide renatured human DNA clones. J. Mol. Biol. 151: 17–33. [Medline] [CrossRef]
6. Drummer, H.E., Wilson, K.A., and Poubiourpios, P. 2002. Identification of the hepatitis C virus E2 glycoprotein binding site on the large extracellular loop of CD81. J. Virol. 76: 11143–11147. [Medline] [CrossRef]
7. Eliot, O. 1971. Bibliography of the tree shrews. Primates 12: 323–414.
8. Fan, Y., Huang, Z.Y., Cao, C.C., Shen, C.S., Chen, Y.X., Fan, D.D., He, J., Hou, H.L., Hu, L., Hu, X.T., Jiang, X.T., Lai, R., Lang, Y.S., Liang, B., Liao, S.G., Mu, D., Ma, Y.Y., Niu, Y.Y., Sun, X.Q., Xia, J.Q., Xiao, J., Xiong, Z.Q., Xu, L., Yang, L., Zhang, Y., Zhao, W., Zhao, X.D., Zheng, Y.T., Zhou, J.M., Zhu, Y.B., Zhang, G.J., Wang, J., and Yao, Y.G. 2013. Genome of the Chinese tree shrew. Nat. Commun. 4: 1426. [Medline] [CrossRef]
9. Fuchs, E., Kramer, M., Hermes, B., Netter, P., and Hienke, C. 1996. Psychosocial stress in tree shrews: clomipramine counteracts behavioral and endocrine changes. Pharmaco1. Biochem. Behav. 54: 219–228. [Medline] [CrossRef]
10. Fuchs, E. and Flügge, G. 2002. Social stress in tree shrews: effects on physiology, brain function, and behavior of subordinate individuals. Pharmaco1. Biochem. Behav. 73: 247–258. [Medline] [CrossRef]
11. Fuchs, E. 2005. Social stress in tree shrews as an animal model of depression: an example of a behavioral model of a CNS disorder. CNS Spectr. 10: 182–190. [Medline]
12. Han, J.B., Zhang, G.H., Duan, Y., Ma, J.P., Zhang, X.H., Luo, R.H., Lü, L.B., and Zheng, Y.T. 2011. [Sero-epidemiology of six viruses natural infection in Tupaia belangeri chinensis]. Zool. Rev. 32: 11–16 (in Chinese). [Medline]
13. Helgen, K.M. Order Scandentia, 2005. Tupaia belangeri. In: Mammal Species of the World: A Taxonomic and Geographic Reference, 3rd ed. (Wilson, D.E., and Reeder, D.M., eds.), Vol. 1, pp. 104–108. John Hopkins University Press.
14. Higginbottom, A., Quinn, E.R., Kuo, C.C., Flint, M., Wilson, L.H., Bianchi, E., Nicosia, A., Monk, P.N., McKeating, J.A., and Levy, S. 2000. Identification of amino acid residues in the E2 glycoprotein of hepatitis C virus-infected chimeric mice in vivo. Hepatology 32: 11–16 (in Chinese). [Medline]
15. Inoue, K., Umehara, T., Ruegg, U.T., Yasui, F., Watanabe, T., Yasuda, H., Dumont, J.M., Scalairo, P., Yoshida, M., and Kohara, M. 2007. Evaluation of a cyclophilin inhibitor in hepatitis C virus-infected chimeric mice in vivo. Hepatology 45: 921–928. [Medline] [CrossRef]
16. Jeulin, H., Velay, A., Murray, J., and Schoevoer, E. 2013. Clinical impact of hepatitis B and C virus envelope glycoproteins. World J. Gastroenterol. 19: 654–664. [Medline] [CrossRef]
17. Kriegs, J.O., Churakov, G., Jurka, J., Brosius, J., and Schmitz, J. 2007. Evolutionary history of 7SL RNA-derived SiNEs in supraprimates. Trends Genet. 23: 158–161. [Medline] [CrossRef]
18. Krayev, A.S., Kramerov, D.A., Skryabin, K.G., Ryskov, A.P.,
Bayev, A.A., and Georgiev, G.P. 1980. The nucleotide sequence of the ubiquitous repetitive DNA sequence B1 complementary to the most abundant class of mouse fold-back RNA. *Nucleic Acids Res.* 8: 1201–1215. [Medline] [CrossRef]

19. Kumar, S. and Hedges, S.B. 1998. A molecular timescale for vertebrate evolution. *Nature* 392: 917–920. [Medline] [CrossRef]

20. Le Gros Clark, W.E. 1924. On the brain of the tree shrew (*Tupaia minor*). In: Proc. Zoool. Soc., London. pp. 1053–1074.

21. Li, Y., Su, J.J., Qin, L.L., Yang, C., Ban, K.C., and Yan, R.Q. 1999. Synergistic effect of hepatitis B virus and aflatoxin B1 in hepatocarcinogenesis in tree shrews. *Ann. Acad. Med. Singapore* 28: 67–71. [Medline]

22. Martin, R.D. 1968. Reproduction and ontogeny in tree-shrews (*Tupaia belangeri*), with reference to the general behaviour and taxonomic relationships. *Z. Tierpsychol.* 25: 409–495. [Medline] [CrossRef]

23. Martin, R.D. 1968. Reproduction and ontogeny in tree-shrews (*Tupaia belangeri*), with reference to their general behavior and taxonomic relationships. *Z. Tierpsychol.* 25: 505–532.

24. Nakagawa, S., Hirata, Y., Kameyama, T., Tokunaga, Y., Nishito, Y., Hirabayashi, K., Yano, J., Ochiya, T., Tateno, C., Tanaka, Y., Mizokami, M., Tsukiyama-Kohara, K., Inoue, K., Yoshida, M., Takaoka, A., and Kohara, M. 2013. Targeted induction of interferon-λ in humanized chimeric mouse liver abrogates hepatotropic virus infection. *PLoS ONE* 8: e59611. [Medline] [CrossRef]

25. Nishihara, H., Terai, Y., and Okada, N. 2002. Characterization of novel Alu- and tRNA-related SINEs from the tree shrew and evolutionary implications of their origins. *Mol. Biol. Evol.* 19: 1964–1972. [Medline] [CrossRef]

26. Okamoto, H., Nishizawa, T., Takahashi, M., Tawara, A., Peng, Y., Kishimoto, J., and Wang, Y. 2001. Genomic and evolutionary characterization of TT virus (TTV) in tupaias and comparison with species-specific TTVs in humans and non-human primates. *J. Gen. Virol.* 82: 2041–2050. [Medline]

27. Quentin, Y. 1994. A master sequence related to a free left Alu monomer (FLAM) at the origin of the B1 family in rodent genomes. *Nucleic Acids Res.* 22: 2222–2227. [Medline] [CrossRef]

28. Ruan, P., Yang, C., Su, J., Cao, J., Ou, C., Luo, C., Tang, Y., Wang, Q., Yang, F., Shi, J., Lu, X., Zhu, L., Qin, H., Sun, W., Lao, Y., and Li, Y. 2013. Histopathological changes in the liver of tree shrew (*Tupaia belangeri* chinesis) persistently infected with hepatitis B virus. *Virol. J.* 10: 333. [Medline] [CrossRef]

29. Schinazi, R., Haf临n, P., Marcellin, P., and Asselah, T. 2014. HCV direct-acting antiviral agents: the best interferon-free combinations. *Liver Int.* 34: 69–78. [Medline] [CrossRef]

30. Sekiguchi, S., Kimura, K., Chiyot, O., Ohtsuki, T., Tobita, Y., Tokunaga, Y., Yasui, F., Tsukiyama-Kohara, K., Wakis, T., Tanaka, T., Miyasaka, M., Mizuno, K., Hayashi, Y., Hishima, T., Matsushima, K., and Kohara, M. 2012. Immunization with a recombinant vaccinia virus that encodes nonstructural proteins of the hepatitis C virus suppresses viral protein levels in mouse liver. *PLoS ONE* 7: e51656. [Medline] [CrossRef]

31. Sun, H., Zhang, A., Yan, G., Piao, C., Li, W., Sun, C., Wu, X., Li, X., Chen, Y., and Wang, X. 2013. Metabolomic analysis of key regulatory metabolites in hepatitis C virus-infected tree shrews. *Mol. Cell. Proteomics* 12: 710–719. [Medline] [CrossRef]

32. Tidona, C.A., Kurz, H.W., Gelderblom, H.R., and Darai, G. 1999. Isolation and molecular characterization of a novel cytotoxicparamyxovirus from tree shrews. *Virolology* 258: 425–434. [Medline] [CrossRef]

33. Walter, P. and Blobel, G. 1982. Signal recognition particle contains a 7S RNA essential for protein translocation across the endoplasmic reticulum. *Nature* 299: 691–698. [Medline] [CrossRef]

34. Walter, E., Keist, R., Niederost, B., Pult, I., and Blum, H.E. 1986. Hepatitis B virus infection of tupaiha hepatocytes in vitro and in vivo. *Hepatology* 24: 1–5. [Medline]

35. Wasley, A. and Alter, M.J. 2000. Epidemiology of hepatitis C: geographic differences and temporal trends. *Semin. Liver Dis.* 20: 1–16. [Medline] [CrossRef]

36. Wang, Q., Schwarzenberger, P., Yang, F., Zhang, J., Su, J., Yang, C., Cao, J., Ou, C., Liang, L., Shi, J., Yang, F., Wang, D., Wang, J., Wang, X., Ruan, P., and Li, Y. 2012. Experimental chronic hepatitis B infection of neonatal tree shrews (*Tupaia belangeri chinesis*): a model to study molecular causes for susceptibility and disease progression to chronic hepatitis in humans. *Virol. J.* 9: 170. [Medline] [CrossRef]

37. Yan, H., Zhong, G., Xu, G., He, W., Jing, Z., Gao, Z., Huang, Y., Qi, Y., Peng, B., Wang, H., Fu, L., Song, M., Chen, P., Gao, W., Ren, B., Sun, Y., Cai, T., Feng, X., Sui, J., and Li, W. 2012. Sodium taurocholate cotransporting polypeptide is a functional receptor for human hepatitis B and D virus. *Elife* 1: e00049. [Medline] [CrossRef]

38. Yan, R.Q., Su, J.J., Huang, D.R., Gan, Y.C., Yang, C., and Huang, G.H. 1996. Human hepatitis B virus and hepatocellular carcinoma. I. Experimental infection of tree shrews with hepatitis B virus. *J. Cancer Res. Clin. Oncol.* 122: 283–288. [Medline] [CrossRef]

39. Yang, M., Li, N., Li, F., Zhu, Q., Liu, X., Han, Q., Wang, Y., Chen, Y., Zeng, X., Lv, Y., Zhang, P., Yang, C., and Liu, Z. 2013. Xanthohumol, a main prenylated chalcone from hops, reduces liver damage and modulates oxidative reaction and apoptosis in hepatitis C virus infected Tupaia belangeri. *Int. Immunopharmacol.* 16: 466–474. [Medline] [CrossRef]

40. Yang, Z.F., Zhao, J., Zhu, Y.T., Wang, Y.T., Liu, R., Zhao, S.S., Li, R.F., Yang, C.G., Li, J.Q., and Zhong, N.S. 2013. The tree shrew provides a useful alternative model for the study of influenza H1N1 virus. *Virol. J.* 10: 111. [Medline] [CrossRef]

41. Xie, Z.C., Riezu-Boj, J.I., Lasarte, J.J., Guillen, J., Su, J.H., Civeira, M.P., and Prieto, J. 1998. Transmission of hepatitis C virus infection to tree shrews. *Virolology* 244: 513–520. [Medline] [CrossRef]