# A risk assessment of a common drug using xenograft model

**Masami Kodama**[1], **Junko Kurokawa**[1,2]

[1]Department of Bio-Informational Pharmacology, Medical Research Institute, Tokyo Medical Dental University, Tokyo, Japan; [2]Department of Bio-Informational Pharmacology, School of Pharmaceutical Sciences, University of Shizuoka, Shizuoka, Japan

*Correspondence to:* Junko Kurokawa, PhD. Department of Bio-Informational Pharmacology, School of Pharmaceutical Sciences, University of Shizuoka, 52-1 Yada, Suruga-ku, Shizuoka 422-8526, Japan. Email: junkokuro@u-shizuoka-ken.ac.jp.

*Provenance:* This is a Guest Commentary commissioned by Section Editor Hongcheng Zhu, MD, PhD (Department of Radiation Oncology, The First Affiliated Hospital of Nanjing Medical University, Nanjing, China).

*Comment on:* van den Driesche S, Macdonald J, Anderson RA, *et al*. Prolonged exposure to acetaminophen reduces testosterone production by the human fetal testis in a xenograft model. Sci Transl Med 2015;7:288ra80.

Submitted Dec 12, 2016. Accepted for publication Dec 19, 2016.
doi: 10.21037/atm.2017.01.59
**View this article at:** http://dx.doi.org/10.21037/atm.2017.01.59

Congenital cryptorchidism is the most common male reproductive disorders that is manifest at birth. It affects 2% to 9% of newborn boys (1) and the incidence have been reported to increase (2,3). Cryptorchidism confers an increased risk of infertility and testicular cancer in adulthood (3-5). The process of testis descent is dependent on hormones secreted by fetal testis. According to the studies using rat, fetal testosterone deficiency during a critical period in masculinization development leads to male reproductive disorders such as cryptorchidism (6-8). However, the environmental/lifestyle factors that reduce testosterone production have remained elusive, since it is not possible at present to decide whether rodent models mimic hormone secretion in human fetal testis.

Whether animal models can be used to predict human response to drugs and other chemicals is apparently a contentious issue. Recently, van den Driesche *et al*. proposed that chronic application of acetaminophen, a pain reliever and a fever reducer, attenuates testosterone production by human fetal testis using an original xenograft model (9). The xenograft model using human fetal testicular tissue can be a possible solution to translate experimental evaluation to clinical drug safety. Actually, some human studies mentioned an association between the use of acetaminophen in pregnant mothers and cryptorchidism in their sons.

Acetaminophen (N-acetyl-p-aminophenol, paracetamol) is a member of the aniline family of analgesics, which is widely available as over-the-counter drug. Because acetaminophen may be generally perceived as a safe drug, it is most commonly taken by pregnant women (1,10). Because acetaminophen is known to freely cross the placenta (2,11), the effects on fetus are needed to be considered for use during pregnancy. To date, several different birth cohort studies have associated maternal use of acetaminophen with increasing risk for reproductive disorders in male offspring (12-14).

Experimental investigation using rats revealed that intrauterine exposure to acetaminophen, with a lowest dose of 150 mg/kg/day, reduces the anogenital distance (AGD) (6), which is dependent on fetal exposure to testosterone in the male offspring (14). Also in *ex vivo* experiments based on the culture system of rat fetal testes (gestational day 14.5) for 1 to 3 days, acetaminophen directly inhibits testosterone production by the rat fetal testes (14,15). However, in the same *ex vivo* experiments using human fetal testis (7–12 weeks), testosterone production was unaffected by exposure to acetaminophen nor its metabolite N-(4-hydroxyphenyl)-arachidonoylethanolamide (AM404) at concentrations relevant to human therapeutic dose (16), although it remains unclear whether the contradiction reflects the differences of species or the differences existing in the culture systems.

Because it is impossible to investigate directly the effects on human fetal testis development for ethical reasons, the development of appropriate model is necessary for study of human fetal testis development. In this context, the xenograft model of human fetal testis was developed as a new approach to investigate human fetal testis development (17).

Small pieces of human fetal testis xenografted under the dorsal skin of male castrated nude mice were maintained for 7 days. The xenografts showed more than 75% survival with normal morphology and produced testosterone when host mice were treated with human chorionic gonadotropin (hCG) to mimic human in utero environment. Using human fetal testes (14 to 20 weeks), van den Driesche *et al*. tried to evaluate the effect of exposure to acetaminophen on testosterone production (9). They tested three different doses and regimens: (I) a single high dose (350 mg/kg) of acetaminophen administered orally daily for 7 days; (II) a therapeutic dose (20 mg/kg) administered orally 3 times a day for 7 days; (III) a therapeutic dose (20 mg/kg) administered orally 3 times a day for a single day. They showed that 1 week's exposure to acetaminophen, even a human-equivalent therapeutic regimen, results in reduced testosterone production by xenografted human fetal testis tissue, whilst short-term (1 day) use does not result in any long-lasting suppression of testosterone production.

We wonder how much impacts these results have on drug safety problem. The purpose of the test using animal models is to predict human response then the tests must be evaluated by how well it conforms to human response. The xenograft model in pseudo-human in utero environment (9) is applicable to improve predictability of reproductive potentials, which has a large impact on drug safety on chronic toxicity tests. At the same time, the authors used a rat model to confirm the difference between acute and chronic exposure of acetaminophen then to unravel the molecular mechanisms, because availability of human tissue is limited. Although the limitation exists, key experiments using the xenograft model must be a robust predictor of human response so far. It has not been determined whether the prediction would be also a potential guide for decision making in drug safety. Further work, especially with retrospective clinical studies, will be necessary to use the xenograft system as a guide for decision making.

## Acknowledgements

*Funding:* This work is supported by a fellowship from JSPS (RPD) and grants from AMED (16mk0104027h1102, 16mk0104007h0003) and MEXT (15H04684).

## Footnote

*Conflicts of Interest:* The authors have no conflicts of interest to declare.

## References

1. Bay K, Main KM, Toppari J, et al. Testicular descent: INSL3, testosterone, genes and the intrauterine milieu. Nat Rev Urol 2011;8:187-96.
2. Virtanen HE, Toppari J. Epidemiology and pathogenesis of cryptorchidism. Hum Reprod Update 2008;14:49-58.
3. Serrano T, Chevrier C, Multigner L, et al. International geographic correlation study of the prevalence of disorders of male reproductive health. Hum Reprod 2013;28:1974-86.
4. Lee PA, Coughlin MT. Fertility after bilateral cryptorchidism. Evaluation by paternity, hormone, and semen data. Horm Res 2001;55:28-32.
5. Prener A, Engholm G, Jensen OM. Genital anomalies and risk for testicular cancer in Danish men. Epidemiology 1996;7:14-9.
6. Welsh M, Saunders PT, Fisken M, et al. Identification in rats of a programming window for reproductive tract masculinization, disruption of which leads to hypospadias and cryptorchidism. J Clin Invest 2008;118:1479-90.
7. Macleod DJ, Sharpe RM, Welsh M, et al. Androgen action in the masculinization programming window and development of male reproductive organs. Int J Androl 2010;33:279-87.
8. van den Driesche S, Scott HM, MacLeod DJ, et al. Relative importance of prenatal and postnatal androgen action in determining growth of the penis and anogenital distance in the rat before, during and after puberty. Int J Androl 2011;34:e578-86.
9. van den Driesche S, Macdonald J, Anderson RA, et al. Prolonged exposure to acetaminophen reduces testosterone production by the human fetal testis in a xenograft model. Sci Transl Med 2015;7:288ra80.
10. Werler MM, Mitchell AA, Hernandez-Diaz S, et al. Use of over-the-counter medications during pregnancy. Am J Obstet Gynecol 2005;193:771-7.
11. Byer AJ, Traylor TR, Semmer JR. Acetaminophen overdose in the third trimester of pregnancy. JAMA 1982;247:3114-5.
12. Jensen MS, Rebordosa C, Thulstrup AM, et al. Maternal use of acetaminophen, ibuprofen, and acetylsalicylic acid during pregnancy and risk of cryptorchidism. Epidemiology 2010;21:779-85.
13. Kristensen DM, Hass U, Lesné L, et al. Intrauterine exposure to mild analgesics is a risk factor for development of male reproductive disorders in human and rat. Hum Reprod 2011;26:235-44.

 

14. Snijder CA, Kortenkamp A, Steegers EA, et al. Intrauterine exposure to mild analgesics during pregnancy and the occurrence of cryptorchidism and hypospadia in the offspring: the Generation R Study. Hum Reprod 2012;27:1191-201.
15. Kristensen DM, Lesné L, Le Fol V, et al. Paracetamol (acetaminophen), aspirin (acetylsalicylic acid) and indomethacin are anti-androgenic in the rat foetal testis. Int J Androl 2012;35:377-84.
16. Mazaud-Guittot S, Nicolas Nicolaz C, Desdoits-Lethimonier C, et al. Paracetamol, aspirin, and indomethacin induce endocrine disturbances in the human fetal testis capable of interfering with testicular descent. J Clin Endocrinol Metab 2013;98:E1757-67.
17. Mitchell RT, Saunders PT, Childs AJ, et al. Xenografting of human fetal testis tissue: a new approach to study fetal testis development and germ cell differentiation. Hum Reprod 2010;25:2405-14.

**Cite this article as:** Kodama M, Kurokawa J. A risk assessment of a common drug using xenograft model. Ann Transl Med 2017;5(4):88. doi: 10.21037/atm.2017.01.59

 