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Synthesis and characterization of novel quick-release propofol prodrug via lactonization

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ABSTRACT

The water-soluble derivatives of propofol have gained attention as a method to increase solubility of propofol. According to the principle of lactonization, the lead compound HX0969 was synthesized first and then the pharmacological features of HX0969 were evaluated in a comparison with those of propofol in the SD rats. Then, HX0969 disodium phosphate monoester (HX0969W) and glycine ester trifluoroacetic acid salt (HX101230) were synthesized, and their pharmacological features were compared with those of Lusedra®, which has been recognized and marketed as a water-soluble prodrug of propofol since 2008. The results showed that HX0969 could produce an anesthetic effect within a few seconds (3.6 ± 3.0 s) and its therapeutic index was 4.66 in the SD rat. The pharmacodynamic characteristics of HX0969W were similar to those of the Lusedra®. HX101230 could still produce an anesthetic effect within 60 s in the rats though its therapeutic index was not so high (TI = 2.96). Therefore, our study has indicated that HX0969 is a potentially useful lead compound of propofol derivative. Its rapid anesthetic effect is probably associated with lactonization.

Propofol is an intravenous anesthetic used frequently in clinical practice. It has characteristics of rapid onset, rapid metabolization and rapid elimination. The raw propofol is insoluble in water, so the researchers have used some pharmaceutical methods to improve its water solubility for intravenous administration.[1] One fat emulsion of propofol named Diprivan® has become a popular formulation.[2] Now, the researchers have been focused on the water-soluble prodrugs of propofol because this fat emulsion has its disadvantages, for example, an injection pain and a bacterial infection as well as some other adverse effects observed in the patient's fat metabolism.[3] Some propofol solutions have successively been created by the medicinal chemistry methods, and Lusedra® is the most outstanding propofol solution, which was firstly marketed in the United States in 2008. Its active ingredient is fospropofol disodium (GPI15715) (Fig. 1).[4] GPI15715 can quickly release propofol, but its by-product, formaldehyde, has a safety risk for infusion. In addition, the intermediate, hemiacetal of propofol, is unstable and actually not available, so it cannot be modified conveniently as a lead compound.

To overcome the disadvantages of Lusedra®, design of a prodrug that can be decomposed quickly in vivo is an effective and efficient strategy, that is, design of a novel quick-release propofol prodrug via lactonization.[5] So, the purpose of our study was to investigate the synthesis and the characterization of this kind of propofol prodrug, that is, HX0969. When HX0969 was synthesized, the stability test of HX0969 in the rat plasma was conducted. The result showed that HX0969 could release propofol quickly in the plasma (Fig. 2) and no decomposition was observed in the water–ethanol and emulsion.

In order to make sure whether lactonization or normal ester hydrolysis is the mechanism for the propofol release from HX0969, we measured butyrolactone, a theoretical breakdown product of HX0969, when HX0969 was decomposed in the plasma. However, butyrolactone was also rapidly metabolized to 4-hydroxybutyraten (GHB) in the body, and the determination of butyrolactone could be interfered by the matrix. If the dose of butyrolactone was not great enough, butyrolactone would be difficult to be determined accurately in the plasma.[6] So, the propofol release mechanism was verified by the medicinal chemistry method in the following procedures: compounds **1** and **2** (analogues of HX0969) were synthesized (Fig. 3), both of which were insoluble in water. Compounds **1**, **2** and HX0969 were prepared as an emulsion. They were compared with Diprivan®, and their pharmacodynamic characteristics were evaluated in the SD rats (Table 1).

HX0969 could produce anesthesia in the SD rat very quickly (onset time = 3.6 ± 3.0 s). Compound **1** injected in a dose of 40 mg/kg through the tail vein could induce systemic convulsions rather than anesthesia effects for 3.8 ± 2.8 min in all the rats, which revealed its severe toxicity. When the dose of compound **1** was decreased to 34 mg/kg, 3 of the 8 rats did not exhibit anesthesia effects or

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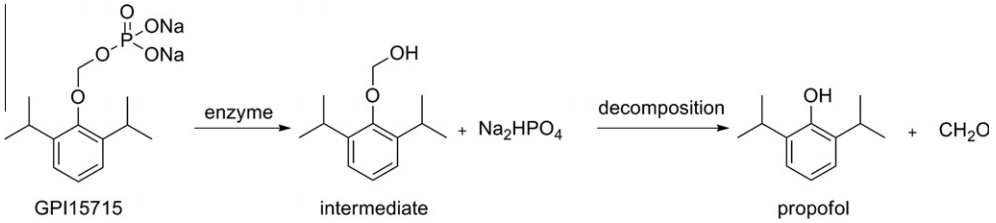


Figure 1. Decomposition of GPI15715.

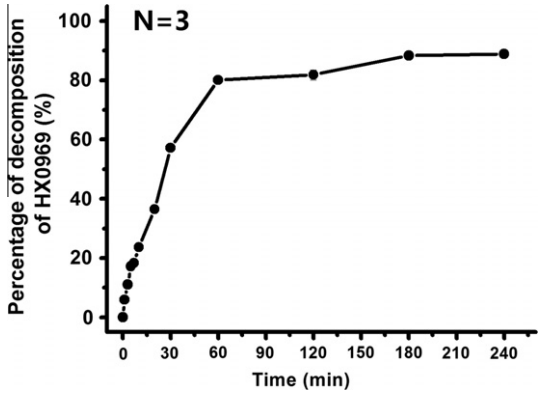


Figure 2. The decomposition rate of HX0969 in the plasma.

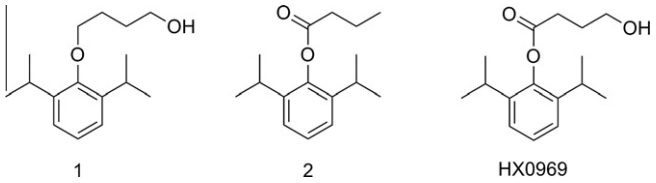


Figure 3. Three propofol derivatives.

Table 1
Pharmacodynamic characteristics of HX0969 and Diprivan®

Compound	ED_{50} (mg/kg)	LD_{50} (mg/kg)	TI	OT[a] (s)	DT[a] (min)
HX0969	17.48 ± 2.11	81.55 ± 9.85	4.67	3.6 ± 3.0	24.36 ± 4.87
Diprivan®	6.57 ± 0.91	27.08 ± 4.25	4.12	0.00[b]	8.41 ± 1.47

Data are presented as mean ± SD in all the eight measurements except TI (Therapeutic index).

[a] OT and DT, respectively stand for the onset time and the duration time, that is, the time from the injection of the drug to the disappearance of the righting reflex and the time from the disappearance of the righting reflex to the recovery of the righting reflex. The dosage was the two-fold of ED_{50} for each of the two drugs when OT and DT were determined, that is, HX0969: 35 mg/kg, and Diprivan®: 13 mg/kg, administered via the tail vein.

[b] The onset during the injection.

seizures, but 5 of the 8 rats still exhibited systemic convulsions. Compound **2** did not produce anesthesia effects when used in a dose

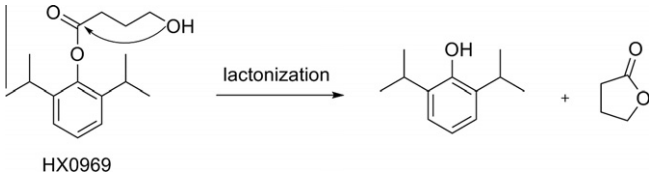


Figure 4. Decomposition of HX0969.

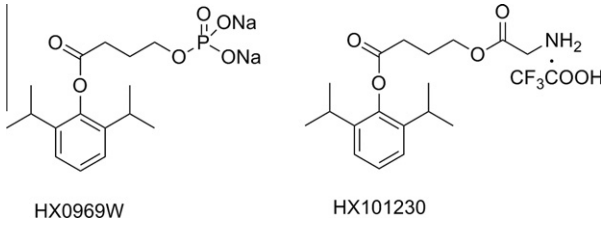


Figure 5. The structure of HX0969 and HX101230.

Table 2
Pharmacodynamic characteristics of HX0969W, HX101230 and GPI15715

Compound	ED_{50} (mg/kg)	LD_{50} (mg/kg)	TI	OT[a] (min)	DT[a] (min)
GPI15715	34.78 ± 4.14	163.11 ± 19.71	4.69	2.17 ± 0.32	31.04 ± 7.90
HX0969W	44.50 ± 5.30	288.47 ± 34.30	6.48	2.11 ± 0.23	27.49 ± 5.73
HX101230	14.42 ± 1.98	42.68 ± 5.90	2.96	0.26 ± 0.16	12.74 ± 2.84

Data are presented as mean ± SD in all the eight measurements except TI.

[a] OT and DT, respectively stand for the onset time and the duration time, that is, the time from the injection of the drug to the disappearance of the righting reflex and the time from the disappearance of the righting reflex to the recovery of the righting reflex, the dose of the drug is the two-fold of ED_{50} (GPI15715: 70 mg/kg, HX0969W: 89 mg/kg, HX101230: 29 mg/kg) injected via the tail vein.

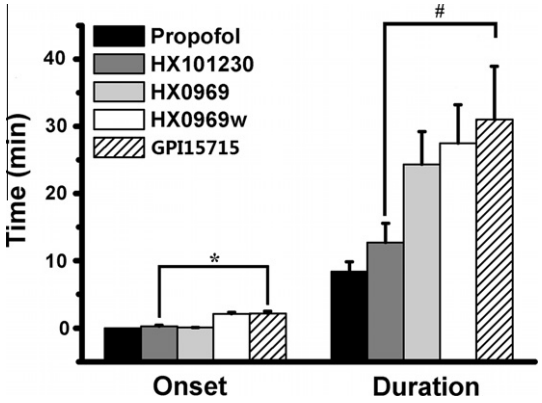


Figure 6. The onset time and the duration time in the rats when the two-fold of ED_{50} of each drug was administered. No significant difference was found in the onset time between HX0969 and Diprivan® ($P = 0.568$). No significant difference was found in the onset time between HX0969W and GPI15715 ($P = 0.568$). No significant difference was found in the duration time between HX0969W and GPI15715 ($P = 0.171$). Both the onset time and the duration time for HX101230 were shorter than those for GPI15715 ($P < 0.01$ for the onset time, $P < 0.001$ for the duration time). $^{*}P < 0.01$, $^{\#}P < 0.001$

of 200 mg/kg. These findings indicated that with no structure of lactonization, compounds **1** and **2** would not be safe or effective in the production of anesthesia. So, the lactonization was probably a mechanism for the rapid release of propofol from HX0969 (Fig. 4).

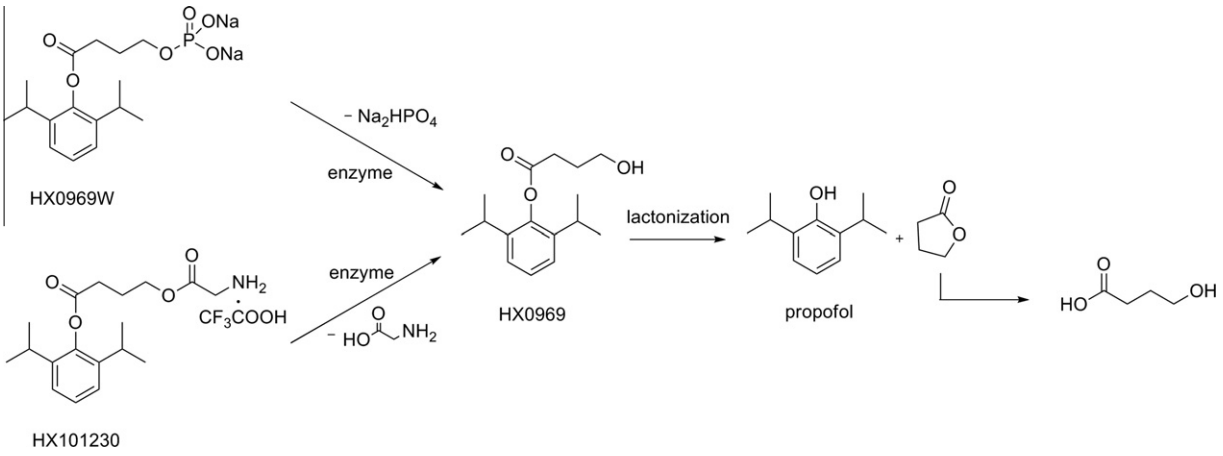
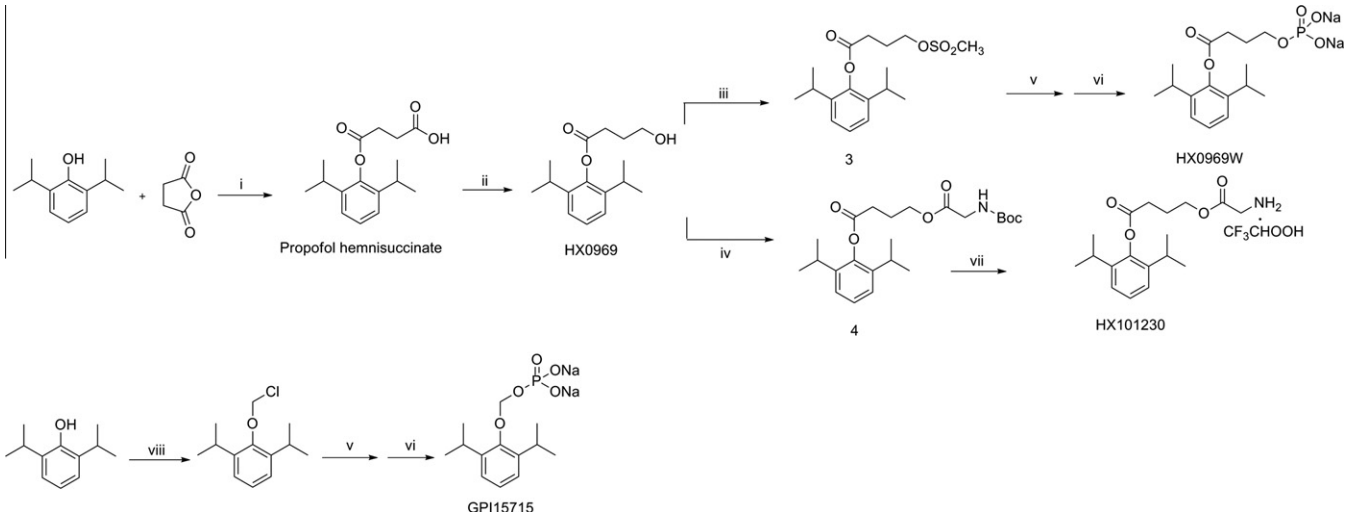


Figure 7. Decomposition of HX0969W and HX101230 in vivo.



Scheme 1. Reaction conditions: (i) Et_3N; (ii) $NaBH_4/I_2$; (iii) MsCl/pyridine; (iv) Gly-Boc/DCC; (v) H_3PO_4/Et_3N; (vi) NaOH; (vii) CF_3COOH; (viii) $ClCH_2I/NaH$.

A phosphate-linked form of HX0969 synthesized in our study could also release propofol quickly, which was its sodium salt, a water-soluble derivative of propofol named HX0969W. Then, the trifluoroacetic acid salt of glycine ester of HX0969 was synthesized, which was named HX101230 (Fig. 5). Both the compounds were soluble in water. HX0969W, HX101230 and GPI15715 were formulated into aqueous solutions and were administered to the SD rats via the tail vein for the investigation on their pharmacodynamic characteristics (Table 2).

As a result, HX0969W was similar to GPI15715 in some of the pharmacodynamic characteristics, including the duration time (DT) (time from disappearance of righting reflex to recovery of righting reflex) ($P = 0.171$). The lead compound HX0969 could produce a very rapid onset of anesthesia but no significant difference was found when compared with Diprivan® ($P = 0.568$). HX101230 produced a significantly more rapid anesthesia effect than GPI15715 ($P < 0.01$) or HX0969W ($P < 0.01$), and its anesthesia duration time was significantly shorter than that by GPI15715 ($P < 0.001$) or by HX0969W ($P < 0.01$), but the therapeutic index of HX101230 was not high (TI = 2.96). The simulation results showed that HX0969 could be modified conveniently to obtained some water-soluble derivatives of propofol (Fig. 6). These derivatives could produce anesthesia quickly in vivo. We speculated that HX0969W and HX101230 were decomposed first to generate

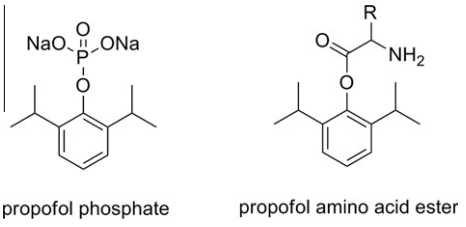


Figure 8. The slow-release derivatives of propofol.

HX0969 under the action of alkaline phosphatase or esterase in vivo; then, HX0969 released propofol by lactonization (Fig. 7).

Propofol hemisuccinate was synthesized from propofol using succinic anhydride.[7] HX0969 was synthesized by the sodium borohydride–iodine system, an excellent method for reduction of carboxylic acid.[8] HX0969W was synthesized from sulfonyl, which was a good leaving group. GPI15715 was synthesized from propofol using chloroiodomethane and phosphoric acid[7] (Scheme 1). Chloroiodomethane was synthesized according to the method previously reported.[9]

The water solubility of propofol prodrugs has received enough attention but not enough attention has been paid on the release rate of propofol. There have been some notable failures, such as

propofol disodium phosphate esters and propofol amino acid ester derivatives[10,11] (Fig. 8). These prodrugs were unable to meet clinical requirements due to their slow propofol release rate. If the propofol release was not rapid enough, the anesthetic dosage had to be increased, which inevitably led to an accumulation of the anesthetic. Thus, the main advantages of propofol, including the rapid recovery, would be lost. As a drug development project, we consider it a failure.

The main reason for the slow release of propofol from the prodrugs obtained by the modification of the phenolic hydroxyl group is the slow hydrolysis rate of the pro-moiety due to the steric hindrance with two ortho-isopropyl groups. When dynamic force is strong enough to promote decomposition of the prodrug, the propofol release rate would become rapid. For example, when GPI15715 was decomposed to a hemiacetal, the unstability of the intermediate would become a strong dynamic force to induce the rapid propofol release. HX0969 can produce anesthesia in the SD rat by a rapid release of propofol. Since the primary hydroxyl group of HX0969 is not severely hindered, a variety of second pro-moieties such as phosphate and amino acid ester can be attached to increase the water-solubility of HX0969. Such pro-moieties can be smoothly hydrolyzed by enzymes in vivo to generate HX0969, which is quickly converted to propofol probably via lactonization.

The breakdown product of HX0969 is butyrolactone in theory, which will be converted to 4-hydroxybutyric acid within a short period of time in vivo, as an endogenous substance in mammals, 4-hydroxybutyric acid has a low toxicity and can be metabolized into the Krebs cycle.[12,13] There has been no report about the acute toxicity of butyrolactone injected through the rat tail vein. However, there has been a report about the effect of butyrolactone on the local cerebral glucose utilization in the rat after its tail vein injection, but no significant changes were found in the rat behavior in the 75 or 150 mg/kg dose group.[14] Additionally, there has been a report about the effect of butyrolactone on the activity of the rat after the administration of butyrolactone 100 or 200 mg/kg, but no toxicity was reported.[15] The release of butyrolactone was only 20 mg/kg when administered with the two-fold of ED_{50} for HX0969w in the rat, which was much lower than the dose of butyrolactone reported in the literature; therefore, we speculated that butyrolactone released from HX0969w might not cause any significant toxicity. Butyrolactone will be transformed to 4-hydroxy-butyric acid in a short time in vivo, the latter is a clinically used drug. A report showed that an intravenous administration of 4-hydroxybutyric acid sodium 200 mg/kg induced sleep in the rat.[16] Another report was focused on the relationship between the sleep duration of the rat and the concentration of 4-hydroxybutyraten (GHB) in the rat body fluid when 400–800 mg/kg of 4-hydroxybutyric acid was administered.[17] The release of 4-hydroxybutyric acid was only 24 mg/kg when administered with the two-fold of ED_{50} for HX0969w in the rat, which was much lower than the dose of 4-hydroxybutyric acid for the sleep-inducing effect on the rat reported in the literature; therefore, we have believed that the by-product of HX0969W used for anesthesia has no toxicity or a lower toxicity.

HX0969 was found to be sensitive to the plasma but no quick decomposition was detected in the emulsion or the aqueous ethanol solutions without enzymes (Supplementary data). But the exact kinds of enzymes that decompose HX0969 or HX0969W remain to be established.

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Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.bmcl.2013.01.034.

References and notes

1. Baker, M. T.; Naguib, M. *Anesthesiology* **2005**, *103*, 860.
2. Sebel, P. S.; Lowdon, J. D. *Anesthesiology* **1989**, *71*, 260.
3. Picard, P.; Tramèr, M. R. *Anesth. Analg.* **2000**, *90*, 963.
4. Schywalsky, M.; Ihmsen, H.; Tzabazis, A.; Fechner, J.; Burak, E.; Vornov, J.; Schwilden, H. *Eur. J. Anaesthesiol.* **2003**, *20*, 182.
5. Gomes, Paula; Vale, Nuno; Moreira, Rui *Molecules* **2007**, *12*, 2484.
6. Palmer, R. B. *Toxicol. Rev.* **2004**, *23*, 21.
7. Hwndler Sheldon, S.; Sanchez Robert, A.; Zielinski, Jan. U.S. Patent 6,254,853, 2001.
8. Bhaskar Kanth, J. V.; Mariappan, P. *J. Org. Chem.* **1991**, *56*, 5964.
9. Altabev, N.; Snith, R. D.; Suratwala, S. I. *Chem. Ind.* **1973**, 331.
10. Banaszczyk, M. G.; Carlo, A. T.; Millan, V.; Lindsey, A.; Moss, R.; Carlo, D. J.; Hendler, S. S. *Anesth. Analg.* **2002**, *95*, 1285.
11. Gallop, M. A.; Xu, F.; Cundy, K. C.; Sasikumar, V.; Woiwode, T. W. U.S. Patent 20,050,004,381, 2005.
12. Waszkielewicz, A.; Bojarski, J. *Pol. J. Pharmacol.* **2004**, *56*, 43.
13. Doherty, J. D.; Roth, R. H. *J. Neurochem.* **1978**, *30*, 1305.
14. Wolfson, L. I.; Sakurada, O.; Sokoloff, L. *J. Neurochem.* **1977**, *29*, 777.
15. Hampel, H.; Hapke, H. J. *Arch. Int. Pharmacodyn. Ther.* **1968**, *171*, 306.
16. Depoortere, H.; Rousseau, A.; Jalfre, M. *Rev. Electroencephalogr. Neurophysiol. Clin.* **1977**, *7*, 153 [Article in French].
17. Roiko, S. A.; Felmlee, M. A.; Morris, M. E. *Drug Metab. Dispos.* **2012**, *40*, 212.