A Novel Time-Controlled Release System Based on Drug–Resin Complexes and Elementary Osmotic Pump

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A novel time-controlled system based on elementary osmotic pump tablet containing a drug–resin complexes (DRCs) core is presented. In the traditional osmotic pump tablets (OPTs), the lag time was always minimized. On the contrary, in the DRCs osmotic pump tablet (DRCOPT), the lag time was increased to achieve time-controlled delivery. The system led to a zero-order drug release after an initial lag time. Polyethylene oxide (PEO) N80 was used as suspension agent and NaCl was applied as ion-exchange, osmotic pressure (electrolyte supplementary) agent, respectively. To examine the mechanism of this system, drug release behaviors were investigated under conditions of various osmotic pressures. A new method of combination of conductivity and HPLC was applied to determine the different fractions of NaCl in producing osmotic pressure, ion-exchange and electrolyte supplement. The pharmacokinetic studies conducted in beagle dogs showed that a steadier and controlled drug release behavior was obtained compared with the traditional formulations. On the basis of prescription of the DRCOPT, a good *in-vitro–in-vivo* correlation (IVIVC, R^2=0.9541) was achieved. In addition, a lag time of 4 h was observed in *in vivo* experiment, which indicated that the DRCOPT can be used in therapeutic regimens with the characteristics of chronotherapy.

Key words drug–resin complexes; osmotic pump tablet; time-controlled delivery; drug release mechanism; propranolol hydrochloride

The osmotic pump tablet (OPT) is an advanced drug-delivery technology that uses osmotic pressure as the driving force to deliver pharmacotherapy, which was developed as an oral drug delivery system. In the historical development of OPT, many achievements for its promotion, including the Rose–Nelson pump,[1] Higuchi–Leeper pump,[2,3] elementary osmotic pump tablet (EOPT),[4] and push-pull system.[5] In recent years, many novel technologies and formulations related have been developed, such as osmotic drug delivery using swellable-core technology,[6] effervescent OPT from traditional Chinese medicine compound recipe,[7] $(SBE)_{7m}$-β-CD in controlled-porosity OPT [8] and other new approaches.

But it is not always desirable that a controlled release formulation has a zero-order release of its contents. Oral dosage forms with time-controlled drug release have become of growing interest in recent years. Pharmaceutical scientists have displayed considerable ingenuity in the development of time-controlled drug delivery systems to meet the emerging chronotherapeutic requirements. Possible applications include the topical treatment of diseases in the gastrointestinal (GI) tract such as ulcerative colitis,[9] or are considered for chronopharmacological aspects as described for asthma, heart diseases or arthritis.[10] It is essential to develop a time-controlled release system to achieve an effective drug level only at the required time. One of the objectives of this study was to develop a novel EOPT for time-controlled release system using the cores of drug–resin complexes (DRCs).

Ion exchange resins (IERs) are cross-linked water-insoluble polymers carrying ionizable functional groups. The resins have been used in various pharmaceutical applications, primarily for taste masking and for controlled release systems in liquid or solid form.[11—13] IERs behave, for some drugs, as reliable controlled drug delivery systems.[14] However, improvements of their release properties can be affected by coating the resin beads and further controlling the rate of drug release.[15—18] In these forms, the pattern of drug release is governed by the degree of cross-linking of the resins and by the properties of the coat.

Propranolol hydrochloride (PNH) was chosen as the model drug, which is a nonselective Beta-adrenergic blocking agent, has been widely used in the treatment of hypertension, angina pectoris, and many other cardiovascular disorders.[19] Patients with cardiovascular disease are at the greatest risk of heart attack and stroke during the early hours of the morning (02:00), and there is a need for adequate control of hypertension during this vulnerable period. It is necessary to develop a time-controlled system to achieve an effective drug level only at the demanded time.

In previous studies, DRCs have been employed in the controlled release and numerous types of materials such as poly (4-vinylpyridine), sulphonic acid cation exchange resin, and sodium polystyrene sulfonate, and there were several reports on PNH–resin complexes, for instance, calcium alginate beads loaded with PNH–resin complexes,[20] poly (acrylic acid) grafted poly (vinylidene fluoride) membrane,[21] and polymeric microparticles.[22] The first report concerning the use of a DRC to modulate osmotic pump tablets was published in 1990.[23] Furthermore, the effects of IERs on the release of PNH-matrix tablets were studied in 1998.[24]

The paper focused on the combination of an OPT and IERs to prepare a novel osmotic pump tablet for time-controlled delivery. Unlike other EOPT systems,[25] this delivery system showed a steady zero-order release profile after an initial lag time, which is the difference between the DRCOPT and other approaches.

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Experimental

Materials Propranolol HCl (Rouz Darou Co.), the cation-exchange resins Amberlite® IRP69 (sodium polystyrene sulfonate) was obtained from Rohm and Haas Company, Philadelphia, U.S.A.; sodium chloride (Tianjin Bo-di Chemical Industry, Tianjin, China), cellulose acetate (CA, Shanghai Chemical Reagent, Shanghai, China), polyethylene oxide (PEO, Dow Chem., New Jersey, U.S.A.), polyethylene glycol 4000 (PEG, Shenyang Chemical Reagent, Shenyang, China) and polyvinylpyrrolidone (PVP, ISP Technologies Inc., U.S.A.) were employed in the experiments. All other reagents used were of analytical grade.

Preparation of the DRCOPT DRCs were prepared by the batch method. In this, 10 g IERs were suspended in 1000 ml aqueous solution with the concentration of 10 mg/ml PNH under magnetic stirring at 30 °C for 4 h. The DRCs were washed free of the unexchanged drug with deionized water. The drug-loaded resin beads thus obtained were dried in a fluid bed drier at 40 °C.

Granules were prepared in the laboratory scale high shear mixer (Micro-Gral®, Collette, Belgium). Absolute alcohol was used as the binder liquid. A powder mixture containing 30% dried drug-loaded resin beads, 55% PEO, 9% sodium chloride and 5% PVP were used as raw material and the total mass for each batch is 100 g. All the components were sieved through 160 mesh before granulating, respectively. The powders were filled into the vessel and premixed for 3 min using the impeller speed of 3000 rpm and the chopper was off during this stage. A precisely determined amount of binder liquid (30 ml) was then added to the powder mix using a titration device (Universal Titronic, Schott, Germany) in 5 min while the impeller and chopper (1000 rpm) were running, and the mass was mixed for a preset period of time after the liquid addition. The granules produced were dried to constant weight in a tray dryer at 40 °C and then stored in sealed bags. To the dried granules (sieved through 20 mesh), 1% sieved magnesium stearate was added and blending continued for 10 min longer. The final powders were compressed using a single-punch tableting machine (Shanghai Huanghai Drug Inspection Instrument, Shanghai, China) with a 9.0 mm bulgy-faced punch to yield a 0.33 g tablet each time. The hardness of the tablets was kept constant (8 kg, hardness tester, idem, Shanghai).

The tablet core was coated (BY300A Coating Machine, Shanghai Huanghai Drug Inspection Instrument, Shanghai) with coating solution composed of 30 g CA dissolved in 970 ml acetone and 6 g PEG 4000 dissolved in 30 ml distilled water to prepare a semipermeable membrane. A 0.6 mm orifice was drilled on the membrane surface mechanically. In this study, tablet was coated to the 8% weight gain of the core.

The Condition of HPLC The chromatographic condition of HPLC was as follow: The HPLC system consisted of a pump (LC-10ATVP HPLC pump, Shimadzu, Japan), a UV detector (SPD-10AVP Spectrometer, Shimadzu, Japan). The chromatography column was an ODS column (Diamonsil C_{18} 200 mm×4.6 mm, 5 μm, Dikma, Beijing). The mobile phase consisted of methanol–potassium dihydrogen phosphate solution–triethylamine (54 : 46 : 0.5, v/v/v) *in vitro* and (44 : 56 : 0.5, v/v/v) *in vivo*, in addition, pH was adjusted to 3.0 by phosphoric acid for both mobile phases. The flow rate was 1.0 ml/min. Detection was performed at a wavelength of 290 nm under a constant temperature at 30 °C. All reagents were of analytical grade.

Dissolution Tests The release of drug from the DRCOPT (equivalent to PNH 40 mg) was determined using the USP paddle method (50 rpm, 37±0.5 °C). The dissolution medium (900 ml) was 0.15 mol/l NaCl solution. Sample of 5.0 ml was withdrawn at 1.0, 2.0, 3.0, 4.0, 6.0, 8.0, 10.0, 12.0, 14.0, 16.0, 24.0 h and replaced with 5.0 ml fresh dissolution media. Analysis of dissolved PNH at each time point was conducted by HPLC method mentioned above.

To investigate the effects of the pH value of the dissolution media on the DRCOPT release behavior, release tests were carried out in 1) simulated gastric fluid (SGF), pH 1.2; 2) simulated intestinal fluid (SIF), pH 6.8; 3) SGF for 2h and SIF for up to 24 h; 4) simulated colonic fluid (SCF), pH 7.4. All of the dissolution media were adjusted to the same ionic strength with NaCl solution.

Sodium chloride solutions with different ionic strength of 0.05 mol/l, 0.10 mol/l, and 0.15 mol/l were prepared to study the effects of ionic strength on release behavior.

Dissolution tests were conducted at different rotation speed of 50 rpm, 75 rpm, and 100 rpm to investigate the effects of rotation speed on release behavior.

Determination of NaCl and PNH Concentration in a Binary System by Conductivity-HPLC Method Based on the principle of electrochemistry, a relationship between conductivity and equivalent concentrations of the components in a binary solution was derived, which was elucidated by the standard curve of conductivity. The concentration in a binary system can be determined from those of their corresponding pure component, therefore, it was easy and fast to evaluate the concentration of one component with a predetermined ion concentration from the other component. This method may be expected to become a useful supplement to chemical analysis or instrumentation.

According to the theory of electrochemistry, the relationship between conductivity of solution and respective ion in the diluted solution consisting of concurrent ions was given below.

$$\lambda = F\sum |Z_i|\, c_i \mu_i \tag{1}$$

Where F is Faraday constant, Z_i, c_i, μ_i is the charge number, concentration and mobility (*i.e.* the motion of ion in the electric field), respectively. As far as a binary system composed of AbBa and CdDc is concerned, whose conductivity is expressed as Eq. 2 according to Eq. 1:

$$\lambda = F(bc_A\mu_A + ac_B\mu_B) + F(dc_C\mu_C + cc_D\mu_D) \tag{2}$$

Because $bc_A = ac_B = N_{AB}$, $dc_C = cc_D = N_{CD}$, Eq. 2 can be simplified as Eq. 3:

$$\lambda = F(\mu_A + \mu_B)N_{AB} + F(\mu_C + \mu_D)N_{CD} \tag{3}$$

Where N_{AB} and N_{CD} is the molar concentration of AbBa and CdDc, respectively. In a specific condition, μ_i (i=A, B, C, D) is only concerned with the property of ion, which can be regarded as being a constant. However, the deionized water is not absolutely pure. Even if there is not any ion in the solution, the conductivity is not equal to zero. As a result, a constant λ_0 should be added, so the Eq. 1 is modified to Eq. 4:

$$\lambda = K_{AB}N_{AB} + K_{CD}N_{CD} + \lambda_0 \tag{4}$$

From Eq. 4, a linear relationship between composite components in a binary system and their molar concentration is obtained. Proportionality factors K_{AB}, K_{CD} are exclusively relevant to the property of pure component, which can not be affected by the existence of the other component. K_{AB} and K_{CD} can be obtained by the independent experiment, especially for the incorporated system composed of monovalence ion, the conductivity of incorporated system is directly proportional to the molar concentration of each component, which is coincident with the behavior of incorporated ion in the charged membrane. According to Eq. 4, concentration of a component can be defined by determining the conductivity of incorporated system and the concentration of other component that can be measured easily.

Unlike in the conventional OPTs, NaCl was only used as osmotic pressure agent. In the core of the DRCOPT, NaCl takes on the contribution in three aspects, *i.e.*, osmotic pressure agent, ion-exchange agent and electrolyte supplementary agent. To determine the different function of sodium chloride in the DRCOPT, the theory expounded above was applied to the determination of NaCl and PNH concentration in solution.

Pharmacokinetic Studies in Dogs Healthy beagle dogs (9.3±1.0 kg, from the Lab Animal Center of Shenyang Pharmaceutical University, Shenyang, China, Grade I) were used. Six dogs were used in this study and separated into 2 groups. They were fed standard laboratory chow with water and fasted overnight before the experiments. The *in vivo* study was approved and performed complying with the guidelines of the Institutional Animal Ethics Committee. The dogs were fasted for 24 h prior to dosing, fed at 12 h post-dosing, and water was available *ad libitum* throughout the study period. Six dogs were given commercially available tablets (propranolol hydrochloride tablet, 10 mg/tablet) and the DRCOPT in a random cross-over design. The dose of reference tablets and the DRCOPT administered was both 120 mg PNH per beagle dog. After a washout period of 7 d, the study was repeated in the same manner to complete the cross-over design.

To 1.0 ml plasma, 1.0 ml internal standard solution (Bisoprolol Fumarate, Beijing Lunarsun Science & Technology Ltd., China) and 0.6 ml 0.03 mol/l NaOH were added and then vortexed uniformly. Thereafter, the resultant sample was extracted by 4 ml skellysolve C–isobutanol mixture (95 : 5, v/v) for 3 min. The organic layer was separated by centrifugation at 4000 rpm for 10 min and then transferred into a centrifuge tube mixed with 1 ml 0.02 mol/l HCl by vortexing for 3 min. The mixture was centrifuged at 4000 rpm for 10 min. The organic layer was discarded, and a 20 μl remnant sample solution was injected into the HPLC with 290 nm as the detection wavelength for analysis. The same procedure was used to determine the recovery and precision in plasma. The lower detection limit by this method was 10 ng/ml. Recovery and accuracy of the method were reasonable.

1.0 ml PNH solutions of different concentrations were added into 1.0 ml blank plasma of Beagle dogs respectively to prepare the plasma sample with

concentrations ranged from 20 to 1000 ng/ml, after this, 1.0 ml internal standard solution was added into the different plasma samples accurately. According to the procedure mentioned above, plasma samples were gained. The concentrations of PNH in plasma (x) were determined from the peak area ratios (y) of PNH to internal standard using the linear regression equation obtained from the calibration curve. The *in vivo* calibration curve of PNH was gained as follow: $y=0.0062x+0.2445$ ($r=0.9970$).

All data were subsequently processed by the computer program DAS 2.0 (Anhui Provincial Center for Drug Clinical Evaluation, China). Overall elimination constant rate K_e was obtained by the means of residuals method. All data were expressed as the mean±S.D. The relative bioavailability (Fr) of the conventional tablet (reference tablet) compared with the DRCOPT (test tablet) was calculated using Eq. 5:

$$\mathrm{Fr}=(AUC_{\mathrm{test}}/D_{\mathrm{test}})/(AUC_{\mathrm{ref}}/D_{\mathrm{ref}})\times 100\% \quad (5)$$

Here, D is the dose.

The Wagner–Nelson method was used to calculate the percent of PNH absorbed, Fa:

$$\mathrm{Fa}=[(C_{(t)}+K_eAUC_{(0-t)})/K_eAUC_{(0-\infty)}]\times 100 \quad (6)$$

Results and Discussion

Evaluation of Release Behavior of the DRCOPT To compare the lag time of the DRCOPT with that of the conventional formulations, different tablet cores were designed. 1) One was the DRCOPT formulation; 2) the same formulation in which only DRCs were replaced with equal PNH content. After being coated with the specific coating solution, both were compared in an *in vitro* drug release test. At the same time, 3) the DRCOPT cores without coating; and 4) DRCs were also examined in the same trial.

Significant differences showed up in Fig. 1 among formulation 1 and other formulations. The onset of the drug release from the DRCOPT took place at the end of a lag time. Lag time is a normal phenomenon in the OPT delivery systems. In the previous designs, the main purpose was always to decrease lag time; on the contrary, the lag time was used to achieve the time-controlled delivery system in this investigation. Compared with the conventional OPT, the DRCOPT gained an advantage in terms of lag time, which was due to two factors. On one hand, the influx of water into tablets customarily required a certain time, on the other hand, the DRCs in cores exchanged drug with sodium chloride also consumed time. As a result, a relatively longer lag time of the DRCOPT compared to that of the conventional OPT occurred.

Figure 1 showed the release profile of different formulations, and the release rates increased according to the sequence of formulation 4, 3, 2 and 1. When the DRCs were exposed to the dissolution medium, the exchange reaction occurred immediately. The cumulative release was 98.43% within 2.5 h and rapid release behavior can be achieved. As for the DRC tablet, a relatively slower drug release rate was observed, because the DRC tablet was a type of matrix tablet with the PEO (N80) as the hydrophilic matrix material.

Further, the drug release from the conventional OPT and the DRCOPT formulations could be controlled, which exhibited better controlled release behavior, and the DRCOPT sustained a longer zero-order drug release period, ranging from 2 to 14 h, than that of the conventional OPT, ranging from 0 to 10 h. Additionally, the DRCOPT had a 2 h lag time in the *in vitro* dissolution test, which would accommodate such diseases as hypertension and angina pectoris occurring during certain periods.

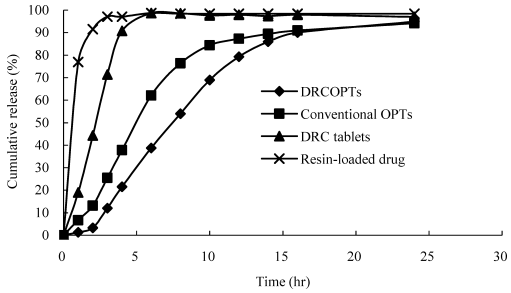

Fig. 1. Dissolution Profile of Different Preparations ($n=3$, Mean)

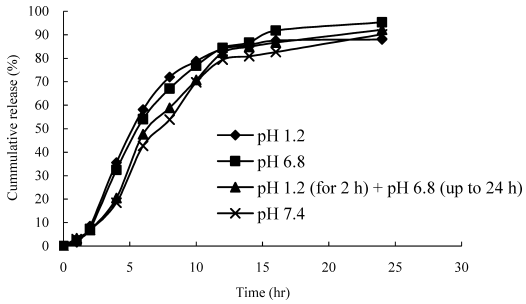

Fig. 2. Dissolution Profile of Propranolol Hydrochloride in the DRCOPT of Different Dissolution Media ($n=3$, Mean)

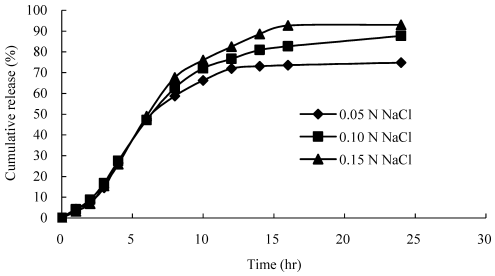

Fig. 3. Dissolution Profile of Propranolol Hydrochloride in the DRCOPT of Different Ionic Strength ($n=3$, Mean)

Effects of Dissolution Medium To elucidate the effects of the dissolution medium on the DRCOPT release characteristics, the release profiles of the DRCOPT in different media were obtained. The dissolution percentage was found to change slightly, the results were not statistically significant ($p<0.05$). As illustrated in Fig. 2, linear correlation of zero-order can be achieved for different pH gradient, indicating the release behavior of the DRCOPT was not affected by the simulated pH environment in the human body, which was consistent with the characteristics of the OPT.

Effects of Ionic Strength The drug release studies were performed in sodium chloride solution of different ionic strength. Based on the results shown in Fig. 3, the drug release exhibited similarity before 6 h, after that the cumulative release increased with the ionic strength increased. A possible reason was given: when water entered the core through the semi-permeable membrane, PEO (N80) swells gradually. During the first 6 h, PEO (N80) acts as an excellent suspension agent, which provided NaCl in core with the proper conditions to exchange PNH. Thereafter, the exchanged drug is released from the orifice under the osmotic pressure. The more water taken in, the more the PEO swelled. A portion of

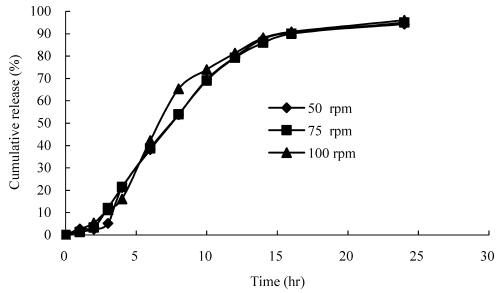

Fig. 4. Dissolution Profile of Propranolol Hydrochloride in the DRCOPT of Different Rotation Speed (n=3, Mean)

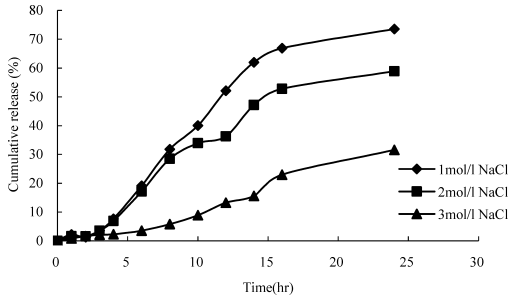

Fig. 5. Dissolution Profile of the DRCOPT in NaCl Solution of Different Concentrations (n=3, Mean)

the DRCs were driven away to the medium solution. It was noted that greater ionic strength provides the DRCs with more opportunities to exchange drug, consequently, the DRCs expelled in higher ionic strength solution manifested a higher release profile than that in lower ionic strength solution.

Effects of Rotation Speed To investigate the effects of stirring speed on the drug release behavior, dissolution experiments with the DRCOPT were carried out at stirring rates of 50, 75 and 100 rpm. As shown in Fig. 4, no significant effect of rotation speed on the drug release profile. Thus, the mobility of the gastrointestinal tract may only slightly affect the drug release of the DRCOPT, meaning that the DRCOPT will remain in the gastrointestinal tract in a reliable and reproducible manner.

Release Mechanism. Investigation of Drug Release Mechanism There are three mechanisms that contribute to the release of active material from OPT,[4] *i.e.*, drug release driven by the mechanism of osmotic pressure, forced by the mechanism of diffusion through orifice and membrane.

The steady-state zero-order release rate (dm/dt) of a drug from an osmotic delivery device can be expressed as Eq. 7

$$dm/dt = AS/hLp\sigma\Delta\Pi + PAS/h \quad (7)$$

Here, A is the OPT surface area, h is the thickness of coating, S is the solubility of drug, $Lp\sigma$ is the fluid permeability of the coating, P is the permeability coefficient of the active ingredient through the coat, $\Delta\Pi$ is the osmotic pressure difference between the core inside and outer circumstances.

To demonstrate the mechanism of osmotic pressure, a dissolution test of the DRCOPT in solution containing different concentrations of sodium chloride was carried out. As manifested in Fig. 5, cumulative release increased with the concentration decreased. As the water hydrated the core, a sodium chloride solution of high concentration was formed that allowed DRCs to exchange PNH with sodium chloride in core. At the same time, the difference of osmotic pressure between core and dissolution medium was relatively great, driven by which PNH exchanged from DRCs released from the orifice. The bigger the concentration contrasted, the more different the osmotic pressure occurred between the core and external environment. As a result, drug release exhibited a quicker tendency in 1 mol/l NaCl solution than that in 2 mol/l and 3 mol/l NaCl solution, which indicated that in release process drug escaped from the core controlled by osmotic pressure mechanism.

When the mechanism of diffusion through orifice dominates the drug delivery, the release rate is proportional to the

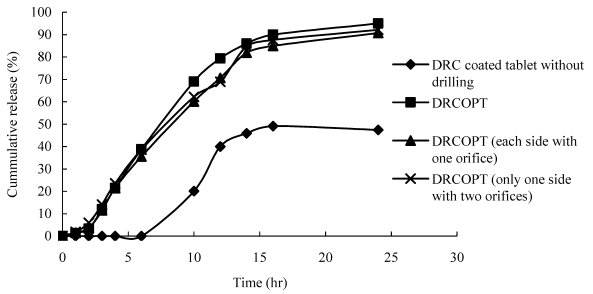

Fig. 6. Dissolution Profile of the Different DRCOPT for Investigating Drug Release Mechanism (n=3, Mean)

size of orifice. Drilling one or two orifices on the membrane has much effect on the surface area, if the diffusion mechanism takes advantage, two orifices are twice than one orifice, so the drug release will change greatly. As shown in Fig. 6 the release profiles of the DRCOPT with one and two orifices were approximately same, that is to says the contribution of diffusion mechanism through orifice to the total drug release was extremely trifling. Furthermore, the slopes of release profiles of the DRCOPT, the DRCOPT (each side with one orifice) and the DRCOPT (one side with two orifices) were 0.0691 mg/ml·h, 0.0627 mg/ml·h and 0.0612 mg/ml·h, respectively. The differences between the release rates were 6.3% and 7.8%, which meant the release rate induced by the diffusion mechanism through orifice was no account.

In order to gain insight into the mechanism of diffusion through membrane, the DRCs with the same excipients was directly compressed into 0.33 g tablets and coated with the same weight gain as the DRCOPT, but no drilling on the tablet surface. The release behavior was shown in Fig. 6, the tablet bursted through expansion at the sixth hour, before that point of time no significant dissolution was observed. The following reasons could be in charge of the phenomenon. Due to the semipermeability of coating membrane, only water and some micro molecule were allowed to go out and come in the core, in addition, the property of active ingredient in core was commonly macromolecule, which was restricted to escape from the core through the membrane. As a result, the mechanism of diffusion through membrane hardly took responsibility for the drug release.

As mentioned above, the mechanism of osmotic pressure prevailed.

Estimation of Function of Sodium Chloride in the Core In *in vitro* solution test, 0.15 mol/l NaCl was acted as solution medium, to avoid its influence on the determination of conductivity, 0.3 mol/l glucose solution was used to substi-

Table 1. Relationship of Conductivity between Mixture Solution and Pure Components

Ratio of PNH to NaCl	Calibration curve of concentration *vs.* conductivity of mixture solution	Experimental value	Theoretical value	Error (%)
1 : 9	$\lambda=55320\cdot C_{Mix}+13.21$	55320	57200	3.29
5 : 5	$\lambda=66677\cdot C_{Mix}+11.49$	66677	66591	0.13
9 : 1	$\lambda=76451\cdot C_{Mix}+11.67$	76451	75982	0.61
Calibration curve of PNH solution	$\lambda=78330\cdot C_{PNH}+10.26$			
Calibration curve of NaCl solution	$\lambda=54853\cdot C_{NaCl}+9.83$			
Calibration curve of PNH and NaCl mixture solution	$\lambda_{theoretical\ value}=78330\cdot C_{PNH}+54853\cdot C_{NaCl}+8.17$ (conductivity of glucose deionized aqueous solution)			

C represents concentration.

tute 0.15 mol/l NaCl. Because glucose is non-electrolyte and 0.3 mol/l glucose solution can produce the same osmotic pressure as 0.15 mol/l NaCl solution does.

Pure components (PNH and NaCl) of different concentration were prepared as 10^{-2} mol/l, 10^{-3} mol/l, 10^{-4} mol/l, 10^{-5} mol/l, 10^{-6} mol/l in glucose deionized aqueous solution, respectively, the calibration curve of concentration *vs.* conductivity was determined. Thereafter, the conductivity of mixture solution was measured. Different ratios of PNH to NaCl in pooled solution were prepared to investigate the relationship of conductivity between the pure components in a binary system. As illustrated in Table 1, the error of conductivity in mixture solution between experimental value and theoretical value was less than 5%, which may be caused by the slight change of temperature. The result indicated that the slope of conductivity in a binary system was replaced by respective slope of pure component existing in this system, which will not bring about too much error. Thus, the relationship between conductivity and concentration in the binary system composing of PNH and NaCl can be expressed as Eq. 8

$$\lambda=78330\cdot C_{PNH}+54853\cdot C_{NaCl}+8.17\ \text{(conductivity of glucose deionized aqueous solution)} \quad (8)$$

In this work the concentration of NaCl delivered from the DRCOPT in dissolution medium was studied by three steps: 1) determination of conductivity in mixture solution by conduct meter (DDS-11A, Shanghai Leici Xinjing Instrument Co. Ltd., Shanghai); 2) determination of PNH concentration by HPLC method; 3) ascertainment of NaCl concentration used Eq. 8. According to the Table 2 and Fig. 7, the PNH detected in the solution was exchanged by NaCl in core, which took responsibility for exchanging drug at the speed of 0.32 mg/ml·h. In addition, concentration of NaCl calculated by Eq. 8 was the part of NaCl escaped from orifice at the rate of 0.065 mg/ml·h, which contributed to generating osmotic pressure and supplementing electrolyte. Osmotic pressure caused by NaCl could provide the drug with enough motivation to release from orifice, at the same time, a certain amount of DRCs were expelled from the core by osmotic pressure into solution medium. PNH will be exchanged by NaCl in solution medium. As a result, the amount of NaCl decreased, and the balance of electrolyte in circumstances was disturbed. However, NaCl acting as osmotic pressure agent extruded the port, and this tendency will not halt until the osmotic pressure between the core and outer circumstances kept identical. The amount of NaCl consumed by exchanging drug outside will be replenished by NaCl released from core, from this point of view, NaCl served as electrolyte supplementary agent. There was 100 mg DRC (40 mg PNH) in the core, 8 mg NaCl (27%) was needed to exchange PNH with DRC in total theoretically, as tested above, 9% NaCl acted as ion exchange agent, the other 18% was given by NaCl existing in dissolution medium. This part of NaCl was supplemented by that liberated from the core into the external medium. Thus, the ratios of NaCl playing the roles of exchanging drug and producing osmotic pressure (electrolyte supplement) were nearly 9% and 90% (among which, 18% as electrolyte supplementary agent), respectively.

Table 2. Cumulative Release of PNH and NaCl Determined by Conductivity-HPLC Method

Time (h)	Conductivity of mixture solution (μs/cm·N)	Concentration of PNH (mg/ml)	Concentration of NaCl (mg/ml)
2	31683	0.032	0.007
4	40665	1.247	0.011
6	51554	1.506	0.016
8	73260	2.187	0.021
10	77879	2.753	0.024
12	91988	3.256	0.025
14	93533	3.456	0.026
24	97480	3.766	0.030

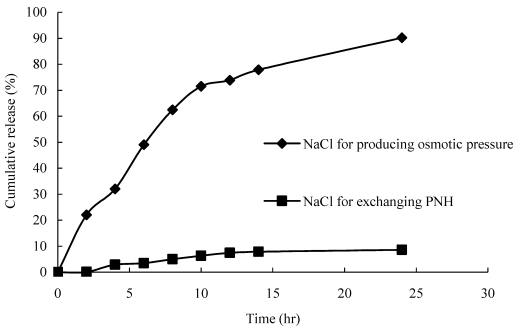

Fig. 7. Dissolution Profile of NaCl as Osmotic Pressure Agent (Electrolyte Supplementary Agent), Ion-Exchange Agent (n=3, Mean)

From Fig. 7, a characteristic was showed that there was no PNH release before 2 h, which was related to the time consumption of exchange between DRCs and NaCl, in this case, it was consistent with the lag time in *in vitro* dissolution test.

***In Vivo* Data Analysis** Pharmacokinetic parameters of PNH, calculated from the plasma concentrations of PNH following oral administration of the conventional tablets and the DRCOPT were listed in Table 3. From Fig. 8, these results demonstrated that the DRCOPT exhibited a lag time of 4 h

and a satisfactory sustained-release effect, with less fluctuation in the plasma levels.

The pharmacokinetic parameters of PNH after oral administration of the DRCOPT, which were compared with those of tablets (propranolol hydrochloride tablet, 10 mg/tablet), provided evidence of a longer t_{max} (11.000 h *vs.* 3.000 h, $p<0.01$), a lower C_{max} (128.246 ng/ml *vs.* 749.582 ng/ml, $p<0.01$), a lower $AUC_{0-\infty}$ (2583.071 ng·h/ml *vs.* 2708.350 ng·h/ml, $p<0.01$) and a longer $t_{1/2}$ (6.559 h *vs.* 3.214 h, $p<0.01$). A significant improvement in all these parameters and a behavior of sustained release were confirmed by these results. The slight decrease in *AUC* suggested that a certain amount of PNH wasn't absorbed after oral administration of the DRCOPT, which might attribute to the incomplete release of DRCs in the core of the DRCOPT. In addition, a relatively long t_{max} and $t_{1/2}$ were achieved, implying a delayed absorption and slow excretion of PNH after oral administration of the DRCOPT. Therefore, it was considerable to investigate their pharmacokinetic properties to explain the pharmacological effects of the conventional tablets and the DRCOPT in *in vivo* situations. Most of all, a lag time of 4 h was obtained, which was well consistent with the phenomenon of lag time observed in *in vitro* release. However, a longer lag time occurred *in vivo* than appeared *in vivo*. There were two reasons that contributed to this phenomenon. Firstly, in *in vitro* experiment, dissolution medium could provide DRCs with enough ion strength to exchange the drug. While in *in vivo* situation, less ion strength was given at corresponding time point as in *in vitro* trail. Secondly, sink condition was easier to reach *in vitro*. In all, the longer lag time could give us a therapeutic regimen for the disease with the characteristics of chronotherapy. We could adapt the time of taking medicine in order to reduce the times of taking medicine and improve the compliance of the patients.

Table 3. Pharmacokinetic Parameters of PNH in Beagle Dogs ($n=6$) after Oral Administration of the Conventional Tablets and the DRCOPT (Each Dose Containing 120 mg PNH)

Parameters	Conventional tablets	DRCOPT
T_{max} (h)	3.000±0.632	11.000±1.095
C_{max} (ng/ml)	749.582±55.40	128.246±10.780
AUC_{0-t} (ng·h/ml)	2634.829±370.849	2088.078±199.520
$AUC_{0-\infty}$ (ng·h/ml)	2708.350±385.545	2583.071±508.047
$T_{1/2}$ (h)	3.214±0.584	6.559±2.858
Lag time (h)	0.000±0.000	4.000±0.400
	Fr=104.484±13.195%	

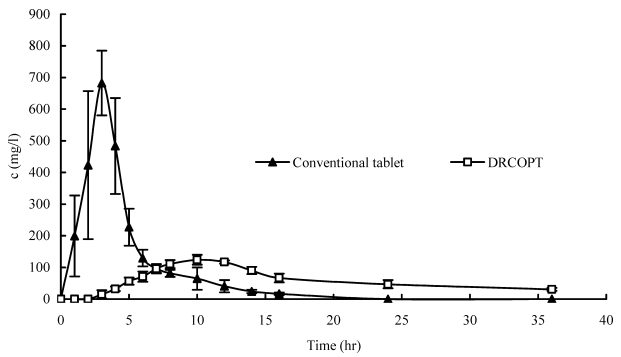

Fig. 8. Plasma Concentration–Time Profile of PNH in Beagle Dogs after Oral Administration of the Conventional Tablets and the DRCOPT (Each Dose Containing 120 mg PNH)

Each Point represents mean±S.D. ($n=6$).

***In Vivo*/*In Vitro* Correlation (IVIVC)** A solution was used to obtain the best fitted compartmental model by Drug and Statistics (DAS) version 2.0, which gave the evidence of one-compartment model for the DRCOPT in beagle dogs. Wagner–Nelson (W-N) method is mainly applied to the pharmacokinetic study of the drug fitted to a one-compartment model. As a result, the data generated in the pharmacokinetic study were used to develop the IVIVC by the W-N method. The relationship between percent of *in vitro* dissolution and the fraction of drug absorbed *in vivo* (Fa) was examined. The Fa was determined using the W-N method by Eq. 2. Linear regression analysis was applied to the IVIVC plots. The values of correlation coefficient (R^2), slope and intercept were calculated, respectively. The results of *in vitro* drug cumulative release and *in vivo* (Fa), together with IVIVC profile, were given in Table 4. A good *in-vitro-in-vivo* correlation (IVIVC, $R^2=0.9541$) was achieved.

Conclusions

A time-controlled release system that met the requirements for chronopharmaceutical drug delivery was presented based on the combination of drug–ion exchange resin complexes and osmotic pump tablet. Compared with other formulations, an obvious lag time was obtained from *in vitro* experiment and a longer lag time was observed in *in vivo* experiment. In addition, a satisfactory controlled release behavior and less fluctuation in the plasma levels were achieved.

The drug release mechanism was investigated in detail. The role of sodium chloride played in the drug release process was also studied. Determination of ionic concentration in a binary system by conductivity-HPLC method offers a useful supplement to chemical analysis or instrumentation. The DRCOPT could not only improve the application of osmotic pump tablet in the direction of time-controlled release by combinating EOPT and IER, but also control the drug release rate. Besides, the lag time could provide a new idea of preparing a novel elementary osmotic pump tablet to develop a time-controlled system. And it could provide a way to achieve an effective drug level only at the demanded time using EOPT.

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Table 4. *In Vitro* Cumulative Release of the DRCOPT and *in Vivo* Absorption Percentage (Fa)

Time (h)	4	6	8	10	12	14	24
In vitro %	0.2120	0.3819	0.5379	0.6947	0.7967	0.8774	0.9426
In vivo %	0.1147	0.3812	0.6469	0.7886	0.8993	0.9092	0.9115

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