

Impact of Azithromycin on Forsythiaside Pharmacokinetics in Rats: A Population Modeling Method*

Jing-chen TIAN^{1,2}, Xuan-ling ZHANG³, Jian-rong CUI⁴, Xin-gang LI^{3#}

¹Department of Pharmacy, Beijing Tiantan Hospital, Capital Medical University, Beijing 100070, China

²School of Chinese Materia Medica, Beijing University of Chinese Medicine, Beijing 100102, China

³Department of Pharmacy, Beijing Friendship Hospital, Capital Medical University, Beijing 100050, China

⁴Department of Pharmacy, Chengdu Seventh People's Hospital, Chengdu 610000, China

© Huazhong University of Science and Technology 2022

[Abstract] Objective: Lianhuaqingwen and Shuanghuanglian are drug treatment options for Corona Virus Disease 2019 (COVID-19). In China, use of traditional Chinese medicine with Shuanghuanglian or Lianhuaqingwen (for them, forsythiaside is the active antiviral and antibacterial component) in combination with azithromycin is common for the treatment of pediatric pneumonia. It is important to understand the reason why the combination of these compounds is better than a single drug treatment. This study aimed to explore the pharmacokinetic interaction between forsythiaside and azithromycin. **Methods:** Twelve male Sprague-Dawley rats were randomly divided into an experimental group (*Forsythia suspensa* extract and azithromycin) and a control group (a single dose of *Forsythia suspensa* extract in 5% glucose solution). Plasma samples were collected at scheduled time points, and the high-performance liquid chromatography combined with ultraviolet method was used to determine the plasma forsythiaside concentration. Non-compartmental analysis and population pharmacokinetic methods were used to investigate the forsythiaside pharmacokinetic difference between the experimental and control group. **Results:** Compared with a single administration, the area under the curve and half-life of forsythiaside increased, and forsythiaside clearance decreased significantly after co-administration with azithromycin. The *in vivo* behavior of forsythiaside could be described by the one compartment model. The forsythiaside clearance decreased when combined with azithromycin. Visual evaluation and bootstrap results suggested that the final model was precise and stable. **Conclusion:** Co-administration of azithromycin can significantly decrease the forsythiaside clearance and increase drug exposure. A lower dose of azithromycin can obtain sufficient forsythiaside concentration to provide antiviral and antibacterial activity.

Key words: azithromycin; forsythiaside; pharmacokinetics; drug interaction; traditional Chinese medicine; Corona Virus Disease 2019; antivirals

As of October 15, 2021, more than 239 million Corona Virus Disease 2019 (COVID-19) cases have been reported across the world. At present, the United States FDA has authorized three COVID-19 vaccines for human coronavirus, and China has approved four COVID-19 vaccines for emergency use. Chinese traditional medicine, such as Lianhuaqingwen capsule^[1-3] and Shuanghuanglian oral liquid^[4] could be additional drug treatment options for COVID-19. In particular, Lianqiao (*Forsythia suspensa* fruit) is an important antiviral ingredient of Lianhuaqingwen capsule and Shuanghuanglian oral liquid.

Forsythia suspensa (Thunb.) is a species of

flowering plant native to Asia and it is one of the 50 fundamental herbs used in traditional Chinese medicine. The fruit of *Forsythia suspensa* is widely used as *Fructus Forsythias* for the treatment of upper respiratory tract infections, acute nephritis, pyrexia, tonsillitis and pharyngitis^[5-7]. This compound is also commonly combined with honeysuckle flower (*Lonicera*) and other ingredients. Forsythiaside is one of the important components in *Fructus Forsythias* and has various biological activities, such as anti-endotoxin effect^[8], antiviral effect^[9], antioxidant and antibacterial activity^[10], etc. However, there are no recent clinical studies on *Forsythia* to provide a basis for dosage recommendations.

Azithromycin is on the World Health Organization's List of Essential Medicines, the most effective and safe medicines needed in a health system^[11]. Combining traditional Chinese medicine and western medicine is

Jing-chen TIAN, E-mail: salfigo@sina.com

#Corresponding author, E-mail: lxg198320022003@163.com

*This project was supported by the Natural Science Foundation of Beijing Municipality (No. 7192060).

popular in China, and therefore co-administration may offer hope for developing new treatments for many diseases^[12–14] such as COVID-19^[15–17]. Azithromycin, a western medicine and traditional Chinese medicine involving Shuanghuanglian or Lianhuaqingwen (forsythiaside, the active antiviral component) are often matched together to treat pediatric pneumonia in China^[18, 19]. Compared with azithromycin alone, the co-administration has an acceptable clinical effect in treating pediatric pneumonia (improving immunologic functions) and has a proven safety record^[18]. The clinical efficacy of co-administration is better than azithromycin monotherapy. In our previous study, co-intravenous administration of azithromycin and Shuanghuanglian can significantly increase both forsythiaside and azithromycin concentration^[20].

It is important to understand the pharmacokinetic interaction of Chinese and western medicine. *Forsythia suspensa* is the major ingredient of Shuanghuanglian and Lianhuaqingwen. To exclude interference from other ingredients, a nonlinear mixed-effects modeling approach was utilized to explore the pharmacokinetic change of *Forsythia suspensa* extract in combination with azithromycin.

1 MATERIALS AND METHODS

1.1 Rats and Dosing Regimens

Twelve male Sprague-Dawley rats (240–260 g) were purchased from Vital River Laboratories (China). *Forsythia suspensa* extract (5.9% of forsythiaside) was obtained from Sichuan Weikete Biological Technology Co., Ltd. (China). The structure of forsythiaside is displayed in fig. 1. Azithromycin for injection was purchased from the northeast pharmaceutical group Shenyang First Pharmaceutical Co., Ltd. (China, production batch number: 131138). All rats were housed at a constant temperature of 23–25°C, and a relative humidity of 50%±10% in an animal breeding room for 7 days before experiments.

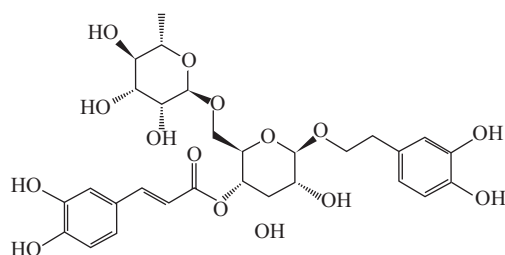


Fig. 1 The structure of forsythiaside

All rats were randomly divided into two groups: a single administration group ($n=6$) and a combined administration group ($n=6$). All animals were fasted for 12 h before administration of any compound. In the single administration group, a single dose of *Forsythia suspensa* extract (containing 3.4 mg of forsythiaside) in

5% glucose solution was administered to rats through intragastric injection. In the co-administration group, *Forsythia suspensa* extract (3.4 mg of forsythiaside) was injected to rats intragastrically, and 13 mg of azithromycin was simultaneously administered intravenously. Animals were euthanized by cervical dislocation at the end of each experiment. All experiments followed the Guide for Care and Use of Laboratory Animals as adopted and promulgated by the Animal Ethics Committee of the Beijing University of Chinese Medicine.

1.2 Sampling and Drug Determination

After administration, approximately 0.4 mL of whole blood samples were collected via the retro-orbital sinus into heparinized microcentrifuge tubes at 5 min, 10 min, 15 min, 30 min, 1 h, 2 h, and 4 h. The plasma was separated through centrifugation (10 000 r/min for 5 min), and plasma samples were stored at -70°C until analyzed for forsythiaside concentration.

Our previously reported high-performance liquid chromatography combined with ultraviolet (HPLC-UV) (Shimadzu Liquid Chromatographic System, Japan) method was used to detect the forsythiaside concentration^[21]. Briefly, the internal standard (IS) was hesperidin (Shanghai Source Leaf Bio-Tech Co., Ltd., China; lot number: 20130820, purity $\geq 98.0\%$). The mobile phase consisted of A (acetonitrile) and B (aqueous solution containing 0.4% acetic acid). Before use, the mobile phase was filtered and degassed. Plasma samples were separated at a flow-rate of 1.0 mL/min, using a gradient elution of 0–10 min with 10% A, 10–20 min with 10%–30% A; 20–25 min with 90% A, and 25–32 min of 100% A. The column eluate was detected at 284 nm and 330 nm.

Forsythiaside and IS extract recovery rates were $>73.81\% \pm 2.96\%$ and $82.23\% \pm 5.07\%$, respectively. No matrix components in plasma produced significant changes in the HPLC-UV responses to forsythiaside or IS. Overall, these results indicated that no endogenous substances significantly influenced the recovery or chromatographic profiles of the IS or analytes^[21]. The related chromatographic profiles are displayed in fig. 2.

1.3 Noncompartmental Analysis

A noncompartmental analysis (NCA) can calculate pharmacokinetic parameters of forsythiaside without using the assumption of a compartmental model. The area under the curve (AUC) from the time of dosing to the time of the last measurable concentration (AUC_{last}) was calculated as follows:

Linear trapezoidal rule

$$AUC_{t_i}^{t_{i+1}} = \delta_i \times \frac{C_{i+1} - C_i}{2} \quad (1)$$

Logarithmic trapezoidal rule

$$AUC_{t_i}^{t_{i+1}} = \delta_i \times \frac{C_{i+1} - C_i}{\ln\left(\frac{C_{i+1}}{C_i}\right)} \quad (2)$$

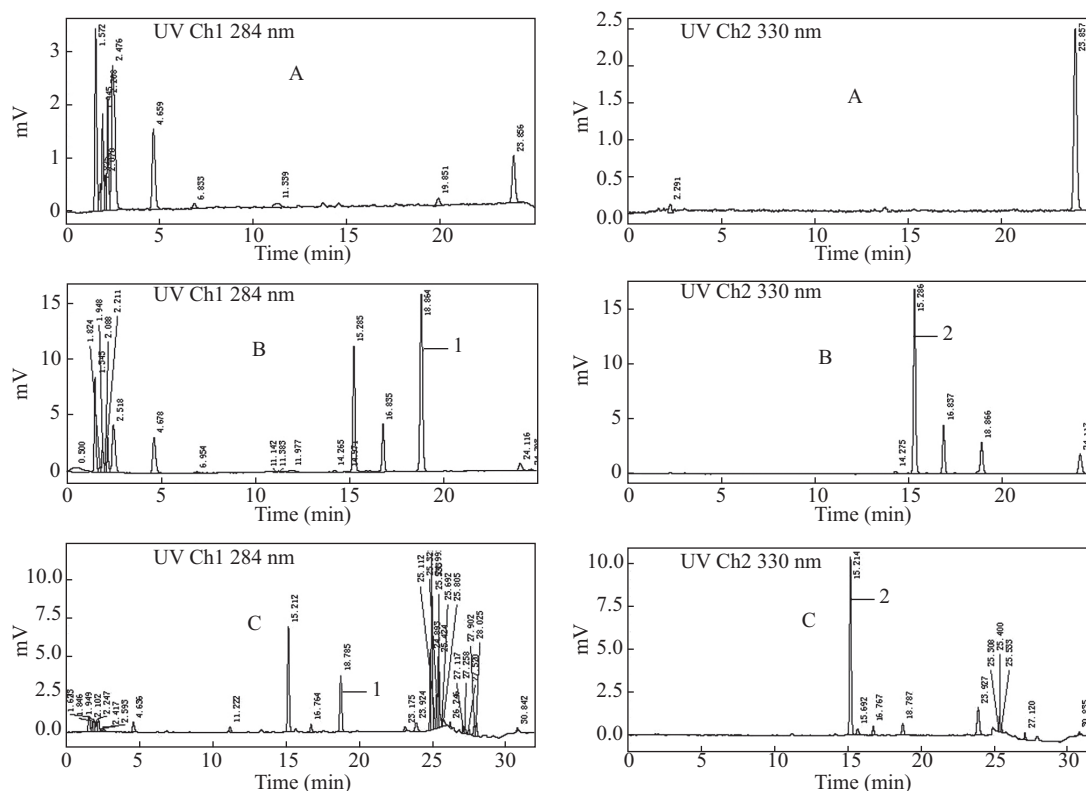


Fig. 2 Specificity of the HPLC-UV method (1: IS; 2: forsythiaside)
 A: blank plasma; B: blank plasma+IS+forsythiaside; C: plasma sample

where δ_i is $t_{i+1} - t_i$, the i^{th} sampling time; t_{i+1} , the $i+1^{\text{th}}$ sampling time). If the logarithmic trapezoidal rule fails in an interval because $C_i \leq C_{i+1}$, then the linear trapezoidal rule will be applied for that interval. The following formula was used to calculate the total body clearance for extravascular administration (CL):

$$CL = \frac{\text{Dose}}{AUC} \tag{3}$$

The maximum observed concentration (Cmax) occurred at time Tmax. Phoenix’s NCA engine computes derived measurements from raw data by using methods appropriate for serially-sampled data. NCA was performed on each animal and then the results averaged. The NCA parameters were compared between single and co-administration groups.

1.4 Population Pharmacokinetics

Several (one, two and three) compartmental models were used to fit the pharmacokinetic data. Based on the objective function value (OFV) and diagnostic plots, a proper model was selected to describe the forsythiaside *in vivo* behavior. The inter-individual variability (IIV) of population parameters was described using the exponential model:

$$P_i = P \times e^{\eta_i} \tag{4}$$

where P_i and P represent the i^{th} individual value and the typical value for the population parameter, respectively. The relationship between P_i and P is described using η_i , which is normally distributed with a mean of 0 and a variance of ω^2 . The multiplicative

error model was selected to describe residual error:

$$C_i = C \times (1 + \epsilon_i) \tag{5}$$

where C_i and C account for observation and prediction, respectively. The parameter ϵ_i indicates the residual error, which is normally distributed with a mean of 0 and a variance of σ^2 .

The Phoenix NLME software (Certara, Inc., USA) was used to fit the pharmacokinetic data. The first-order conditional estimation method with the η - ϵ interaction option (FOCE-ELS) was used throughout the model development process.

1.5 Model Validation

Model validation checks the accuracy of the model’s representation of the observation through a goodness-of-fit (GOF) plot, bootstrap, and visual predictive check (VPC). GOF plots include scatter plots of observations and predictions *versus* time, observations *versus* predictions, and conditional weighted residuals (CWRES) *versus* predictions and time. Additionally, the Normal Quantile-quantile (QQ) plot was also used to evaluate how well the distribution of CWRES matches a standard normal distribution^[22]. A bootstrap sample was generated by repeated random re-sampling with replacement from the original dataset 1000 times. The final population pharmacokinetic model was determined by repeatedly testing the 1000 bootstrap samples. The median parameter estimates obtained from replication and their 95% confidence intervals (95%CI) were compared with those estimated

from the original dataset. If no significant difference between the data was observed, one may confirm that the estimates for the final population pharmacokinetic model were stable and precise. VPCs were constructed based on 1000 Monte Carlo simulations. Out of the 1000 median values originating from the model simulations, a nonparametric 90% prediction interval (90%PI) was calculated (5th and 95th percentile). Then the observed concentration-time data were graphically superimposed on the median values, and the 5th and 95th percentiles of the simulated concentration-time profiles. The model is precise if the observations were approximately distributed within 90%PI.

2 RESULTS

2.1 NCA

Using the NCA method, the AUC_{last} , CL and C_{max} , T_{max} (time at the C_{max}), V_z (apparent volume of distribution) were calculated and are presented in table 1. The parameters are presented as the mean±standard deviation (SD). After merging azithromycin into *Forsythia suspensa* extract, the AUC_{last} increased significantly (P -value=0.002), meanwhile, the CL obviously decreased compared with single administration (P -value=0.036). Regarding C_{max} , there were also obvious differences between the single and co-administration groups (P =0.028). These results indicate co-administration could improve forsythiaside exposures through slowing drug clearance.

2.2 Population Pharmacokinetics

The concentration of forsythiaside drops rapidly *in vivo*, and the drug concentration is only 1/30th or less of the peak concentration after 4 h. Considering the sensitivity of the detection method and the lower

limit of quantitation ($0.2 \mu\text{g/mL}$)^[21], we only tested drug concentrations within 4 h after administration. The obtained data can be fitted with a one-compartment model (OFV=360.026). Although the two-compartment (OFV=359.396) and three-compartment (OFV=359.002) models fit well with the above data, there was no significant increase in the goodness-of-fit of the population model, and the decline in OFV was not significant ($P>0.05$). Finally, we chose a one-compartment model for subsequent group analysis. After covariate selection, CL was markedly affected by the co-administration of azithromycin, and the final model was presented as follows:

$$K_{ai}=6.891 \times e^{\eta_i} \text{ (h}^{-1}\text{)} \quad (6)$$

$$V_i=152.259 \times e^{\eta_i} \text{ (mL)} \quad (7)$$

$$CL_i=158.104 \times e^{\eta_i} \text{ (mL} \cdot \text{h}^{-1}\text{)} \text{ [single]} \quad (8)$$

$$CL_i=158.104 \times e^{-0.214 \times \eta_i} \text{ (mL} \cdot \text{h}^{-1}\text{)} \text{ [co-administration]} \quad (9)$$

where K_{ai} is the individual first-order absorption rate constant and 6.891 h^{-1} is the typical value. The parameter V_i indicates the individual distribution volume, and the typical value is equal to 152.259 mL . The value of $158.104 \text{ mL} \cdot \text{h}^{-1}$ is the typical value of CL_i when single *Forsythia suspensa* extract was administered, and the coefficient is -0.214 , indicating the relationship between co-administration of azithromycin and CL_i . According to equation 9, CL_i decreased when combining azithromycin into *Forsythia suspensa* extract. The final population pharmacokinetic parameters, their IIV and residual errors are summarized in table 2. The IIVs are lower, which may be due to rats having similar physiological features. All the pharmacokinetic parameters were estimated with an acceptable precision [relative standard error (RSE)% <20.921%].

Table 1 Forsythiaside pharmacokinetic parameters obtained from noncompartmental analysis (mean±SD)

	AUC_{last} ($\mu\text{g} \cdot \text{mL}^{-1} \cdot \text{h}^{-1}$)	CL ($\text{mL} \cdot \text{h}^{-1}$)	C_{max} ($\mu\text{g} \cdot \text{mL}^{-1}$)	T_{max} (h)	V_z (mL)
Single	20.84±5.50	175.56±77.78	14.06±11.70	0.17	194.10±100.44
Co-administration	29.85±2.78	111.54±9.35	19.34±13.98	0.33	118.53±35.90
Significance (P value)	0.002*	0.036*	0.028*	–	0.112

AUC_{last} , area under the curve from the time of dosing to the time of the last measurable concentration; CL , total body clearance for extravascular administration; C_{max} , maximum observed concentration. * P value <0.05

2.3 Model Validation

Fig. 3 displays the GOF plots of both structural models (fig. 3A–3E) and the final population pharmacokinetic model (fig. 3A'–3E'). These plots do not show a systematic bias for both structural and final pharmacokinetic model predictions. After co-administration was incorporated into the pharmacokinetic model, the predictions were closer to observations and the diagnostic plots improved significantly (fig. 3A and 3B vs. 3A' and 3B'). CWRES are a new diagnostic tool testing for model misspecification, and are calculated as the FOCE-approximated difference between an individual's data and the model

prediction of that data, divided by the root of the covariance of the data given the model^[23]. Compared with a structural model (fig. 3C and 3D), CWRES in the final model were closer to the zero line (fig 3C' and 3D'). CWRES were scattered evenly around the zero line (without any apparent systematic bias). A QQ plot is an exploratory tool used to assess the similarity between the distribution of one numeric variable and a normal distribution. A significant improvement in the predictive performance of the final pharmacokinetic model (fig. 3E') was achieved compared to the basic one (fig. 3E).

A total of 986 re-samplings of the original dataset

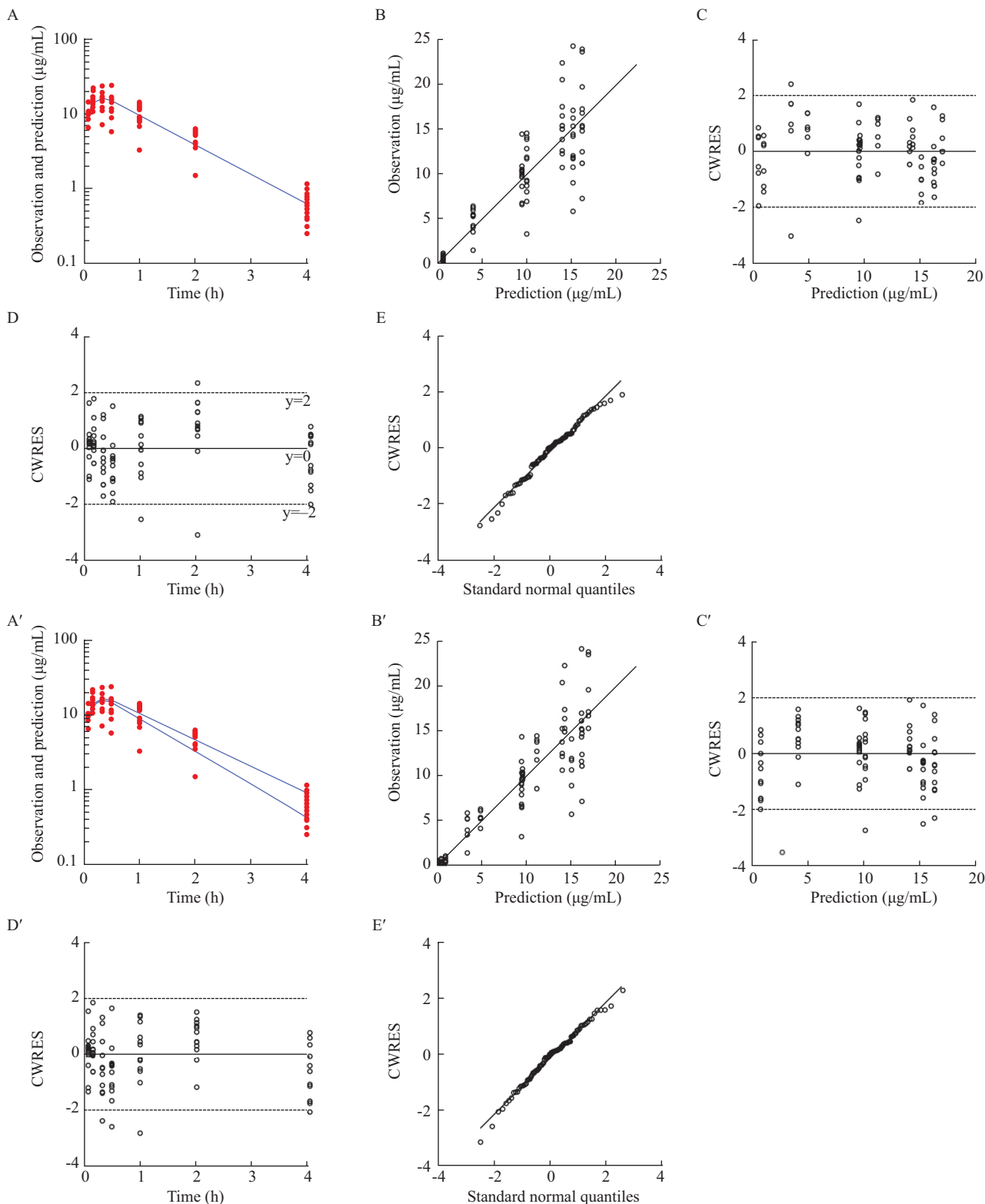


Fig. 3 Goodness-of-fit plots of forsythiaside base (A–E) and final (A’–E’) pharmacokinetic model
 A and A’: prediction (solid lines) and observation (dots) against time; B and B’: observation *versus* prediction, and lines are the lines of unity $Y=X$; C and C’: conditional weighted residuals (CWRES) against prediction; D and D’: CWRES against time; E and E’: normal quantile-quantile plots of CWRES

were successfully performed in the process of bootstrap evaluation, indicating qualified stability for the final population pharmacokinetic model. The typical values of final pharmacokinetic model parameters were close to the parameter estimates for bootstrap replicates

and were contained within their 95% CIs, indicating the robustness and stability of the final population pharmacokinetic model (table 2). The VPC plot displayed the distribution of 1000 simulated data-time curves (median and 95% PI) and the comparison

Table 2 Estimated parameters of forsythiaside final population pharmacokinetic model

Parameter (unit)	Model estimate			Bootstrap	
	Estimate	RSE%	IIV (CV%)	Median	95%CI
K_a (h^{-1})	6.891	14.242	10.869	6.567	[4.976, 7.812]
V (mL)	152.259	10.180	15.049	149.443	[103.052, 184.725]
CL (mL/ h^{-1})	158.104	6.447	0.102	153.143	[118.152, 191.434]
f_{co-cl}	-0.214	16.565	–	-0.211	[-0.312, -0.181]
Residual variability					
ϵ	0.250	20.921		0.241	[0.196, 281]

CL , clearance; V , volume of distribution; f_{co-cl} , coefficient between co-administration and clearance; RSE, relative standard error; IIV, inter-individual variability; CV, coefficient of variation; 95%CI, 95% confidence interval

with the observations (fig. 4). Most observations were located in the 90% PI, indicating nice predictive properties of our model. Overall, the validation of the final population pharmacokinetic model by bootstrap and VPC confirmed satisfactory results.

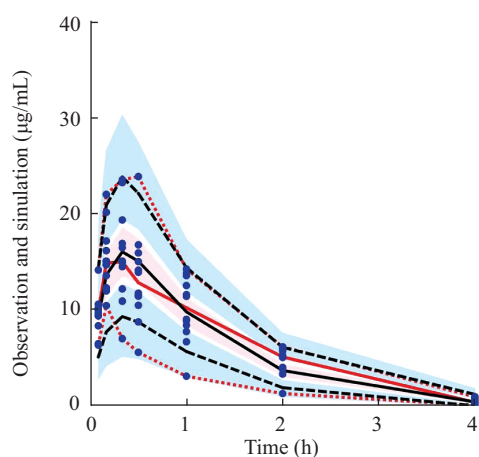


Fig. 4 Visual predictive check plots of final forsythiaside pharmacokinetic model

Dots, observations; black solid line, predicted 50th percentile; black dashed lines, 5th and 95th percentiles from the simulation; shadow, 95% confidence band; area between the 5th and 95th percentiles, 90% prediction interval; red dotted lines, observed 5th and 95th percentiles; red solid line, 50th percentiles

3 DISCUSSION

The use of traditional Chinese medicine and western medicine in the treatment of diseases is a major focus of pharmacotherapy in China^[24–26]. Since many Chinese medicines are over-the-counter drugs, patients often purchase them from pharmacies on their own, making the combined use of Chinese medicine and western medicine widespread in China. Many tertiary-level hospitals in China have clinics specializing in integrated Chinese and western medicine to allow patients to use both types, a practice that has been widely accepted by patients. The combination of azithromycin and Shuanghuanglian or Lianhuaqingwen in the treatment of pneumonia in children and community-acquired pneumonia is very common, and its efficacy has also been demonstrated by clinical observations^[18, 19]. The main antiviral active ingredient

of both Shuanghuanglian and Lianhuaqingwen is forsythiaside in the *Forsythia suspensa*. In order to eliminate the interference of honeysuckle and *Scutellaria baicalensis* against forsythiaside, we only tested forsythia extract. The combination with azithromycin was studied. When used in practice, the drug combination is accomplished through a sequential administration^[18] because direct mixing of the two drugs *in vitro* results in precipitation. In addition, intravenous administration may result in embolism, and thus we chose intragastric injections in the current work. This study aimed to investigate the pharmacokinetic basis of the combination of the two drugs from the perspective of pharmacokinetics. To the best of our knowledge, this study reports for the first time the pharmacokinetic interactions between forsythiaside and azithromycin.

NCA revealed that the use of azithromycin significantly increased the *in vivo* exposure of forsythiaside compared with forsythiaside alone, with the AUC_{last} increasing from 20.84 to 29.85, an increase of 43%. The increase in AUC_{last} was mainly due to a decrease in CL (from 175.56 to 111.54 mL/h) and an increase in C_{max} (from 14.06 to 19.34 μ g/mL). For these two parameters, there was a significant difference between the two groups ($P < 0.05$). Nevertheless, NCA does not describe the details of the drug's *in vivo* processes, i.e. the parameters obtained from NCA do not accurately predict drug concentration and drug concentration-time curves at different time points. Therefore, we also analyzed the data through a compartmental model analysis.

The results of the one-compartment model still need to be verified by further tests for three reasons. First, it may be necessary to accurately determine whether the *in vivo* process of the drug belongs to the one-compartment or multi-compartment model; an intravenous injection is preferred because extravascular administration involves absorption. This may obscure the rapid distribution phase of the drug and it is easy to misfit the multi-compartment model into a one-compartment model^[20]. Second, the distribution of blood collection points should be as dense as possible, especially for drugs with short half-life. It is easy to misfit the multi-compartment model as a one-compartment model with sparse blood collection time

points. Finally, the lower limit of the detection method should be as low as possible so that the drug *in vivo* process can be observed for a longer time, and so that the drug compartment analysis is more accurate.

As we know, macrolides inhibiting P450 have been widely studied, and may be one of the mechanisms of interaction with azithromycin. There will be some flocculent precipitates produced by the direct *in vitro* mixing of the two drugs. Although the two drugs were injected into different dosing sites to avoid their direct contact, we speculated that there may be interactions between the two drugs after their absorption into the blood. This may change the solubility of forsythiaside and result in an inhibition of its elimination process, thereby increasing the drug exposure in the blood. In addition, a reduced adverse reaction is associated with a lower dose, which can also lead to sufficient drug exposure to provide antibacterial activity. The co-administration of the two compounds is a potential approach to decrease adverse drug reactions while increasing therapeutic efficiency. However, excessive drug interactions may increase the number of precipitates and increase the risk of vascular embolism. Therefore, it is necessary to study the dose ratio of the combined drugs achieving an increase in drug exposure without an increase in the risk of adverse reactions.

The following limitations should be addressed. First, the mechanism of interaction with azithromycin is still unclear, and this should be further studied. Second, rats were used as experimental subjects in this research. However, the possibility of extrapolation from animals to humans is unknown since the mechanism is unclear. A clinical study is therefore needed to validate this result in the future. Additionally, since herb/plant extracts are considered to be effective in multiple components, the impact of other components on pharmacokinetics and efficacy is still unclear. Finally, it will be more profound if we create a control/sham group in which rats are given glucose solution or water.

To sum up, after the combined use of forsythia extract and azithromycin, azithromycin will slow the elimination of forsythiaside and increase its peak concentration, thereby increasing its exposure *in vivo*. These results were in accordance with our previous study^[20]. From the perspective of pharmacokinetics, this study provides certain animal experimental support for the combined application of traditional Chinese medicine and western medicines in practice. However, the efficacy and safety of these drugs for COVID-19 still need to be further confirmed by clinical experiments.

Conflict of Interest Statement

The authors declare that there is no conflict of interest with any financial organization or corporation or individual that can inappropriately influence this work.

REFERENCES

- 1 Runfeng L, Yunlong H, Jicheng H, *et al*. Lianhuaqingwen exerts anti-viral and anti-inflammatory activity against novel coronavirus (SARS-CoV-2). *Pharmacol Res*, 2020,156:104761
- 2 Wang L, Xu X, Ruan J, *et al* Quadruple therapy for asymptomatic COVID-19 infection patients. *Expert Rev Anti Infect Ther*, 2020,18(7):617-624
- 3 Lu H. Drug treatment options for the 2019-new coronavirus (2019-nCoV). *Biosci Trends*, 2020,14(1): 69-71
- 4 Ni L, Zhou L, Zhou M, *et al* Combination of western medicine and Chinese traditional patent medicine in treating a family case of COVID-19. *Front Med*, 2020,14(2):210-214
- 5 Wang F, Cao GS, Li Y, *et al* Characterization of forsythoside A metabolites in rats by a combination of UHPLC-LTQ-Orbitrap mass spectrometer with multiple data processing techniques. *Biomed Chromatogr*, 2018,32(5):e4164
- 6 Zhang S, Shao SY, Song XY, *et al* Protective effects of Forsythia suspense extract with antioxidant and anti-inflammatory properties in a model of rotenone induced neurotoxicity. *Neurotoxicology*, 2016,52:72-83
- 7 Wu ZQ, Liu GH, Yan LJ, *et al*. Experimental study on anti-influenza virus infection with yinqiao-decoction by orthogonal design. *Zhonghua Shi Yan He Lin Chuang Bing Du Xue Za Zhi (Chinese)*, 2010,24(6):427-429
- 8 Zeng XY, Yuan W, Zhou L, *et al* Forsythoside A exerts an anti-endotoxin effect by blocking the LPS/TLR4 signaling pathway and inhibiting Tregs *in vitro*. *Int J Mol Med*, 2017,40(1):243-250
- 9 Law AH, Yang CL, Lau AS, *et al* Antiviral effect of forsythoside A from Forsythia suspensa (Thunb.) Vahl fruit against influenza A virus through reduction of viral M1 protein. *J Ethnopharmacol*, 2017,209:236-247
- 10 Qu H, Zhang Y, Wang Y, *et al* Antioxidant and antibacterial activity of two compounds (forsythiaside and forsythin) isolated from Forsythia suspensa. *J Pharm Pharmacol*, 2008,60(2):261-266
- 11 Bakheit AH, Al-Hadiya BM, Abd-Elgalil AA. Azithromycin. *Profiles Drug Subst Excip Relat Methodol*, 2014,39:1-40
- 12 Ma WG, Jia JM, The effects and prospects of the integration of traditional Chinese medicine and Western medicine on andrology in China. *Asian J Androl*, 2011,13(4):592-595
- 13 Lampe H, Halle B, Freund M. Western and Chinese medicine in oncology and hematology. *Forsch Komplementmed*, 2011,18(4):185-191
- 14 Zhang C, Wang Y, Zhao X, *et al*. Prediction of Recurrent Stroke or Transient Ischemic Attack After Noncardiogenic Posterior Circulation Ischemic Stroke. *Stroke*, 2017,48(7):1835-1841
- 15 Qing GC, Zhang H, Bai Y, *et al*. Traditional Chinese and Western Medicines Jointly Beat COVID-19 Pandemic. *Chin J Integr Med*, 2020,26(6):403-404
- 16 Ren JL, Zhang AH, Wang XJ. Traditional Chinese medicine for COVID-19 treatment. *Pharmacol Res*, 2020,155:104743
- 17 Yang Y, Islam MS, Wang J, *et al*. Traditional Chinese Medicine in the Treatment of Patients Infected with

- 2019-New Coronavirus (SARS-CoV-2): A Review and Perspective. *Int J Biol Sci*, 2020,16(10):1708-1717
- 18 Liu N, Chen X, Yin L. Clinical effect of azithromycin sequential therapy combined with shuanghuanglian oral solution on pediatric mycoplasma pneumonia and the impact on serum inflammatory cytokines levels and immunologic function. *Pract J Cardiac Cereb Pneum Vasc Dis*, 2016,24(9):94-97
- 19 Wang H. Efficacy of shuanghuanglian oral liquid only for children combined with azithromycin in the treatment of mycoplasma pneumonia in children. *Chin Foreign Med Res*, 2018,16(7):25-27
- 20 Tian J, Sun S, Zhao Z, *et al.* Pharmacokinetic interaction between shuanghuanglian and azithromycin injection: a nonlinear mixed-effects model analysis in rats. *Xenobiotica*, 2019,49(11):1344-1351
- 21 Tian JC, Lin LF, Li XC, *et al.* Forsythiaside stability in pretreated rat plasma and its pharmacokinetics after i.v. administration. *Anal Methods-Uk*, 2015,7(5):1809-1815
- 22 Wang H, Li X, Sun S, *et al.* Population Pharmacokinetics and Dosing Simulations of Ceftazidime in Chinese Neonates. *J Pharm Sci*, 2018,107(5):1416-1422
- 23 Hooker AC, Staats CE, Karlsson MO. Conditional weighted residuals (CWRES): a model diagnostic for the FOCE method. *Pharm Res*, 2007,24(12):2187-2197
- 24 Zhang L, Yu J, Zhou Y, *et al.* Becoming a Faithful Defender: Traditional Chinese Medicine against Coronavirus Disease 2019 (COVID-19). *Am J Chin Med*, 2020,48(4):763-777
- 25 Li T, Lu H, Zhang W. Clinical observation and management of COVID-19 patients. *Emerg Microbes Infect*, 2020,9(1):687-690
- 26 Ho LTF, Chan KKH, Chung VCH, *et al.* Highlights of traditional Chinese medicine frontline expert advice in the China national guideline for COVID-19. *Eur J Integr Med*, 2020,36:101116
- (Received Jan. 28, 2021; accepted Oct. 21, 2021)