Pharmacokinetics of single- and multiple-dose roflumilast: an open-label, three-way crossover study in healthy Chinese volunteers

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Purpose: To determine the pharmacokinetic properties of the common tablet of roflumilast administered in single and multiple oral doses in Chinese subjects.

Subjects and methods: Both the single- and multiple-dose studies included 12 adults (6 males and 6 females). In this single-center, open-label study, single doses of 0.25, 0.375, and 0.5 mg were administered using a randomized, three-way crossover design, and then, the 0.375 mg dose was continued for 11 days once daily. The pharmacokinetic parameters for roflumilast and roflumilast N-oxide were determined and the safety evaluation included adverse events assessed by monitoring, physical examination, vital sign tests, and clinical laboratory tests.

Results: After every single dose, the time to the maximum concentration ($C_{\text{max}}$) of roflumilast ($T_{\text{max}}$) was 0.25–2.0 hours; thereafter, the concentration declined, with a mean half-life ($t_{1/2}$) of 19.7–20.9 hours over the range of 0.25–0.50 mg. As for roflumilast N-oxide, the mean $T_{1/2}$ was 23.2–26.2 hours. The area under curve from the beginning to 24 hours (AUC$_{0-24}$), the AUC until infinity (AUC$_{\infty}$), and the $C_{\text{max}}$ of roflumilast and roflumilast N-oxide increased in a dose-proportional manner. After multiple doses, the accumulation index (R$_{1}$) on the 11th day of the steady state was ~1.63 for roflumilast and 3.20 for roflumilast N-oxide. No significant sex differences were observed in the pharmacokinetic parameters of roflumilast and roflumilast N-oxide. In addition, there were no serious adverse events across the trial.

Conclusion: Roflumilast was safe and well-tolerated in healthy volunteers, and a linear increase in its $C_{\text{max}}$ and AUC values was observed at doses ranging from 0.25 to 0.50 mg.

Keywords: pharmacokinetics, roflumilast, roflumilast N-oxide, healthy volunteer, phosphodiesterase 4 inhibitor

Introduction

Roflumilast is a benzamide compound designed for the treatment of asthma and COPD.1–5 A phosphodiesterase 4 (PDE4) inhibitor (Figure 1A) is extensively metabolized by I (cytochrome [CYP]450) and II phase (conjugation) reactions.6 The only major metabolite of roflumilast found in human plasma is roflumilast N-oxide (Figure 1B), whose formation is mainly catalyzed by CYP3A4. Although roflumilast is three times more potent than roflumilast N-oxide upon the inhibition of the PDE4 enzyme in vitro, the plasma area under the curve (AUC) of roflumilast N-oxide on average is ~10-fold greater than that of roflumilast.7 After oral administration of a 0.5 mg dose of roflumilast, its absolute bioavailability was ~80%. The maximum concentration ($C_{\text{max}}$) for roflumilast and roflumilast N-oxide is usually observed at ~1 hour (range 0.5–2 hours) and 8 hours (range 4–13 hours) after administration, respectively. Food intake does not affect the total inhibitory activity of PDE4,8 but delays the time to the $C_{\text{max}}$ of roflumilast ($T_{\text{max}}$).
by 1 hour and decreases $C_{\text{max}}$ by $\sim40\%$. However, the $C_{\text{max}}$ and $T_{\text{max}}$ of roflumilast $N$-oxide are not influenced at all. The effective half-life ($t_{1/2}$) ranges from 8 to 31 hours after dosing of 0.5 mg roflumilast; thus, these findings support a daily dosing regimen of roflumilast.

However, to our knowledge, there is little literature about the pharmacokinetics of roflumilast and roflumilast $N$-oxide in healthy Chinese subjects. As a result, it is important to investigate the pharmacokinetics of these two compounds. This study was designed to evaluate the pharmacokinetics of roflumilast and roflumilast $N$-oxide in healthy Chinese volunteers.

**Subjects and methods**

**Eligibility**
A total of 12 healthy Chinese volunteers including six males and six females were screened for inclusion. The volunteers were aged between 18 and 45 years, with a body mass index (BMI) of 19–24 kg/m$^2$. The minimum body weight was 45 and 50 kg for females and males, respectively. None of the volunteers had significant cardiac, hepatic, renal, pulmonary, neurologic, gastrointestinal, or hematologic disorders, as confirmed by performing a physical examination. Besides, those who had abused alcohol in the past 6 months or those who smoked heavily (more than five cigarettes a day) 3 months prior to the study were excluded.

The study was conducted at a Phase I clinical center in the Third Xiangya Hospital of Central South University. All participants were informed of the aim and risks by a clinical investigator, and each submitted written informed consent before participating in the study.

**Study drug**
Roflumilast tablets (0.25 mg, No 150101C, expiration date: January 2017; 0.375 mg, No 150101B, expiration date: January 2017; 0.5 mg, No 150101A, expiration date: January 2017) were manufactured and provided by Sichuan Baili Pharmaceutical Industry (Sichuan, P.R. China).

**Study design and drug administration**
This was a randomized, single-center, open-label trial based on a flowchart (Figure 2) wherein the pharmacokinetics of single and multiple oral administration of roflumilast were studied. A single-dose pharmacokinetic test was in the form of $3 \times 3$ Latin square crossover design. The subjects were admitted to the I phase ward one night before each cycle trial and fasted for more than 10 hours before the treatment. Then, a single dose of 0.25, 0.375, and 0.5 mg was administrated in the morning on the first day of each study period. The single dose was 0.25, 0.375, or 0.5 mg coupled with 250 mL of boiled water. The patients were not allowed to drink water 2 hours before and after dosing. Four and 10 hours after dosing, standard lunch and dinner, respectively, were allowed. After the completion of the single-dose test, multiple doses of 0.375 mg were administrated once daily for 11 days continuously.

**Blood collection**
For the single-dose test, blood samples were collected from the cubital vein before dosing and 0.25, 0.5, 0.75, 1, 1.5, 2.5, 3, 4, 6, 8, 10, 12, 14, 24, 30, 36, 48, 72, and 96 hours after dosing. The washout period for each group was 10 days. For the multiple-dose test, blood samples were collected before

![Figure 1](image_url)
dosing and 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 4, 6, 8, 10, 12, 14, 24, 30, 36, 48, 72, 96 hours after dosing on the 11th day.

Sample preparation
A volume of 5 mL of blood was collected into heparinized tubes and centrifuged at 2,700 rpm at 4°C for 10 minutes. The collected plasma was transferred to Eppendorf tubes, which were maintained at −70°C until the concentration analysis.

Analytical assays
High-performance liquid chromatography–tandem mass spectrometry was used to determine the concentration of N-oxide in the samples at different time points after administration. An LC–MS/MS quantitation method was developed to simultaneously determine roflumilast and roflumilast N-oxide in human plasma with rather low limits of quantitation (0.02 ng mL⁻¹ for roflumilast and 0.04 ng mL⁻¹ for roflumilast N-oxide). Human plasma samples were prepared by solid-phase extraction, which ensured high recovery and slight matrix effect for both analyses. In each validation batch, the calibration standards were analyzed and the calibration curve was linear within the range of 0.02–10 ng mL⁻¹ and 0.04–50 ng mL⁻¹ for roflumilast and roflumilast N-oxide, respectively. The regression coefficients of all the calibration curves were >0.99. The average bias among lower quality control (LQC), middle quality control (MQC), and higher quality control (HQC) samples was <15% compared with the nominal concentration, and the coefficient of variation (CV) of each concentration level was <15% as well, both for inter-run precision and intra-run precision. The developed method was successfully applied for the pharmacokinetic research in Chinese healthy volunteers after oral administration of 0.25, 0.375, and 0.5 mg roflumilast tablet.

Safety assessment
Safety was assessed according to interviews and adverse event (AE) monitoring. Vital sign tests, physical examinations, clinical laboratory tests, and electrocardiography were performed for each subject at screening and at study completion. AEs were assessed by direct observation, by using spontaneous

Figure 2 Flowchart of the study design. Blank indicating three phases of single-dose trial and multiple-dose trial until sample collection.
Notes: The blue arrow indicates 12 volunteers divided into randomized three doses groups, experiencing 10-day washout. Yellow arrow indicates 12 subjects entering into multiple trial after 10-day washout with the last single-dose administration.
reports, and by nonspecific inquiry and their clinical significance was determined by the monitoring physician. All information, including symptoms and signs before and after dosing were recorded in case report forms by investigators regardless of whether a relationship with the study drug was suspected.

Pharmacokinetic analysis
First, the pharmacokinetic analysis was performed by noncompartmental analysis using WinNonlin version 6.3 (Pharsight Corporation, Mountain View, CA, USA). Plasma roflumilast and roflumilast N-oxide concentrations vs time data after single-dose administration included \( C_{\text{max}} \) and \( T_{\text{max}} \). The AUC from the beginning to the last values (AUC\(_{\text{last}}\)) was determined using the linear trapezoidal method as was the AUC\(_{0-24\text{h}}\), which indicated the AUC from the beginning to 24 hours. The first-order rate decline constant (\( \lambda_z \)) for roflumilast plasma concentrations in the terminal phase of the concentration–time curve was determined using log-linear regression, and the \( t_{1/2} \) was estimated from \ln2/\( \lambda_z \). AUC\(_{\text{int}}\) was determined using the following equation: AUC\(_{\text{last}}\) + AUC\(_{\text{int}}\)/\( \lambda_z \). Clearance rate (CL/F) was estimated as dose divided by AUC\(_{\text{int}}\), and the volume of distribution (V/F) was determined by dividing the apparent CL/F by \( \lambda_z \). The multiple-dose study provided \( T_{\text{max}} \) at steady state (\( T_{\text{max},2} \)), \( C_{\text{max}} \) at steady state (\( C_{\text{max},2} \)), and minimum plasma concentration at steady state (\( C_{\text{min},2} \)). The accumulation index was determined as AUC\(_{0-24\text{h}}\)/(AUC\(_{0-24\text{h}}\) \( \times \) day 1). The mean plasma concentration vs time profiles for roflumilast plasma concentrations in the terminal phase of the concentration–time curve was determined using log-linear regression, and the \( t_{1/2} \) was estimated from \ln2/\( \lambda_z \). AUC\(_{\text{int}}\) was determined using the following equation: AUC\(_{\text{last}}\) + AUC\(_{\text{int}}\)/\( \lambda_z \). Clearance rate (CL/F) was estimated as dose divided by AUC\(_{\text{int}}\), and the volume of distribution (V/F) was determined by dividing the apparent CL/F by \( \lambda_z \). The multiple-dose study provided \( T_{\text{max}} \) at steady state (\( T_{\text{max},2} \)), \( C_{\text{max}} \) at steady state (\( C_{\text{max},2} \)), and minimum plasma concentration at steady state (\( C_{\text{min},2} \)). The accumulation index was determined as AUC\(_{0-24\text{h}}\)/(AUC\(_{0-24\text{h}}\) \( \times \) day 1). The mean plasma concentration vs time profiles for roflumilast plasma concentrations in the terminal phase of the concentration–time curve was determined using log-linear regression, and the \( t_{1/2} \) was estimated from \ln2/\( \lambda_z \). AUC\(_{\text{int}}\) was determined using the following equation: AUC\(_{\text{last}}\) + AUC\(_{\text{int}}\)/\( \lambda_z \). Clearance rate (CL/F) was estimated as dose divided by AUC\(_{\text{int}}\), and the volume of distribution (V/F) was determined by dividing the apparent CL/F by \( \lambda_z \). The multiple-dose study provided \( T_{\text{max}} \) at steady state (\( T_{\text{max},2} \)), \( C_{\text{max}} \) at steady state (\( C_{\text{max},2} \)), and minimum plasma concentration at steady state (\( C_{\text{min},2} \)). The accumulation index was determined as AUC\(_{0-24\text{h}}\)/(AUC\(_{0-24\text{h}}\) \( \times \) day 1). The shape of the curves was similar between the various dose groups. As shown in Table 2, the mean \( T_{\text{max}} \) values for roflumilast among the three dose groups were within 0.25–2.0 hours. The mean \( t_{1/2} \) values of roflumilast and roflumilast N-oxide were ~20 and 25 hours, respectively. The differences in \( t_{1/2} \) and CL/F among these dose groups were not statistically significant (P>0.05).

Table 1 Baseline demographic characteristics of study volunteers (n=12)

|          | Mean±SD (relative standard deviation) |          |          |          |          |
|----------|--------------------------------------|----------|----------|----------|----------|
|          | Age (years) | Height (cm) | Weight (kg) | BMI (kg/m²) |
| Males (n=6) | 24±1 (4.0%)  | 171±4 (2.33%) | 62.4±7.1 (11.4%) | 21.3±2.2 (10.3%) |
| Females (n=6) | 21±4 (19.0%) | 157±5 (3.18%) | 53.7±6.7 (12.5%) | 21.7±2.2 (10.1%) |

**Abbreviations:** BMI, body mass index; n, sample size.

Statistical analyses
Statistical analyses were performed using SPSS version 18.0. All data were expressed as mean ± SD values. Log-transformed pharmacokinetic parameters AUC\(_{0-24\text{h}}\), AUC\(_{\text{int}}\), and \( C_{\text{max}} \) were analyzed to determine dose proportionality using the power model, PK=A×(dose)\(^{\beta} \), where PK was the pharmacokinetic parameter, A was the intercept, and \( \beta \) was the dose-proportionality coefficient. In case the 90% CI for the dose-proportionality parameter \( \beta \) was included in the estimation interval, the pharmacokinetic parameters would correspond to perfect dose proportionality. Pharmacokinetic parameters were compared among dose levels by using ANOVA, including sex-related differences. A P-value of <0.05 was considered significant.

Results
Demographic characteristics
Twelve healthy Chinese volunteers (Table 1) with the following characteristics were enrolled in this study: age, 21 ± 3 years (range, 18.0–27.0 years); weight, 61.3 ± 5.4 kg (range, 47.0–73.5 kg); and height, 171 ± 4 cm (range, 153–176 cm). All participants completed the study and were included in the PK analysis.

Noncompartmental analysis of single-dose administration
The mean plasma concentration vs time profiles for roflumilast and roflumilast N-oxide after administration of a single dose of 0.25, 0.375, or 0.50 mg were shown in Figure 3. The shape of the curves was similar between the various dose groups. As shown in Table 2, the mean \( T_{\text{max}} \) values for roflumilast among the three dose groups were within 0.25–2.0 hours. The mean \( t_{1/2} \) values of roflumilast and roflumilast N-oxide were ~20 and 25 hours, respectively. The differences in \( t_{1/2} \) and CL/F among these dose groups were not statistically significant (P>0.05). Furthermore,
Figure 3  Mean (SD) plasma concentration–time curves of roflumilast and roflumilast N-oxide following 0.25, 0.375, and 0.5 mg single dose in ordinary coordinates and semi-log coordinates.

Notes: (A) Ordinary coordinates curves for roflumilast. (B) Semi-log coordinates curves for roflumilast. (C) Ordinary coordinates curves for roflumilast N-oxide. (D) Semi-log coordinates curves for roflumilast N-oxide.

Table 2  Pharmacokinetic parameters of the roflumilast and roflumilast N-oxide after single dose

| Analytes              | Parameters | Dose       |
|-----------------------|------------|------------|
|                       |            | 0.25 mg    | 0.375 mg  | 0.5 mg    |
| Roflumilast           | AUC<sub>0–24</sub> (h ng ml<sup>-1</sup>) | 24.1 (8.2) | 35.70 (12.0) | 47.9 (16.3) |
| Roflumilast           | AUC<sub>inf</sub> (h ng ml<sup>-1</sup>) | 33.8 (17.3) | 48.70 (23.4) | 65.1 (34.4) |
| Roflumilast           | AUC<sub>last</sub> (h ng ml<sup>-1</sup>) | 33.4 (15.8) | 49.4 (22.8) | 67.5 (35.0) |
|                       | CL/F (L h<sup>-1</sup>) | 9.2 (4.5) | 9.3 (4.0) | 9.5 (4.3) |
|                       | C<sub>max</sub> (ng ml<sup>-1</sup>) | 6.4 (1.1) | 9.2 (1.9) | 12.5 (2.9) |
|                       | t<sub>1/2</sub> (h) | 0.041 (0.0267) | 0.042 (0.0247) | 0.0429 (0.0251) |
|                       | T<sub>max</sub> (h) | 0.75 (0.50–1.50) | 0.50 (0.25–2.00) | 0.75 (0.50–1.50) |
|                       | V/F (L) | 243.0 (94.7) | 237.0 (82.3) | 234.0 (70.4) |
| Roflumilast N-oxide   | AUC<sub>0–24</sub> (h ng ml<sup>-1</sup>) | 128.0 (30.4) | 192.0 (32.1) | 266.0 (54.7) |
| Roflumilast N-oxide   | AUC<sub>inf</sub> (h ng ml<sup>-1</sup>) | 333.0 (103.0) | 493.0 (148.0) | 649.0 (189.0) |
| Roflumilast N-oxide   | AUC<sub>last</sub> (h ng ml<sup>-1</sup>) | 320.0 (75.1) | 458.0 (107.0) | 630.0 (172.0) |
|                       | C<sub>max</sub> (ng ml<sup>-1</sup>) | 6.7 (1.7) | 10.4 (2.1) | 14.7 (3.7) |
|                       | t<sub>1/2</sub> (h) | 0.031 (0.00986) | 0.0284 (0.0083) | 0.0317 (0.0075) |
|                       | T<sub>max</sub> (h) | 4.00 (2.50–24.00) | 4.00 (3.00–24.00) | 4.00 (2.50–24.00) |

Note: *T<sub>max</sub>*: median (min–max), the other parameters of pharmacokinetic: mean (SD).

Abbreviations: AUC, area under the curve; AUC<sub>inf</sub>, AUC until infinity; AUC<sub>0–24</sub>, AUC from the beginning to 24 hours; AUC<sub>last</sub>, AUC from the beginning to the last values; C<sub>max</sub>, maximum concentration; t<sub>1/2</sub>, half-life; T<sub>max</sub>, the time to the C<sub>max</sub> of roflumilast; CL/F, clearance rate; V/F, volume of distribution.
the differences in these main pharmacokinetic parameters between male and female patients were not statistically significant (P > 0.05) (Table 3).

As shown in Table 4, the power model estimating the linearity of a single dose ranging from 0.25 to 0.50 mg was analyzed. The 90% CIs for the slope of the relational equation were 0.869–1.103 (C_max), 0.930–1.099 (AUC_0–24 h), and 0.890–1.085 (AUC_inf) for roflumilast, whereas those for roflumilast N-oxide were 1.020–1.198 (C_max), 0.984–1.116 (AUC_0–24 h), and 0.852–1.059 (AUC_inf), all of which were within the estimation interval (0.8, 1.25). The pharmacokinetic parameters of roflumilast in the human body at a dose range of 0.25–0.50 mg conformed to the linear pharmacokinetic process.

Noncompartmental analysis of multiple-dose administration

After multiple doses of roflumilast 0.375 mg was administered over 11 days, the pharmacokinetic parameters T_max (0.75 vs 0.5, P > 0.05), CL/F (8.6 vs 9.4, P > 0.05), and C_max (11.4 vs 9.2, P > 0.05) for roflumilast were similar to those observed in the single-dose study. For roflumilast N-oxide, some pharmacokinetic parameters were relatively higher, such as AUC_0–24 h (618.0 vs 192.0, P < 0.05), AUC_inf (1,320.0 vs 493.0, P < 0.05), AUC_inf (1,410.0 vs 458.0, P < 0.05), and C_max (35.4 vs 10.4, P < 0.05). In addition, the accumulation index (R_inf) was around 1.63 for roflumilast and 3.20 for roflumilast N-oxide. The DF was 4.85 for roflumilast and 0.56 for roflumilast N-oxide (Table 5 and Figure 4).

Compartmental analysis of single- and multiple-dose administration

According to statistical analysis and “goodness-of-fit” criteria, a weighted (1/Y^2) two-compartment pharmacokinetic model was adopted to describe roflumilast activity in plasma, and a one-compartment pharmacokinetic model was adopted to describe roflumilast N-oxide activity in plasma following three single oral doses of 0.25, 0.375, and 0.5 mg as well as multiple oral doses of 0.375 mg. The compartment pharmacokinetic parameters are summarized in Table 6.

Safety and tolerability

As seen in Table 7, during the course of the single- and multiple-dose studies, a total of 48 treatment-related adverse events were noted, of which 19 were considered as certainly related to treatment; 5, probably related to treatment; and 24, possibly related to treatment. Seven TRAEs were reported by

Table 3 The comparison of pharmacokinetic parameters in different treatments and genders

| Factors | Parameters | Roflumilast | Roflumilast N-oxide |
|---------|------------|-------------|-------------------|
|         | AUC_0–24 h | C_max       | T_1/2             |
| Dose    | —          | 0.905       | 0.870             |
| Gender  | AUC_0–24 h | 0.824       | 0.668             |
|         | —          | 0.402       | 0.197             |

Abbreviations: AUC, area under the curve; AUC_0–24 h, AUC from the beginning to 24 hours; C_max, maximum concentration; T_1/2, half-life.

Table 4 Dose proportionality following 0.25–0.5 mg doses

| Analytes | Parameters | Slope β (90% CI) | Criterion |
|----------|------------|-----------------|-----------|
| Roflumilast | AUC_0–24 h | 1.011 (0.930, 1.099) | (0.800, 1.250) |
|          | AUC_ss     | 0.983 (0.890, 1.085) | (0.800, 1.250) |
|          | C_max      | 0.979 (0.869, 1.103) | (0.800, 1.250) |
| Roflumilast N-oxide | AUC_0–24 h | 1.048 (0.984, 1.116) | (0.800, 1.250) |
|          | AUC_ss     | 0.950 (0.852, 1.059) | (0.800, 1.250) |
|          | C_max      | 1.105 (1.020, 1.198) | (0.800, 1.250) |

Abbreviations: AUC, area under the curve; AUC_0–24 h, AUC until infinity; AUC_ss, AUC from the beginning to 24 hours.

Table 5 Pharmacokinetic parameters of the roflumilast and roflumilast N-oxide after multiple doses

| Parameters | 0.375 mg |
|------------|----------|
| Roflumilast | AUC_0–24 h (h ng ml^-1) | 59.3 (32.7) |
|            | AUC_ss (h ng ml^-1)     | 81.4 (49.7) |
|            | AUC_inf (h ng ml^-1)    | 94.0 (70.7) |
|            | CL/F (L h^-1)           | 8.6 (4.0)   |
|            | C_max (ng ml^-1)        | 2.5 (1.4)   |
|            | C_inf (ng ml^-1)        | 11.40 (3.6) |
| DF         |                       | 4.85 (1.95) |
|            | R_inf (h^-1)           | 0.0298 (0.00952) |
|            | t_1/2 (h)              | 25.6 (8.5)  |
|            | t_max (h)              | 0.75 (0.50–2.50) |
|            | V/F (L)                | 329.0 (269.0) |
| Roflumilast N-oxide | AUC_0–24 h (h ng ml^-1) | 618.0 (223.0) |
|            | AUC_ss (h ng ml^-1)    | 1,320.0 (624.0) |
|            | AUC_inf (h ng ml^-1)   | 1,410.0 (696.0) |
|            | C_max (ng ml^-1)       | 25.8 (9.3)  |
|            | C_inf (ng ml^-1)       | 35.4 (13.8) |
| DF         |                       | 0.56 (0.13) |
|            | R_inf (h^-1)          | 0.0286 (0.00636) |
|            | t_1/2 (h)             | 25.2 (5.1)  |
|            | t_max (h)             | 3.00 (1.00–4.00) |

Note: T_max, median (min–max), the other parameters of pharmacokinetic: mean (SD).

Abbreviations: AUC, area under the curve; AUC_0–24 h, AUC until infinity; AUC_0–24 h, AUC from the beginning to 24 hours; AUC_inf, AUC from the beginning to the last values; C_max, the average steady-state drug concentration during multiple-dosing intervals; C_inf, maximum concentration; DF, degree of fluctuation; R_inf, the accumulation index; t_1/2, half-life; T_max, the time to the C_max of roflumilast; V/F, volume of distribution.
Figure 4 Mean (SD) plasma concentration–time curves of roflumilast and roflumilast N-oxide following 0.375 mg multiple doses.

Notes: (A) Ordinary coordinates curves for roflumilast. (B) Semi-log coordinates curves for roflumilast. (C) Ordinary coordinates curves for roflumilast N-oxide. (D) Semi-log coordinates curves for roflumilast N-oxide.

Four subjects while receiving the 0.25 mg single dose, six by six subjects while receiving the 0.375 mg single dose, 10 by six subjects while receiving the 0.50 mg single dose, and 25 by 12 subjects while receiving 0.375 mg multiple doses. Dizziness (53.8%), nausea (53.8%), diarrhea (30.8%), and headache (23.1%) were the more common TRAEs. Forty-six TRAEs were of mild degree, whereas only one TRAE presenting as diarrhea was of severe degree, and one TRAE presenting as acute gastroenteritis was of moderate degree. All these TRAEs resolved without sequelae.

Table 6 The parameters of the two-compartment model of roflumilast and one-compartment model of roflumilast N-oxide

| Analytes                  | Parameters | Single dose | Multiple doses |
|---------------------------|------------|-------------|----------------|
|                           |            | 0.25 mg     | 0.375 mg       | 0.5 mg         | 0.375 mg       |
| Roflumilast               | Kᵦ (h⁻¹)  | 12.746 (13.6)| 15.270 (13.44)| 15.196 (16.558)| 14.782 (10.327)|
|                           | Kᵦ (h⁻¹)  | 0.259 (0.092)| 0.258 (0.124)| 0.254 (0.115)| 0.205 (0.057)|
|                           | Kᵦ (h⁻¹)  | 0.324 (0.112)| 0.310 (0.141)| 0.300 (0.101)| 0.292 (0.168)|
|                           | Kᵦ (h⁻¹)  | 0.101 (0.033)| 0.096 (0.038)| 0.097 (0.036)| 0.082 (0.036)|
|                           | Tᵦ (h)     | 0.304 (0.108)| 0.296 (0.129)| 0.241 (0.033)| 0.283 (0.103)|
|                           | V₁/F (L)   | 33.148 (6.036)| 35.031 (8.793)| 35.02 (8.804)| 35.721 (10.918)|
| Roflumilast N-oxide       | Kᵦ (h⁻¹)  | 1.486 (0.499)| 1.748 (1.058)| 1.440 (0.631)| 2.422 (1.166)|
|                           | Kᵦ (h⁻¹)  | 0.020 (0.012)| 0.021 (0.012)| 0.021 (0.012)| 0.024 (0.008)|
|                           | Tᵦ (h)     | 0.260 (0.112)| 0.246 (0.150)| 0.213 (0.082)| 0.232 (0.182)|
|                           | V₁/F (L)   | 40.13 (12.686)| 37.566 (9.235)| 37.402 (10.657)| 29.230 (8.373)|

Notes: Parameters: mean (SD). Kᵦ: rate constant from the absorption chamber into the central chamber; Kᵦ: elimination rate constant of the central chamber; Kᵦ: transportation rate constant from the central chamber into the peripheral chamber; Kᵦ: transportation rate constant from the peripheral chamber into the central chamber; Tᵦ: lag time; V₁/F: apparent volume of the central compartment.
COPD is a crucial factor causing morbidity and mortality and has a substantial global economic and social burden. The present approach for the management of COPD involves initial treatment with bronchodilators, followed by inhaled steroids when required. However, many patients still experience exacerbations despite access to the currently available therapy.

A previous study confirmed that oral roflumilast at doses of 0.10, 0.25, and 0.50 mg once daily showed statistically significant effects with dose-related increases from baseline based on forced expiratory volume in 1 second in patients with asthma. It is clear that roflumilast at 0.50 mg once daily is more effective than lower doses with well tolerance. In a randomized double-blind controlled trial of roflumilast treatment for acute exacerbations of COPD, patients treated with roflumilast over 4 weeks experienced a statistically significant greater reduction in the percentage of sputum neutrophils and sputum myeloperoxidase concentration, which tended to have beneficial effects on lung function.

As seen in Table 8, Bethke et al used a two-period, two-sequence crossover study in 15 subjects receiving immediate-release tablets of roflumilast 0.25 or 0.5 mg to investigate the single-dose pharmacokinetics of the drug. However, that study was based on Caucasians. Moreover, the previous literature, which described a parallel-group study conducted in healthy Chinese subjects, did not show considerable dose-proportional pharmacokinetic characteristics because of interindividual variability. This article used a 3×3 Latin square crossover design to investigate single-dose pharmacokinetics while reducing interindividual differences. The report showed that after a single oral dose of 0.25 and 0.5 mg roflumilast, the Cmax and AUCinf of roflumilast and roflumilast N-oxide increased along with the dose of the drug. In our study, after single oral administration of 0.25, 0.375, and 0.5 mg of roflumilast, the Cmax values were 6.38, 9.24, and 12.50 ng mL⁻¹ and the AUCinf values were 33.80, 48.7, and 65.10 h ng mL⁻¹ for roflumilast, respectively, while the Cmax values were 6.74, 10.4, and 14.7 ng mL⁻¹ and AUCinf values were 333.0, 493.0, and 649.0 h ng mL⁻¹ for roflumilast N-oxide, respectively, confirming the linear pharmacokinetic characteristics of the drug. The Cmax of roflumilast was attained within 1.0 hours in healthy volunteers and the AUCinf of roflumilast N-oxide is around 10 times more than that of roflumilast, which is in accordance with the previous literature.

It appears that after single oral administration of roflumilast, the degree of roflumilast exposure in Chinese patients was slightly higher than that in the foreign population. Population pharmacokinetic models have demonstrated

### Table 7 Number of subjects (%) reporting treatment-related adverse events (TRAEs) following single and multiple doses of roflumilast

| Condition                        | Single dose (n=12/13) | Multiple doses (n=12) | Total TRAEs (n=48) | Total subjects (n=13) |
|----------------------------------|-----------------------|-----------------------|--------------------|-----------------------|
|                                  | 0.25 mg (n=12)        | 0.375 mg (n=13)       | 0.50 mg (n=13)     |                       |
| Any adverse event                |                       |                       |                    |                       |
| Dizziness                        | 4 (33.3)              | 6 (46.2)              | 8 (61.5)           | 12 (100)              |
| Nausea                           | 3 (25)                | 2 (15.4)              | 5 (38.5)           | 5 (41.7)              |
| Headache                         | 1 (8.3)               | 1 (7.7)               | 1 (7.7)            | 1 (7.7)               |
| Diarrhea                         |                       |                       |                    |                       |
| Upper respiratory infection      | 1 (8.3)               | 1 (7.7)               | 1 (7.7)            | 1 (7.7)               |
| Increased total bilirubin        | 1 (8.3)               | 1 (7.7)               | 1 (7.7)            | 1 (7.7)               |
| Chest distress                   | 1 (8.3)               | 1 (7.7)               | 1 (7.7)            | 1 (7.7)               |
| Elevated creatine kinase         | 1 (8.3)               | 1 (7.7)               | 1 (7.7)            | 1 (7.7)               |
| Potassium reduction              |                       |                       |                    |                       |
| Fatigue                          |                       |                       |                    |                       |
| Loss of appetite                 | 1 (7.7)               | 1 (7.7)               | 1 (7.7)            | 1 (7.7)               |
| Elevated uric acid               |                       |                       |                    |                       |
| Acute gastroenteritis            | 1 (7.7)               | 1 (7.7)               | 1 (7.7)            | 1 (7.7)               |
| Proteinuria                      | 1 (7.7)               | 1 (7.7)               | 1 (7.7)            | 1 (7.7)               |
| Back pain in the right side      |                       |                       |                    |                       |
| Related to drug                  |                       |                       |                    |                       |
| Certainly                        | 19                    |                       |                    |                       |
| Probably                         | 5                     |                       |                    |                       |
| Possibly                         | 24                    |                       |                    |                       |
Table 8 Comparison of the pharmacokinetic parameters after the oral administration of roflumilast

| Pharmacokinetic parameters | 0.25 mg | 0.375 mg | 0.50 mg |
|----------------------------|---------|----------|---------|
|                            | Test    | Ref 1(1) | Ref 2(2) | Test    | Ref 1(1) | Ref 2(2) |
| Roflumilast                |         |          |         |         |          |         |
| C<sub>max</sub> (ng ml<sup>-1</sup>) | 6.38 (1.13) | 2.92 (2.0, 4.25) | 5.01 (1.34) | 9.24 (1.95) | 5.21 (1.41) | 12.50 (2.91) |
| AUC<sub>C<sub>max</sub></sub> (h ng ml<sup>-1</sup>) | 33.80 (17.30) | 18.1 (11.1, 29.7) | 245 (9.1) | 48.70 (23.40) | 32.8 (10.1) | 65.10 (34.40) |
| T<sub>max</sub> (h) | 0.75 (0.50–1.50) | 1.00 (0.50, 2.00) | 0.50 (0.17) | 0.50 (0.25–2.00) | 1.0 (0.5) | 1.25 (0.50, 2.00) |
| t<sub>1/2</sub> (h) | 20.90 (7.62) | 13.56 (7.26, 25.31) | 21.3 (0.60) | 19.70 (6.38) | 20.8 (7.8) | 19.90 (7.69) |
| Roflumilast N-oxide        |         |          |         |         |          |         |
| C<sub>max</sub> (ng ml<sup>-1</sup>) | 6.74 (1.72) | 4.50 (3.50, 5.79) | 6.05 (1.48) | 10.4 (2.08) | 7.56 (1.15) | 14.70 (3.74) |
| AUC<sub>C<sub>max</sub></sub> (h ng ml<sup>-1</sup>) | 333 (103) | 178.6 (115.8, 275.2) | 301 (11.13) | 493 (14.8) | 397 (111) | 649 (189) |
| T<sub>max</sub> (h) | 4.00 (2.50–24.00) | 12 (2.00, 12.00) | 4.4 (3.4) | 4.00 (2.00–24.00) | 5.7 (5.9) | 4.00 (2.50–24.00) |
| t<sub>1/2</sub> (h) | 20.90 (7.62) | 22.13 (14.58, 33.58) | 26.5 (10.11) | 19.70 (6.38) | 27.7 (6.5) | 19.90 (7.69) |

Note: Superscript figures indicate comparing the pharmacokinetic parameters with the cited study.

Abbreviations: AUC, area under the curve; AUC<sub>inf</sub>, AUC until infinity; C<sub>max</sub>, maximum concentration; T<sub>max</sub>, the time to the C<sub>max</sub> of roflumilast; t<sub>1/2</sub>, half-life.
(46.7%), nausea (20%), and abdominal discomfort (20%). Thus, the results presented in the literature were consistent with those obtained in our paper.

It is worth mentioning that our study only included 12 volunteers, which a drawback. Second, reasonable markers should be used to describe dose–response, and the results obtained remain to be tested in patients with COPD and asthma, because healthy volunteers are not representative of a patient population. Finally, the influence of renal impairment on the pharmacokinetics of oral roflumilast in Chinese subjects should be studied.

Conclusion
Roflumilast was safe and well-tolerated in healthy Chinese volunteers and showed linear characteristics for \( C_{\text{max}} \) and AUC values at single doses ranging from 0.25 to 0.5 mg. No significant sex differences were observed in the pharmacokinetic parameters of roflumilast and roflumilast \( N \)-oxide.

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Disclosure
The authors report no conflicts of interest in this work.

References
1. Karish SB, Gagnon JM. The potential role of roflumilast: the new phosphodiesterase-4 inhibitor. Ann Pharmacother. 2006;40(6):1096–1104.
2. Taegtmeyer AB, Leuppi JD, Kullak-Ublick GA. Roflumilast – a phosphodiesterase-4 inhibitor licensed for add-on therapy in severe COPD. Swiss Med Wkly. 2012;142:w13628.
3. Pinner NA, Hamilton LA, Hughes A. Roflumilast: a phosphodiesterase-4 inhibitor for the treatment of severe chronic obstructive pulmonary disease. Clin Ther. 2012;34(1):56–66.
4. Spina D. Phosphodiesterase-4 inhibitors in the treatment of inflammatory lung disease. Drugs. 2003;63(23):2575–2594.
5. Lipworth BJ. Phosphodiesterase-4 inhibitors for asthma and chronic obstructive pulmonary disease. Lancet. 2005;365(9454):167–175.
6. FDA. DALIRESP (roflumilast) tablets (Initial U.S. Approval: 2011). Available from: http://www.accessdata.fda.gov/drugsatfda_docs/label/2015/022522s006lbl.pdf. Accessed March 31, 2015.
7. Susuki-Miyata S, Miyata M, Lee B-C, et al. Cross-talk between PKA-C\(\beta\) and p65 mediates synergistic induction of PDE4B by roflumilast and NTHI. Proc Nat Acad Sci. 2015;112(14):E1800–E1809.
8. Hermann R, Nassr N, Lahu G, et al. Steady-state pharmacokinetics of roflumilast and roflumilast N-oxide in patients with mild and moderate liver cirrhosis. Clin Pharmacokinet. 2007;46(5):403–416.
9. Hauns B, Hermann R, Hünnemeyer A, et al. Investigation of a potential food effect on the pharmacokinetics of roflumilast, an oral, once-daily phosphodiesterase 4 inhibitor, in healthy subjects. J Clin Pharmacol. 2006;46(10):1146–1153.
10. Li Q, Wang Y, Liu L, Ma P, Ding L. Pharmacokinetics of roflumilast and its active metabolite roflumilast N-Oxide in healthy Chinese subjects after single and multiple oral doses. Eur J Drug Metab Pharmacokinet. 2017;42(3):371–381.
11. Bethke TD, Böhmer GM, Hermann R, et al. Dose-proportional intraindividual single- and repeated-dose pharmacokinetics of roflumilast, an oral, once-daily phosphodiesterase 4 inhibitor. J Clin Pharmacol. 2007;47(1):26–36.
12. World Medical Association. World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. JAMA. 2013;310(20):2191–2194.
13. European Agency for the Evaluation of Medicinal Products (EMEA). International Conference on Harmonisation World Health Organization. Guideline for Good Clinical Practice [EMEA Web site]. ICH topic E6 (R1). Geneva, Switzerland: WHO; 2002. Available from: http://www. emea.europa.eu/pdfs/human/ich/013595en.pdf. Accessed September 26, 2007.
14. State Food and Drug Administration. Good Clinical Practice Guideline. Available from: http://www.sda.gov.cn/WS01/CL0053/24473.html. Accessed August 6, 2003.
15. Cui X, Huang J, Zheng X. Simultaneous determination of roflumilast and its metabolite in human plasma by LC–MS/MS: Application for a pharmacokinetic study. J Chromatogr B Analyt Technol Biomed Life Sci. 2016;1029–1030:60–67.
16. Hurst JR, Vestbo J, Anzueto A, et al; Evaluation of COPD Longitudinally to Identify Predictive Surrogate Endpoints (ECLIPSE) Investigators. Susceptibility to exacerbation in chronic obstructive pulmonary disease. N Engl J Med. 2010;363(12):1128–1138.
17. Bateman ED, Izquierdo JL, Harnett U, et al. Efficacy and safety of roflumilast in the treatment of asthma. Ann Allergy Asthma Immunol. 2006;96(5):679–686.
18. Mackay AJ, Patel ARC, Singh R, et al. Randomized double-blind controlled trial of roflumilast at acute exacerbations of chronic obstructive pulmonary disease. Am J Respir Crit Care Med. 2017;196(5):656–659.
19. Lahu G, Hünnemeyer A, Diletti E, et al. Population pharmacokinetic modelling of roflumilast and roflumilast \( N \)-oxide by total phosphodiesterase-4 inhibitory activity and development of a population pharmacodynamic-adverse event model. Clin Pharmacokinet. 2010;49(9):589–606.
20. Lahu G, Nass N, Hünnemeyer A. Pharmacokinetic evaluation of roflumilast (Daliresp) for COPD. Expert Opin Drug Metab Toxicol. 2011;7(12):1577–1591.
21. Roflumilast (Daliresp) for COPD. Med Lett Drugs Ther. 2011;53:59–60. Available from: https://secure.medicalletter.org/article-share?a=1369b&p=tml&title=Roflumilast%20(Daliresp)%20for%20COPD&canotaccessit=1. Accessed November 6, 2018.
22. Kim K, Johnson JA, Derendorf H. Differences in drug pharmacokinetics between East Asians and Caucasians and the role of genetic polymorphisms. J Clin Pharmacol. 2004;44(10):1083–1105.
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