American Journal of Clinical and Experimental Medicine
2015; 3(6): 368-371
Published online December 28, 2015 (http://www.sciencepublishinggroup.com/j/ajcem)
doi: 10.11648/j.ajcem.20150306.18
ISSN: 2330-8125 (Print); ISSN: 2330-8133 (Online)

Pharmacokinetics and Relative Bioavailability of Ambroxol Hydrochloride Aerosol and Injection

Jing Zhang[1], Na Yi[1], Lidao Bao[1, 2], Tengfei Yu[1, *]

[1]School of Pharmacy, Inner Mongolia Medical University, Hohhot, China
[2]Department of Pharmacy, Affiliated Hospital of Inner Mongolia Medical University, Hohhot, China

Email address:
349881704@qq.com (Tengfei Yu)

To cite this article:
Jing Zhang, Na Yi, Lidao Bao, Tengfei Yu. Pharmacokinetics and Relative Bioavailability of Ambroxol Hydrochloride Aerosol and Injection. *American Journal of Clinical and Experimental Medicine*. Vol. 3, No. 6, 2015, pp. 368-371. doi: 10.11648/j.ajcem.20150306.18

Abstract: To compare the pharmacokinetics and relative bioavailability of ambroxol hydrochloride (AH) aerosol and injection, the volunteers were subjected to single-dose crossover inhalation (injection) of 100 mg AH aerosol (injection). The drug concentrations in plasma were determined by HPLC. The areas under curve (AUC) of the two formulations were compared by the three-factor analysis of variance and the bi-directional one-side t test. The C_{max}, t_{max}, t1/2β and AUC values of the aerosol group and the injection group were (154.75±26.12) and (157.39±26.09) ng/ml, (1.12±0.34) and (1.29±0.33) h, (6.98±1.62) and (7.75±1.26) h, and (1593.02±290.45) and (1438±132.46) h·ng/ml, respectively. The bioavailabilities of AH aerosol and injection were the same, and the relative bioavailability of the aerosol was (96.52±11.44)%.

Keywords: Ambroxol Hydrochloride, Bioavailability, Pharmacokinetics, HPLC

1. Introduction

Ambroxol hydrochloride (AH), which is also known as bromo-cyclohexylamine alcohol hydrochloride, is a new generation of phlegm dissolver that facilitates bronchial cilia evacuation and contributes to the clearance of airway secretions. Therefore, AH has been mainly used to treat acute and chronic respiratory diseases, especially to eliminate the phlegm of chronic bronchitis patients [1, 2]. This study established a HPLC method to determine the AH concentrations in blood and investigated the pharmacokinetics of AH aerosol (made in China) and injection (imported) in healthy human body. The bioavailabilities of the two formulations were compared by the three-factor analysis of variance and the bi-directional one-side t test, which will provide evidence for clinical use [3].

2. Materials and Methods

2.1. Reagents

AH aerosol (trade name: Mucosolvan; approval number: H20030360) that was manufactured by Boehringer Ingelheim (Shanghai) pharmaceuticals Co., Ltd. was used for the control group, and AH injection (300mg/100ml, approval number: H19980178) that was produced by Jiangsu Hengrui Medicine Co., Ltd. was used for the trial group. AH reference substance (content: 99.6%) was also provided by Jiangsu Hengrui Medicine Co., Ltd. Methanol, acetonitrile and tetrahydrofuran were of HPLC grade. Distilled water was used throughout the experiment. Ethyl ether, potassium dihydrogen phosphate, disodium hydrogen phosphate, hydrochloric acid, sodium hydroxide, potassium chloride, and phosphoric acid were of AR grade. The prepared buffer was filtered through a 0.50 μm membrane. Preparation of 0.1mol/L pH 9.0 PBS buffer: 10.284 g potassium chloride was added to 8.528 g phosphoric acid, to which was added water till 1000 ml. Then 500 ml of the resulting solution was taken out, to which were added 476 ml of 0.1mol/L NaOH and water until 1000 ml. Preparation of 0.1 mol/L pH 7.0 PBS buffer: 0.48 g potassium dihydrogen phosphate and 0.97 g disodium hydrogen phosphate were dissolved in 1000 ml of water.

2.2. Determination of AH Concentrations in Blood

AH concentrations in blood were determined by HPLC.

2.2.1. Apparatus and HPLC Conditions

Hitachi 655 HPLC, including 655A-12 type pump, 655A-22 type variable-wavelength UV detector and 655B-71 type microprocessor; Rheodyne 87125 type injection valve

equipped with a 50 μl quantitative tube; Resolve C18 HPLC column (Dalian Institute of chemical physics, Chinese Academy of Sciences, 4.6mmx250mm, 5 μm); mobile phase: acetonitrile: methanol: 0.1mol/L pH 7.0 phosphate buffer: tetrahydrofuran (300:300:200:20), flow rate: 2.5ml/min, sensitivity: 0.01 AUFs, detection wavelength: 242 nm.

2.2.2. Blood Sample Treatment

1.0ml of plasma was transferred into a 10ml plugged glass centrifuge tube, to which were added 0.5ml of 0.1mol/L pH 9.0 PBS buffer and 5.0ml of ethyl ether. Then the sealed tube was volutedly mixed for 100 s and centrifuged at 3000 r/min for 6 min. Then the ether layer was transferred to another 10 ml clean plugged glass centrifuge tube, to which was added 250 μl of 0.1 mol/L HCl. The resulting product was volutedly mixed for 100 s and centrifuged at 3000 r/min for 6 min. Thereafter 35 μl of the acid layer was injected after discarding the ether layer.

2.2.3. Plasma Standard Curve and Detection Limit

Experimental plasma samples at 10, 20, 40, 80, 160 and 320 ng/ml were prepared by adding AH standard solution into 6 healthy human plasma samples. Then the concentrations were linearly regressed by the peak heights recorded after injecting the samples. The results show that the detection of AH concentrations in plasma had good linearity from 20 to 320 ng/ml. The regression equation was H=2.352+1.486c (r=0.998), the detection limit of AH in plasma was 4.5 ng/ml (S/N=2.8).

2.2.4. Recovery and Precision

AH plasma standard solutions at 20, 80, 160 and 320 ng/ml were precisely prepared utilizing healthy human plasma samples. Then the solutions were injected according to the injection methods of blood samples. Absolute recoveries (ratio of peak height to the concentration of standard solution) and relative recoveries (ratio of the calculated concentration to the real one) of the 4 four solutions were calculated as (83.98±2.15)%, (82.77±1.74)%, (83.16±2.27)% and (83.46±2.23)%, as well as (100.9±6.68)%, (103.4±5.75)%, (102.7±4.84)% and (99.7±5.29)%, respectively. AH plasma standard samples at 20, 80, 160 and 320 ng/ml were precisely prepared, and the detections were repeated for 5 times within one day, yielding the intraday precision. The detections were continuously performed (once per day) for 7 days, yielding the day to day precision. The intraday and day to day RSD values of the samples at the 4 concentrations were 5.39%, 3.46%, 2.67% and 2.17%, as well as 6.38%, 4.21%, 3.14% and 2.98%, respectively.

2.3. Selection of Volunteers and Sample Collection

10 healthy male volunteers aging 18-32 years old (average: 19.46±6.91) and weighing 63-75 kg (average: 67.17±6.39) were selected. All volunteers were confirmed normal by the routine examinations of liver, kidney and urine and electrocardiogram. They did not take any drugs 1 month before the trial, and they had signed consent forms. The trial had been approved by the Hospital Ethics Committee. The 10 volunteers were administered with 100 mg AH by inhalation and intravenous injection. Aerosol plus 5 ml of saline were inhaled by an air compression pump. The inhalation each time lasted for 2-3 min, and the injection was given 4-6 ml (10mg/1ml) per time, bid. The volunteers ate simultaneously 2.5 h after medication, the venous bloods (5.0ml) of which were collected before and 0.33, 0.67, 1.0, 1.5, 2, 2.53, 4, 6, 9, 12, 15 and 24 h after medication, respectively. Then the samples were centrifuged at 3000 r/min for 6 min after heparin anticoagulation, from which the plasma was separated. Thereafter the samples were stored at -20°C. Two trials were separated by 15 days.

2.4. Calculation of Pharmacokinetics Parameters and Bioavailability

The pharmacokinetics parameters were calculated automatically by the iteration fitting of program 3P87 utilizing the AH concentrations in blood determined at each time interval. AUC values were estimated by the ladder method using Mucosolvan as the reference to calculate the relative bioavailability of AH aerosol [4].

2.5. Statistical Analysis

The AUC values of the two administration methods were compared by the three-factor analysis of variance and the bi-directional one-side t test utilizing SPSS 15.0, aiming to evaluate their bioavailabilities [5].

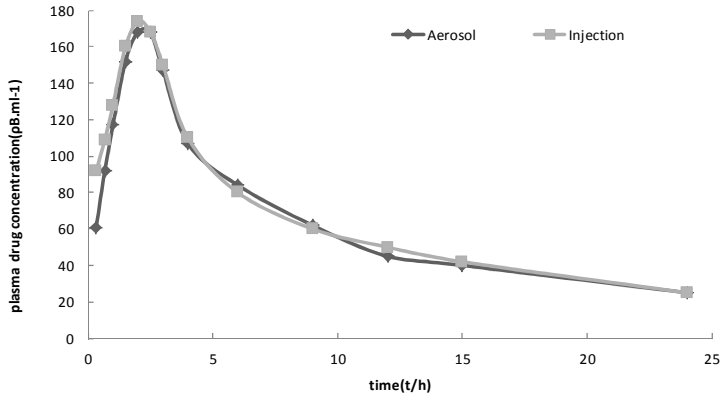

Figure 1. *Dependences of the average AH concentrations in blood on time.*

3. Results

3.1. AH Concentrations in Blood

The volunteers were subjected to single-dose crossover inhalation (injection) of 100 mg AH aerosol (injection). The dependences of the average AH concentrations in blood on time are shown in Figure 1.

3.2. Pharmacokinetics Parameters

The automatic iteration fitting reveals that the pharmacokinetics of AH in healthy human body followed the two-compartment open model [6]. The relative parameters are listed in Table 1, and the AUC values are summarized in Table 2.

Table 1. *Pharmacokinetics parameters (n=10, x±s).*

Sample	C_{max}(PB/ng.ml^{-1})	t_{max}(t/h)	t1/2ka(t/h)	t1/2α(t/h)	t1/2β(t/h)	CL(L/h)	AUC(A/h.ng.ml^{-1})
Aerosol	154. 75 ±26.12	1.12±0.34	0.53±0.19	1.46±1.10	6.98 ±1.62	215.67±101.7	1593.02±290.45
Injection	157.39 ±26.09	1.29±0.33	0.71±0.16	1.10±0.12	7.75±1.26	269.3±40.5	1438±132.46

Table 2. *AUC values.*

	Aerosol		Injection		F(%)
No.	P*	AUC1 (A/h·ng·ml^{-1})	P*	AUC2 (A/h.ng.ml^{-1})	AUC1/AUC2
A	2	1 317. 29	1	1 510. 21	87.23
B	2	1 402. 77	1	1 315. 12	106.67
C	1	1 272. 49	2	1 237. 65	102.82
D	1	928. 05	2	1 219. 34	76.11
E	2	1 564. 95	1	1 401. 62	111.65
F	1	1 432. 53	2	1 529. 61	93.65
G	2	1 641. 51	1	1 457. 46	112.62
H	1	1032.26	2	1108.21	93.15
I	1	1263.28	2	1191.37	106.03
J	2	879. 96	1	1 168. 59	75.3
x±s	96.523±11.44				

3.3. Statistical Analysis of Bioavailability

The three-factor analysis of variance shows that the AUC values between different formulations, periods and individuals did not differ significantly (P>0.07). The bi-directional one-side t test suggests that the bioavailabilities of AH aerosol and injection were identical [7, 8].

4. Discussion

This reserch to compare the pharmacokinetics and relative bioavailability of ambroxol hydrochloride (AH) aerosol and injection, the volunteers were subjected to single-dose crossover inhalation (injection) of 100 mg AH aerosol (injection). The drug concentrations in plasma were determined by HPLC. The areas under curve (AUC) of the two formulations were compared by the three-factor analysis of variance and the bi-directional one-side t test.

The experimental results of AH concentrations in blood indicate that AH was easily absorbable in both formulations. The pharmacokinetics parameters are consistent with those reported before. The t1/2β and c_{max} of the two formulations did not differ significantly, whereas the t_{max} and t1/2ka of aerosol were apparently shorter than those of injection, suggesting aerosol was more easily absorbed [9].

The three-factor analysis of variance and the bi-directional one-side t test of the AUC values of the two formulations did not differ significantly, indicating that both formulations could be similarly absorbed gastrointestinally and were of the identical bioavailability ((96.52±11.44)%) [10].

5. Conclusion

Evaluation of hydrochloric acid ammonia bromine by the effectiveness of the aerosol inhalation medication, to hydrochloric acid ammonia bromine line by atomizing inhalation medication provides the theoretical basis of acute or chronic respiratory diseases.

References

[1] Hayashi K, Hosoe H, Kaise T, Ohmori K (2000) Protective effect of erdosteine against hypochlorous acid-induced acute lung injury and lipopolysaccharide-induced neutrophilic lung inflammation in mice. J Pharm Pharmacol 52(11): 1411-1416.

[2] Wiessmann KJ, Niemeyer K (1978) [Clinical results in the treatment of chronic obstructive bronchitis with ambroxol in comparison with bromhexine (author's transl)]. Arzneimittelforschung 28(5a): 918-921.

[3] Lee HJ, Joung SK, Kim YG, Yoo JY, Han SB (2004) Bioequivalence assessment of ambroxol tablet after a single oral dose administration to healthy male volunteers. Pharmacol Res 49(1): 93-98.

[4] Ren YC, Wang L, He HB, Tang X (2009) Pulmonary selectivity and local pharmacokinetics of ambroxol hydrochloride dry powder inhalation in rat. J Pharm Sci 98(5): 1797-1803.

[5] Ballantyne BT, O'Hare SJ, Paschall JL, et al. (1993) Electromyographic activity of selected shoulder muscles in commonly used therapeutic exercises. Phys Ther 73(10): 668-677; discussion 677-682.

[6] Amantea M, Newman MS, Sullivan TM, Forrest A, Working PK (1999) Relationship of dose intensity to the induction of palmar-plantar erythrodysesthia by pegylated liposomal doxorubicin in dogs. Hum Exp Toxicol 18(1): 17-26.

[7] Rutland BE, Nachreiner RF, Kruger JM (2009) Optimal testing for thyroid hormone concentration after treatment with methimazole in healthy and hyperthyroid cats. J Vet Intern Med 23(5): 1025-1030.

[8] Yuen O, Caligiuri MP, Williams R, Dickson RA (1996) Tardive dyskinesia and positive and negative symptoms of schizophrenia. A study using instrumental measures. Br J Psychiatry 168(6): 702-708.

[9] Cerretani D, Roviello F, Pieraccini M, et al. (2002) Pharmacokinetics of intraarterial mitomycin C in hypoxic hepatic infusion with embolization in the treatment of liver metastases. Vascul Pharmacol 39(1-2): 1-6.

[10] Villacampa J, Alcántar F, Rodríguez JM, Morales JM, Herrera J, Rosete R (2003) Pharmacokinetic properties of single-dose loratadine and ambroxol alone and combined in tablet formulations in healthy men. Clin Ther 25(8): 2225-2232.