Supplementary materials

Supplementary Table 1 The details of adverse reactions in control, low-dose, medium-dose and high-dose groups.

	Adverse reactions
Control n=98	ALT increased2（2.04%）,APTT prolonged1（1.02%）,AST increased1（1.02%）,Dyspnea1（1.02%）,Ecze（1.02%）,PT prolonged1（1.02%）,Pulmonary infection1（1.02%）, γ-GPT increased1（1.02%）
Low-dose n=102	AST increased2（1.96%),ALT increased1（0.98%）,Cerebellar infarction1（0.98%）,CK-MB increased1（0.98%）,Decreased serum fibrinogen1（0.98%）,Decreased serum potassium1（0.98%）, Dizzy1（0.98%）,Erythra1（0.98%）,Headache1（0.98%）,lipid elevated1（0.98%）,Palpitation1（0.98%）,Rhinobyon1（0.98%）,Serum creatinine elevation1（0.98%）
Medium-dose n=101	AST increased4（3.96%）,ALT increased3（2.97%）,CK-MB increased3（2.97%）,Abdominal discomfort1（0.99%）,Acid indigestion1（0.99%）,Decreased appetite1（0.99%）,Dyspnea 1（0.99%）,Erythra1（0.99%）,Eosinophile granulocyte decreased1（0.99%）,Hemoglobin decreased1（0.99%）,Leukopenia decreased1（0.99%）,Pectoralgia1（0.99%）,Pruritus1（0.99%）, RBC decreased1（0.99%）,Serum creatinine elevation1（0.99%）,Thrombocytopenia decreased1（0.99%）
High-dose n=99	AST increased4（4.04%）,ALT increased3（3.03%）,Pruritus3（3.03%）,Dyspnea2（2.02%）,Erythema2（2.02%）,Acid indigestion1（1.01%）,Acute renal injury1（1.01%）,Allergic dermatitis1（1.01%）,Arthralgia1（1.01%）,Constipation1（1.01%）,Dermatitis1（1.01%）,Erythema perineum1（1.01%）,Erythra1（1.01%）,Hyperlipidemia1（1.01%）,Hypoglycemia1（1.01%）,Hypokalemia1（1.01%）,Leukocyte positive in urine1（1.01%）,Liver injur1（1.01%）,Palpitation1（1.01%）, Serum albumin1（1.01%）

Supplementary Table 2 Summary of pharmacokinetic studies of edaravone, (+)-borneol or compound edaravone in SD rats and Beagle dogs

<table>
<tr><th rowspan="2">Species</th><th colspan="2">Intravenous administration</th><th colspan="3">Comparative pharmacokinetics</th></tr>
<tr><th>Edaravone or (+)-borneol alone</th><th>Compound Edaravone (Edaravone: borneol=4:1)</th><th>PK Parameters</th><th>Distribution *</th><th>Metabolism and Excretion*</th></tr>
<tr><td rowspan="2">SD rats</td><td>edaravone
0.60, 1.20, 2.40, 7.20 mg/kg</td><td rowspan="2">0.75, 1.50, 3.00, 9.00 mg/kg</td><td>• Linear dose-dependent AUC of edaravone:
✓ $AUC_{0-\infty}$:827.7, 2383.5, 3878.7, 10759.9 ng/mL·h respectively for dosing of edaravone;
✓ $AUC_{0-\infty}$: 784.6, 2103.4, 3636.2, 11743.0 ng/mL·h respectively for dosing of compound edaravone.
• No significant differences of PK parameters</td><td>• Rapid distribution and elimination for each tissue
• No distinct accumulation
• No significant difference in vitro tissues
• No effect of (+)-Borneol on Edaravone</td><td>• Glucuronide conjunction and sulfate conjunction were major metabolites and excretion form
• mostly excreted in urine and bile
• approximately 80.3% and 82.5% respectively after 72 hours
• no significant difference in excretion ratios
• no apparent impact of (+)-borneol on the excretion of edaravone.</td></tr>
<tr><td>(+)-borneol
0.15, 0.30, 0.60, 1.80 mg/kg</td><td>• Linear dose-dependent AUC of (+)-borneol:
✓ $AUC_{0-\infty}$: 12.5, 31.1, 65.9, 194.0 ng/mL·h respectively for dosing of (+)-borneol;
✓ $AUC_{0-\infty}$: 21.7, 39.2, 73.3, 246.3 ng/mL·h respectively for dosing of compound edaravone.
• No significant differences of PK parameters</td><td>• Rapid distribution and elimination for most tissues
• No distinct accumulation
• No significant difference in vitro tissues
• No effect of edaravone on (+)-borneol</td><td>• glucuronide conjunction was major metabolite, camphor as trace
• mostly excreted in urine and bile
• approximately 20.4% and 21.2% respectively after 72 hours (glucuronide conjunction was not quantitatively analyzed)
• no significant difference in excretion ratios</td></tr>
<tr><td rowspan="2">Beagle dogs</td><td>edaravone
2.4, 4.8, 9.6 mg/kg</td><td rowspan="2">3.0, 6.0, 12 mg/kg</td><td colspan="3">• A dose dependent non-linear increase in AUC values:
✓ $AUC_{0-\infty}$: 30977.6, 51648.3, 65479.8 ng·h/ml for dosing of edaravone;
✓ $AUC_{0-\infty}$: 28341.4, 48593.2, 60077.6 ng·h/ml for dosing of compound edaravone
• No differences in pharmacokinetic parameters between single and repeated 7-day dose
• No effect of (+)-borneol on the PK profile of edaravone</td></tr>
<tr><td>(+)-borneol
0.6, 1.2, 2.4 mg/kg</td><td colspan="3">• A dose dependent linear increase in AUC values of (+)-borneol:
✓ $AUC_{0-\infty}$: 99.8, 197.9, 501.2 ng·h/ml for dosing of (+)-borneol;
✓ $AUC_{0-\infty}$:114.9, 207.3, 432.1 ng/mL for dosing of compound edaravone
• No differences in pharmacokinetic parameters between single and repeated 7-day dose
• No effect of edaravone on the PK profile of (+)-borneol
• A slight accumulation trend after repeated 7-day administration</td></tr>
<tr><td colspan="6">Other pharmacokinetic characteristics</td></tr>
</table>

Compounds	Plasma protein binding	CYP enzyme inhibition	CYP Enzyme Induction
edaravone/ compound edaravone	• Species differences: dog was lower than rat and human • Saturation phenomenon at 50000 ng/mL • No impact of (+)-borneol on edaravone.	• CYP1A2, CYP2C9, CYP2C19, CYP2D6 and CYP3A4 IC50 ＞ 100 μmol/L for edaravone, (+)-borneol and compound edaravone	• 0.15, 0.45, 1.50 mg/kg of (+)-borneol, 0.60, 1.80, 6.00 mg/kg of edaravone, 0.75, 2.25 7.50 mg/kg of compound edaravone (4:1) • Tested in rats after 7-day of intravenous administrations • No apparent induction of CYP1A2, CYP2C9, CYP2C19 and CYP3A4
(+)-borneol/ compound edaravone	• No apparent species differences (60.8-77.1%) • No saturation phenomenon at 5000 ng/mL • No impact of edaravone on (+)-borneol		

***Note:** 2.4 mg/kg Edaravone, 0.60 mg/kg (+)-borneol and 3.0 mg/kg compound edaravone (edaravone: (+)-borneol=4:1) were used for distribution and excretion study.

Abbreviation：PK, Pharmacokinetic; SD, Sprague Dawley； AUC, area under the curve; CYP, cytochrome P450

Supplementary Table 3 Summary of preclinical toxicology Studies of compound edaravone

Type of study	Species/Strain	Route	Dose mg/kg	n/Sex/Group	GLP	Observed Maximum Nonlethal Dose (mg/kg)	NOAEL mg/kg	Noteworthy findings
Acute toxicity Study (combo)	SD rats	i.v.	25, 50, 100	10	GLP	>100	25	• Red urine in rats of 50mg/kg group
Dose range finding study(combo)	Beagle dogs	i.v.	50, 100, 200	2	GLP	>200	50	• Red urine and tubular degeneration in kidney in rats of 100 or 200mg/kg group
Repeat-dose toxicity study	SD rats	i.v.	10, 20, 40	15 11(TK)	GLP	20	20	• Red urine, correlated decrease in RBC and HGB, and increase in Retic in 6.4% Propylene Glycol or 40 mg/kg group
	Beagle dogs	i.v.	25, 50, 100	6 4(TK)	GLP	50	50	• Adverse effects in the kidneys and injection site in 100 mg/kg group
Reproduction Toxicity Segment Ⅰ	SD rat	i.v.	6.25, 12.5, 25	24/sex	expl			• No apparent toxicity in fertility or early embryonic development • Red urine, a slight decrease in body weights and food consumption
Reproduction Toxicity Segment Ⅱ	SD rat	i.v.	6.25, 12.5, 25	25(or 26)	expl			• No embryo-fetal developmental toxicity • Red urine
Reproduction Toxicity Segment Ⅱ	New Zealand rabbit	i.v.	2.5, 5, 10	15	expl			• No apparent parental or embryo-fetal developmental toxicity
Active systemic anaphylaxis	Guinea pig	i.v.	2, 5	3	expl			• No anaphylactic reaction
Passive cutaneous anaphylaxis	SD rat	i.v.	2.5, 12.5	3	expl			• No passive cutaneous anaphylaxis
Vascular irritation	New Zealand rabbit	i.v.	3.75mg(10ml)/kg	2	expl			• No apparent local irritation
Intramuscular irritation	New Zealand rabbit	i.m.	1 ml (0.375 mg)/site	2	expl			• No apparent or severe muscular irritation

Hemolysis	Rabbit red blood cells	*In vitro*	0.1, 0.2, 0.3, 0.4, 0.5ml	-	expl			• No hemolysis or agglutination
CV and Respiratory System	Beagle dog	i.v.	25, 50, 100	3	expl			• A transient and slight increase in blood pressure. No effect on the function of respiratory system or body temperature
CNS	ICR mice	i.v.	8, 25, 75	5	expl			• A decrease in spontaneous activities and a positive and negative synergistic effect with subthreshold hypnotic dose of pentobarbital

Abbreviation：SD, Sprague Dawley；NOAEL, no observed adverse effect lever；TK, toxicokinetics；CV, cardiovascular；CNS, central nervous system; GLP, good laboratory practice；expl, exploration, i.v., intravenous; i.m., intramuscular.

Supplementary Table 4 Summary of genetic toxicology studies of compound edaravone

Studies	GLP	Route	Dose	-S9	+S9
Reverse mutation study in *Salmonella Typhimurium*	expl	-	250, 25, 2.5, 0.25, 0.025 μg/plate	N	N
In vitro chromosome aberration in cultured Chinese Hamster Lung Cells	expl	-	200, 100, 50, 25μg/ml	N	N
In vivo mouse bone marrow micronucleus test	expl	i.v.	31.25, 62.5, 125mg/kg	N	N/A

Abbreviation: GLP, good laboratory practice；expl, exploration；N, negative, N/A, not applicable.