Original article

Toxicity of Five Herbs in *Aconitum* L. on *Tetrahymena thermophila* Based on Spectrum–effect Relationship

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**ABSTRACT**

**Objective** To explore the active components with toxic effects in five *Aconitum* L. herbal medicines on *Tetrahymena thermophila*. **Methods** The fingerprints of five *Aconitum* L. herbal medicines were established by ultra-high performance liquid chromatography (UPLC) and the toxicity was evaluated by using a TAM Air Isothermal Calorimeter on *Tetrahymena thermophila* SB110. **Results** By analyzing the spectrum–effect relationships between UPLC fingerprints and toxic effects, the active components which had the toxic effects were obtained. **Conclusion** This work provides a general model of the combination of UPLC and microcalorimetry to study the spectrum–effect relationships of the five *Aconitum* L. herbal medicines, which could be used to evaluate the toxic effects and analyze the principal toxic components of the five *Aconitum* L. herbal medicines. On the whole, this result provides the experimental basis for the safe use of the five *Aconitum* L. herbal medicines in clinic.

**Key words** *Aconitum* L. herbal medicines; microcalorimetry; spectrum–effect relationships; *Tetrahymena thermophila*; ultra-high performance liquid chromatography

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1. **Introduction**

*Aconitum* L. is a genus of flowering plants belonging to Ranunculaceae family which is an important class of medicinal plants of Chinese materia medica (CMM) (Fu, 2000). As *Aconitum* L. herbal medicines, *Aconiti Radix* (AR), *Aconiti Kusnezoffii Radix* (AKR), *Aconiti Singularis Radix* (ASR), *Aconiti Lateralis Radix Preparata* (ALRP), and *Aconiti Brachypodi Radix* (ABR) are commonly used in the treatment of joint pain, rheumatoid arthritis, and other inflammations in clinic (Wu, 2011; Zhou et al, 2003). Accompanied with good efficacy of the five *Aconitum* L. herbal medicines, the toxicity, such as arrhythmia, respiratory paralysis, vagus nerve exciting, etc, simultaneously occurred occasionally (Fu, 2004). Therefore, the five *Aconitum* L. herbal medicines have already been recorded as high risk...
medicines (Ono et al, 2009; Voss et al, 2008; Ye et al, 2011) which lead to great challenges for the safe use of them in practice. Furthermore, efficiently assessing the toxicity of the five Aconitum L. herbal medicines has been a big problem and has been paid more attention to.

Tetrahymena thermophila is a freshwater ciliate, and its growth, reproduction, behavior, and metabolic synthesis reaction could be easily influenced by external substances, showing the sensitive characteristics to the toxic substance (Li, 2011; Sauvant et al, 1999). As a useful model organism, T. thermophile has been chosen as a standard living for toxicity detection and was used as a pharmacological tool in different bioassay techniques to detect toxicants (Monika et al, 2010; Chen et al, 2004; Fu et al, 2005; Li et al, 2012).

In this paper, UPLC was used to establish the fingerprints of the five Aconitum L. herbal medicines and analyze the active components. As an important analytical method, UPLC had been employed in the qualitative and quantitative analysis of chemical constituents in herbal medicines by examining the contents, analyzing the constituents of herbs, and getting the fingerprints of the drug, food, and other materials (Xiao et al, 2006; Proestos et al, 2008; Schaneberg et al, 2003; Lau et al, 2004). However, although UPLC has been proved to be a mature method for the detection of chemical constituents in drugs, the chemical components alone could not represent the toxicity or efficacy of drugs. So, it is necessary to combine the UPLC method and the biological techniques to determine the toxicity of the five Aconitum L. herbal medicines.

Due to the good sensitivity, reproducibility, and accuracy, microcalorimetry was widely used in assessing the toxicity of many substances, such as drugs, metal, soil, and foodstuffs in multiple living matrices (Buttiglieri et al, 2010; Kong et al, 2010; Zhou et al, 2009; Zhuang et al, 2011). The technique of microcalorimetry used for toxicity study was based on the measurement of microbial heat output (Kong et al, 2009). The toxicity and grading of toxicity of drugs could be assessed according to the heat production (Chen et al, 2010; Kong et al, 2012). In this paper, microcalorimetry was applied to evaluating the toxic effects of the five Aconitum L. herbal medicines on T. thermophila (SB110). By analyzing the spectrum-effect relationships between UPLC fingerprints and toxic effects, the active components with the toxic effects could be obtained.

Both UPLC and microcalorimetry have been widely used as important and useful methods. However, the combination of them to assess the toxic effects of the five Aconitum L. herbal medicines on T. thermophile and to analyze the toxic components of them has not been reported. So, in this study, microcalorimetry was selected to evaluate the toxic effects of the five Aconitum L. herbal medicines on T. thermophile, and UPLC was applied to searching for and analyzing the active components which had the toxic effects on T. thermophile. On the whole, the aim of this work is to evaluate the toxicity of the five Aconitum L. herbal medicines on T. thermophile and to analyze the toxic components in them so as to provide theoretical basis for their safety assessment and accurate use in practice.

2. Materials and methods

2.1 Materials

Aconiti Radix (batch No. 120322), Aconiti Singularis Radix (batch No. 120322), Aconiti Lateralis Radix Preparata (Aconitum carmichaeli Debx., batch No. 120322), Aconiti Kusnezoffii Radix (A. kusnezoffii Rehb., batch No. 110203), and Aconiti Brachypodi Radix (A. brachypodum Diels, batch No. 110910) were all purchased from Anguo Medicine Market (Baoding, China) and the crude materials were authenticated by Prof. Xiao-he Xiao (China Military Institute of Chinese Materia Medica, 302 Military Hospital of China). AR (100 g) was macerated in 1000 mL of water for 30 min and decocted with water twice, for 1.5 h each time. The filtrates from each decoction were blended and concentrated to a thick solution using a rotary evaporator. The concentrated sample was dried in vacuum oven and dried powders were got. AKR, AR, ALRF, and ABR were decocted and dried with the same procedure. Standard substances of aconitine (AC, 110720-200410), hyaconitine (HA, 110799-200505), and mesaconitine (MA, 110798-200805) were obtained from National Institute for Food and Drug Control. The purity of all the compounds was more than 98%. All other chemicals used were of analytical grade and locally available.

Ammonium acetate was from Fluka (Buchs, Switzerland). Acetonitrile and methanol of UPLC grade were from Fisher Chemicals (Pittsburg, USA). Water was purified using Milli-Q Water Purification System (Milipore, USA).

Tetrahymena thermophila (SB110) was provided by Institute of Hydrobiology, Chinese Academy of Sciences (Wuhan, China). T. thermophila (SB110) was grown axenically in modified SSP medium containing 2% proteose peptone, 0.1% yeast extract, and 0.2% glucose, supplemented with 0.003% ferric citrate. And the double distilled water was used to prepare the medium in the experiment and the medium solution was sterilized in high-pressure steam at 121 °C for 30 min.

2.2 Instruments

UPLC fingerprints of the five Aconitum L. herbal medicines were performed using Waters Acquity UPLC TM System (USA), consisting of binary solvent delivery pump, auto sampler manager, column compartment, and photo diode array detector, connected to Waters Empower 2 software. T. thermophila SB110 cells were cultivated in constant temperature incubator at 28 °C. A Thermal Activity Monitor Air Isothermal Microcalorimeter (Stockholm, Sweden) was used to measure the metabolic power-time curves of T. thermophila SB110 growth. The microcalorimeter was thermostated at 28 °C and the power-time curves were recorded by a computer. Wadsö (2002) has reported more details of the performance and structure of the instrument.

2.3 Microcalorimetric measurements

Initially, T. thermophila SB110 was cultivated in the
incubator at 28 °C, and then inoculated in the prepared 5 mL culture medium in 10 mL glass ampoule. The initial population density was $4.5 \times 10^3$ cells/mL. Each kind of Aconitum L. herbal medicines at different concentration was added into each glass ampoule. Then, the glass ampoules were sealed with a cap and put into microcalorimeter. The growth process was monitored continuously and its thermogenic curve was obtained. The experiments above were all carried out with aseptic technique.

2.4 UPLC analysis

2.4.1 Preparation of reference solution

The mixed reference solutions containing each 5 μg/mL of AC, HA, and MA were prepared by adding an accurately weighed amount of each standard stock into a volumetric flask and dissolved with 10 mL MeOH, and then filtered through 0.22 μm membrane to yield the mixed reference solutions.

2.4.2 Preparation of sample solution

The powder (10.0 g) of each kind of Aconitum L. herbal medicines was extracted with 30 mL methanol by ultrasonic extraction for 50 min. After filtering, the residue was dissolved with 3 mL methanol. The dissolved solution was prepared by the method of weight relieving, by which the weight loss in the dissolving procedure was compensated, and then filtered through 0.22 μm film to yield the sample solution at the concentration of 0.11 g/mL for UPLC to get the UPLC fingerprints.

2.4.3 UPLC conditions

The chromatographic separation was performed using Waters Acquity BEH C18 column (50 mm × 2.1 mm, 1.7 μm) and operated at 30 °C. The mobile phase was composed of solvent A (acetonitrile-water solution of 40 mmol/L ammonium acetate 65:35, pH 10 adjusted with aqua ammoniae) and solvent B (water solution of 40 mmol/L ammonium acetate, the pH 10 adjusted with aqua ammoniae) as the mobile phase with a linear gradient as follows: 0–1 min, 1.5%–4.6% A; 1–3.5 min, 4.6%–35% A; 3.5–7.5 min, 35%–50% A; 7.5–11 min, 50%–60% A; 11–16 min, 60%–68% A; 16–20 min, 68%–100% A, with a mobile flow rate of 0.2 mL/min. The detection wavelength was set at 235 nm with the sample injection volume of 5.0 μL.

3. Results

3.1 Microcalorimetric measurement

3.1.1 Power-time curves of T. thermophila SB110 growth

The power-time curve of T. thermophila SB110 growth in the absence of drug is shown in Figure 1. It was a typical power-time curve of T. thermophila SB110 and could be divided into lag phase, exponential phase, stationary phase, and decline phase. Then, the corresponding power-time curves of growth in the presence of the five Aconitum L. herbal medicines with different concentration are shown in Figure 2. It could be seen that these curves produced some changes when the five Aconitum L. herbal medicines were added, illustrating that the T. thermophila SB110 growth was influenced. By analyzing the power-time curves, the quantitative parameters, such as maximum heat-production rate ($P_{\text{max}}$), appearance time ($t$), and total heat-production ($Q$) are obtained and shown in Table 1.

3.1.2 Effect of five Aconitum L. herbal medicines on T. thermophila SB110 growth

The power-time curves of T. thermophila SB110 growth affected by Aconitum L. herbal medicines with different concentration are shown in Figure 2. As seen from the profiles of the curves, the growth of T. thermophila SB110 was significantly influenced by Aconitum L. herbal medicines. The highest peak of T. thermophila SB110 growth was all depressed gradually with the concentration of the Aconitum L. herbal medicines increasing compared with the control, illustrating that the metabolism of T. thermophila SB110 growth was inhibited. The $Q$ value of T. thermophila SB110 growth degraded gradually with the concentration of the Aconitum L. herbal medicines increasing.

The relationship between $Q$ and $C$ is shown in Figure 3, illustrating that the Aconitum L. herbal medicines with different concentration had different toxic effects on T. thermophila SB110 growth. The growth inhibitory ratio ($I$) could be calculated on the basis of $Q$ (Wang et al, 2010). Inhibitory ratio is another necessary and important parameter in assessing the toxicity of many toxic substances, and it could be defined as follows:

$$I/\% = (Q_0 - Q) / Q_0 \times 100\%$$

Where $Q_0$ and $Q$ represent the total heat-production in the whole metabolic progress of T. thermophila SB110 growth without and with different concentration of the five Aconitum L. herbal medicines, respectively.

Based on the values of $Q_0$ and $Q$ in Table 1, the $I$ values of the five Aconitum L. herbal medicines on the T. thermophila SB110 growth were calculated and the relationships between $I$ and $C$ of the five Aconitum L. herbal medicines were established and shown in Figure 4.
Figure 2  Power-time curves of *T. thermophila* SB110 growth affected by different concentration of AKR (A), ASR (B), ABR (C), AR (D), and ALRP (E)

Table 1  Thermokinetic parameters of *T. thermophila* SB110 growth affected by AKR, ASR, ABR, AR, and ALRP with different concentration

<table>
<thead>
<tr>
<th>Samples</th>
<th>$C_i$ ($\mu$g·mL$^{-1}$)</th>
<th>$t$ / min</th>
<th>$P_i$ / $\mu$W</th>
<th>$Q_i$ ($\times 10^{-4}$ J)</th>
<th>$I_i$ / %</th>
<th>$IC_{50}$ / (μg·mL$^{-1}$)</th>
</tr>
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<tbody>
<tr>
<td>AKR</td>
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<td>442.30</td>
<td>119.27</td>
<td>5.47</td>
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<td>208</td>
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<td></td>
<td>200</td>
<td>447.38</td>
<td>78.43</td>
<td>2.77</td>
<td>49.36</td>
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<tr>
<td></td>
<td>400</td>
<td>472.51</td>
<td>74.38</td>
<td>2.36</td>
<td>56.86</td>
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<tr>
<td></td>
<td>800</td>
<td>499.54</td>
<td>59.27</td>
<td>1.11</td>
<td>79.71</td>
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</tr>
<tr>
<td></td>
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<td>635.16</td>
<td>54.77</td>
<td>0.79</td>
<td>85.56</td>
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<tr>
<td>ASR</td>
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<td>561.54</td>
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<td>5.43</td>
<td>0</td>
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<td>1.34</td>
<td>77.21</td>
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<td>ALRP</td>
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<td>94.45</td>
<td>5.67</td>
<td>0</td>
<td>1166</td>
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<td>733.47</td>
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<td>3.89</td>
<td>31.39</td>
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<tr>
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<td>854.40</td>
<td>77.51</td>
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<td>72.39</td>
<td>2.32</td>
<td>59.08</td>
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</tbody>
</table>
Figure 3  $Q - C$ (A) and $P_m - C$ (B) relationships between thermokinetic parameters and concentration of AKR, ABR, AR, ASR, and ALRP

Figure 4  Relationships between $I$ and $C$ of AKR, ABR, AR, ASR, and ALRP

When $I$ is equal to 50%, the corresponding half-inhibitory concentration of the inhibitor could be represented as IC$_{50}$. This could be regarded as the inhibitory concentration causing a 50% decrease of the growth rate constant. From the relationship between $I$ and $C$ in Figure 4, IC$_{50}$ values of the five *Aconitum* L. herbal medicines on the *T. thermophila* SB110 growth were obtained and shown in Table 1.

3.2 UPLC experiment

**UPLC fingerprints and similarities among five Aconitum L. herbal medicines**

The results of methodology validation showed that the relative standard deviation (RSD) for precision was in the range of 2.01%–2.19%, for the reproducibility was less than 1.10% and the storage stability was 2.03%–2.42%. All the results indicated that the method of UPLC for the fingerprint analysis was valid and satisfactory. So, the UPLC fingerprints of the five *Aconitum* L. herbal medicines were obtained (Figure 5). The reference fingerprint is generated in Figure 5.

Peaks, which existed in all of the 15 chromatograms of the five *Aconitum* L. herbal medicine samples, with large areas and good segregation from consecutive peaks, were regarded as “common peaks”, which indicated the similarity among various samples. Sixteen common peaks were found from the samples by comparison of their ultraviolet spectra and UPLC retention time. The comparison process will not be illustrated in detail, while the detailed description was similar to the reference (Chen et al, 2007; Chen et al, 2008). By the comparison between the chromatogram of sample and reference solution, we could find peaks 14, 13, and 15 were
AC, MA, and HA, respectively, and peaks 1–12 were unknown compounds.

Peak 13 was MA at the retention time of 12.562 min. By the comparison between the chromatogram of sample and reference solution, peak 13 was selected as the reference peak to calculate the relative retention time of AC and HA, and the coefficient of variance (CV) could be calculated as follows:

$$CV = \frac{\sigma}{\mu}$$

where $\sigma$ was the standard deviation, $\mu$ was the average value of the peak area.

The CV values of the peak area showed that the content of the three constituents varied significantly in different samples, which were 97.3%, 119.3%, and 93.8%, respectively.

### 3.3 Analysis of spectrum-effect relationship

The results of microcalorimetric measurement showed that the five *Aconitum* L. herbal medicines had toxic effect on *T. thermophila* SB110, and the sequence of the toxic effect of the tested five *Aconitum* L. herbal medicines were obtained: AKR > ABR > AR > ASR > ALRP. Depending on the relevant assay for the results of UPLC and microcalorimetry of the five *Aconitum* L. herbal medicines, we could obtain the content of AC, MA, and HA in the five *Aconitum* L. herbal medicines associated with the toxicity of them. The AC had the greatest toxic impact on *T. thermophila* SB110, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, while the highest content of HA in ALRP had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact. As a result, the highest content of AC in AKR had the strongest toxicity on *T. thermophila*, and HA had the minimal toxic impact.

### 4. Discussion

Due to the high risk, the toxicity of *Aconitum* L. species should be assessed comprehensively and objectively by sensitive and reliable methods. In the study, UPLC method provided the chemical profile of active components of the five *Aconitum* L. species, the microcalorimetric method was applied to assessing and comparing the toxic effects of the five *Aconitum* L. herbal medicines and the toxicity sequence of them was obtained as AKR > ABR > AR > ASR > ALRP. UPLC and microcalorimetry were combined for the first time to investigate the toxic effects of the five *Aconitum* L. herbal medicines and analyze the toxic components.

This study showed that the different toxic effects of the five *Aconitum* L. herbal medicines were related with the main chemical components, such as MA, AC, and HA. The investigation of UPLC fingerprints and toxic effects relationships of the five *Aconitum* L. herbal medicines based on UPLC and microcalorimetry could provide a tool to evaluate the toxicity differences of the five *Aconitum* L. herbal medicines, and an experimental foundation and model for the study on the toxicity of the herbal medicines. The results showed that the effectiveness on a combination of microcalorimetry and UPLC for assessing the toxic effects of the similar herbal medicines and gave some indications for the safety assessment and accurate use of the five *Aconitum* L. herbal medicines in practice.

### References


chemical fingerprinting: Application to Ephedra. Phytochemistry 62: 911-918.

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*Chinese Herbal Medicines* (CHM) is an official international journal sponsored by Tianjin Institute of Pharmaceutical Research and Institute of Medicinal Plant Development, Chinese Academy of Medical Sciences.

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