

A review on toxicity effects on *Aconitum carmichaelii* Debx (Chuan wu and Fuzi) and TCM processing approach 'Pao zhi' in reducing/ eliminating toxicity

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ABSTRACT

Aconitum carmichaelii Debx. (Chuan wu & Fuzi) is one of the most widely used Chinese traditional medicinal herbs and is listed in the Chinese Pharmacopoeia. It has been used for the treatment of various conditions rheumatism being among them. However, the high toxicity risks and narrow therapeutic ranges limited the medicinal application on a larger scope, due to the poisonous aconitine alkaloids present in it. Therefore, this review article mainly focuses on the principle components causing toxicity effects, and the main Traditional Chinese medicine (TCM) processing method (Pao zhi) & its mechanisms in reducing or eliminating the toxicity associated with rootstocks of *Aconitum carmichaelii* Debx.

Keywords: *Aconitum carmichaelii* Debx; toxicity; toxicity effect; aconitine alkaloids; Pao zhi.

1. INTRODUCTION

Plants which are commonly used in traditional medicine are frequently promoted as natural and, therefore, harmless. This assessment is based on their usage in the treatment of diseases over centuries [1, 2]. However, there had been occasional few reports on adverse reactions associated with herbal consumption [3, 4] such as *Aconitum carmichaelii*, commonly known as Chinese aconite [5]. Same case as Western pharmaceuticals, some herbs are toxic and must be used with caution. Generally, herbal intoxication is more difficult to manage, due to inadequate knowledge about the safety and efficacy of the Chinese medicine. For example in the case of Taiwan, it was reported that 47% of the potentially toxic effects of Chinese medicine were either unknown or could not be found in the literature. [6] Hence, toxicological problems associated with the use of herbal medicines may not be readily recognized. [7] So, despite the growing market demand for herbal medicines, there are still concerns associated with not only their use, but their safety. Less than 10% of herbal products in the world market are truly

standardized to known active components and strict quality control measures are not always diligently adhered to [8]. Consequently, in response to public health concerns, research that focuses on deficiencies in the knowledge about medicinal plants and their potential toxicities is highly encouraged by many official medical and scientific organizations [9–12] and by complementary and alternative medicine (CAM) researchers and practitioners [13, 14]. Toxicity is defined as “the potential of a substance to exert a harmful effect on humans or animals, and a description of the effect and the conditions or concentration under which the effect takes place” [15].

Toxic effects are defined as “harmful responses of a biological system to a toxic compound, and death of cells or the whole organism are the major response.” [16] In all the cases, the toxic effects are usually manifested either in an acute or a chronic manner, and occur mostly as a result of an acute or chronic exposure to toxic compound by oral ingestion, inhalation or absorption following skin contact [16]. The toxic effects

are seen as (a) signs or reflection of a disturbance of the normal activities of enzymes that perform essential biochemical roles in all forms of life; (b) alteration of the normal activities of plasma membrane that regulate the exchange of nutrients and metabolites between the cell and its surroundings and (c) the disturbances of other normal cell activities, e.g. RNA and DNA synthesis, growth, division and general metabolism at all levels of organization from sub-cellular to organ and organ system [16, 17].

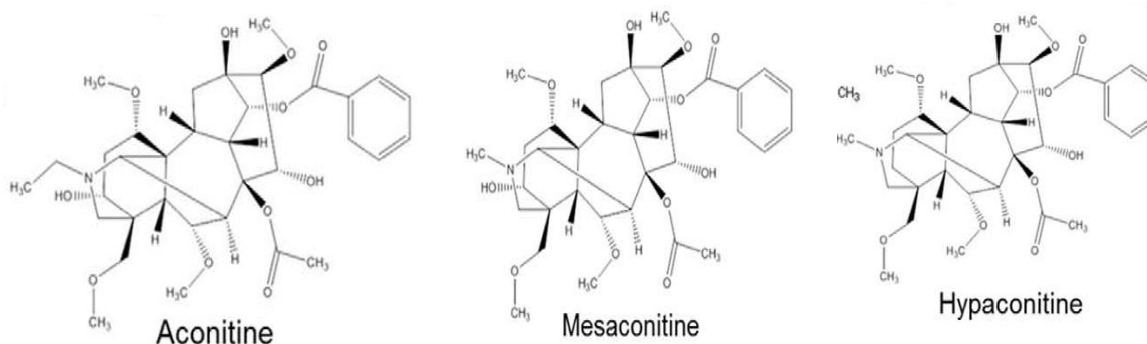
Aconitum carmichaelii is a species of flowering plant of the genus *Aconitum*, of which the genus is over 250 species of flowering plants belonging to the family *Ranunculaceae*. Most species are extremely poisonous [18] and must be dealt with carefully. It is native to East Asia and eastern Russia. It is commonly known as Chinese aconite, Carmichael's monkshood or Chinese wolfsbane. It is known in Mandarin as *Wu Tou* (meaning tuberous mother root, or root tuber). [5] In Chinese Pharmacopoeia (CP) 2010, *Aconitum carmichaelii* Debx is recorded, extensively distributed in Sichuan Province of China. The mother root of *Aconitum carmichaelii* Debx is named "Chuanwu" (CW), while the daughter or lateral root is "Fuzi" (FZ). [19, 20] The plant is excavated in late June to early August, removed above ground stem leaves, and leave rhizome after picked daughter roots and removed fibrous roots and soil, then sun dried [21]. The medicinal property of the herb is considered pungent and bitter in flavor, warm in nature, extremely toxic and attributive to the

heart, spleen, liver and kidney meridians [22]. Although CW has a limited therapeutic range, it is commonly used to treat various diseases such as collapse, syncope, rheumatic fever, painful joints, gastroenteritis, diarrhea, edema, bronchial asthma, various tumors, and some endocrinal disorders [23, 24]. However, many cases of accidental and intentional intoxication with this plant have been reported; some of these have been fatal. [19, 20] It was thus important to develop a more efficient method to lowering the toxicity of Aconite root. [25] The toxicity effects of this herb was mainly attributed due to aconitine alkaloids e.g. aconitine, mesaconitine, hypoconitine etc, that are presents in *Aconitium spp.* [26]. Therefore, this paper reviews the toxicity studies associated with *Aconitium carmichaelii* Debx plant, Chinese herbal treatment in aconite toxicity, and a concrete processing method used in TCM to prevent (i.e. reduce/eliminate) the toxicity, for safe clinical use.

2. Chinese Aconite toxicity

2.1 Main components causing toxicity in aconite rootstocks

Aconite rootstocks of *Aconitum carmichaelii* (CW & FZ) is commonly used as a key ingredient in many herbal prescriptions in the orient for rheumatism, heart failure, contracture of limbs and pain joints. Whereby, the principle active ingredients are alkaloids with C19-diterpenoid skeleton, including aconitine, mesaconitine, and hypoconitine [26, 27].



These alkaloids in rootstocks of *Aconitum carmichaelii* are toxic and have very narrow safety range because; they could easily induce ventricular tachycardia and fibrillation even at therapeutic dose level [27,28] This so due to the content of aconitine alkaloids which varies according to their part of the plant, growth region, season and species [29-36], and the content in roots is higher than that of stems and leaves; the level of aconitines in roots was considered about ten times that in stems [33] and about three hundred times that in leaves. [32] This is supported in a research case study on aconite poisoning on inpatients ingested aconite roots compared with those of leaves ingestion; thus, it was noted that the Area under curve (AUC) i.e. a pharmacokinetic parameter, in the patient whom ingested leaves was low compared to that of roots thus,

this indicated that the level of toxic aconitine alkaloids content are lower in leaves than that of roots. [50] Still in the same case study research done, there was a patient had AUC lower than other four patients admitted on aconite poisoning; yet, the patient ingested the root part of the plant like others. But it was observed that the main alkaloid was different from the other patient in which hypoconitine was detected, while jesaconitine was not detected yet the patient ingested the root part of aconite. [50].

Therefore, the level of aconitine, hypoconitine, jesaconitine and mesaconitine in aconite vary according to the part of the plant, the growing region, the aconite species and the season [29-36]. The LD₅₀ value of aconitine in mice was reported to be 1.8mg/kg

when given orally [37], and the lethal dose of aconitine for humans was estimated to be 1-2mg. [38, 39].

2.2 Pharmacological study on aconitines in aconite rootstocks

Many pharmacological studies of aconitines have been done [40-46], and it was noted that, the toxicity of these molecules was known to be due to their action on Na⁺ channels in excitable membranes [41-44]. Aconitines were also seen to be distributed in high concentration to the liver and kidney [47]. This is so, because it has been reported that aconitines can be detected in urine until 7 days after ingestion, but that they are not detectable in serum 24 hours after ingestion [48]. Thus, here kidney have shown to be involved pathway, hence more supportive information is required to explore the plant in renal function involvement. Similarly, in animal experiments done, it had been reported that the toxic symptoms in aconite poisoning are dependent on the dose of aconitines [49]. This was supported in a case study done (in Iwate medical university, Japan) was found that, the extent of toxic symptoms of aconites was not only affected by amount of exposure to aconites but also by time of exposure [50]. Where by patient whom had hemodynamic instability such as low cardiac output was seen to affect the half-life i.e. increase in half life. In this case study it was noted that, there was an increase in toxicokinetic parameters such as Area under curve (AUC) and Mean residence time (MRT). Since AUC reflects the amount of exposure while MRT reflects to the time of exposure, it was then depicted that, increase of these two parameters lead to larger concentration of aconites in the body, thus acted in sodium channel for long time hence, lead to ventricular tachycardia and ventricular fibrillation [50].

2.3 Clinical symptoms of aconite intoxication

Aconite (*Aconitum carmichaelii* Debx.) poisoning is probably the most common cause of cardiac toxicity among people taking Chinese herbs. Clinical presentation typically includes numbness and paresthesia beginning in the mouth and spreading to the limbs, as well as dizziness, nausea, vomiting, weakness, sweating, and palpitations. The conscious state may be reduced. With severe poisoning, hypotensive sustained ventricular tachyarrhythmias may develop, often complicated by acidosis and hypokalemia [51]. Since aconite toxicity is common, it can be manifested as; involuntary salivation, nausea, vomiting, diarrhea, dizziness, blurred vision, dry mouth, vertigo, numbness of the body and extremities, slowed pulse, difficulty in breathing, twitching of the limbs, convulsions, disorientation, urinary and fecal incontinence, decrease in blood pressure and temperature, arrhythmia, tachyarrhythmia, bradycardia and possible death [52, 53]. Whereby, death occurred in aconite poisoning was due to the cardiac arrest which is caused by fatal arrhythmia such as ventricular fibrillation (Vf). Thus, it was noted that, a great importance to understand arrhythmias that are caused by aconite poisoning was required [50].

2.4 Herbal Treatment of aconite intoxication

Aconite poisoning, especially causing cardiac toxicity which is associated with arrhythmias may be reversible by supportive therapy [51]. Consequently, these aconite toxic effects manifestations can usually be reversed by Chinese herb *Rou Gui* (Cortex *Cinnamomi*), this is usually used to reverse during an early- stage overdose, i.e. within 4-6 hours of *fuzi* ingestion. There is a report, where by acute *fuzi* poisoning in 14 patients were treated by oral ingestion of *Rou Gui* tea, which was prepared by soaking 5 to 10 grams of the herbs in hot water. Hence, the *Rou Gui* tea do induce vomiting of the toxin within 5-15 minutes and thus, relieved overall symptoms within 15 to 30 minutes. And if the toxic symptoms persist, then, the process was repeated again by using 3 to 5 grams of *Rou Gui* in tea. Henceforth, the resolution treatment of the poisoning is characterized by warmth at the extremities, increased contractility of the heart and gradual sensory recovery from numbness of the mouth, lips and extremities [52, 54].

The overdose of aconite can also be treated with a decoction containing Chinese herbs; *sheng jiang* (Rhizoma *Zingiberis* Recens), *Gancao* (Radix *Glycyrrhizae*), *Ganjiang* (Rhizoma *Zingiberis*), *Lu Dou* (Semen *Phaseoli* Radiati), *Hei Dou* (Semen *Glycine* Max), *Huang Lian* (Rhizoma *Coptidis*), *Ren Shen* (Radix *Ginseng*) and *Huang Qi* (radix *Astragali*). Where by the dosage of each of the herb will vary depending on the specific condition of the patient [52].

Moreover, the specific overdose which is characterized by irregular heartbeat can be treated with a decoction of 20 grams of *Ku Shen Gen* (Radix *Sophorae* Flavescentis) and 10 grams of *Gancao* (Radix *Glycyrrhizae*). While the overdose which is characterized by extreme coldness of the extremities, extremely weak pulse and shortness of breath, can be treated with a decoction of *Ren Shen* (Radix *Ginseng*), *Gancao* (Radix *Glycyrrhizae*) and *Gan jiang* (Rhizoma *Zingiberis*) [52]. Therefore, the herbal treatments of these toxicities associated with CW and FZ have not been fully researched to show the pharmacological mechanism that is associated with the management of search toxicities in case of accidental ingestion of such herbs.

3. Preventive measures of Chinese aconite toxicity

3.1 TCM approach in elimination/reduce toxicity of Aconite

Pao zhi is a common approach that is used to strengthen the effect, eliminate or reduce the toxicity, facilitate the preparation and storage of drugs [55]. During processing, secondary plant metabolites are transformed, thus helping to exert maximal therapeutic efficacy [56] and reduce toxicity, and altering their effects [55]. For example, processing form of *Chuanwu* and *Fuzi* converted into *Zhichuanwu* and *Yanfuzi* preparation respectively, whose forms were recorded in CP 2010. From this preparation, there are more than 20 commonly used proprietary herbal formulas from both historical literature and modern clinical reports

containing processed medicine that include the processed *Yanfuzi* or *Zhichuanwu* as a main ingredient, such as “*Yin Chen Si Ni Tang*”, “*Fuzi Lizhong Wan*”, “*Guifulizhong Wan*”, “*Jinguishenqi Wan*”, and “*Shenfu injection*” [57, 58]. The main indications for these products are muscular disorders, joint pain and arthritis [59, 60].

Therefore, as a detoxifying measure, Pao zhi is necessary to remove the poisonous Aconitum alkaloids (i.e. in *CW* and *FZ*) mainly deriving from the diester diterpene alkaloids (DDAs) including aconitine, mesaconitine and hyaconitine [55] to relatively less toxic monoester diterpene alkaloids [61-63]. For example, before used in proprietary herbal products, the toxicity of these alkaloids can be lowered by hydrolysis into much less poisonous benzoyleaconines which are the products of deacetylation of the 8 β -acetoxy [55]. Like in an analytical research done, it was noted that; the contents of three alkaloids in *processed chuanwu* samples were lower than those in *raw chuanwu* samples; this is so because, aconite alkaloids in *raw chuanwu* were hydrolyzed to their corresponding semi-hydrolyzed products during treating process [64]. Moreover, evaluating toxicological differences between *raw* and *processed* rootstocks of *Aconitium carmichaelii* Debx was also depicted in an experimental study, which revealed that *raw chuanwu* and *fuzi* could lead to a serious myocardial and liver damage, when compared to the *processed products* which exhibit weak toxicity. [65].

This was supported in Histopathology, electron micrograph and serum biochemical experiment in rats, which showed that in *raw products*, serum creatinine kinase (CK), lactate dehydrogenase (LDH) and aspartate transferase (AST) level were increased significantly (P <0.05) when compared to the control group, while that of *processed products* there was a marked decrease of CK, LDH, and AST when compared to their *raw products group* [65]. While in histopathology *raw chuanwu* and *fuzi* produced more severed pathological damages compared to their *processed products* respectively, i.e. inflammatory cells and confluent areas were present in the cardiac muscle tissue, edema and rupture of striated muscle tissue were also observed, and these were compared to control group. While the *processed CW* and *FZ* were compared to their *raw products* respectively [65].

Furthermore, in electron micrograph still supported the serum biochemical test, whereby in *raw CW* and *FZ* group showed apparent swelling of endothelium with a large number of caveolae in endothelial cells and prominent edema in the vascular surrounding, dramatic injury in cardiac muscle cells, which was manifested as disrupted myofibrils and ruptured mitochondria. While in control and *processed CW* and *FZ*, cardiac myofibrils stood regularly arranged with well-preserved myofilaments and mitochondria occupied the cytoplasm between myofibrils with densely packed cristae [65].

Henceforth, the Histopathology and electron micrograph still supported the serum biochemical test, showing more serious toxicity in *raw products* than in *processed products*, which showed weak toxicity. Thus, from the experiment results it was depicted that traditional Chinese processing approach “*Paozhi*”, can decompose the poisonous drugs into less or nontoxic derivatives and thus, guaranteeing clinical safety and low toxicity in decoction [65]. However, the specific change process and metabolite conversion mechanism of Pao zhi were still unknown. Thus, this lead to a research further to analyze the change in metabolic markers for better understanding in mechanism of attenuated toxicity caused by Pao zhi [65].

3.2 Mechanism of Tcm processing approach ‘paozhi’ used in lowering aconite toxicity

Despite the fact that, Pao zhi can decompose drugs into less or nontoxic derivatives, several metabolomic studies have been used to evaluate the differences in toxicological and pharmacological actions of aconite products, so as a clear understanding of mechanism is evaluated for drug safety [59, 60]. For examples key metabolite markers used are lysophosphatidylcholine (LPC), dihydrosphingosine, phytosphingosine and tryptophan [65]. However, in a metabolomic study research done, it was observed that Pao zhi was detoxifying by regulating sphingolipid metabolism, glycerophospholipids metabolisms, aminoacyl-tRNA biosynthesis and tryptophan metabolism [65]. Whereby, *raw chuanwu* and *fuzi* were seen to decrease the level of lysophosphatidylcholine (LPC), dihydrosphingosine, phytosphingosine and tryptophan metabolites in serum when compared to the normal group. Unlike the *processed Chuanwu* and *fuzi*, which showed an increased in this metabolites when compared to their *raw products*. [65].

Therefore, as it is known that Dihydrosphingosine and Phytosphingosine are classified as Sphingolipids; they do play important roles in sphingolipids biosynthesis and metabolism [66, 67]. While decrease in serum lysophosphatidylcholine (LPC) metabolite in *raw products* was owing to increase autotoxin activity within serum. Hence, leading to the rapid conversion of lysophosphatidylcholine (LPC) into lysophosphatidic acids (LPAs), thus interfering with sphingolipids biosynthesis [65]. Since, sphingolipids are the membrane components of cells and emerged as key mediators of stress responses by canonical (protein kinase activation or inhibition and G-coupled receptor signaling) and non canonical (components of lipid platforms and initiation of membrane pore formation) paradigms, [68] therefore, interfering with its biosynthesis lead to cell stress [65]. Also, intensive investigation in past decade has firmly established the role of sphingolipids in controlling signal pathways for cell survival and aging [69, 70]. Hence, it was concluded that Pao zhi is detoxifying by regulating sphingolipid metabolism, glycerophospholipids metabolisms, aminoacyl-tRNA biosynthesis and tryptophan metabolism [65].

4. CONCLUSION

Aconitum carmichaelii Debx (CW and FZ) plant is highly rich in aconitine alkaloids, which are highly poisonous and are the causative agents in causing toxicity of this plant. TCM processing approach Pao zhi is an efficient method in reducing toxicity of *Aconitum carmichaelii* Debx. However, still more information is needed to understand the cardiac arrhythmias associated with *Aconitum carmichaelii* Debx, as we have seen more serious poisoning is in the myocardium and even lead to death. Also, we need to explore the plant further in observing the toxicity effects when used in combinations with other Chinese herbs. As it is known that, use of a single herb often possesses extensive effects, but if some of these effects could not match the situation of a specific disease, it may be harmful to patients [71].

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