scientific reports

Check for updates

OPEN

Epidemiological patterns and therapeutic approaches of toad toxin poisoning in a retrospective case study

Yuhang Liu, Jinlong Zhang, Binbin Wang, Feifei Zheng & Jie Yan✉

Toad toxin, a bioactive compound revered in traditional Chinese medicine, has been employed therapeutically for centuries. Recent studies have increasingly confirmed its pharmacological benefits, including cardioprotection, anesthetic effects, anti-inflammatory properties, enhancement of sexual function, and antineoplastic activities. This toxin is applied in the treatment of diverse medical conditions such as chronic bronchitis, pharyngitis, and colon cancer. Nonetheless, the consumption of toad-related substances—such as flesh, eggs, gallbladders—or the medicinal use of toad toxin frequently leads to poisoning incidents, some of which are fatal. This paper comprehensively reviews the principal features of toad toxin poisoning, encompassing clinical symptoms, therapeutic approaches, and other relevant factors to aid in the diagnosis and management, as well as the forensic evaluation of lethal cases. We advocate for further research into the cardiotoxic and neurotoxic effects of toad toxin to deepen our understanding of its poisoning mechanisms and pharmacological profile. Future efforts should focus on regulatory standardization of treatment practices and public education to mitigate the risks associated with toad toxin exposure.

Keywords Retrospective analysis, Toad toxin, Poisoning, Clinical manifestations, Clinical treatment

Toads, encompassing a broad taxonomic category within the order Anura and the family Bufonidae, are represented by over 300 species across 26 genera. Notable species include *Anaxyrus americanus* in the United States and Canada, *Bufo bufo* and *Bufotes viridis* in Eurasia and northwestern Africa, *Duttaphrynus melanostictus* in Southeast Asia, *Rhinella marina* in South America and Australia, and *Bufo gargarizans* in China[1–7] (Fig. 1). Historically documented in ancient Chinese medical texts, such as the "List of Famous Doctors," toads have long been valued for their medicinal properties[8]. The dried secretion from the parotoid and dorsal skin glands, known as Venenum Bufonis or "Chan su," has been effectively used in traditional Chinese medicine to treat abscesses, sore throats, toothaches, and other ailments[9–11]. Venenum Bufonis has shown considerable therapeutic effects against various cancer cell types, including those from liver, colon, lung, and gastric cancers, positioning it as a promising candidate for novel anticancer drug development[12–17]. The desiccated body of the toad, referred to as "toad-cortex," is employed in traditional practices for treating childhood sores, chronic bronchitis, and abscesses. Its recent applications extend to cancer treatment, either alone or in combination with conventional therapies, where it enhances therapeutic efficacy and alleviates side effects while improving hematological parameters[15,16].

In China, over 100 types of patented medicines contain toad ingredients, including notable products such as Liushen Pill, Chansu Pill, and Chan Su Analgesic Cream[18–20]. In the United States, toad toxin is also formulated into various aphrodisiac products like "Rock Hard" and "Black Cube"[21].

While folk remedies and certain studies have highlighted potential medicinal benefits of toad toxins, there remains a lack of definitive guidelines regarding their safe usage and dosage (Qi et al. 2018). According to the "Chinese Pharmacopoeia," the recommended dosage for internal use of Venenum Bufonis decoction ranges from 0.015 to 0.03 g. For external applications, an appropriate amount of Venenum Bufonis should be ground into powder for direct application or mixed into an ointment. It can also be formulated into various medicinal preparations such as pills or agentia. Content limits for certain formulations are specified, such as musk in Shexiang Baoxin pills and Musk Tongxin dropping pills, with ranges set at 18–56 μg/pill and 35–70 μg/pill respectively, while other formulations only stipulate minimum content requirements. Research indicates that doses between 4.86 and 120 mg/kg of Venenum Bufonis may result in acute poisoning or death in animals,

Department of Forensic Science, School of Basic Medical Science, Central South University, Changsha, Hunan, China. ✉email: wills212156@csu.edu.cn

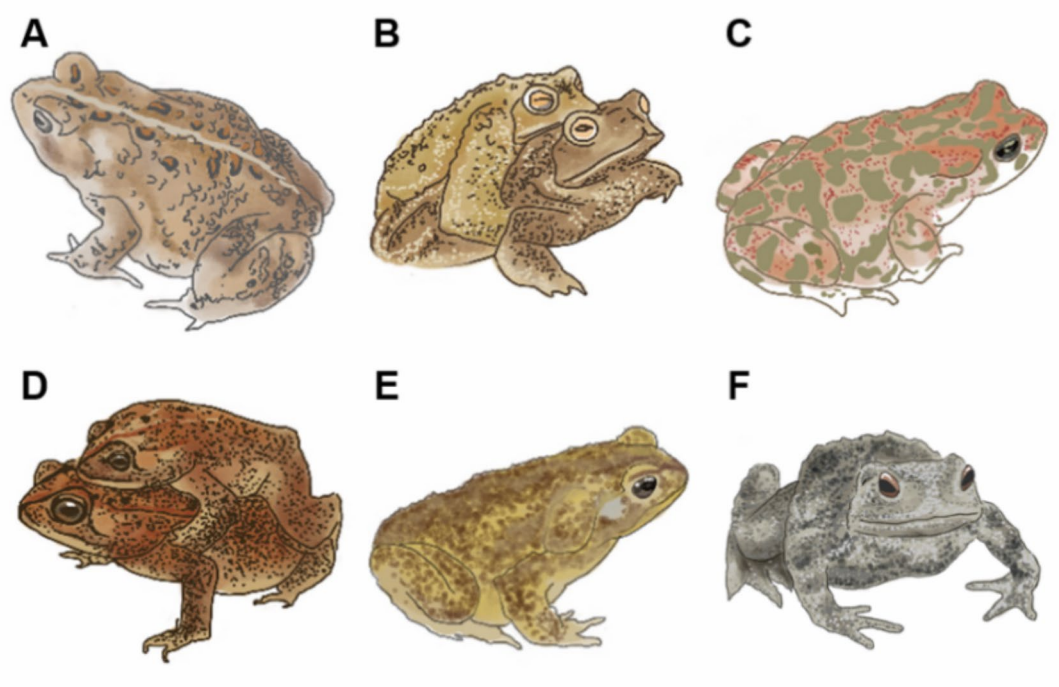

Fig. 1. Pictures of different types of toads. (**A**) *Anaxyrus americanus*. (**B**) *Bufo bufo*. (**C**) *Bufotes viridis*. (**D**) *Duttaphrynus melanostictus*. (**E**) *Rhinella marina*. (**F**) *Bufo gargarizans*.

corresponding to a human equivalent dose of approximately 62-585.4 mg/day (Qi and Li 2007; Song et al. 2005). Clinical reports have documented fatalities from ingesting doses as low as 1.5 g and as high as 6 g, though there are also accounts of recovery from poisoning after consuming doses ranging from 0.58 g to 10 g (Wei and Chen 1993; Zhou and Liu 2004; Zhou and Liu 1988; Jiang 2002).

Despite the recognition of toad toxin's toxicological profile, standardized treatment protocols for toad poisoning remain undefined. This study compiles cases of toad toxin poisoning from literature over recent decades for a retrospective analysis, summarizing symptomatology, treatment strategies, and prognosis factors, to improve clinical management outcomes.

Materials and methods

Data source

Database and search strategy

Multiple scientific databases were utilized for case collection, including Web of Science (WOS), PubMed, VIP Journal of Chinese Science (VIP), WAN FANG Database (WF), China National Knowledge Infrastructure (CNKI), and China Biomedical Literature Service System (Sinomed). Two independent investigators conducted a systematic literature search from database inception to September 30, 2023. Search terms were applied either singly or in combination: "Toad toxin", "Toad egg", "Toad gallbladder", "Venenum Bufonis", "Chan su", and "poisoning". The search was conducted without language restrictions. This retrospective study was approved by Ethics Committee of School of Basic Medical science, Central South University (No.2021-KT10) and the informed consent requirement was waived.

Inclusion and exclusion criteria

Inclusion criteria included studies that: (1) reported poisoning case reports involving ingestion of toads or toad-related products; (2) provided demographic information on the affected individuals; (3) detailed clinical presentations, diagnostic approaches, therapeutic interventions, and outcomes; and (4) were published adhering to national and international guidelines ensuring case authenticity. Exclusion criteria excluded: (1) studies involving animal experimentation; (2) studies with incomplete medical records or data unobtainable from other sources; (3) reports unrelated to medical science, such as studies on environmental toxicants.

Literature selection

Literature was independently screened by two investigators based on the inclusion and exclusion criteria. They independently selected and verified the studies, reconciling any discrepancies through discussion with a third investigator in cases of disagreement.

Data extraction

Data was systematically extracted using a standardized form capturing: patient age, gender, year of poisoning, purpose of ingestion, ingestion route of toad toxin, clinical manifestations post-poisoning, interval from ingestion to hospital admission, duration of hospitalization, administered treatments, and patient outcomes.

Analysis method

General information

We conducted a temporal analysis of poisoning incidents, quantifying trends based on annual data. Patients were stratified into four age groups, with the final group encompassing individuals aged 60 years and older, and the remaining groups segmented into 20-year intervals. Additionally, gender distribution and the underlying causes of poisoning were assessed. Symptomatology associated with toad toxin exposure was meticulously cataloged from the medical records and organized by the affected anatomical systems. Therapeutic interventions administered to the patients were systematically classified and quantified for further analysis.

Death case analysis

Despite recovery in some cases of toad toxin poisoning, approximately one-third of these incidents have been fatal. To understand these fatal outcomes, we analyzed the ages of the deceased, the respective years of death, and the specific modes of toad toxin ingestion. This analysis aimed to identify critical factors contributing to mortality and to guide preventive strategies and therapeutic approaches.

All experiments were performed in accordance with the relevant guidelines and regulation.

Results

Description of included studies

We identified 1542 relevant publications initially across multiple databases. After eliminating duplicates, 447 unique publications remained. Upon full-text review based on our inclusion and exclusion criteria, 126 publications were excluded, leaving 72 that met all criteria for detailed analysis. From these, we identified 180 cases of toad toxin poisoning (Supplementary Fig. 1).

General data, clinical symptoms and treatment

Out of the 180 cases, detailed timelines were available for 129, all reported within China. These cases span from 1961 to 2020, divided into 12 five-year intervals for analysis. The number of toad toxin poisoning incidents over these periods indicated an initial increase, peaking between 2001 and 2005, followed by a subsequent decline (Fig. 2). Patient ages ranged from 4 to 82 years, with a significant number of cases occurring among the elderly and children. The gender distribution showed 108 males (65.06%) and 58 females (34.94%), highlighting a higher incidence in males.

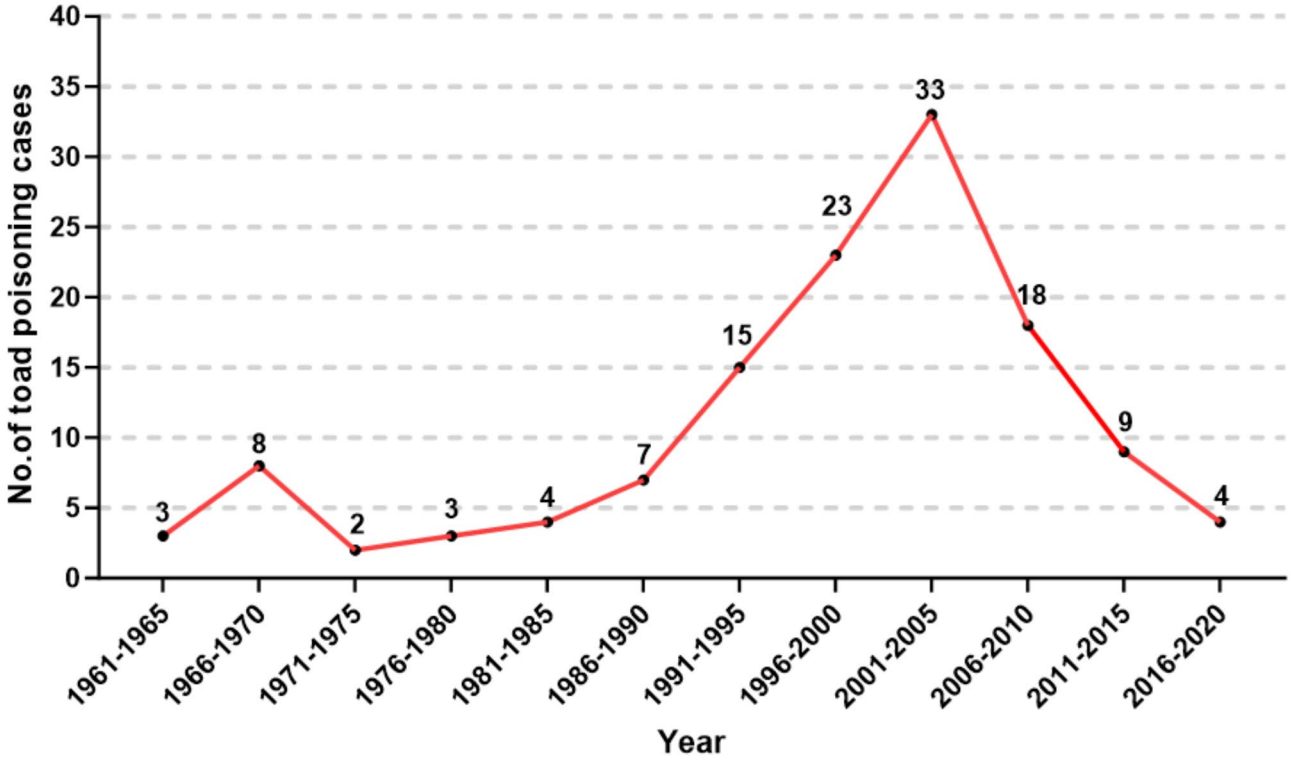

Fig. 2. Trends in toad toxin poisoning cases from 1961 to 2020.

Types of edible toads and toad-related products	Sex distribution (cases)			Total (cases)
	Male	Female	Unknown	
Toad meat	59	30	29	118
Toad egg	10	8	9	27
Liquor containing toad skin	0	0	3	3
Toad gallbladder	2	0	0	2
Venenum Bufonis	6	5	0	11
Chinese patent drugs containing toad toxin	5	0	0	5
Aphrodisiac-containing toad toxin	7	0	0	7
Tadpole	0	0	3	3
Toad skin secretions	0	0	2	2
Toad skin	0	2	0	2
Total	89	45	46	180

Table 1. Types of ingesting toads and toad-related products, sex distribution of the poisoned.

Anatomical system	Symptom	Positive case	Positive rate (%)
Digestive system	Emesis	171	94.91%
	Nausea	104	39.44%
	Stomachache	91	50.55%
	Diarrhea	33	18.33%
Respiratory and circulatory system	Chest tightness	58	32.22%
	Palpitations	15	8.33%
	Shortness of breath	14	7.77%
	Cyanochroia	1	0.55%
Nervous system	Headache	32	17.77%
	Dizziness	17	9.44%
	Sleepiness	15	8.33%
	Sweating	14	7.77%
	Acro-anesthesia	24	13.33%
	Fatigue	15	8.33%

Table 2. The manifestations of poisoned patients.

Given the recognized medicinal value of toads and the variety of derived preparations, individuals are exposed to toad toxins through multiple routes. Among the 180 documented cases of toad toxin poisoning, a diverse array of exposure pathways was noted. The predominant route was via consumption of toad meat, accounting for 65.55% of cases. Other notable routes included ingestion of toad eggs (15%), consumption of alcohol infused with toad skin (1.67%), and intake of toad bile (1.11%). Additionally, traditional Chinese medicine formulations such as "Chansu" were implicated in 6.11% of cases, and Chinese patent medicines, including "Liu Shen Wan", "Shexiang Baoxin Wan", and "Chansu Wan", accounted for 2.77%. Further exposure occurred through aphrodisiacs derived from toads (3.88%), consumption of tadpoles (1.67%), toad skin secretions (1.11%), and direct consumption of toad skin (1.11%) (Table 1).

In the analysis of the 180 cases, the majority of incidents involved patients who mistakenly consumed toads, confusing them with frogs, leading to poisoning. Some children inadvertently ingested toad eggs during outdoor activities, resulting in toxicity. Additionally, a portion of the cases were due to intentional consumption of toads or toad-derived products for medicinal purposes. Specific ailments treated included respiratory system diseases (6 cases), cardiovascular diseases (2 cases), digestive system diseases (3 cases), tumors (6 cases), skin diseases (5 cases), immune system disorders (3 cases), oral diseases (3 cases), use as aphrodisiacs (7 cases), and other diseases (2 cases) (Supplementary Table 1).

The latency period for toad toxin effects ranged from 30 min to 1 h, with the vast majority (95.5%) of cases manifesting symptoms within 5 h. The median onset time, denoting the interval from ingestion to symptom emergence, was established at 1 h (IQR = 1). Predominant clinical symptoms included emesis (94.91%), nausea (57.77%), abdominal pain (50.55%), dizziness (17.77%), diarrhea (18.33%), limb numbness (13.33%), and headache (9.44%) (Table 2). The digestive system was the most frequently affected, followed by impairments to the respiratory and circulatory systems. The nervous system also showed considerable effects, with 57 cases (31.66%) reporting neurological damage. This sequence underscores the widespread impact of toad toxin across multiple body systems, with vomiting and severe effects on the respiratory and circulatory systems being particularly prominent.

Toad toxin is characterized by a rapid onset of toxic effects, emphasizing the need for immediate medical intervention upon exposure. In clinical settings, poisoned patients typically receive standard detoxification treatments such as gastric lavage and hemoperfusion to eliminate the toxin. Given the toxin's propensity to cause severe cardiac conduction disturbances, careful monitoring and adjustment of treatments are crucial. Specifically, if a patient's heart rate falls below 60 beats per minute, the administration of higher doses of atropine or alteration of the therapeutic regimen should be considered. A heart rate below 40 beats per minute raises concerns for asystole, necessitating immediate preparations for defibrillation, pacing, and tracheal intubation. In instances where atropine fails to resolve atrioventricular block, isoproterenol may be administered intravenously. The management of vital signs and the prompt treatment of shock, electrolyte imbalances, and cardiac arrhythmias are critical to improving outcomes in cases of toad toxin poisoning (Table 3). This approach underscores the urgency and complexity of treating such severe toxicological emergencies.

Prognosis

The toxic effects of a compound are generally influenced by its dosage and duration of action[22,23]. In this study, detailed quantification of the toads consumed (both in number and weight) was available for only a few cases. Complicating matters further, several patients consumed products derived from toad toxins, not the toads themselves, which obscures the exact toxic dosage received.

The majority of patients (70.55%) with toad toxin poisoning recovered with a hospital stay ranging from 1 to 15 days, averaging around 5 days. Interestingly, the mortality rate among patients in rural areas was higher compared to those in urban settings. Additionally, the mortality rate decreased significantly after 2010, with only one death reported post-2010. Prognosis varied with age; most young and middle-aged patients recovered post-treatment, whereas children under 15 years and seniors over 60 experienced a higher mortality rate of 56.75%.

Death cases analysis

Of the 180 cases studied, 53 resulted in death (29.44%). The majority of these fatalities (56.75%) occurred in individuals either over 60 or under 15 years of age, with males comprising 66.66% of the deaths. Clinical symptoms such as nausea, emesis, abdominal pain, diarrhea, dizziness, headache, drowsiness, sweating, and numbness did not directly cause fatalities. The primary causes of death were severe symptoms like chest tightness, palpitations, cyanosis, arrhythmias, cardiac conduction blockages, and respiratory and circulatory failure, leading to death.

Analysis of autopsy findings

Autopsy results from four children who succumbed to poisoning revealed significant findings in the gastrointestinal tract, including the presence of a greenish or egg-drop soup-like turbid fluid, congested and hemorrhagic gastric mucosa, and fecal residue. This mixture, comprised of vomit, blood, gastric acid, and bile, highlights the severe impact of toad toxins on the gastrointestinal mucosa and its motility. Pulmonary findings were equally concerning, with notable pulmonary edema, blood-tinged froth in the nasal cavity and trachea, hemorrhagic spots on the lung surface, and signs of congested edema in lung tissue cross-sections. Additionally, hemorrhagic episodes beneath the pericardium, likely due to increased vascular permeability triggered by acute circulatory failure induced by the toxin, were observed.

Discussion

This retrospective analysis examines the trends in toad toxin poisoning cases over the past six decades, primarily within China. The number of reported cases increased initially, peaking between 2001 and 2005. This trend may reflect the growth of China's publishing industry, which likely improved the documentation and public awareness of such incidents. As medical and scientific knowledge expanded and living conditions improved, public reliance on traditional folk remedies involving toads diminished. This shift, coupled with better access to medical services, likely contributed to the decline in toad toxin poisoning cases and the associated mortality rates post-2005. Notably, individuals under 20 years old were found to be particularly vulnerable to toad toxin poisoning, with adolescents being the most affected group. Additionally, a higher incidence of toad toxin poisoning and fatalities among males may be linked to cultural or behavioral factors that predispose males to such risks.

Types of treatment	Therapies	Positive case	Positive rate (%)
Hemopurification	Hemoperfusion	8	4.44%
	Continuous renal replacement therapy	1	0.55%
Internal toxin excretion	Gastric lavage	94	52.22%
	Inducing diarrhea	15	8.33%
	Inducing vomiting	7	3.88%
	Diuresis	2	1.11%
Humoral environmental regulation	Adjust electrolyte balance	14	7.77%
Drug therapy	Atropine	119	66.11%
	Isoproterenol	61	33.88%
Other	Supportive treatment	71	39.44%

Table 3. Treatment methods for poisoned patients.

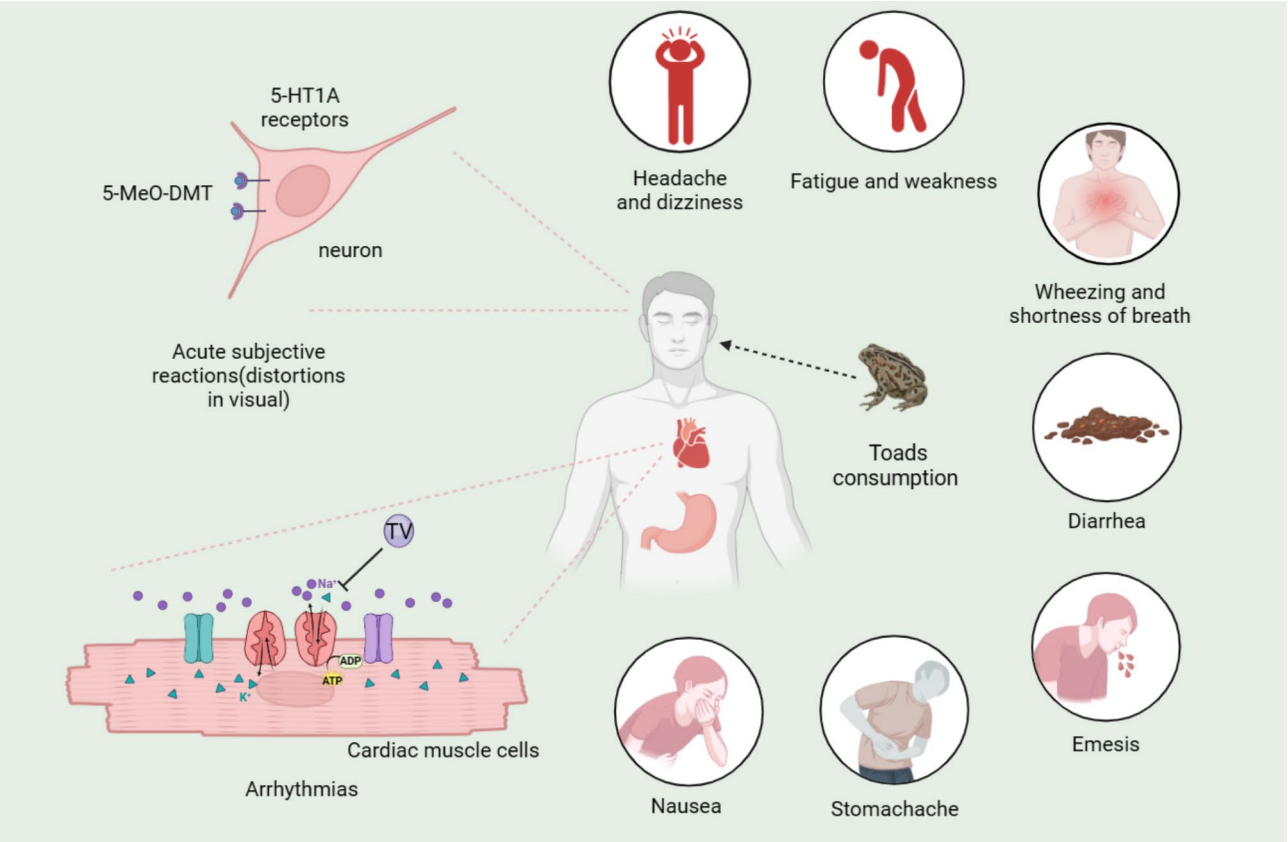

Fig. 3. The toxicity of toad venom on human body. Toad venom acts on gastrointestinal smooth muscle, enhancing peristalsis and thereby inducing nausea and vomiting. It also amplifies the action of the neurotransmitter norepinephrine, which stimulates the central nervous system and can lead to headaches. 5-MeO-DMT, a component of toad venom, interacts swiftly with the 5-HT1A receptors in the central nervous system. This interaction triggers a range of acute subjective reactions, including distortions in visual, auditory, and temporal perception, alongside intense emotional experiences and memory impairments. Additionally, toad venom targets the NKA on myocardial cell membranes, inhibiting their activity, which may result in arrhythmias. *TV* Toad Venom.

The chemical composition of toad toxin is highly complex, comprising primarily bufadienolides[24] and indole alkaloids. To date, 142 bufadienolide compounds have been isolated from toad toxin[16]. Prominent among these are gamabufotalin, telocinobufagin, bufotaline, cinobufotalin, bufalin, cinobufagin, and resibufogenin, which together constitute over 85% of the bufadienolides found in toad toxin[25–28]. Bufalin, cinobufagin, and resibufogenin are now recognized as benchmark indicators for quantifying the bufadienolides content in toad toxins[29]. Additionally, more than ten types of indole alkaloids have been identified, including notable substances such as bufotenidine, 5-Methoxy-N, N-dimethyltryptamine (5-MeO-DMT), and bufotenine[30–34]. These findings underscore the potent and diverse pharmacological profile of toad toxins.

The predominant symptoms of toad toxin poisoning primarily affect the digestive, nervous, respiratory, and circulatory systems[35]. Oral ingestion of the toxin frequently damages the gastrointestinal mucosa and disrupts motility, leading to prevalent digestive symptoms such as nausea, vomiting, diarrhea, and abdominal pain[35]. The indole alkaloids in toad toxins act on the central nervous system and exhibit hallucinogenic effects akin to lysergic acid diethylamide (LSD), with substances like bufotenine and 5-MeO-DMT being particularly notable. Bufotenine has been implicated in drug-facilitated sexual assaults[34,36], While 5-MeO-DMT binds to 5-HT1A receptors, dampening sympathetic nerve activity, which in turn reduces blood pressure and heart rate. This can elicit a range of acute subjective reactions including distortions in visual, auditory, and temporal perception, intense emotional experiences, and memory impairments[37–39].

Cardiovascular symptoms are primarily driven by bufadienolides[40]. At therapeutic doses, bufadienolides can be beneficial in treating congestive heart failure and arterial hypertension due to their positive inotropic effects, like those of digitalis[41,42]. These compounds bind to the Na^+/K^+-ATPase (NKA) on the myocardial cell membrane, inhibiting its activity and leading to elevated intracellular Na^+ levels. This action facilitates Ca^{2+} entry into myocardial cells via the Na^+/Ca^{2+} exchanger, prompting premature myocardial depolarization, an increase in diastolic pressure, and enhanced myocardial contractility[41,42]. The pronounced cardiac effects include simultaneous increases in myocardial contraction and diastolic peaks[43,44]. However, inappropriate usage of

bufadienolides may lead to severe cardiac toxicity due to their high selectivity and affinity for the NKA "digitalis receptor" site[45] (Fig. 3).

The dosage of poison typically serves as a critical factor in toxicology research and informs clinical treatment strategies[46]. Generally, larger toads tend to contain greater quantities of toxins, increasing the risk of poisoning. However, the absence of detailed information on the size and weight of toads consumed makes it challenging to determine the precise toxic dose of the toad toxin. Notably, toad toxins are distributed not only in the parotoid and dorsal skin glands but are also found in other anatomical parts such as the muscles, limbs, liver, and ovaries[47]. Additionally, the composition of bufotoxin and bufogenin varies significantly between fresh and dried toad toxins; fresh specimens typically have higher levels and a greater variety of bufotoxin but lesser and fewer bufogenin, whereas dried toad toxins exhibit the opposite characteristics[16,48]. The complex nature of toad toxin composition, coupled with the diverse methods by which people consume toads and the different parts ingested, poses significant challenges in establishing a precise toxic dosage.

Prompt medical intervention following toad toxin exposure significantly enhances the prognosis. Typically, individuals poisoned by toad toxin begin exhibiting toxic reactions within 30–60 min post-ingestion, implying the acute nature of this toxic substance. In cases of acute oral poisoning, immediate detoxification is crucial and can markedly reduce morbidity rates[49–51]. Early diagnosis and rapid treatment are paramount, with the initial hour post-exposure being critical, and ideally, emergency interventions should begin within the first 10 min.

Effective early interventions include the administration of emetics, gastric lavage, and stabilization of vital signs, along with timely management of shock, electrolyte imbalances, and cardiac arrhythmias. These measures are critical for favorable outcomes[35]. Gastric lavage, commonly used in treating toad toxin poisoning, should be performed promptly while concurrently maintaining vital signs. This procedure must ensure normal blood oxygen saturation levels, with oxygen supplementation provided as necessary[52]. Hemoperfusion stands out as an important treatment modality for toad toxin poisoning, capable of rapidly and effectively removing various toxins from the body, thereby minimizing toxic effects on organs and tissues. Continuous renal replacement therapy (CRRT) is another effective option[53]. The comprehensive elimination of toad toxin is best achieved through a combination of treatments such as gastric lavage, catharsis, and CRRT. Employing multiple therapeutic strategies simultaneously, like combining gastric lavage with hemoperfusion or CRRT, can significantly improve the success rates in treating toad toxin poisoning.

The mortality rate associated with toad toxin poisoning is high at 29.44%, which is greater than the 5.85% for fish bile poisoning and 18.75% for cantharidism[54]. Several factors influence the mortality rates in cases of toad toxin poisoning: (a) Timing of exposure: The incubation period for toad toxin is brief. In rural areas, where access to hospitals is limited or transportation is inadequate, delayed medical intervention can result in a higher mortality rate. (b) Age: In China, the mortality rate for individuals under the age of 15 and over 60 who suffer from toad toxin poisoning is 65.62%. This demographic exhibits a poorer prognosis, potentially due to weaker immune systems and reduced physiological resilience. (c) Underlying health conditions: Often, individuals consume toad toxins seeking therapeutic benefits for various ailments, inadvertently increasing their risk. The mortality rate among these patients is higher, likely exacerbated by pre-existing health conditions that can weaken immune responses and compromise physiological stability, particularly in those with cancer or other severe illnesses. (d) Treatment methods: Historically, the primary treatment for toad toxin poisoning has involved the rapid elimination of the toxin from the body[51]. Despite the application of gastric lavage and pharmacological interventions, the mortality rate remained elevated in the past due to limited medical resources and a lack of specific knowledge about effective treatments for toad toxin exposure. Since 2010, the introduction and use of hemoperfusion or CRRT in treatment protocols have led to a significant improvement in survival rates, suggesting that failure to effectively remove the toxin is a critical factor contributing to fatalities[55]. This analysis underscores the urgent need for prompt, effective medical interventions and highlights the importance of advancing healthcare accessibility and treatment methods to reduce mortality from toad toxin poisoning.

Conclusion

This study summarized the key features of toad toxin poisoning, focusing on symptoms, treatment strategies, and factors influencing prognosis and mortality. The findings provide insights for clinical diagnosis and treatment of toad toxin poisoning, as well as for forensic determinations of cause of death. There is a need for enhanced drug safety education and clear regulatory guidelines on the therapeutic use of toad-related products.

Data availability

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

Received: 18 November 2024; Accepted: 7 February 2025
Published online: 15 February 2025

References

1. Lan, Y. et al. Comparative transcriptomes of three different skin sites for the Asiatic toad (*Bufo gargarizans*). *PeerJ* **10**, e12993 (2022).
2. Masta, S. E. et al. Molecular systematics, hybridization, and phylogeography of the Bufo americanus complex in Eastern North America. *Mol. Phylogenet. Evol.* **24** (2), 302–314 (2002).
3. Elmberg, J. Amphibians and reptiles in North Sweden: distribution, habitat affinities, and abundance (classes: Amphibia and Reptilia). *Zootaxa* **5301** (3), 301–335 (2023).
4. Guzmán-Markevich, K. et al. Cytogenetic analysis in the toad species *Bufo spinosus*, *Bufotes viridis* and *Epidalea calamita* (Anura, Bufonidae) from the Mediterranean Area. *Genes (Basel).* **13**(8). (2022).

5. Othman, S. N. et al. Impact of the mid-pleistocene revolution and anthropogenic factors on the dispersion of Asian black-spined toads (*Duttaphrynus melanostictus*). *Anim. (Basel)*, **10**(7). (2020).
6. Slade, R. W. & Moritz, C. Phylogeography of *Bufo marinus* from its natural and introduced ranges. *Proc. Biol. Sci.* **265** (1398), 769–777 (1998).
7. Hu, Y. L. et al. Population genetics and phylogeography of *Bufo gargarizans* in China. *Biochem. Genet.* **45** (9–10), 697–711 (2007).
8. Yang, Q. et al. Angel of human health: current research updates in toad medicine. *Am. J. Transl. Res.* **7** (1), 1–14 (2015).
9. Zhakeer, Z. et al. Bufalin inhibits the inflammatory effects in Asthmatic mice through the suppression of Nuclear factor-Kappa B Activity. *Pharmacology* **99** (3–4), 179–187 (2017).
10. Wen, L. et al. Anti-inflammatory and antinociceptive activities of bufalin in rodents. *Mediat. Inflamm.* **2014**, 171839 (2014).
11. Wei, W. L. et al. Venenum bufonis: an overview of its traditional use, natural product chemistry, pharmacology, pharmacokinetics and toxicology. *J. Ethnopharmacol.* **237**, 215–235 (2019).
12. Dong, Q. et al. Bufadienolides from the Bufo viridis toad venom exert cytotoxic effects on cancer cells by inducing cell apoptosis and cell cycle arrest. *Toxicol. Vitro*. **89**, 105566 (2023).
13. Zhou, S. W. et al. Bufadienolides from the Eggs of the Toad Bufo bufo gargarizans and their antimelanoma activities. *J. Nat. Prod.* **84** (5), 1425–1433 (2021).
14. Zhang, D. M. et al. Arenobufagin, a natural bufadienolide from toad venom, induces apoptosis and autophagy in human hepatocellular carcinoma cells through inhibition of PI3K/Akt/mTOR pathway. *Carcinogenesis* **34** (6), 1331–1342 (2013).
15. Zhang, H., Jian, B. & Kuang, H. *Pharmacol. Eff. Cinobufagin Med. Sci. Monit.*. **29**, e940889 (2023).
16. Li, F. J. et al. Toad venom: a comprehensive review of chemical constituents, anticancer activities, and mechanisms. *Arch. Pharm. (Weinheim)*. **354** (7), e2100060 (2021).
17. Wang, J. et al. Cinobufacini inhibits colon cancer invasion and metastasis via suppressing Wnt/β-Catenin signaling pathway and EMT. *Am. J. Chin. Med.* **48** (3), 703–718 (2020).
18. Zhang, X. et al. Analysis of therapeutic mechanism of Liushen Wan against colitis-associated colorectal cancer based on network pharmacology and validation in mice. *Nan Fang Yi Ke Da Xue Xue Bao*. **43** (7), 1051–1062 (2023).
19. Wang, Z. et al. Integrated network pharmacology and fecal metabolomic analysis of the combinational mechanisms of Shexiang Baoxin pill against atherosclerosis. *Mol. Omics*. **19** (8), 653–667 (2023).
20. Datta, P. & Dasgupta, A. Interactions between drugs and Asian medicine: displacement of digitoxin from protein binding site by bufalin, the constituent of Chinese medicines Chan Su and Lu-Shen-Wan. *Ther. Drug Monit.* **22** (2), 155–159 (2000).
21. Brubacher, J. R. et al. Treatment of toad venom poisoning with digoxin-specific Fab fragments. *Chest* **110** (5), 1282–1288 (1996).
22. Yang, X. et al. Redox regulation in hydrogen sulfide action: from neurotoxicity to neuroprotection. *Neurochem. Int.* **128**, 58–69 (2019).
23. Lu, S. et al. Antioxidant cascades confer neuroprotection in ethanol, morphine, and methamphetamine preconditioning. *Neurochem. Int.* **131**, 104540 (2019).
24. Asrorov, A. M. et al. Toad venom bufadienolides and bufotoxins: an updated review. *Drug Dev. Res.* **84** (5), 815–838 (2023).
25. Yu, Z. et al. Gamabufotalin, a bufadienolide compound from toad venom, suppresses COX-2 expression through targeting IKKβ/NF-κB signaling pathway in lung cancer cells. *Mol. Cancer*. **13**, 203 (2014).
26. Ma, X. et al. Telocinobufagin inhibits osteosarcoma growth and metastasis by inhibiting the JAK2/STAT3 signaling pathway. *Eur. J. Pharmacol.* **942**, 175529 (2023).
27. Zhang, W. et al. Bufotalin induces ferroptosis in non-small cell lung cancer cells by facilitating the ubiquitination and degradation of GPX4. *Free Radic. Biol. Med.* **180**, 75–84 (2022).
28. Cao, Y. T. et al. Correlative study of bufogenin constituents in venom of Bufo bufo gargarizans. *China J. Chin. Materia Med.* **44** (9), 1850–1856 (2019).
29. Chen, W. J. et al. Research progress on biotransformation and biosynthesis of bufadienolides. *Chin. Tradit. Herb. Drugs*. **52** (15), 4741–4751 (2021).
30. Yang, M. et al. Identification of protein quality markers in Toad Venom from Bufo gargarizans. *Molecules*. **28**(8). (2023).
31. Qu, T. et al. Content of indole alkaloids and bufadienolides contained in toad medicines. *Zhongguo Zhong Yao Za Zhi*. **37** (20), 3086–3091 (2012).
32. Chen, Y. L. et al. Two new indole alkaloids from Toad Venom of Bufo bufo gargarizans. *Molecules*. **25**(19). (2020).
33. Uthaug, M. V. et al. A single inhalation of vapor from dried toad secretion containing 5-methoxy-N,N-dimethyltryptamine (5-MeO-DMT) in a naturalistic setting is related to sustained enhancement of satisfaction with life, mindfulness-related capacities, and a decrement of psychopathological symptoms. *Psychopharmacol. (Berl).* **236** (9), 2653–2666 (2019).
34. Navarro Escayola, E., Vega Vega, C. & Hernando Torrecilla, C. Bufotenine use in drug facilitated sexual assult. *Med. Clin. (Barc).* **159** (10), e66–e67 (2022).
35. Trakulsrichai, S. et al. Toad poisoning: clinical characteristics and outcomes. *Ther. Clin. Risk Manag.* **16**, 1235–1241 (2020).
36. McBride, M. C. Bufotenine: toward an understanding of possible psychoactive mechanisms. *J. Psychoact. Drugs*. **32** (3), 321–331 (2000).
37. Reckweg, J. T. et al. The clinical pharmacology and potential therapeutic applications of 5-methoxy-N,N-dimethyltryptamine (5-MeO-DMT). *J. Neurochem.* **162** (1), 128–146 (2022).
38. Albert, P. R. & Vahid-Ansari, F. The 5-HT1A receptor: signaling to behavior. *Biochimie* **161**, 34–45 (2019).
39. Barsuglia, J. et al. Intensity of mystical experiences occasioned by 5-MeO-DMT and comparison with a prior psilocybin study. *Front. Psychol.* **9**, 2459 (2018).
40. Isoardi, K. et al. Bufadienolide toxicity in a child following cane toad egg ingestion. *Emerg. Med. Australas.* **30** (5), 728–729 (2018).
41. Laursen, M. et al. Structures and characterization of digoxin- and bufalin-bound Na+,K+-ATPase compared with the ouabain-bound complex. *Proc. Natl. Acad. Sci. U S A*. **112** (6), 1755–1760 (2015).
42. Prassas, I. & Diamandis, E. P. Novel therapeutic applications of cardiac glycosides. *Nat. Rev. Drug Discov.* **7** (11), 926–935 (2008).
43. Ma, H. et al. Interactions between bufadienolides derived from toad venom and verapamil in langendorff-perfused guinea-pig hearts. *Toxicol. Vitro*. **27** (1), 396–401 (2013).
44. Puschett, J. B., Agunanne, E. & Uddin, M. N. Emerging role of the bufadienolides in cardiovascular and kidney diseases. *Am. J. Kidney Dis.* **56** (2), 359–370 (2010).
45. Sousa, L. Q. et al. Bufadienolides from amphibians: a promising source of anticancer prototypes for radical innovation, apoptosis triggering and na(+)/K(+)-ATPase inhibition. *Toxicon* **127**, 63–76 (2017).
46. Guo, L. M. et al. RIP3/MLKL-mediated neuronal necroptosis induced by methamphetamine at 39°C. *Neural Regen Res.* **15** (5), 865–874 (2020).
47. Tóth, Z. et al. Changes in toxin quantities following experimental manipulation of Toxin reserves in Bufo bufo tadpoles. *J. Chem. Ecol.* **45** (3), 253–263 (2019).
48. Wang, Z. Y. et al. Comparison of chemical composition between fresh and processed Bufonis Venenum by UPLC-TQ-MS. *Zhongguo Zhong Yao Za Zhi*. **40** (20), 3967–3973 (2015).
49. Lu, S. et al. Current status and potential role of circular RNAs in neurological disorders. *J. Neurochem.* **150** (3), 237–248 (2019).
50. Yang, X. et al. The main molecular mechanisms underlying methamphetamine-induced neurotoxicity and implications for pharmacological treatment. *Front. Mol. Neurosci.* **11**, 186 (2018).
51. Lin, G. et al. Continuous venovenous hemofiltration in the management of paraquat poisoning: a meta-analysis of randomized controlled trials. *Med. (Baltim).* **96** (20), e6875 (2017).

52. Dai, Y. H. et al. A new indole alkaloid from the Toad Venom of Bufo bufo gargarizans. *Molecules* **21** (3), 349 (2016).
53. Song, Z. P. & Wang, P. The electrocardiogram characteristics and management of toad poisoning. *J. Militaa Suryeon Southwest. China*. **p. 22** (2), 165–168 (2020).
54. Yan, W. et al. Analysis of factors related to prognosis and death of fish bile poisoning in China: a retrospective study. *Basic. Clin. Pharmacol. Toxicol.* **127** (5), 419–428 (2020).
55. Li, L. et al. Hemoperfusion plus continuous veno-venous hemofiltration in the treatment of patients with multiple organ failure after wasp stings. *Int. J. Artif. Organs*. **43** (3), 143–149 (2020).

Acknowledgements
Thanks to Siqi Chen for the contribution of image drawing.

Author contributions
J.Y. designed and supervised this study. J.L.Z. collects data. Y.H.L. prepared the manuscript. B.B.W. and F.F.Z. organized the data. Y.H.L. and J.Y. contributed to the manuscript revision with contributions from all of the other authors.

Funding
This work was supported by the National Natural Science Foundation of China (82171494).

Declarations

Competing interests
The authors declare no competing interests.

Additional information
Supplementary Information The online version contains supplementary material available at https://doi.org/10.1038/s41598-025-89809-0.

Correspondence and requests for materials should be addressed to J.Y.

Reprints and permissions information is available at www.nature.com/reprints.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Open Access This article is licensed under a Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License, which permits any non-commercial use, sharing, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if you modified the licensed material. You do not have permission under this licence to share adapted material derived from this article or parts of it. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit http://creativecommons.org/licenses/by-nc-nd/4.0/.

© The Author(s) 2025