INVASIVE TOAD’S SECRETIONS ACTS AS ANTICANCER AGENTS

[A REVIEW]

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ABSTRACT

The cane toad was introduced on the believe that it would act as pest control but it failed to do so and itself became an invasive species. This invasive species is over humiliated but as every coin has two sides it too has another side i.e it can be useful in many ways as its secretion, which is mild poisonous, shows traces of having pharmacological and therapeutic significance. The main concern of this article would be the chief secretion (poison) which the cane toad use as a defense for its own protection, the bufadienolides – cinobufagin and bufalin. The concern is also associated with the cancers-prostate cancer, etc which needs attention for chemoprevention and chemical controls/regulation. These bufadienolides are much satisfactory and could be used as a drug of choice as it possess anticancer properties. Besides their anticancer nature they show diverse effects too which shall be later discussed in this article in detail.

1. Classification

Kindom- Animalia
Phylum- chordata
Class- Amphibia
Order- Anura
Family – Bufonida
Genus – Rhinella
Species– R. Marinus.[1,2]

KEYWORDS: The bufadienolides – cinobufagin and bufalin.
2. INTRODUCTION

The cane toad is found in Central and South America and also in Northern Australia, Oceania and Caribbean. It is also called or known as Rhinella marina, giant neotropical toad and marine toad.\cite{3} It has poison glands and most of the animals are highly affected by toxic tadpoles if ingested. As a method of agricultural pest control it has been introduced to many regions of the Caribbean islands and Pacific, from its use against the cane beetle (Dermolepida albohirtum). The cane toad is now considered as pest and invasive species in many of its introduced regions, due to its toxic skin secretion, which kills many animals and native predators if ingested. The common name cane toad arised as it was used to eradicate pest from sugar cane.\cite{4} The cane toad can adapt to the local environment.\cite{5} It is estimated to have a critical thermal environment of 40-42 °C (104-108 F) and a minimum of 10 –15°C (50 – 59 F).\cite{6} Some studies showed that the cane toad has a high tolerance to water loss; some can withstand a 52.6% loss of body water, allowing them to survive outside tropical environments.\cite{7} The adult cane toad’s skin is toxic, as well as the enlarged parotoid glands behind the eyes and other glands across their back, when they are threatened, their glands secrete a milky white fluid known as bufotoxin.\cite{8} The components of it are toxic to many animals.\cite{9} Human deaths have been reported due to consumption of cane toads.\cite{10} Bufotenin, one of the chemicals excreted by the cane toad is classified as a class 1 drug under Australian law alongside heroin and cannabis. The effects of bufotenin are thought to be similar to those of mild poisoning, the stimulation, which includes mild hallucinations, which lasts less than an hour.\cite{11}
3. USES

The cane toad has been employed in a number of commercial and non-commercial applications, other than the previously mentioned uses as biological pests. Traditionally, in South America, the Embra Wounaan would milk the toads for their toxin, which was used as arrow poison. The toxin was also used as entheogen by the Olmec people, in Peru it is hunted for prey. And it is eaten only after the removal of skin and protoid glands.[12] The toads toxin is also used in many ways, the recent ones are, bufotenin, which has been used in Japan as an aphrodisiac and a hair restorer, and also in cardiac surgery in China to lower the heart rates of the patients.[13] New research has suggested that the cane toad’s poison may have some application in treating prostate cancer.[14] Pregnancy testing is one of the modern use of the cane toad, the testing was conducted in the mids 20th century, it is performed by injecting the woman’s urine into a male toad’s lymph sacs, and if spermatozoa or sperms appeared in the toad’s urine, then the patients pregnancy test was confirmed, if no sperm appears then the pregnancy test is negative.[15] Cane toads are also used to make leather and novelty items and even the fertilizers have been tried to make out of its body.[16] The mucous glands and granular glands secretion of toad can sometimes be poisonous.[17-19] Toxins produced by granular glands are more toxic secretions than mucous glands. The toxins or secretions of granular glands can be classified into four categories as Biogenic amines, Bufadienolides, Alkaloids and Steroids, Peptides and Proteins.[20] Chemically, other biogenic amines and peptides, granular secretions of toads may contain nearly 86 types of bufadienolides,[21] with also other components like, Bufotenine, bufotoxin, bufagin.[22-24] Some of the important bufadinolides are, Cinobufagin, Marinobufagin, Resibufagin Bufalin, Bufogenin, Bufotalin. Other major bufadienolides isolated from Bufo species are – Arenobufagin, Arenobufagin hemiesuberate, Arenobufotoxin, Argentinogenin, Bufalin, Bufalin hemiesuberate, Bufalitoxin, Bufotalin, Bufotalinin, Bufotalone, Cinobufagin, Cinobufagin hemiesuberate, Cinobufagino, Cinobufotoxin, Cinobufotalin, Cinobufotalitoxin, Desacetylcinobufotalin, Gamabufotalin, Gamabufotalitoxin, hellebrigenin, Hellebritoxin, Marinobufagin, Marinoic acid, Marinosin, Resibufogin, Resibufogenin, Resibufotoxin, Telocinobufogin, Vulgarobufotoxin. Bufadienolide consists of two double bonds present in the side chain of bufonolide, when combined with nitrogenous compounds like arginine, bufogenin in a free state is called as bufotoxin. Bufonolides are the derivatives of steroids which are present in conjugated as well as non-conjugated forms.[25-26] The conjugation may be cause due to the attachment at carbon number 3, resulting in sulfates, amino acid-dicarboxylic esters, dicarboxylic esters. Bufonin is a compound/component secreted by toad`s skin glands, which is a weak poison but it is
oxidized to form a bufanolide called bufotalin \([C_{34}H_{46}O_{10}]\) as soon as it comes in the contact with air, it imparts a milky appearance to the toad secretions.\(^{[27]}\) The toads protect itself from predators from the venoms which are actually the toxins secreted by it, it is also used to protect the toad from microbe-laden hostile habitat.\(^{[28]}\) The various physiological effects are seen on higher vertebrates as well as on lower vertebrates due to the action of the toad’s secretion. The effects when the granular secretion enter in the stomachs of higher vertebrates are nausea, weakening of respiration and muscular paralysis, they produce serious inflammation when comes in the contact with eyes.\(^{[29]}\) Toad secretion may also boast of adrenalin.\(^{[30]}\) Due to activity of bufogen and bufotalin the secretions of \(b\).\(marinus\) are cardioactive showing clinical symptoms like dermatitis, hypotension and severe arrhythmia,\(^{[31]}\) it is also been found to show second degree Wenckebach atrioventricular block and T-wave change.\(^{[32]}\) Other activities associated rather than heart, some secretions of toads have also been to found some neurological activities, the sleep inducing factors (SIF), which induce sleep probably by alteration of brain biogenic amine levels, monoamine oxidase (MAO) and tryptophan hydroxylase (TH) activity is due to the extraction of skin secretion of \(B.\)\(melanostictus\).\(^{[33]}\) The increase in the myocardial contractile force without affecting the heart rate is achieved by the bufotalin, which inhibits the potent \(Na^+K^+\) ATPase activity.\(^{[34]}\) Marinobufagenin is also a potent vasoconstrictor, resibufagenin shows electrophysiological properties similar to acetylstrophanthidin (AS), which suggests similarly to family of digitalis like drugs.\(^{[35]}\) Bufotenine blocks the action of serotonin and is also a indole-hallucinogen.\(^{[36]}\)

4. **Toad-Toxins Pharmacological and Therapeutic Significance**

The pharmacological uses of toads skin secretion was known since ancient times, as the ancient medical practitioners of some Indian and Chinese medical procedures used the toad’s secretion in many forms. The digoxin like effect is show by the traditional Chinese medicine “Kyushin”.\(^{[37]}\) “Chan’Su”, a Chinese term for a mixture of various toad-toxins specially, bufalin and cinobufagin, shows cardiotonic effect. The only problem arise is that the difference of the effect due to the quality and quantity of toxins used. It can also be hazardous if taken in overdose.\(^{[38]}\) The rubbing of toad on swollen areas of animal (cattle) throat for healing and removal of throat obstruction arising due to swelling is an ancient practice used in India.\(^{[39]}\) Toad skin secretions are source of diverse kind of pharmacologically and therapeutically significant compounds. These secretions contain components that can be used for production of painkillers, antimicrobial drugs, anti-viral drugs and even anti-cancerous
drugs. These compounds could also prove to be useful in treatment of cardiovascular ailments and neurological problems. Toad secretions have been shown to contain components, which exhibit analgesic and pain killing effects. There are various kind of pharmacologically and therapeutically significant compounds, these compounds contains the components which can be used for the production of pain killers, antimicrobials drugs, anti-viral drugs and also anti-cancerous drugs. Toad secretion also shows the effects which can be used in the treatment of cardiovascular and neurological problems. The analgesic effect of the Bufalin is used in the hepatic-cancerous pain; the bufalin is a toxin, which is Na+/K+ ATPase inhibitor. The experimental result of bufalin action is explained as it increases the hepatic-blood circulation, which decreases the amount of stagnating blood and hence reducing the pain. The drugs now days used commonly and frequently as pain killers are aspirin and morphine. But aspirin shows some side effects, such as bleeding in stomach, Reye’s disease, urticaria and asthmatic attacks. Even morphine shows some ill-effects, in this cases, the substitution of the aspirin and morphine can be made by bufalin, which is a potent pain relief drug, it also diminishes the anxiety and induces sleep which means bufalin is also sleep inducing agent with analgesic effect so for some extent it can be used as general anaesthetics. The substitution of aspirin can be made by bufalin in the treatment of arthritis and the anti-cancerous properties have been recently exhibited by a number of bufadienolides. The bufalin shows its anticancerous property on leukemia cells-lines. It was found to induce apoptosis in human leukemia cells by altering the apoptotic genes c-myc, and bcl-2. The resemblance of topoisomerase inhibitors is shown by the bufalin on cell cycle of leukemia cells. The concentration about which bufalin induced apoptosis in HL-60 cells was 10-8M, which is comparable to drug camptothecin but lower than the anti-tumor drugs like cisplatin, lower than all transretinoic acid. The treatment of action and granulocytopenia during combined chemotherapy is shown by another bufadienolid, namely Cinobufagin. The risk of infection and degree as well as duration of granulocytopenia associated with malignant blood disease was reduced by it. Other than bufalin and cinobufagin certain other toad skin secretion components like 3-formyloxyresibufogenin, 19-oxobufalin, 19-oxodesactylcinobufagin, 6-hydroxycinobufagin, and 1-hydroxycinobufalin have been recently found to exert inhibitory effects on KB, HL-60 and MH-60 cancer cell lines. The skin secretions of Bufo bufo gargarizans like the derivatives of bufogenin and also some synthetic derivatives of 20, 21-epoxy-resibufogenin have been found to exhibit interleukin-6 (IL-6) antagonistic activity due to their growth inhibitory activities on IL-6 dependent MH-60 cells. However, anti-cancerous properties of toad skin secretions can be debatable especially those on skin cancers because while they
seem to inhibit cancer proliferation in vitro, toad themselves have been reported to be affected by skin cancers. This raises doubts on anti-cancerous activity of these secretions in vivo.\[47\]

\[\text{Cinobufagin.} \quad \text{Bufalin.}\]

5. CONCLUSION
The can toad was introduced chiefly to control the pest i.e. cane beetle in Caribbean islands, and Pacific but it failed to do so and itself became an invasive pest. But the cane toads secretion which it secrets for its prevention is found to be useful in many ways. The cane toad secretes the venoms from the granular glands and mucous glands, venom of cane toad i.e. its secretion contains steroids, peptides like proteins, bufadienolides and biogenic amines. Its secretion is been used since ancient days in Asian countries, China, the major preparation Chan Su’ an ancient Chinese mixture of bufalin and cinobufagin, shows cardiotonic effects. The secretions of cane toad nearly contain about 86 types of bufadienolides. It is useful in various ways, for the pregnancy testing, as a hair restorer, it increases the myocardial contractile force without affecting the heart rate, used in cardiovascular ailments and neurological problems, it shows atrioventricular block and it is also used as pain killers, antiviral drugs, anti-microbial drugs and anti-cancerous drug. It is a potent Na\(^{+}/\)K\(^{+}\) ATPase inhibitor.

Conflict of interest
We declare that we have no conflict of interest.

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