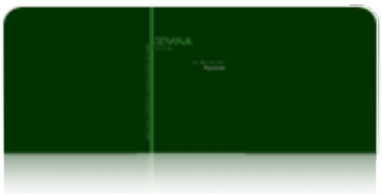

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Ethanol extract of *Nandina domestica* Thunb. leafs: effect on *Pomacea canaliculata* and growth of *Orzya sativa* seedlings

Extrato etanolico de folhas de *Nandina domestica* Thunb.: efeitos sobre *Pomacea canaliculata* e crescimento de mudas *Orzya sativa*

Hu Ye[1]; Liu Xuan[2]; Ye Meng[3]; Nong Xiang[4]*; Yang Yaojun[4]; Hu Qiang[5]; Huang Lanying[6]; Huang Ying[6].

Abstract

Alien species may adversely affect not only human health, agriculture, and fisheries but also the native ecosystem. *Pomacea canaliculata* is one of alien harmful species (IUCN). Forest residues are a kind of renewable, cheap and secure source of primary energy. *Nandina domestica* Thunb. is a commonly and widely cultivated plant for city landscape construction in China. After being pruned, a large of *N. domestic*a leaves are usually abandoned. In this work, extracts of ethanol Soxhlet (SE), ethanol immerse (EE), and water immerse (WE) were obtained from leaves of *N. domestica* Thunb. cv. Firepower. The molluscicidal effects of the three extracts from the leaves were evaluated against black *P. canaliculata*. In sand pot culture condition and pot culture with paddy soil condition, SE at concentration of 4000 mg L^{-1} did not affect seedlings normal growth. Plant height, seedling fresh weight (SFW), seedling dry weight (SDW) and Chlorophyll content of SE (4000 mg L^{-1}) were not significantly different from that in DS (distilled water + seedlings). Besides, in pot culture with paddy soil condition, the SE treatment at concentration of 4000 mg L^{-1} inhibited *P. canaliculata* chewing rice seedlings effectively, which was not statistically different from the positive control {NSP, niclosamide (according to field dosage, 0.086 g m^{-2}) + seedlings + *P. canaliculata*)}. Results from this study indicated that SE has molluscicidal effect against *P. canaliculata* and SE did not affect the normal growth of rice seedlings. Thus, we suggest further chemical and toxicological studies of SE.

Key words: Plant extracts. Molluscicidal effect. Vegetative parameters

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Resumo

As espécies exóticas podem afetar negativamente não apenas a saúde humana, a agricultura e a pesca, mas também o ecossistema nativo. *Pomacea canaliculata* é uma das espécies exóticas prejudiciais (UICN). Os resíduos florestais são um tipo de fonte renovável, barata e segura de energia primária. *Nandina domestica* Thunb. é uma planta comumente cultivada em paisagens urbanas na China. Depois de ser podada, uma grande quantidade de folhas de *N. domestica* são geralmente abandonadas. Neste trabalho, extratos, Soxhlet de etanol (SE), folhas mergulhadas em etanol (EE) e imersas em água (WE) foram obtidas a partir de folhas de *N. domestica* Thunb. cv. Firepower. Os efeitos moluscicidas dos três extratos das folhas foram avaliados contra *P. canaliculata* preta em cultivo de arroz em potes de areia e cultivo em potes com solo compactação, SE foi utilizado na concentração de 4000 mg L^{-1} e não afetou o crescimento normal das mudas. A altura da planta, (o peso fresco das mudas) SFW, (peso seco das mudas) SDW e o teor de clorofila do tratamento SE (4000 mg L^{-1}) não foram significativamente diferentes dos observados no tratamento controle DS (água destilada + mudas). Além disso, no cultivo de arroz em pote na condição de compactação, o tratamento SE na concentração de 4000 mg L^{-1} inibiu a alimentação de *P. canaliculata* em mudas de arroz, e não foi estatisticamente diferente do controle positivo {NSP, niclosamida (de acordo com a dosagem de campo, 0.086g m^2) + mudas + *P. canaliculata*)}. Os resultados deste estudo indicaram que SE tem efeito moluscicida contra *P. canaliculata* e não afetou o crescimento normal das mudas de arroz. Assim, sugerimos outros estudos químicos e toxicológicos da SE.

Palavras-chave: Extratos de plantas. Efeito moluscicida. Parâmetros vegetativos.

Introduction

Alien species may adversely affect not only human health, agriculture, and fisheries but also the native ecosystem (MAEZONO; MIYASHITA, 2003). *Pomacea canaliculata* (Mesogasropoda: Pilidae) is one of alien harmful species in Red List of International Union for Conservation of Nature (IUCN). *P. canaliculata*, the golden apple snail, is a large freshwater snail native to tropical and subtropical South America. Since the 1980s, this snail has become a serious cash crops pest in most Southeast, East Asian and North America countries, because the snail feeds on tender leaves of cash crops (DING et al., 2011; HARLWRT, 1994; NAYLOR, 1996; WADA et al., 1999). Chewing crops' leaves results in dead seedlings, inhibiting *tiller* and decrease of yield (COWIE, 2002; JOSHI, 2005). Meanwhile, this snail damages ecological environment (WANG et al., 2009). Moreover, *Angiostrongylus cantonensis* and *Echinostoma revolutum* use *Pomacea canaliculata* as hosts. *A. cantonensis* and *E. revolutum* are the most common causes of eosinophilic meningitis worldwide (DONG, 2006). The overuse of chemical molluscicide is a worldwide issue. Niclosamide is the only recommended molluscicide by World Healthy Organization (WHO). However, niclosamide is with low toxicity to fishes. Chemical molluscicides are serious threats to green agricultural production (WANG, 2003). Anthelmintic resistance and environmental protection have stimulated the search for new alternatives of treatment, including the use of medicinal plants (JACKSON; MILLER, 2006).

Forest residues are a kind of renewable, cheap and secure source of primary energy. Forestry residues provide many functions. Those include acting as bio-energy, biological agents, as well as habitats for organisms (LIU et al., 2012; MALINEN; PESONEN, 2001). In China, annual output of forestry residues is 16 billion kilograms (LIU et al., 2017). China is a developing country so re-utilization of forestry residues is not only for protecting plant resource, but also for improving environment effectively. *Nandina domestica* Thunb.(Berberidaceae) is a broad-leaf evergreen shrub which widely distributed and cultivated in Yangtze river basin, India and Japan (EDITORIAL COMMITTEE OF CHINESE ACADEMY OF SCIENCES PLANT, 1979). It

has ornamental, ecological and medicinal value. With the development of urbanization, *N. domestica* is a commonly and widely cultivated plant for city landscape construction in China. After being pruned, a large of *N. domestica* leaves are usually abandoned. Thus, it not only causes environmental pollution, but also causes waste of *N. domestica* resource. Some chemical components of leaves are identified, including magnoline, amentoflavone, nantenoside A, nantenoside and hydrogen cyanide (LIU, 2004). The color of the leaves are dark green. Growing in strong light, autumn or winter, the color of the young leaves are red. The contents of anthocyanins and soluble sugar in red leaves were higher than those in green leaves while green leaves had a higher content of chlorophyll (WEN et al., 2005). The leaves are bitter taste, sexual cold and a mild toxicity. Due to their functions in dispelling pathogenic wind, removing dampness and astringing lung to stop cough, the leaves are used for treating cough, asthma, malaria, jaundice with damp-heat pathogen and other diseases (EDITORIAL BOARD OF CHINESE MATERIAL MEDICAL OF STATE ADMINISTRATION OF TRADITIONAL CHINESE MEDICINE, 2015). Recently, methanol, ethanol, acetone and ethyl acetate extracts of *N. domestica* leaves and fruits have varying degrees of antimicrobial activities against *Rhizoctonia solani*, *Alternaria solani*, *Alternaria porri*, *Fusarium oxysporum f. sp. vasinfectum*, *Coniothyrium diplodiella* and *Magnapothe grisea* (LIU et al., 2004). Besides, organic extracts (hexane, chloroform, ethyl acetate and methanol) of *N. domestica* leaves can control food-borne pathogenic and spoilage bacteria, such as *Listeria monocytogenes* ATCC8739, *Bacillus subtilis* ATCC6633, *Salmonella typhimurium* KCTC2515, *Salmonella enteridis* KCCM12021, *Enterobacter aerognes* KCTC2190, *Escherichia coli* 0157-Human, *E. coli* 057: H7 ATCC43888 and *E. coli* ATCC8739 (BAIPAI et al., 2008).

Therefore, the aim of this study was to evaluate molluscicidal effects of extracts from *N. domestica* leaves. Meanwhile, vegetative parameters of *O. sativa* seedlings were measured. Besides, the chemical components of the extracts were analyzed by GC-MS. The results of molluscicidal experiments offered new promising scenario in utilizing forest residues for the biological control of *P. canaliculata*.

Materials and Methods

Plant material

Leaves of *N. domestica*, which have been collected from artificial cultivated adult plants in Leshan Municipality, Sichuan province {Latitude (LAT): 29°, 56′ N, longitude (LONG):103°, 74′ E, altitude (ALT): 372 meters} in December, 2016. The sample was identified as a single and ripe leaf of *Nandina domestica* Thunb. cv. Firepower by Professor Hu Chao. Prior to experiments, the original materials were dried naturally. before they were analyzed, the original materials were crushed and pulverized to a size of <0.25 mm.

Firstly, approximately 200 grams of *N. domestica* leaves were crushed and soaked for a week in water. The solvent was dried by distillation to obtain water extract (WE). Secondly, approximately 200 grams of samples were crushed and soaked for a week in ethanol at normal temperature. The solvent was evaporated using a rotary evaporator to obtain ethanol extract (EE). Thirdly, approximately 200 grams of samples were extracted for 6 hour using a Soxhlet extractor. Next add the ethanol (95%). The solvent was evaporated using a rotary evaporator to obtain extract with Soxhlet extractor (SE). The three extracts were stored in tightly closed dark vials at 4º C until use.

Animal

Healthy yellow *Pomacea canaliculata* samples, which lived in a farmland of Long-dang Village, Xindian Town, the Zigong City, Sichuan province (LAT: 29°, 23′ N, LONG: 104°, 55′ E, ALT: 415

meters) and were obtained in March, 2017. The height of a shell was from 30 mm to 40 mm.

Healthy black *P. canaliculata* samples, which lived in a wetland of Leshan Municipality, Sichuan province (LAT: 29°, 56′ N, LONG: 103°, 74′ E, ALT: 372 meters) and were obtained in July, 2017. The black *P. canaliculata* samples had 2-3 spirals.

All samples were identified by methods of Sun et al. (2011). Methods for feeding the animals were in the ways of Zhao et al. (2014).

Tests

Molluscicidal test was performed according to the methods of the WHO guidelines for laboratory molluscicidal test. The three kinds of extracts were dissolved in propanetriol with distilled water (volume ratio,1:1) and formulated as solutions at concentrations of 800, 1000, 1200, 1400, 1800, 2000 mg L^{-1}. Per 700 ml solution was added into 1000 ml white plastic container. Ten *P. canaliculata* samples were put in a container. Niclosamide (20 mg L^{-1}) and distilled water with propanetriol (volume ratio, 1:1) were set as positive control and negative control, respectively. Five times of repeats were treated at each treatment. All samples were submerged. Gauze was taped over the mouth of a container. According to the methods of Zhang et al. (2012) for judging the deaths of *P. canaliculata* samples, the number of deaths were counted at 1 -5d (HU et al., 2009), respectively. The test temperature varied from 20 to 25ºC.

In sand pot culture condition, effect of SE on rice seedling growth was performed by Hu et al. (2009). Plot trial was using a randomized block design with five replicates. Each pot had ten rice seedlings (2-3 leaf stage). Seedlings were growing in the solution (SE) at concentrations of 1000, 2000, 4000 mg L^{-1}. Distilled water was set as the control group. Every plot were put in light incubator with daily illumination 12 hours, light intensity 12000 lx{light intensity unit, 1 lx equals 1 flow (lumen, LM) in the area of 1 square meters of illuminance distribution on the flux uniformity} and temperature 25±1 °C. Following vegetative parameters were measured after seven days: seedling height, seedling fresh weight (SFW), seedling dry weight (SDW) and Chlorophyll (HU et al., 2009). SDW was determined gravimetrically after drying to 60^{o}C.

In pot culture with paddy soil conditions, effect of SE on rice seedling growth and black *P. canaliculata* was performed by Hu et al. (2009). Plot trial was using a randomized block design with five replicates. Each pot had ten rice seedlings (2-3 leaf stage) and 5 black *P. canaliculata* samples (2-3 spirals). seedlings were growing in the solution (SE) at concentrations of 1000, 2000, 4000 mg L^{-1}. DS (Distilled water + seedlings), DSP (Distilled water + seedlings + *P. canaliculata*), and NSP{niclosamide (according to field dosage, 0.086 g m^{-2}) + seedlings + *P. canaliculata*}were set as control group. Every plot were put in light incubator with daily illumination 12 hours, light intensity of 12000 lx and temperature 25±1 °C. The number of rice seedlings were counted per 12 hours. Following vegetative parameters were measured after seven days: seedling height, seedling fresh weight (SFW), seedling dry weight (SDW) and Chlorophyll. Seedling dry weight was determined gravimetrically after drying at 60^{o}C.

Analysis

The crude extracts of leaves were analyzed using a Agilent 7890B Gas Chromatography (GC) apparatus equipped with a Agilent 5977, a mass selective detector, and a HP-5 Mass Spectrometer (MS) capillary column (30 m × 0.25 mm; film thickness, 0.25 μm). The contents of components were calculated with peak area normalization method.

One-way analysis of variance (ANOVA) and Duncan's multiple range tests were used to analyze tests' data. The results were expressed as a mean percentage ± standard deviation, and differences

were considered significant when $p < 0.05$. Probit method (SPSS 19.0 for Windows) was used to calculate the mean lethal concentration (LC_{50}).

Results and Discussion

The Molluscicidal effects of different extracts from leaf of N. domestica against P. canaliculata

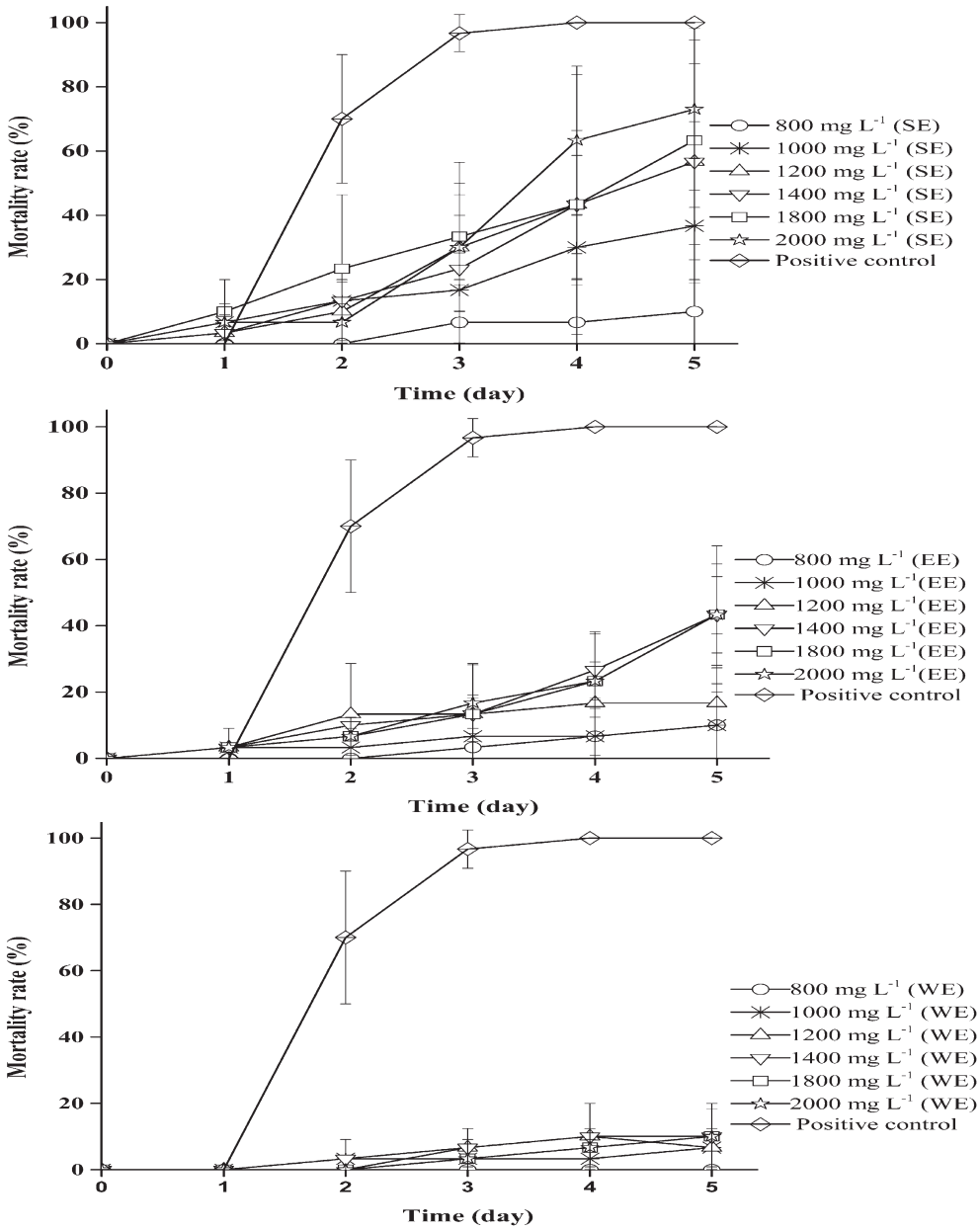
In laboratory immersion test, the mortality ratios of crude extracts from *N. domestica* leaf against *P. canaliculata* were presented in Table 1. The results showed that SE and EE had molluscicidal effects on *P. canaliculata*. Of all treatments, the mortality ratio of *P. canaliculata* was 73% at the concentration of 2000 mg L^{-1} SE, which was no significantly different from the positive control ($p<0.05$). At the concentration of 1200, 1400 and 1800 mg L^{-1} SE, the mortality ratios were 56.67%, 56.67% and 63.33%, respectively. LC_{50} is an important parameter to measure toxicity. The molluscicidal effect of SE was dose-dependent, demonstrating an LC_{50} of 1339.354 mg L^{-1}. The mortality ratio was 43.33 % at the concentration of 1000 mg L^{-1} EE, which was significantly different from the positive control and not significantly different from 1000 mg L^{-1} SE. Treatment with both 1000 mg L^{-1} EE and 2000 mg L^{-1} WE had not molluscicidal effect. Besides, mortality rates increased slowly with the concentration of those extracts solutions (Figure 1). The mortality ratio reached over 50% at the concentration of 2000 mg L^{-1} SE. The mortality ratios reached over 50% at the concentrations of 1200, 1400, 1800 and 2000 mg L^{-1} (SE) after fifth day.

Table 1. Mean efficacy (percentage±SD) of *N. domestica* leaves extracts on the molluscicidal effect of *P. canaliculata* after 5d in laboratory immersion test.

concentration(mg L^{-1})	Extracts(%)		
	Soxhlet	Ethanol immersion	Water immersion
100	$3.33±5.77^{eA}$	$3.33±5.77^{cA}$	$0.00±0.00^{cA}$
200	$6.67±5.77^{eA}$	$6.67±5.57^{cA}$	$0.00±0.00^{cA}$
400	$3.33±5.77^{eA}$	$6.67±5.77^{cA}$	$0.00±0.00^{cA}$
800	$10.00±10.00^{deA}$	$10.00±10.00^{cA}$	$6.67±5.77^{bcA}$
1000	$36.67±5.77^{cdA}$	$10.00±17.32^{cB}$	$6.67±11.55^{bcB}$
1200	$56.67±30.55^{bcA}$	$16.67±20.82^{cAB}$	$10.00±10.00^{bB}$
1400	$56.67±37.86^{bcA}$	$43.33±15.28^{bA}$	$10.00±0.00^{bA}$
1800	$63.33±5.77^{bcA}$	$43.33±20.82^{bA}$	$10.00±0.00^{bB}$
2000	$73.00±25.17^{abA}$	$43.33±11.55^{bA}$	$10.00±0.00^{bB}$
Propanetriol with distilled water (volume ratio, 1:1)	$0.00±0.00^{eA}$	$0.00±0.00^{cA}$	$0.00±0.00^{cA}$
niclosamide (20mg L^{-1})	$100.00±0.00^{aA}$	$100.00±0.00^{aA}$	$100.00±0.00^{aA}$

Small letters compare mean between lines and capital letters between columns ($p < 0.05$).

Figure 1. Mortality rates of yellow *Pomacea canaliculata* over the days after 20 mg L^{-1} niclosamide (positive control) and aqueous dilutions (50, 100, 200, 400, 800, 1000 mg L^{-1}) of three extracts from *Nandina domestica Thunb*.cv. Firepower leaves were immersed. The three kinds of extracts included Soxhlet extract (SE), ethanol immersion extract (EE), and water immersion extract (WE).
Error bars: Data from replicates of each concentration (10 yellow *P. canaliculata* samples per replicate)



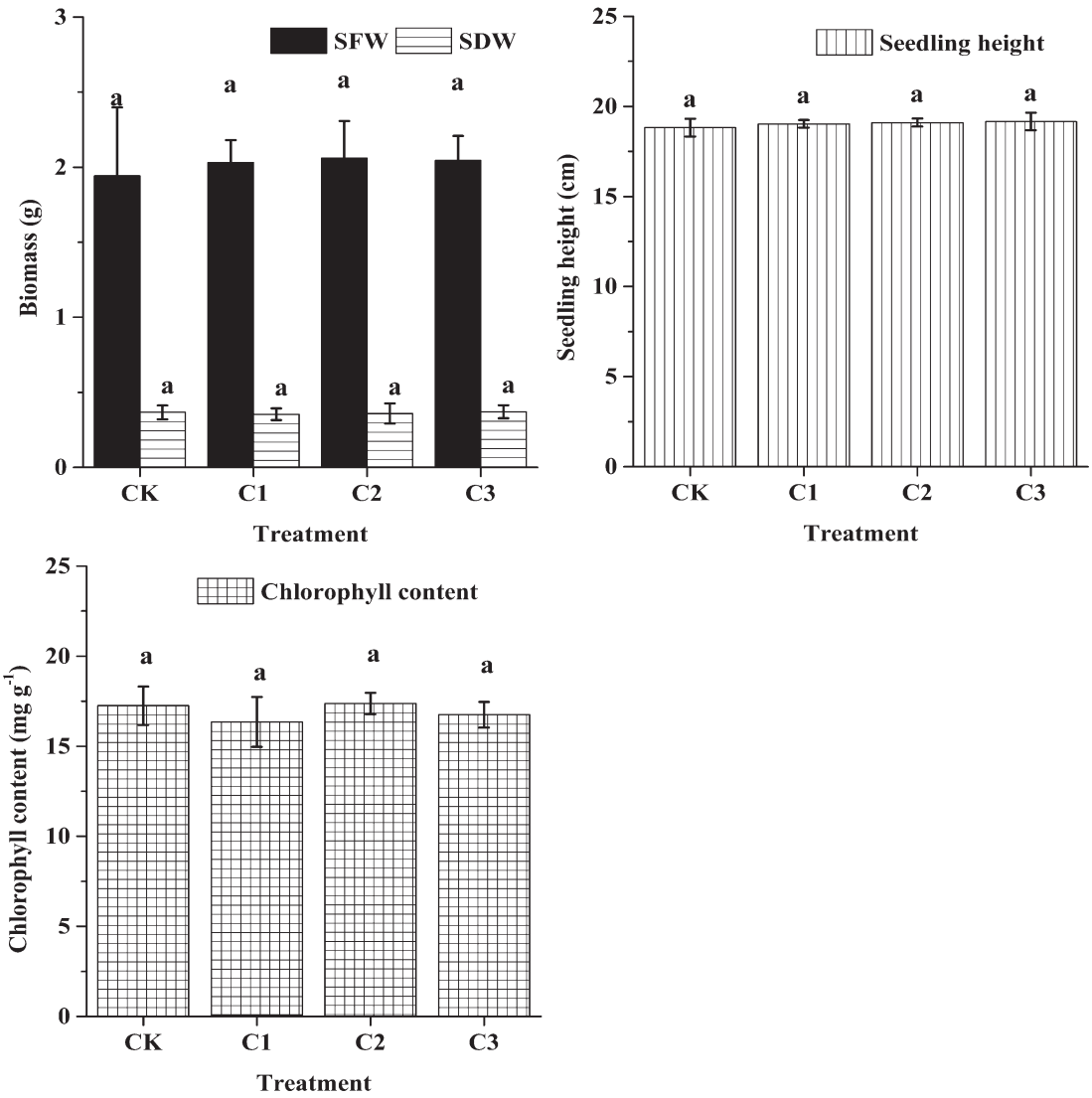
According to table 1, in laboratory immersion test, the mortality ratio of *P. canaliculata* was 73% at the concentration of 2000 mg L^{-1} SE. In pot culture with paddy soil condition, we observed that the mortality ratio of *P. canaliculata* was only 20% at the concentration of 2000mg L^{-1} SE. In other word, in the pot culture with paddy soil condition, the mortality ratio of *P. canaliculata* was decreased obviously. Hu et al. (2009) inferred there were two factors: one factor was that soils had certainly absorption effect on molluscicidal components; the other one was that *P. canaliculata*

closed operculum rapidly, stopped moving and tried to chew little food. The second factor must be paid more attentions due to increasing of resistance. In this experiment, four behavioral responses could be observed respectively: open operculum, climb the wall, close opereulum, and dead. Based on field investigation of cultivating rice, in order to effectively kill *P. canaliculata*, the actual amount of pesticides were 3-5 times the field dosage.

SE effects on the growth of O. sativa seedlings and *P. canaliculata*

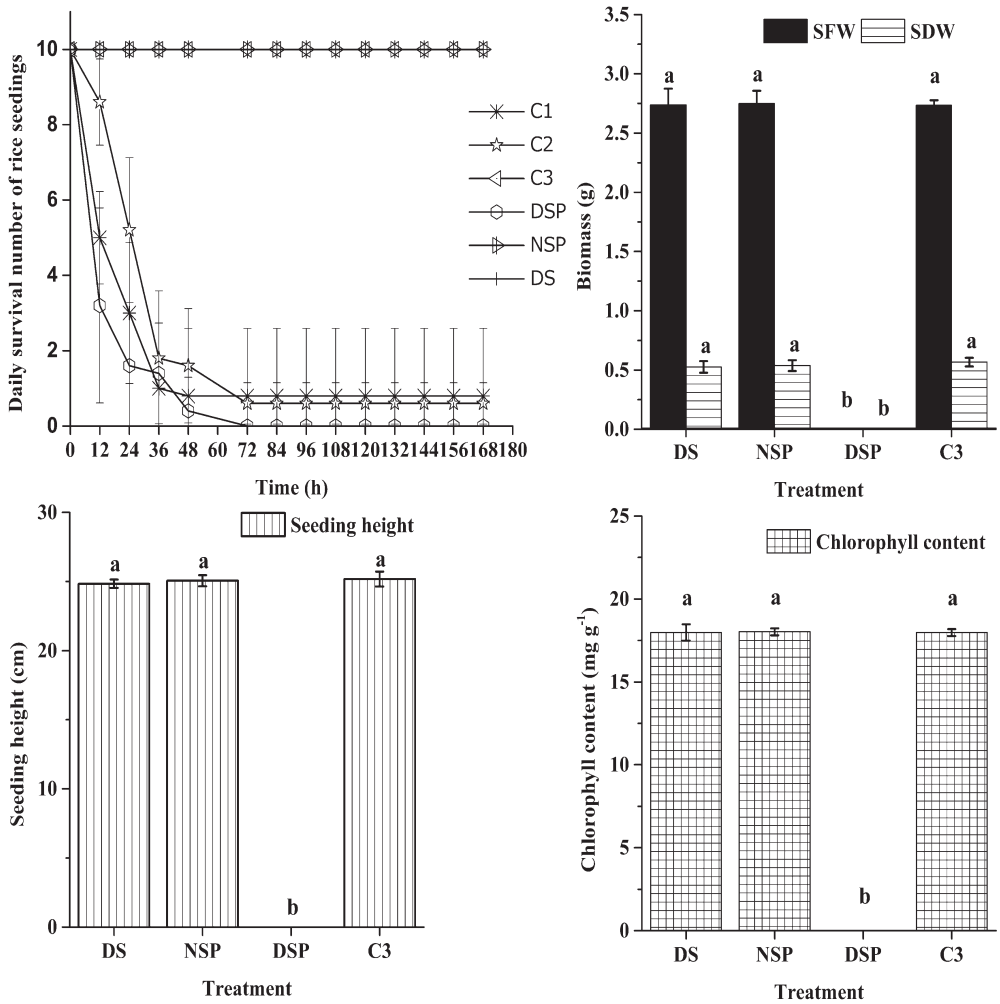
According to figure 2, in sand pot culture condition, the values of seedling height, SFW, and SDW of all treatments were higher than control group (distilled water). The Chlorophyll content of all treatments were not significantly different from that in control group (distilled water).

Figure 2. In sand pot culture condition, the values of seedling height, SFW, SDW, Chlorophyll content of all treatments: CK (Distilled water), C1 (1000 mg L^{-1} SE), C2 (2000 mg L^{-1} SE), C3 (4000 mg L^{-1} SE).SFW: seedling fresh weight; SDW: seedling dry weight; SE: extract with Soxhlet extractor from *Nandina domestica Thunb*.cv. Firepower leaves Error bars: Data from replicates of each concentration (10 seedlings per replicate)



According to figure 3, in pot culture with paddy soil conditions, the SE at concentration of 4000 mg L^{-1} inhibited *P. canaliculata* chewing rice seedlings effectively, which was not statistically different from the positive control {NSP, niclosamide (according to field dosage, 0.086 g m^{-2}) + seedlings + *P. canaliculata*)}. At 60h, *P. canaliculata* ate up all rice seedlings in the condition of DSP (Distilled water + seedlings + *P. canaliculata*). Moreover, at 12h, SE at concentration of 2000 mg L^{-1} inhibited *P. canaliculata* effectively. After 12h, food intake was increased. Furthermore, after 7d, the death ratio of *P. canaliculata* was 96% at the concentration of 4000 mg L^{-1}SE. Besides, plant height, SFW, SDW and Chlorophyll content of SE (4000 mg L^{-1}) were not significantly different from that in DS (distilled water + seedlings). In previous studies, *N. domestica* leaf had allelopathic inhibition on some plants, such as wheat (HAN; LUO, 2011). In pot culture with paddy soil conditions, SE at concentration of 4000 mg L^{-1} did not affect seedlings normal growth.

Figure 3. In pot culture with paddy soil conditions, the values of daily survival number of rice seedlings, seedling height, SFW, SDW, Chlorophyll content of all treatments. DS: Distilled water + seedlings; DSP : Distilled water + seedlings +*P. canaliculata*; NSP: niclosamide (according to field dosage, 0.086 g m^{-2}) + seedlings + *P. canaliculata*}; C3: 4000 mg L^{-1}SE; SFW: seedling fresh weight; SDW: seedling dry weight; SE: extract with Soxhlet extractor from *Nandina domestica Thunb*.cv. Firepower leaves.
Error bars: Data from replicates of each treatment (10 seedlings per replicate)



In order to simulate demonstration of efficacy under field-like conditions, this paper adopt pot culture with paddy soil conditions. In pot culture with paddy soil conditions, SE at concentration of 4000 mg L^{-1} did not affect seedlings normal growth (Figure 3). Meanwhile, the SE at concentration of 4000 mg L^{-1} inhibited *P. canaliculata* chewing rice seedlings effectively, which was not statistically different from the positive control {NSP, niclosamide (according to field dosage, 0.086 g m^{-2}) + seedlings + *P. canaliculata*)}(Figure 3). Consequently, this indicated that ethanol extract of *N. domestica* leaf could be used for controlling *P. canaliculata* in paddy fields. It is known that *N. domestica is* widely distributed and cultivated in Yangtze river basin, India and Japan (EDITORIAL COMMITTEE OF CHINESE ACADEMY OF SCIENCES PLANT, 1979). In city landscape construction of China, a large amount of *N. domestica* leaves are usually pruned and abandoned. Thus, a large of *N. domestica* leaves are easily to harvest. Meanwhile, in this paper, SE was extract with Soxhlet method. The method is simple. Moreover, Chinese government has highly growing policy supports on utilizing agroforestry residues, protecting the environment and developing biological pesticides. "2017 China International agroforestry waste energy utilization technology and Development Summit Forum" focused on utilization technology of agroforestry residues. Recently, 322 issues that had been supervised by the Ministry of environmental protection of People's Republic of China since 2017. In Sichuan province, Companies invest 442 million RMB for the production of biological pesticides (ZHANG, 2017). It was feasible that *N. domestica* leaves might be as a kind of plant materials for molluscicides. SE was the most promising extract.

Main toxic compositions of the three kinds of paste by GC-MS analysis

The main toxic compositions of the three kinds of paste were analyzed by GC-MS. The results were listed in Table 2. The paste (SE) contained some kinds of high and moderate toxic compositions. The paste (SE) was characterized by high quantities of high toxic compositions (3.07 %), with furfural (3.03 %) and 2-Furanmethanol (0.04 %) as the constituents. The paste (SE) was characterized by moderate toxic compositions (0.42 %), with Benzyl alcohol (0.32 %) and Phenylethyl alcohol (0.10 %) as the constituents. Moreover, we identify some kinds of high and moderate toxic compositions of the past (EE). The paste (SE) was characterized by high quantities of high toxic compositions (3.07 %), with furfural (2.24 %) and 2-Furanmethanol (0.03 %) as the constituents. The paste (EE) was characterized by moderate toxic compositions (0.36 %), with Benzyl alcohol (0.28 %) and Phenylethyl alcohol (0.08 %) as the constituents. Furthermore, the paste (WE) contained a kind of moderate composition (Benzofuran, 0.18 %) and a kind of low toxic composition (1,3- Propanediol, 0.23 %).

In molluscicidal test, the effect of SE was obviously better than that of EE and WE. The results of CG-MS analysis showed the three extracts had different types and contents of toxic components (Table 2). The total content of high toxic component in SE (3.07 %) was higher than that in EE (2.27 %). The total content of moderate component in SE (0.42 %) was higher than that in EE (0.36 %) and WE (0.18 %). Therefore, we inferred that the molluscicidal effect of the extract might be related to the types and contents of toxic components. In fact, Kong et al. (1998) and Hu et al. (2009) showed that plants could inhibit harmful organisms due to the combined action of their chemical compounds, such as *Ageratum conyzoides* (L.) and *Ipomoea cairica* (L.). Consequently, we inferred that the combined action of chemical compounds might be one of the essential differences between chemical molluscicides and botanical molluscicides. In this paper, the contents of components were calculated with peak area normalization method. The components of crud extracts were identified by comparing their GC retention indices (RI), NIST14

mass spectral search program (National Institute of Standards and Technology), the content (greater than or equal to 0.01 %), and match factor (greater than 90 %). Then, the next work was that the toxic compounds of SE would be confirmed by standard compounds further.

Table 2. Analysis of toxic compositions of the crude extract (GC-MS).

Extract	Toxic components, content, %	RI	Match factor,%	Experimental data of Chemical Book – Chemical Search Engine
SE	Furfural (3.03%)†††	833	99.43%	Rat, take orally, medial lethal dose 50 (LD50): 65 mg/kg. Mice, take orally, LD50: 400 mg/kg. Rabbit, moderate irritation to eyes: 100 mg/24h, moderate irritation to skin: 20 mg/24h.
	2-Furanmethanol (0.04%)†††	859	94.49%	Rat, take orally, medial lethal dose 50 (LD50): 177 mg/kg. Mice, take orally, LD50: 160 mg/kg. Rabbit, moderate irritation to eyes: 100 mg/24h.
	Benzyl alcohol (0.32%)††	1036	96.56%	Rat, take orally, LD50: 1230 mg/kg. Mice, take orally, LD50: 1360 mg/kg. Rabbit, severe irritation to eyes: 0.75 mg, mild irritation to skin: 10 mg/24h.
	Phenylethyl alcohol (0.10%)††	1116	96.23%	Rat, take orally, LD50: 1790 mg/kg. Rabbit, severe irritation to eyes: 0.75 mg/24h, mild irritation to skin: 100 mg/24h
EE	Furfural (2.24%)†††	833	99.67%	Rat, take orally, medial lethal dose 50 (LD50): 65 mg/kg. Mice, take orally, LD50: 400 mg/kg. Rabbit, moderate irritation to eyes: 100 mg/24h, moderate irritation to skin: 20 mg/24h.
	2-Furanmethanol (0.03%)†††	859	93.52%	Rat, take orally, LD50: 177 mg/kg. Mice, take orally, LD50: 160 mg/kg. Rabbit, moderate irritation to eyes: 100 mg/24h.
	Benzyl alcohol (0.28%)††	1036	97.43%	Rat, take orally, LD50: 1230 mg/kg. Mice, take orally, LD50: 1360 mg/kg. Rabbit, severe irritation to eyes: 0.75 mg, mild irritation to skin: 10 mg/24h.
	Phenylethyl Alcohol (0.08%)††	1116	94.81%	Rat, take orally, LD50: 1790 mg/kg. Rabbit, severe irritation to eyes: 0.75 mg/24h, mild irritation to skin: 100 mg/24h.
WE	1,3-Propanediol (0.23%)†	781	92.18%	Rat, take orally, LD50: 10000 mg/kg.Mice, take orally, LD50: 4773 mg/kg
	Benzofuran (0.18%)††	1004	95.02%	Mice, intraperitoneal injection, LD50: 500 mg/kg

The contents of components were calculated with peak area normalization method. RI: Retention Indices. SE: extract with Soxhlet extractor from *N. domestica* leaves; EE: ethanol extract from *N. domestica* leaves; WE: water extract from *N. domestica* leaves;††† means high toxicity; †† means moderate toxicity; † means low toxicity.
Reference: Chemical registration center of the Ministry of environmental protection of China - CRCMEP.

Conclusion

All together these results indicate that SE from *N. domestica* leaves is toxic to *P. canaliculata*. Thus, we suggest further chemical and toxicological studies of SE.

References

BAIPAI, V. K.; RAHMAN, A.; KANG, S. C. Chemical composition and inhibitory parameters of essential oil and extracts of *Nandina domestica* Thunb. to control foodborne pathogenic and spoilage bacteria. *International Journal of Food Microbiology*, Torino, v. 125, n. 2, p. 117-122, 2008.

CHEMICAL REGISTRATION CENTER OF THE MINISTRY OF ENVIRONMENTAL PROTECTION OF CHINA - CRCMEP. CHINESE EXISTING CHEMICAL INVENTORY – IECSC. Beijing, 2013. Available at: <http://cciss.cirs-group.com/more-info/>. Access at: 4 apr. 2018.

COWIE, R. H. Apple snails (Ampullariidae) as agricultural pests: their biology, impacts and management. In: BARKER, G. M. (Ed.). *Molluscs as crop pests*. Wallingford: CAB International, 2002. p. 145-192.

DING, H.; XU, H. G.; QIANG, S.; MENG, L.; HAN, Z. M.; MIAO, J. L.; HU, B. S.; SUN, H. Y.; HUANG, C.; LEI, J. C.; LE, Z. F. Status and trends of biological invasion in China. *Journal of Ecology and Rural Environment*, Nanjing, v. 27, n. 3, p. 35-41, 2011.

DONG, C. L. *Studies on bioecology of the golden apple snail (Pomacea canaliculata) and Its status of distribution and damage in Guangxi.* 2006. Master (Thesis) - Guangxi Normal University, Guangxi.

EDITORIAL COMMITTEE OF CHINESE ACADEMY OF SCIENCES PLANT - ECCASP. Higher plants of Chinese (second volumes). Beijing: Science Press, 1979. 125 p.

EDITORIAL BOARD OF CHINESE MATERIAL MEDICAL OF STATE ADMINISTRATION OF TRADITIONAL CHINESE MEDICINE - EBCMMSATCM. Chinese material medic. Shanghai: Shanghai Science and Technology Art Press, 2015. 327 p.

HAN, J.; LUO, X. Y. *Study on herbicidal-active materials of Nandina domestica Thunb*. Qingdao: Qingdao Agricultural University, 2011. 85 p.

HARLWRT, M. The golden apple snail *Pomacea canaliculata* in Asian rice farming systems: present impact and future threat. *International Journal of Pest Management*, Abingdon, v. 40, n. 4, p. 199-206, 1994.

HU, F.; ZENG, K. Y.; ZHANG, J. Y.; CHEN, J. J.; TANG, X. R. Ethanol extract of *Ipomoea cairica* toxic effects on *Pomacea canaliculata* and its effect on the *Orzya sativa* seedling. *Acta Ecologica Sinica,* Beijing, v. 29, n. 10, p. 5471-5477, 2009.

JACKSON, F.; MILLER, J. Alternative approaches to control quo vadit? *Veterinary Parasitology*, Amsterdam, v. 139, n. 2, p. 371-384, 2006.

JOSHI, R. Managing invasive alien molluscan species in rice. *International Rice Research Notes*, Indianapolis, v. 30, n. 2, p. 5-12, 2005.

KONG, C. H.; XU, T.; HU, F. Study on interactions among allelochemicals of *Ageratum conyzoides*. *Acta Phytoecologica Sinica*, Beijing, v. 22, n. 5, p. 403-408, 1998.

LIU, J. C. Study on the utilization and exploitation of *Nandina domestica* Thunb. Resource. *Chinese Wild Plant Resource*, Nanjing, v. 23, n. 6, p. 22-23, 2004.

LIU, X.; HU, Y.; XIAO, Y. Risk management for rural energy industry of Sichuan Province in China. *Renewable and Sustainable Energy Reviews*, Washington, v. 69, n. 3, p. 1029-1044, 2017.

LIU, X.; YE, M.; PU, B.; TANG, Z. K. Risk management for *jatropha curcas* based biodiesel industry of Panzhihua Prefecture in Southwest China. *Renewable and Sustainable Energy Reviews*, Washington, v. 16, n. 3, p. 1721-1734, 2012.

MAEZONO, Y.; MIYASHITA, T. Community-level impacts induced by introduced large-mouth bass and bluegill in farm ponds in Japan. *Biological Conservation*, Barking, v. 109, n. 2, p. 111-121, 2003.

MALINEN, J.; PESONEN, M. Potential harvest for wood fuels (energy wood) from logging residues and first thinning in Southern Finland. *Biomass and Bioenergy*, Oxford, v. 20, n. 3, p. 189-196, 2001.

NAYLOR, R. Invasions in agriculture: assessing the cost of the golden apple snail in Asia. *Ambio*, Oslo, v. 25, n. 7, p. 443-448, 1996.

SUN, Z. J.; ZENG, Q. X.; ZHANG, B.; PENG D. J.; QU, Z. Y.; YANG, X.; ZHAN, X. M.; WU, Y. Comparison on main meteorologic characters for different varieties of *Pomacea canaliculata* in China. *Chinese Journal of Zoonoses*, Fuzhou, v. 27, n. 3, p. 184-189, 2011.

WADA, T.; ICHINOSE, K.; HIGUCHI, H. Effect of drainage on damage to direct-sown rice by the apple snail *Pomacea canaliculata* (Lamarck) (Gastropoda:Ampullariidae). *Applied Entomology and Zoology*, Tokyo, v. 34, n. 3, p. 365-370, 1999.

WANG, F. H.; HAN, J. Y.; ZHANG, F. *Study on biological invasion in China.* Beijing: Science Press, 2009. 316 p.

WANG, R. Tribune of snail control (1) - the history and current situation of the research on snail control drugs. *Chinese Journal of Schistosomiasis Prevention and Control*, Wuxi, v. 15, n. 6, p. 478-481, 2003.

WEN, G. Q.; HE, D. W.; HE, Z. Relationships between colors and some physiological and biochemical indexes of leaves from *Nandina domestica*. *Subtropical Plant Science*, Xiamen, v. 34, n. 4, p. 38-40, 2005.

ZHANG, X. L.; LI, F.; CHEN, B.; WANG, M. K. The molluscicidal activity of different extracts from *Jatropha curcas* L. against *Pomacea canaliculata. Journal of Anhui Agricultural Science*, Anhui, v. 40, n. 6, p. 3349-3350, 2012.

ZHANG, Y. B. *New development of pesticide in the world.* Beijing: Chemical Industry Press, 2017. 55 p.

ZHAO, L. L.; PU, S.; YANG, S.; YANG, L.; LIU, Q.; ZHAO, J. Q.; YAN, T. M.; ZHANG, J. E. A comparison study on immune function characterization of two shell colours of golden apple snail (*Pomacea canaliculata*). *Journal of Fisheries of China*, Shanghai, v. 38, n. 6, p. 896-903, 2014.