# **Biology and Control of Snail** Intermediate Host of Schistosoma japonicum in The People's Republic of China

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#### Abstract

Schistosomiasis caused by *Schistosoma japonicum* is a severe parasitic disease in The People's Republic of China and imposed considerable burden on human and domestic animal health and socioeconomic development. The significant achievement in schistosomiasis control has been made in last 60 years. *Oncomelania hupensis* as the only intermediate host of *S. japonicum* plays a key role in disease transmission. The habitat complexity of the snails challenges to effective control. In this review we share the experiences in control and research of *O. hupensis*.

# 1. INTRODUCTION

Schistosomiasis is a severe parasitic disease that imposes considerable burden on human and domestic animal health and socioeconomic development (Chen, 2014; Gryseels et al., 2006). Of the six Schistosoma species known to infect humans, namely Schistosoma japonicum, Schistosoma mansoni, Schistosoma haematobium, Schistosoma intercalatum, Schistosoma mekongi and Schistosoma malayensis, the former three are of primary importance. The feature and geographical distribution of intermediate snail host exhibits differences in line with different species of schistosomes (Mao, 1990; Zhou, 2005). Oncomelania hupensis is the only intermediate host of S. japonicum, distributed in The People's Republic of China, Japan, the Philippines and Indonesia (Attwood et al., 2015).

The geographical distribution of *O. hupensis* in The People's Republic of China coincides with the endemic areas of schistosomiasis in 12 southern provinces (municipalities, autonomous regions). A great achievement in schistosomiasis control has been made after national control programmes were implemented since mid-1950s (Collins et al., 2012; Xu et al., 2016). Snail control has played a key role in the achievements. According to national statistics, the total area of snail habitats in The People's Republic of China has decreased by 74.48%, from 14,321 km² in 1956 to 3654.68 km² in 2013. In this paper, we review the current status of *O. hupensis* research and control in The People's Republic of China.



# 2. BIOLOGY OF ONCOMELANIA SNAILS

# 2.1 Taxonomy

Oncomelania hupensis (Gredler, 1881) was first discovered in Hubei Province, The People's Republic of China in 1981 and not considered as the intermediate host of S. japonicum until 1913. The taxonomy of O. hupensis has been a subject of debate due to its wide distribution, complex habitat environment and variable morphological characters (Kang et al., 1958; Mao and Li, 1954). Oncomelania hupensis was usually classified into eight subspecies (Zhou, 2005) (Table 1). However, this classification was questioned by some authors. For example, subspecies (Zhou et al., 2009). A 2014 study showed that geographical distance contributes more to genetic divergence than environmental isolation, and among all variables that were

**Table 1** Geographical distribution of *Oncomelania* spp. in the World. **Subspecies Distribution region** 

Oncomelania (Gredler, 1881)	
O. minima (Bartsch, 1936)	Japan
O. hupensis (Gredler, 1881)	The People's Republic of China
O. h. hupensis (Gredler)	Middle and lower reach of Yangtze River (MLY)
O. h. hupensis fausti strain	Mountains and hills along MLY
O. h. hupensis hupensis strain	Marshlands and lakes along MLY
O. h. tangi (Bartsch)	Southeast coast areas in The People's Republic of China
O. h. hupensis tangi strain	Fujian
O. h. hupensis guangxi strain	Guangxi
O. h. hupensis subei strain	North Jiangsu
O. h. robertsoni (Bartsch)	Southwest China
O. h. hupensis yunnan strain	Yunnan
O. h. hupensis sichuan strain	Sichuan
O. h. formosana (Pilsbry and	Taiwan
Hirase, 1905)	
O. h. chiui (Habe and	Taiwan
Miyazaki, 1962)	
O. h. nosophora (Robson,	Japan
1915)	
O. h. quadrasi (Mollendorff,	The Philippines
1895)	
O. h. lindoensis (Davis and	Sulawesi island in Indonesia
Carney, 1973)	

analysed, wetland showed the strongest correlation with the genetic pairwise distances, in The People's Republic of China (Liang et al., 2014). *Oncomelania hupensis* distribution is strongly linked to the presence of wetlands, and the distribution pattern of both *O. hupensis* and schistosomiasis might be altered due to the changed wetland resulting from water development projects such as the Three Gorges Dam and the South-to-North water transfer project (Liang et al., 2014; Yang et al., 2016).

Since the 1990s, the numerical taxonomy technique has been applied to morphology of *O. hupensis* from different areas in The People's Republic of China. The studies, however, showed that the classification of *O. hupensis* only based on epigenetic characteristics was unsound, and hence *O. hupensis* in The People's Republic of China can be classified into three geographical strains at genetic level, namely the Hunan, Hubei and Anhui/Jiangxi strains (Zhou, 2005). Furthermore, there was a geographical strain in Sichuan and Yunnan and another in Fujian and Taiwan (Zhou, 2005).

New progress of Oncomelania classification has been made with the development and application of molecular biology. SSR-PCR technique was used to analyse 19 populations of O. hupensis from eight provinces in mainland The People's Republic of China and found that the average genetic distance of population was 0.22 (Zhou et al., 2007). The study divided those specimens into four groups by applying cluster analysis; 16 populations along the middle-lower Yangzi River basin in five provinces (ie, Hunan, Hubei, Jiangxi, Anhui and Jiangsu) constituted one group and the remaining three groups were from Guangxi, Fujian and Yunnan, respectively. Therefore, O. hupensis in mainland The People's Republic of China probably can be accordingly divided into four subspecies, that is, O. h. hupensis, O. h. robertsoni, O. h. tangi and O. h. formosana. In addition, landscape genetic analysis based on ITS1, ITS2 and 16s genes also indicated four groups that located in four different landscapes, namely the middle and lower reach of Yangtze River, the mountainous areas of Yunnan and Sichuan, the inland hill areas of Guangxi and the inshore hill areas of Fujian (Li et al., 2009).

# 2.2 Susceptibility to different geographical strains of S. japonicum

The susceptibility of *O. hupensis* to schistosomes from different endemic areas and even different habitats within the same area may be variable considerably (Hong et al., 1995; Lin et al., 1994). When exposed to a strain of *S. japonicum*, the infection rate of local snails is much higher than that of snails from other areas. For example, the infection rate of snail from Guichi,

Anhui Province was 84% when infected with local strain, while it was only 32% and 28% for snails from Bixian, Sichuan Province and Dali, Yunnan Province, respectively, when exposed to the same strain (Huang et al., 2002; Xu and Ni, 1987). Resistance to nonlocal strains of schistosomes was also observed (Cao et al., 2009). *Oncomelania hupensis* from marshland areas was found highly susceptible to schistosome strains from hilly areas, while *O. hupensis* from hilly areas showed the low susceptibility even when exposed to local strains of schistosome (Wang et al., 2014) (Table 2).

Interestingly, infectivity of miracidia hatched from eggs isolated from various hosts may be variable. In a study where *O. hupensis* was exposed to miracidia obtained from rabbit, buffalo and mouse, the infection rates were 1.42%, 8.67% and 19.87%, respectively (Tian et al., 2011). There was a significant difference between invisibility of miracidiae; *O. hupensis* was more susceptible to miracidia from mice.

# 2.3 Factors influencing infection

Experiments indicated that the number of miracidia, distance from the release point, temperature, frequency, time, sex and age of snail may influence infection. The infection rate of snails increase with the number of miracidia in a proper range; excess exposure leads to mortality of snails (He et al., 1989, 1994; Xu et al., 1993).

The infection rate of snails is inversely proportional to the distance from the release point of miracidia, the volume and depth of water (He et al., 1989; Jiang et al., 1989; Wang et al., 2011). The optimum temperature for infection is 10–20°C; the infection rate of snails declined greatly below 10°C and above 20°C (Sun et al., 2004; Yang et al., 2007). In another study where snails were exposed to miracidia of various release time, the infection rate was highest for miracidia 1–4 h post release, while infection rates of snails exposed to miracidia 8 or 12 h post-release were lower (He et al., 1994).

Table 2 Compatibility between Oncomelania hupensis and Schistosoma japonicum Infection rate of snails (No. snails shedding cercariae/No. snails survived)

Strains of	54.11.54,							
S. japonicum	Anhui	Hubei	Guangxi	Sichuan	Yunnan	Fujian		
Anhui	42.9(249/580)	43.8(56/128)	20.0(36/180)	1.5(4/270)	0(0/173)	25.0(1/4)		
Hubei	40.9(110/269)	33.1(47/142)	_	0(0/126)	0(0/71)	_		
Guangxi	30.7(59/192)	_	9.4(12/128)	0(0/67)	0(0/12)	0(0/66)		
Sichuan	4.5(40/881)	10.2(25/244)	_	1.4(5/360)	0.4(1/229)	_		
Yunnan	10.8(43/397)	33.6(46/137)	14.9(7/47)	1.0(2/198)	8.3(8/96)	4.0(1/25)		

He et al., 1990.

Female snails aged between 3 and 4 months are more susceptible to infection than male snails of the same age (He et al., 1994). However, the male and female snails above 5 months showed the similar susceptibility and duration of cercariae release. Susceptibility of adult snails was higher than that of immature snails (He et al., 1994).

# 2.4 Factors influencing release of cercariae from snails

Many factors may influence the development and release of cercariae from infected snails, of which temperature is the most notable (Mao, 1990). According to the degree-day model, the accumulative temperature for development of cercariae in snail is  $1793.93 \pm 232.45$  degree-days (Sun et al., 2001). The temperature range  $20-35^{\circ}$ C is optimal for cercariae release, but 13% of infected snails still can release cercariae at the water temperature of 1°C, which implies that people can become infected during winter months. In addition, pH of the water may influence cercariae release; pH in the range 6.6-7.8 seems optimal for cercariae release (Liang et al., 2004). The lifetime of cercariae is not affected by pH in the range 4.6-9.8 (Liang et al., 2004).

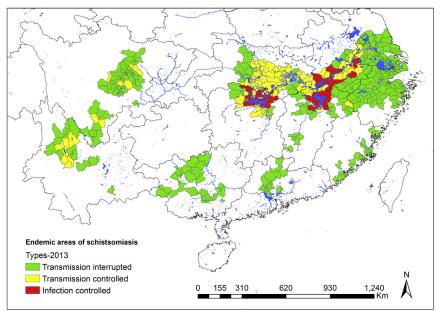
Cercariae release is closely related to light. A circadian rhythm can be observed; the release of cercariae from infected snails mainly occurs in day time (Mao, 1990). Observation of cercariae release from naturally infected snails showed that the peak time is at 7–10 am in marshland areas where cattle were the main infection source, and at 5–9 pm in hilly areas where field rats were the main infection source (Lu et al., 2009; Su et al., 2013).



# 3. DISTRIBUTION AND SURVEY OF *O. HUPENSIS* IN THE PEOPLE'S REPUBLIC OF CHINA

## 3.1 Distribution characteristics

Oncomelania hupensis is distributed in 12 southern provinces with an accumulated snail infested area of 14.32 billion m<sup>2</sup> (Fig. 1). The range meets an annual average temperature of over 14°C and an annual average precipitation of over 750mm, which indicates that mild climate and abundant rainfall are necessary for snail (Zhou, 2005). The geographical distribution of O. hupensis is consistent with that of schistosomiasis endemic areas in The People's Republic of China (Liu et al., 2016; Shi et al., 2016; Wang et al., 2016). It is possible that potential snail habitats can extend further



**Figure 1** Distribution map of *Oncomelania hupensis* and schistosomiasis in The People's Republic of China, 2013.

to the north in the context of global warming and hence cause the expansion of endemic areas of schistosomiasis (Zhou et al., 2004).

Based on geomorphological features, the snail habitats are classified into three types, namely, marshland and lake area, mountain and hill area and plain water network area (Mao, 1990). These types of snail habitats can be commonly observed in a single county (Mao, 1990).

### 3.1.1 Marshland and lake areas

A large number of marshlands are formed on both sides of the middle-to-lower reaches of the Yangtze River and around the large lakes connected to the river, which are mainly located in four provinces, namely, Hunan, Hubei, Jiangxi and Anhui (Zhang et al., 2016b). Currently, O. hupensis is mainly distributed in these areas, accounting for 96.49% in the whole country (Table 3) (Lei et al., 2014). Marshland is characterized by a large seasonal fluctuation of water level and hence is flooded during summer and remerges during winter. Mild climate, abundant rainfall, fertile and humid soil and overgrown grassy vegetation contribute to favourable conditions for snail breeding (Mao, 1990). Snails in these areas generally grow larger than those in mountainous and hilly areas as well as the plain water

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**Table 3** Snail habitats area and mollusciciding area in The People's Republic of China, 2013

Marshland endemic area (hm²)

				Marsillaria eric	define area (iiii )			
Province	No. township with snails	No. village with snails	Total area (hm²)	Inside embankment	Outside embankment	Plain water network endemic area(hm²)	Mountain and hill endemic area (hm²)	
Shanghai	8	17	0.93	0.00	0.00	0.93	0.00	
Jiangsu	69	190	3293.74	0.00	3056.57	202.97	34.20	
Zhejiang	98	369	94.21	0.00	0.00	12.48	81.73	
Anhui	211	962	27,396.49	0.00	24,480.08	0.00	2916.41	
Fujian	9	14	1.51	0.00	0.00	0.00	1.51	
Jiangxi	140	601	78,794.33	3.87	76,952.04	0.00	1838.42	
Hubei	352	2604	76,485.70	19,231.60	54,818.65	0.00	2435.45	
Hunan	199	922	175,116.23	854.50	173,230.07	0.00	1031.65	
Guangdong	0	0	0.00	0.00	0.00	0.00	0.00	
Guangxi	5	5	4.96	0.00	0.00	0.00	4.96	
Sichuan	361	1850	2600.39	0.00	0.00	0.00	2600.39	
Yunnan	56	248	1679.51	0.00	0.00	0.00	1679.51	
Total	1508	7782	365,468.00	20,089.98	332,537.41	216.38	12,624.23	

network areas, usually over 7.5 mm in height and 3.2 mm in width, with thick and ribbed shell.

Depending on the water level control system, marshlands and lake areas can be classified into four subtypes, namely, 'island', 'marsh', 'embankment' (area from dike towards the marsh/lake) and 'inside embankment' (area protected from flooding by the dike) (Chen, 1980). The distribution of snails exhibit high spatial heterogeneity in these vast marshlands. Two distribution features of snails can be observed, that is, 'two lines' (the highest and lowest elevation lines where snails can be found), thus defining 'three zones' (upper and lower zones with few snails, middle zone with high density) (Li et al., 2006b; Zhou, 2005).

The distribution of snails in marshland is related to micro-ecological factors, such as submerging time in flooding season, groundwater level, soil moisture, vegetation and light intensity (Cao et al., 2015). Prolonged submerging, high groundwater level and moist soil lead to increase in the snail density. Of these factors, the groundwater level had the dominant effect on the snail intensity (Wang et al., 2007). A study revealed that snail density and the percent of frames with snail was highest when the groundwater level was around 32 cm, and soil moisture content of 28-38% was optimal for snail breeding (Zhang et al., 1999). Water-level fluctuation can affect the macro- and micro-environment of snail habitats as well as snail's life activity and distribution status, which in turn, affect schistosomiasis transmission (Li et al., 2010a; Ma et al., 2011). Snails are few in low-elevation marshlands that are submerged over 8 months, or high-elevation marshlands that are submerged less than 3 months. Snail habitat is often characterized by the dominant plant species, such as Cyperus rotundus, Carex tristachya and Phragmites communis.

More than 93% of infected snails in marshland are found in irrigation ditches or marshes within 500 m from residential areas. Density of infected snail shows negative correlation with distance to residential areas (Su et al., 2011). Owing to frequent activities of humans and livestock in snail-occupied ditches and marshlands near the residential areas, the snail habitats were contaminated with faeces, and this led to schistosomiasis transmission (Peng et al., 2007). Although the density of snails in large ditches (width 1–3 m) might be lower than in small ditches (width < 1 m), the prevalence of schistosome in snails in big ditches could be higher than small ones (Guan et al., 1999). It was explained that more frequent activities of humans and livestock occurred around or in the big ditches than in the small ones. In a different location, however, the density of infected snails was not positively correlated

with the density of snail, but was related with frequency water contact activity (Wei et al., 2001). In another similar environment, there was the positive correlation between the mean density of snails and the prevalence of infection in snails (Wei et al., 2001).

### 3.1.2 Mountain and hill areas

This kind of snail habitat can be found in all endemic provinces except Shanghai, but it only accounted for 3.45% in the whole country (Lei et al., 2014; Liu et al., 2016) (Table 3). Complex landscapes due to diverse elevation lead to many small fragments of snail habitat (Mao, 1990). In hill area, the distribution of snails is restrained in specific zones and is strikingly variable. For example, snails can be found at one side of a mountain while none at the other side (Qiu et al., 2014). In the same water system, snails can live both in ditches and farmlands along the entire length of water courses (Qiu et al., 2014). In mountain area, snails usually live in the undergrowth with patchy distribution or along water course with linear distribution. Some snails can inhabit the mountain side and foot. A few snails can survive in spring water on the top of mountain and form the source of the snail distribution (Mao, 1990). In the basins in mountain area, O. hupensis is distributed mainly in ditches and farmlands, similar to plain network areas (Mao, 1990).

Less rainfall and lower temperature in mountain and hill areas are the factors affecting the development and breeding of snail, resulting in the snail's smaller size in these areas than in marshland and plain areas with the height of 5.8–6.9 mm and width under 2.85 mm (Zhou, 2005; Zhou et al., 2006). The snail shell is smooth without ribs or with only thin rib lines (Zhou, 2005; Zhou et al., 2006).

In the mountain and hill areas, the highest density of snails is found along river banks, followed by irrigation ditches, farmlands and wild grass ground, where it is the lowest (Xu et al., 2010; Zhang et al., 2007b). The sand content and humidity in soil are the most important factors determining snail presence and survival (Xu et al., 2010; Zhang et al., 2007b). Infected snails are mainly found in ditches, followed by river shore land, pond and river course successively (Xu et al., 2010; Zhang et al., 2007b).

### 3.1.3 Plain water network area

Plain water network area is mainly located in the Yangtze River delta, covering Shanghai and the part of Jiangsu and Zhejiang provinces (Shi et al., 2016). In this area water courses are well developed and form network

with slow water currency, stable water level, weeds in the shore. By the end of 2013, plain water network area with snail only accounted for 0.06% of the national total area (Lei et al., 2014). The snail is medium sized with a height of 6.5–7.5 mm and transverse ribs. *Oncomelania* snails are often distributed along irrigation canals and the density varies among locations (Wang, 2006a). Ponds connected to river or canals, dikes of ridge field, bridge pillars, gap between stone or bricks can be also occupied by *Oncomelania* snails (Mao, 1990).

# 3.2 Methods for survey of O. hupensis

The purpose of snail survey is to obtain information of snail distribution and density as well as characteristics of habitats, and hence help to develop snail control plan, choose control methods and evaluate control effect (Mao, 1990). The targeted areas normally include known snail habitats where snail occurs at present or historically, suspected environments adjoining known snail habitats and potential environments which facilitate the spread of snails. The timing and frequency of survey is determined according to the habitat characteristics and the aim of the survey. Four methods can be selected, namely (1) systematic sampling, (2) environmental sampling, (3) systematic sampling combined with environmental sampling method and (4) thorough investigation (Mao, 1990; Zhou, 2005) (Table 4). Snail sampling is performed using a square frame of 0.1 m<sup>2</sup> and all snails within the frame are collected manually. In systematic sampling, the frames are laid out at an interval of 10-20 m along a line. In the survey area one or several parallel lines should be set at an interval of 10-50 m. The interval between frames or lines can be adjusted according to the size of the area surveyed. In environmental sampling, the frames are placed in locations that are most likely to harbour snails. In a thorough sampling, the frames are not used but the entire area is subjected to a careful visual inspection for snails.

# 3.3 Snail monitoring

In areas where schistosomiasis transmission has been interrupted, it is necessary to monitor the *Oncomelania* snails in existing and historical snail habitats and to prevent snail spread and consolidate achievements of schistosomiasis control (Dang et al., 2014; Wen et al., 2011). In the existing snail habitats, snail survey is conducted in spring or autumn every year. In the historical snail habitat where snail re-emerged within the last 3 years, one snail survey should be carried out during spring and double surveys should be carried out in important environments (eg, close to village). Thorough snail survey is

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Type of snail habitat	urvey in different habitat types Method of survey	Applicable environment	Timing of survey
Marshland and lake area	Systematic sampling	High snail density and scattered distribution	Mar., Apr., May or Sep., Oct., Nov.
	Environmental sampling	Historical snail habitat where snail cannot be found by systemic sampling method.	Mar., Apr., May or Sep., Oct., Nov.
	Thorough survey	Snail has been eliminated recently and small area with complex environment where the systematic sampling method is difficult to use.	Mar., Apr., May or Sep., Oct., Nov.
Plain water network	Systematic sampling	River, ditches, pond	Apr. to Oct.
area	Environmental sampling	Historical snail habitat where snail cannot be found by systemic sampling method	Apr. to Oct.
	Thorough survey	Suspected habitat to be confirmed, or areas where snails were eliminated and small area with complex environment where the systematic sampling method is difficult to use.	Apr. to Oct.
Mountain and hill area	Systematic sampling	River, ditch, pond	Apr. to Oct.
	Environmental sampling	Special environment such as hill land, grave mound, bamboo forest	Apr. to Oct.
	Thorough survey	Suspected habitat to be confirmed, or areas where snails were eliminated and small area with complex environment where the systematic sampling method is difficult to use	Apr. to Oct.

performed once every 3 years in the historical habitats where snails have not been found in last 3—9 years, and once every 5 years in those habitats where snails have not been found over 10 years. For areas where snail never was recorded, monitoring site should be established in suspected environment and one or two snail surveys each year for successive 2 years (Chen et al., 2012a; Wang et al., 2013).

## 3.3.1 Monitoring snails in water bodies

Oncomelania snails are prone to stick to floating matters in water bodies. Therefore, the snails can be collected using floating traps. The traps are usually made of a 0.1 m<sup>2</sup>-sized curtain woven by rice straw. Such traps are put into water near the bank of rivers or canals equidistantly for 3 days. The retrieved traps are scrubbed twice by clean water and the Oncomelania snails are carefully screened in the filtered water. The method shows simple operation, low labour intensity and better performance than systematic sampling (Cao et al., 1990; Zhang et al., 1989). An experiment using two methods, that is, systematic sampling and attracting snail with rice straw, was conducted along 22 rivers from June to August. The results showed 13.48 snails per 0.1 m<sup>2</sup> (trap of rice straw) while 4.67 snails per 0.1 m<sup>2</sup> (frame in systematic sampling) (Mao, 1990). Furthermore, the former method is more sensitive to young snails; the number of young snails on traps (9.41 snails) is 11.2 times higher than that in frame (0.84 snails) in systematic sampling. The results also indicated that traps collected more snails than did systematic sampling during May—August and October, although the counts were similar in September. This seasonal variation of two methods was mainly affected by variation of movement capability of snail (Cao et al., 1990; Zhang et al., 1989).

# 3.3.2 Monitoring snails carried by travelling animals, plants and transportation vehicles

In areas where schistosomiasis transmission is interrupted or where nonendemic areas connect endemic area by water courses, potential source of infections (eg, aquatic animals and plants, trees, boats) from schistosomiasis endemic areas should be monitored to avoid the spread of snails (Cao et al., 2016). In 2002, acute schistosomiasis cases were reported in two villages around the marshlands of the Gan river basin in Jiangxi Province where *Oncomelania* snails were never recorded (Zeng et al., 2007). A subsequent survey confirmed the presence of *Oncomelania* snail in the marshlands. Furthermore, 177 fishing boats in this area were investigated. The results showed that all boats carried *Oncomelania* snails with an average number

of 13.69 per boat (Zeng et al., 2007). In addition, 90.9% crayfish pots and 75.0% fishing nets in those boats also carried snails (Zeng et al., 2007).

In nonendemic areas connecting snail habitat areas by water courses, floating matters on the river water should be monitored during the flood season or after the withdrawal of water (Zhang et al., 2016a). The method may be net—dredge washing, that is using a net to dredge and collect floating matters including aquatic plants to screen for snails (Yin et al., 1993; Zuo et al., 2010).

# 3.3.3 Remote sensing technology for snail surveillance

Remote sensing provides a new approach for identifying the potential distribution of *Oncomelania* snails by predicting the suitable habitats (Feng et al., 2016; Zhou et al., 1999). It is widely acknowledged that the flooding time is an important factor for the spread and establishment of *Oncomelania* snail in the lake areas, and species and distribution of vegetation is different due to various flooding days of each year (Yang et al., 2013; Zhang et al., 2013). Cluster analysis of TM images can identify and extract vegetation areas that are suitable for snail presence, then two spatial models can be established, that is, distribution of snail habitat and spatial spread of snail, so that the area of snail habitats and the potential spreading area of snail can be predicted directly (Yang et al., 2013; Zhang et al., 2013).

Landset ETM+ images have the potential to identify the snail habitats in marshlands along rivers (Zhang et al., 2005). It was found that the snail habitats in these areas can be classified into four types of landscapes, and the density of snail was higher in two of them, which was confirmed through field survey. Similarly, Landsat TM images were also used to classify the snail habitats in mountain areas (He et al., 2010). The sensitivity and specificity of the classified snail habitats to correctly predict the actual snail habitats was 79.82% and 85.58%, respectively. By using RS techniques, vegetation features and the ground survey data of actual snail distribution can be overlaid on the classified satellite images. Hence, the snail habitats can be identified precisely and quickly, which facilitates snail survey and control (Li et al., 2010b; Yang et al., 2005). GIS and GPS special analysis and map overlap analysis reveals that epidemic scale of schistosomiasis is closely related to climate and environmental factors including temperature, altitude, and precipitation (Wu et al., 2014; Zhou et al., 1999). Therefore it is possible to predict potential schistosomiasis transmission areas by analysing these climate factors (Wu et al., 2014; Zhou et al., 1999). RS technique also provides a feasible way for snail surveillance, and can be applied as a supplementary tool for snail detection (Wang et al., 2016). The predictive accuracy for the distribution of *O. hupensis* was up to 86.2%, and it was identified that 82.77% of suspicious snail habitats were located in ditches (Xiao et al., 2009; Yang et al., 2008a).

## 3.3.4 Early identification of infection in snails

Loop-mediated isothermal amplification (LAMP) is a relatively new nucleic acid amplification technique (Chen et al., 2016). LAMP has been successfully used for detecting schistosome-infected *O. hupensis* due to its high specificity, sensitivity, convenience and efficiency (Kumagai et al., 2010; Tong et al., 2015; Yu et al., 2011).

It is well known that it takes more than two months from miracidia invasion to cercariae release (Mao, 1990). The conventional microscopic examination by crushing snail can only find daughter sporocyst and cercariae of schistosome in snail. Instead, LAMP technique based on targeting nucleic acid can detect not only daughter sporocyst and cercariae but also DNA targets at even earlier development stage, and hence the detection rate of schistosome-infected snail can be improved (Deng et al., 2009; Wang et al., 2010).

Infections in snail can be detected by LAMP as early as one week after the snails were exposed to miracidia, and the time of detection can be also shortened from 6 h by routine PCR to 2 h (Yu et al., 2011). In addition, LAMP was more sensitive to infections (Deng et al., 2014; Yu et al., 2008). Tong et al. (2015) employed LAMP to examine snails collected from 28 snail habitats in Hunan, Hubei and Anhui provinces, and mapped high risk areas of schistosomiasis transmission by using ArcMap. Those results could provide basic data for schistosomiasis surveillance and intervention strategy.

# 4. CONTROL OF O. HUPENSIS

The People's Republic of China has developed a series of snail control methods in the last 60 years, including environment modification and chemical mollusciciding (Zhou, 2005). Environment modification include burial of surface soil harbouring snails, cement-lining of irrigation ditches, conversion of paddy fields into dry crop farming, utilization of marsh land for vegetables and crops, blocking river branch for aquaculture, reclaiming uncultivated land, weeding for manure collection and so on. Especially, tree planting in *O. hupensis* snail habitats is a novel snail control method

(see cross-reference in this volume). Chemical mollusciciding is the primary measure in areas where snail habitat modification cannot be implemented temporarily (Zhou, 2005). Table 5 compares the advantages and disadvantages of these two snail control methods. In 2013, the total area where chemical molluscicide was applied was up to 134,423.49 hm<sup>2</sup> with 71,968.41 hm<sup>2</sup> of actual molluscicide area, and the area of environment modification for snail control was 5852.21 hm<sup>2</sup> (Lei and Zhou, 2015).

## 4.1 Environment modification for snail control

Snail habitat modification has significant effects on schistosomiasis control in The People's Republic of China (Lei and Zhou, 2015). Lots of snail habitats in various endemic areas have been changed by agricultural practice, water

Table 5 Comparise Method	on of two snail control methods Advantage	Disadvantage			
Snail habitat modification	<ol> <li>More effective than molluscicides and has a long-term effect</li> <li>Saving resources:         Reclaiming abandoned fields can improve the     </li> </ol>	1. Large one-time investment is needed when reclaiming land, construction of flood control project and irrigation ditches			
	local agricultural structure, and make better use of local resources (labour force, materials)	2. Effect on snail control is slower than chemical molluscicides and needs to be assessed for a long time			
	<ul> <li>3. Public participation: public awareness on schistosomiasis can be improved by community participation</li> <li>4. No environmental pollution</li> </ul>	3. Less scope of application			
Chemical molluscicides	<ol> <li>Wide application</li> <li>Easy operation, saving time and labour</li> <li>Immediate effect</li> <li>Can be used repeatedly</li> </ol>	<ol> <li>High price</li> <li>Toxic to nontarget organisms (eg, fish, aquatic animals and plants)</li> <li>Possible environmental</li> </ol>			
		pollution 4. Need to be used repeatedly			

conservancy and urbanization, which have rendered such environments unsuitable for the snails (Yang et al., 2016).

In marshland and lake areas, environmental modification play an important role in schistosomiasis transmission control, such as building snail retention reservoirs at the entrance of major irrigation canals (water goes to irrigation canal through the reservoir where the snails are retained and sunk), embanking the lowland for aquaculture, and enclosing marshland for planting. A field observation on the effect of a snail retention reservoir in Gongan, Hubei Province showed that the percent of frame with living snails declined from 11.83% in 2004 to 8.81% in 2011 after the reservoir was built in 2005 (Fang et al., 1998). Meanwhile, the mean density of snails dropped from 1.36 per 0.1 m<sup>2</sup> to 0.35 per 0.1 m<sup>2</sup> accordingly. Another comprehensive modification project was carried out in Jixing village in 2003 employing multiple measures, such as cement-lining of irrigation canals, changing production structure, improving water and sanitation, health education and tree planting (Fang et al., 1998). The snail habitat area went down from 41.75 hm<sup>2</sup> in 2003 to 0.83 hm<sup>2</sup> in 2011 and the percentage of frames with living snails and mean density of living snails dropped by 81.52% and 95.03%, respectively. The proportion of infected snails decreased from 0.92% to 0 (Chen et al., 2012b). This study indicated that the comprehensive control strategy with multiple measures has a better effect than the single measure for snail control. In the 1990s, a project of snail habitat modification was implemented in the Dongting Lake area in Hunan Province, which is a typical marshland endemic area with high prevalence of infection in people and high density of snails and infected snails (Fang et al., 1998). Depending on topographical features of specific marshlands, four measures were used, that is embanking the lowland for aquaculture, enclosing marshland for tree, enclosing marshland for reed, reclaiming and cultivation of marshland. Three years later, the density of living snails and infected snails significantly declined. Of these measures, the first one can eliminate snails completely as seen in Table 6 (Fang et al., 1998).

Environmental modification also has significant effect on snail control in plain water network areas. Seven localities in Hubei Province were selected as the observing sites to investigate the effect of 57 previous projects on habitat modification between 1997 and 2001 (Wang et al., 2002). Measures, such as water storage for aquaculture, digging new and filling old small irrigation canals, reclaiming and cultivation, burying soil with sands and cement-lining of irrigation canals, were taken for snail control. Five years later, the area infested with snails, the area potentially suitable as snail habitat,

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 Table 6
 Effect comparison of different measures for snail habitat modification in the Dongting Lake basin

		Den	sity of living sn	ails	Density of infected snails		
Measure	Location	Before (one/0.1 m <sup>2</sup> )	3 years later (one/0.1 m <sup>2</sup> )	Rate of decline (%)	Before (one/0.1 m <sup>2</sup> )	3 years later (one/0.1 m <sup>2</sup> )	Rate of decline (%)
Water storage for fishing in low lying land	Choutang village, Xiangyin County	1.800	0.000	100.0	0.0190	0.0000	100.0
Enclosing marshlands and not allowing grazing	Junshan Farm	1.505	0.056	96.3	0.0454	0.0026	94.3
Enclosing marshlands for reed planting	Qilihu, Chen County	1.808	1.129	37.6	0.0032	0.0000	100.0
Reclaiming and cultivation without enclosing marshlands	Shangfang village, Zhao County	0.638	0.013	98.0	0.0072	0.0000	100.0

the density of snails and infected snails decreased by 71.7%, 90.5%, 96.5% and 95.9%, respectively (Wang et al., 2002). Of the five measures, burying soil with sands and cement-lining of irrigation canals were particularly effective in controlling snails. In another study, a programme of snail control, combined with comprehensive agriculture development, was implemented in Xichahu village, Hubei Province to establish agricultural, forestry and fishing facilities by changing snail habitat condition (Zhang et al., 2007a). In addition, irrigation canals were expanded and facilities, such as bridges, culverts and snail retention reservoirs were built at the sources of snail populations to reduce the risk of snail spread after floods (Zhang et al., 2007a). The study showed that snails were under effective control after 4 years of the programme. The area infested with snail, the infection rate of residents and cattle dropped by 98.01%, 90.60% and 87.16%, respectively (Zhang et al., 2007a). The local economic development was rapid with a 42% increase in agricultural productivity and a 11.53% increase in rural per capita net income (Zhang et al., 2007a).

A comprehensive snail control project was carried out along Yangtze River in Huangshi, Hubei Province during 1996-2000 (Hu et al., 2001). Measures included reclaiming land for cultivation, covering marshland with sand, tree planting, crop planting in woodland, urban construction and development (building park and wharf along the river) and using chemical molluscicides. Five years later, the total area with snail dropped by 97.18%. The number of infected snail went down to 0. The mean density of living snails declined by 95.19%. The effect of environment modification for snail control was higher than chemical drug (Hu et al., 2001). In Ruizhou, Jiangsu Province, the snail area along Yangtze River accounted for 93.7% of the district's entire snail area. A programme of snail habitat modification was conducted in one of such areas, including building low banks and reclaiming the marsh land for mechanical cultivation (Tao et al., 2012). In the same year, the density of living snails decreased by 99.75%, and living snails were not found in survey in 2010 and 2011 (Tao et al., 2012).

Modification in hilly endemic areas may be cement-lining of irrigation canals, burying snail habitat, digging new canals and burying old ones, dam expansion, reclaiming land and cultivation and returning abandoned land to forestry (Li et al., 2011). Jurong, Jiangsu Province is a typical hilly endemic area, where a project of snail habitat modification was carried out from 2001 to 2010 (Li et al., 2011). After the project, no snails were found in 12 hot spots, and the residual snail area decreased to 1.2 hm<sup>2</sup> in

other three hot spots with rapid dropping of snail area and density. Especially, the rates of decline of snail area and density were 100% when measures of digging new ditches and burying old ones and water storage on banks were taken (Table 7) (Li et al., 2011).

Tree planting for snail control is mainly carried out in marshland and lake areas. Planting poplars in marshlands at relatively high altitude can decrease the density of snail by changing vegetation structure of marshland and make it unsuitable for snail habitat. After planting trees on areas with snail, the surviving snails lack nutrition and therefore cannot reproduce primarily due to reduced lactate dehydrogenase and succinic dehydrogenase caused by the decline of glycogen content (Peng, 2001; Zhang et al., 2013). During 1986-2000, 16 projects of tree planting for snail control had been implemented in five provinces located in the middle and lower branches of Yangtze River. As a result of the 15 years of study, the percentage of frames with living snails stably dropped every year, from 19.03% in the fourth year of the project to 3.61% in 2000; the mean density of snail declined from 2.7 per 0.1 m<sup>2</sup> to 0.5 per 0.1 m<sup>2</sup>. The established forestry prevented the activities of people and livestock on marshlands and hence decreased the risk of infection transmission. The mean density of infected snail went down to 0 in the 3rd year of the project from 0.004 per 0.1 m<sup>2</sup> at the beginning (Huang et al., 2011; Peng, 2001).

Water conservancy projects for schistosomiasis control aim at changing snail habitats by conducting a series of measures, such as hardening banks of river and lake, constructing isolated canals, culvert and sluice gates, snail retention reservoirs, cement-lining of irrigation ditches and others (Mao et al., 2010; Yang et al., 2016; Zhu et al., 2009). Twenty-eight water conservancy projects were implemented in endemic areas of seven provinces between 2007 and 2011. The systematic evaluation showed that all these projects had effect on snail populations (Table 8). However, the studies also implied that snails could re-inhabit if cement-lined irrigation canals were not continuously maintained and managed. In this case, the cement surface would be damaged by mud deposition and growth of aquatic plants, which may facilitate formation of snail habitats. Once snails were carried by water from noncemented ditches, they can breed. Therefore, its effect in snail control may be compromised, especially in long term (Zhu and Zhang, 2012).

The Three Gorges Dam operating since 2003 had a significant influence on snail habitats (McManus et al., 2010; Seto et al., 2008; Zhu et al., 2008). Monitoring results in recent years showed that the density and area infested by snails were decreasing in Dongting Lake, Poyang Lake and the

 Table 7 Comparison of effect of different measures of snail habitat modification in hilly endemic areas

·		Before n	nodification	After n	nodification		
Measure	Project	Area with snail (hm²)	Density of snail (one/0.1 m <sup>2</sup> )	Area of snail (hm²)	Density of snail (one/ 0.1 m <sup>2</sup> )	Rate of decline of snail area (%)	Rate of decline of snail density (%)
Digging new ditches and	Changlong Moutain	0.28	0.42	0.00	0.00	100.00	100.00
burying old ones	Kongqing erdao Basin	0.40	0.35	0.00	0.00	100.00	100.00
	Gaolun Basin	0.48	0.32	0.00	0.00	100.00	100.00
	Shanggan Basin	0.95	0.32	0.00	0.00	100.00	100.00
	Daxi Moutain	4.95	4.20	0.00	0.00	100.00	100.00
	Lutang Basin	0.80	3.60	0.00	0.00	100.00	100.00
Reservoir	Pochong Basin	0.70	0.45	0.00	0.00	100.00	100.00
construction	Kongqing Village	0.07	0.58	0.00	0.00	100.00	100.00
Digging fishpond	Fangshan Basin	7.07	4.50	0.00	0.00	100.00	100.00
	Daxintang marshland	1.44	2.40	0.40	0.36	72.22	85.00
Comprehensive	Yucheng Basin	2.20	3.20	0.00	0.00	100.00	100.00
measures in small	Wuqi Basin	1.65	1.30	0.00	0.00	100.00	100.00
watershed	Dongshanchong, Lutang Basin	0.26	0.80	0.00	0.00	100.00	100.00
	Lita Basin I	3.60	3.40	0.50	0.64	86.11	81.18
	Lita Basin II	4.70	3.20	0.30	0.72	93.62	77.50

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	•	Area with snails (hm²)			Percent of frames with snails(%)			007-2011 Density of living snails (one/0.1 m <sup>2</sup>		
Type of project	Location of project	Before	After	Rate of decline (%)	Before	After	Rate of decline (%)	Before	After	Rate of decline (%)
Water sto	rage for fishing in low bank									
	Caipeng lake in Duchang county, Jiangxi	496,000	0	100.00	0.00	0.00	_	0.00	0.000	100.00
	Xiwei bank in Xinzi county, Jiangxi	655,000	32,000	95.11	0.20	0.00	100.00	0.00	0.000	100.00
	Nanxin Village in Wuhu city, Anhui	20,000	0	100.00	2.17	0.00	100.00	0.04	0.000	100.00
Letention	reservoirs									
	Dongza in Junshan district, Hunan	0	0	100.00	9.40	0.00	100.00	0.11	0.000	100.00
	Majiazui in Gongan county, Hubei	8400	8400	0.00	29.50	21.80	26.10	1.21	0.280	76.86
	Jiuxia Canal in Gongan County, Hubei	16,000	16,000	0.00	18.70	7.50	59.89	1.09	0.910	16.51
	Xiagan Canal in Gongan County, Hubei	12,000	12,000	0.00	18.70	7.00	62.57	1.09	0.290	73.39
	Irrigation ditch in Hujiatan, Jiangxi	_	0	100.00	13.43	0.00	100.00	0.99	0.000	100.00
	Irrigation ditch of Hujiatan, Jiangxi	_	0	100.00	15.76	0.42	97.34	0.76	0.010	98.68
	Irrigation station, Fenghuangjing, Wuwei county, Anhui	0	0	100.00	0.06	0.00	100.00	0.00	0.000	100.00
	Touqiao Floodgate of Xia River, Ganjiang District, Jiangsu	3800	0	100.00	31.17	0.00	100.00	1.20	0.000	100.00
	Xiangyang sluice gate, Xinjiang District, Jiangsu	0	0	100.00	26.42	18.50	29.98	0.91	4.580	-403.30
	Liugong sluice gate, Xingjiang District, Jiangsu	0	0	100.00	0.00	0.00	100.00	0.00	0.000	100.00
Cement li	ning of canals									
	Trunk canal, Yueyang, Hunan	8100	0	100.00	36.58	0.00	100.00	1.68	0.000	100.00
	Ditches, Junshan District, Hunan	66,300	0	100.00	50.30	19.57	61.09	2.66	1.370	48.50
	Jingyang Village, Xuanzhou	28,400	8500	70.07	22.28	0.00	100.00	0.75	0.000	100.00

District, Anhui

	Xintao Village, Nanling, Anhui	12,000	12,000	0.00	0.20	0.00	100.00	0.00	0.000	100.00
	Bianming River, Jurong, Jaingsu	41,490	0	100.00	0.36	0.00	100.00	0.00	0.000	100.00
	Panjia River, Yizheng, Jiangsu	27,398	0	100.00	0.00	0.00	100.00	0.00	0.000	100.00
	Irrigation areas Huanglianggen	1300	32	97.54	0.00	0.00	_	0.00	0.000	100.00
	reservoir, Dongpo, Sichuan									
	Irrigation areas Meiwan reservoir,	3300	1064	67.76	5.86	0.00	100.00	0.14	0.000	100.00
	Danling, Sichuan									
	Dongfeng ditch, Dongpo, Sichuan	1015	306	69.85	22.60	13.20	41.59	1.60	0.280	82.50
	Maqiao Village, Meishan, Sichuan	1975	25	98.73	80.30	19.70	75.47	2.30	0.650	71.74
	Simen Village, Guanghan, Sichuan	7900	160	97.97	30.00	17.10	43.00	1.63	0.920	43.56
	Shajing, Dali, Yunnan	1000	960	4.00	15.80	0.19	98.80	1.00	0.000	100.00
	Gaojia Village, Lijiang, Yunnan	988	436	55.87	34.90	9.93	71.55	1.10	0.160	85.45
Pumping station										
	Western sluice gate, Chiangjiang	20,000	0	100.00	11.20	0.30	97.32	0.42	0.004	99.05
	Village, Junshan, Hunan									
	Lianhu farm, Danyang, Jiangsu	500,000	0	100.00	10.80	1.31	87.87	0.37	0.024	93.51

middle-lower reaches of Yangtze River. For example, out of five monitoring sites set up in Dongting Lake in 2005, two sites became snail-free by 2010 and the other three sites showed decline of 93%, 24% and 34% in the density of living snail, respectively (Zhu et al., 2011). Similarly, according to the 8-year continuous monitoring in 12 sites in the Poyang Lake areas, the mean density of snail and the density of infected snail decreased by 94.21% and 98.97%, respectively (Chen et al., 2013). Operation of the Three Gorges Dam leads to a considerable decline of water level as well as change of fluctuation dynamics in the two lake areas. Such change disturbed the natural breeding rhythm of snails and thereby caused the decline of the snail density (Chen et al., 2013).

Modification of snail habitats, combined with comprehensive development of agriculture, can not only achieve significant effects in snail control but also produce economic benefit (Duan and Ji, 2004). A comprehensive agriculture project in Hunan Province, including excavation of fishponds, embankment of low marshland for aquaculture, conversion of paddy fields into dry crops farm, cultivation of winter crops, was carried out in marshlands of Dongting Lake (Qing et al., 2003). The density of infected snail in these areas decreased by 93.5% five years later, meanwhile the annual economic benefit reached 80.6 million RMB (Qing et al., 2003). Between 1998 and 2002, an agriculture structure adjustment was implemented in Lipu, Hubei Province, a plain water network and marshland area, including conversion of paddy field into dry crop farm, establishment of big vegetable farms, planting Italian poplars in marshlands, excavation of pond for fishing and seeding (Duan and Ji, 2004). The agriculture project led to decrease of about 91% of snail-infested areas and produced favourable socioeconomic benefits with the net income per capita of 800 RMB (Duan and Ji, 2004).

### 4.2 Molluscicides

Since the 1950s, a variety of chemicals have been used for mollusciciding in The People's Republic of China, for example, nicotinanilide, calcium cyanamide, unslaked lime, trichlorphon, metaldehyde, benzene hexachloride, carbamide, naphthalene, acetbromamide and sodium pentachlorophenate (Table 9). Currently, WHO recommends niclosamide for snail control since its toxicity is lower than others.

### 4.2.1 Niclosamide

Niclosamide was researched and developed by the Bayer (Germany) Company initially. Gonnert and Schranfstatler first reported the molluscicidal

**Table 9** Benefits and side effects of chemicals used in The People's Republic of China for mollusciciding

Molluscicides	Benefit	Side effects	Reference
NaPCP	High effect on mature and young snails as well as eggs	High toxicity to human, livestock, aquatic animals with teratogenic, carcinogenic and mutagenic effect; serious environmental pollution	Zhao and Gao (1997)
Acetabromamide	High effect on mature and young snails as well as eggs; water soluble; low toxicity to human, fishes and shrimps	Easily deliquescent; unstable to heat, acid and alkali; causing dermatitis	Zhu and Yin (1986)
Nicotinanilide	High effect; lower toxicity to fish and no harmfulness to human and livestock; convenient	Low solubility in water; low effect on eggs and climbing snails; expensive; low output	Chen et al. (1991)
Metaldehyde	High effect	Prone to clot; difficult to spray evenly; toxic to human liver and kidney	Gao et al. (1992)

effects of such chemical compound at the 6th International Tropical Medicine and Malaria Conference in 1959 and named it Bayer-73. Chinese scientists synthesized niclosamide in laboratory in 1960 and developed it into a 50% paste. Because of its agglomeration during storage, niclosamide was not widely used in the field until the initiation of World Bank loan project for schistosomiasis control in The People's Republic of China in 1992. This project designated niclosamide, a 50% wettable powder of niclosamide ethanolamine salt (WPN), as the only chemical molluscicide.

To date, wettable powder and suspension of niclosamide have been developed in The People's Republic of China and applied for large-scale

snail control by immersion or spraying methods. The sensitivity of *O. hupensis* to niclosamide remains high even more than two decades of repeated, extensive application for snail control in the main endemic areas of The People's Republic of China (Dai et al., 2014). An experiment of WPN (wettable powder niclosamide) lethal effect on adult snails collected from 37 sites covering all the three types of endemic regions showed that all snails could be killed at 1 mg/L WPN after 24-h immersion or at 0.5 mg/L after 48-h immersion (Dai et al., 2005b). In another study niclosamide also showed high toxicity to eggs and young snails with 100% of mortality at 0.25 mg/L after 24 h (Dai et al., 2005a).

The formulation of niclosamide has been improved according to the environmental features, which makes the application more convenient, effective and cheaper (Table 10) (Yang et al., 2012). WPN shows excellent suspensibility in water and is highly toxic to snails by immersion or spraying methods. Water is normally needed as carrier and hence WPN is used in environments with abundant water. A field study indicated that the mortality rate of snails was 81.1% at 2.0 mg/m<sup>2</sup> using spraying method after 15 days (He et al., 2007). However, WPN is prone to clot during storage and application, which leads to the decline of drug effect and the damage of spraying machine (Dai et al., 2003). Wang (Wang, 2006b) applied WPN at 2 g/m<sup>2</sup> in the field and found that the mortality rates of snails after 3, 5, 7, 15 and 30 days were 71.58%, 62.80%, 69.03%, 90.16% and 95.48%, respectively. Dai et al. (Dai et al., 2003) applied a 25% niclosamide suspension for mollusciciding in the field by spraying method with the concentration of 0.5 mg/m<sup>2</sup>, and found the mortality rates of snails to be 95.77%, 99.07% and 97.29% after 3, 7, 15 days, respectively. When the suspension was used with the concentration of 0.5 mg/L by immersion method, the mortality rate of snails in soil was 95% after 24, 48 and 72 h (Dai et al., 2003). In another study, the 4% WPN by powdering method was used in dried snail habitats and at  $25 \text{ g/m}^2$ ,  $50 \text{ g/m}^2$  and  $75 \text{ g/m}^2$  of this chemical, the mortality rates of snails after 3 days were 84.56%, 93.56% and 98.02%, respectively (Bao et al., 2006).

Dai et al. (2005b) observed in a study that the 25% niclosamide suspension performed better than the 50% WPN; even its active component reduced by half. It was found that the suspension adsorbs on the surface of snail and this improves its lethal effect, and the formulation can be widely used in the complex environments, such as mountain areas or river marshlands, due to its low toxicity, stability, cheap price and convenient operation.

Yang et al. (2012) compared niclosmide suspension emulsion with wettable powder and suspension formulations of niclosamide. When

**Table 10** Comparison of different formulations of niclosamide applied in The People's Republic of China for snail control

Formulation	Active component	Application environment	Using method	Advantages	Disadvantages	Reference
Wettable powder	50% niclosamide ethanolamine salt	Areas with abundant water	Immersion, spraying	Better suspension in water; Excellent effect	Prone to clot; inconvenient; High toxicity to nontarget organism	He et al. (2007)
Powder	4% niclosamide ethanolamine salt	Water-deficient areas	Powdering	Convenient application without water limitation	Optimum effect depending on weather (raining); toxicity to nontarget organism	Dai et al. (2003)
Suspension	25% niclosamide	Complex environments (eg, mountains and river marshlands)	Immersion, spraying	High effect; low toxicity, stable; cheap; convenient	Toxicity to nontarget organism	Dai et al. (2008)
Suspension emulsion	50% niclosamide ethanolamine salt	Large-scale spraying in the field	Immersion, spraying	High effect; soluble in water	Organic solvent; expensive; environment pollution; toxicity to nontarget organism	Dai et al. (2005b)
Naro preparation	25% niclosamide	Water-accessible areas	Immersion, spraying	High effect; soluble in water; applicable in cold weather	Toxicity to nontarget organism	Jiang et al. (2007) and Guo et al. (2006)
Slow release preparation	5%, 10%, 50%, 70% niclosamide ethanolamine salt	Water-deficient areas	spraying	Long-term effect; convenient	Environmental pollution due to slow degradation; toxicity to nontarget organism	Kenawy and Rizkel (2004)

suspension emulsion of niclosamide with the concentration of 0.12 mg/L was applied, its effect was higher than that of wettable powder, while there was no difference with that of the suspension. The suspension emulsion of niclosamide is suitable for use in large-scale field spraying due to its excellent mollusciciding effect and water solubility. The effect of niclosamide naro-preparation was similar to WPN, and it can be used without the temperature limitation (Guo et al., 2006; Zhang et al., 2007b). The active time of slow-release preparations of niclosamide was longer to 16 days than that of the common WPN, and the mollusciciding effect went up with the decline of water hardness and rise of water pH value (Kenawy and Rizkel, 2004).

### 4.2.2 Novel molluscicides for snail control

Although niclosamide shows good molluscicidal efficacy, its toxicity to fish and other aquatic animals as well as environmental pollution is considerable. Therefore, development of environment-friendly, highly effective and low toxic chemical molluscicides is always the priority (Xia et al., 2014). Many plant species have been confirmed to contain components of molluscicidal activity against *O. hupensis* (Chen et al., 2012a; Guo et al., 2011; Xin et al., 2006; Yang et al., 2008b). Plant molluscicides, such as Rongbao and Luowei, show high lethal effect on snails and overcome the disadvantages of niclosamide.

### 4.2.2.1 Rongbao

'Rongbao' is a kind of alkaline nitrogen waste and appears as black and grey powder. The active component is calcium cyanamide. 'Rongbao' has the similar lethal effect on snails as niclosamide but a better long-term efficacy than niclosamide. Furthermore, the molluscicide shows low toxicity to fish and other aquatic animals (Liu et al., 2007; Lu et al., 2006). A laboratory experiment showed that all Oncomelania snails died when immersed in a solution containing 80 mg/L Rongbao for 2 days or sprayed with the molluscicide of 30 g/m<sup>2</sup> for 1 day. Field study also showed good effect; the mortality rate of snails was 97.18% when the snails were immersed with Rongbao of 50 mg/L for seven days, without observing the death of other aquatic animals. When 30 g/m<sup>2</sup> of Rongbao was applied by spraying and powdering in the field, the mortality rates of snails after 15 days were 74.15% and 81.11%, respectively (Li et al., 2006a). Due to the high cost of Rongbao, niclosamide still is the only molluscicide widely used in The People's Republic of China for snail control. However, because of the low toxicity to fish, Rongbao can be applied in small scale in aquaculture areas (Gu et al., 2008).

### 4.2.3 Luo-wei

'Luo-wei' is a novel plant molluscicide obtained by extracting triterpenoid compound from camellia. A 4% yellow 'Luo-wei' powder (TDS) was developed in The People's Republic of China. The active component is saponins (Tang et al., 2010). The application of the powder in different types of endemic areas by either immersion or spraying is highly toxic to O. hupensis, similar with that of niclosamide ethanolamine salt (WPN). After 1, 3, 7, 15 days of TDS application, the mortality rates of snails increased over time, and the density of living snails at 15 days was significantly lower compared with density before the application (Jia et al., 2013; Sun et al., 2010; Tang et al., 2010). Toxicological test indicated that TDS is highly toxic to fish but low to birds. Decapods are highly sensitive to TDS spraying, while the risk of immersion is low. Hence, the mollusciciding methods should be adopted according to the distribution of fish, bird and decapod in snail-occupied areas. The spraying method on dry land is recommended due to its low toxicity (Sun et al., 2010). However, 'Luo-wei' needs more toxicological tests before it can be used for snail control in field in The People's Republic of China.

# 5. CHALLENGES AND PRIORITIES

A great achievement in schistosomiasis control has been made in The People's Republic of China in the past six decades (Zheng et al., 2013). By the end of 2014, out of a total 453 schistosome-endemic counties, 313 had reached the criteria of transmission interruption, 135 reached transmission control and only five are still at the epidemic control stage (Lei and Zhou, 2015). Around 8000 patients confirmed by stool examination were reported with only two acute cases in 2014 (Lei and Zhou, 2015). However, the snailinfested area is still as large as 3.64 billion m<sup>2</sup> (Lei and Zhou, 2015). During the implementation of a 10-year national programme of schistosomiasis control since 2004, the snail-infested area in The People's Republic of China only decreased by 5.23% from 384,599.97 hm<sup>2</sup> in 2004 to 365,468.00 hm<sup>2</sup> in 2013 (Hao et al., 2005; Lei et al., 2014). Although the decline of snailinfested area is not significant, snail control programmes played an important role to reduce the density of snails in endemic areas and thus significantly lowered the risk of schistosomiasis transmission. According to national statistics in 2013, the area of chemical mollusciciding and environment modification for snail control were 1.344 billion m<sup>2</sup> and 5800 hm<sup>2</sup>, respectively.

Wide distribution of snail and low coverage of chemical mollusciciding are the main reasons for the current status, which are the key factors to hinder the process of schistosomiasis elimination in The People's Republic of China. The People's Republic of China still faces big challenges to achieve the goal of schistosomiasis elimination nationally, and the following research and application priorities should be strengthened to provide scientific support for that goal (Lei and Zhou, 2015; Zheng et al., 2013).

# 5.1 Development of innovative techniques

O. hupensis, the only intermediate host of S. japonicum, plays a vital role in schistosomiasis transmission. One key index for schistosomiasis elimination in the newly issued 'criteria for control and elimination of schistosomiasis' is that no infected snail can be found for successive 5 years. Therefore, monitoring snail habitats or high risk zones should be emphasized in the endemic surveillance system towards schistosomiasis elimination in The People's Republic of China. During the last 10 years The People's Republic of China implemented an integrated strategy with a focus on the infection sources control. This strategy successfully reduced patients and ill livestock and thus weakened the role of humans and livestock in the schistosomiasis transmission. However, several dozens of wild animals can serve as reservoirs of S. japonicum and their capability in transmission is not yet evaluated in field. Moreover, population migration is more and more frequent in The People's Republic of China. All these factors increase the possibility of snail infection with schistosome (Liu et al., 2013; Xiong et al., 2014). In addition, owing to lack of modern tools, snail survey in The People's Republic of China still relies on manual work with the disadvantages of high labour intensity, high cost and low coverage. Monitoring and warning systems should be based on sensitive early detection and modern information techniques. Therefore, more practical and sensitive techniques of snail survey need to be developed to improve monitoring level on snail habitats, especially simple and rapid methods for detection of infected snails. And a dynamic monitoring and warning system and technique on snail, which can identify, simulate, predict and evaluate influence of environmental factors and infection sources on snail distribution, also need be established to improve the sensitivity and efficiency of snail detection and surveillance in epidemic areas.

# 5.2 Research on control strategy

Killing of snails is one of the key strategies for schistosomiasis transmission control as well as one of the measures of The People's Republic of China's

national antischistosomiasis programme. Environmental modification is the fundamental measure for snail control (Yang et al., 2016). However, it has its shortcomings of huge one-time investment and less scope of application. With the development of social economy and the construction of new rural villages in The People's Republic of China, more and more ecological methods for snail control will appear, for example, the projects of agriculture, forestry, water conservation and tourism for snail control (Zheng et al., 2013). At present, the application of chemical molluscicides is still the main method for snail control in The People's Republic of China, but these chemicals need to be used repeatedly, and have toxicity to aquatic animals causing economic loss and affecting the development of aquaculture industry (Dai et al., 2007; Sun et al., 2010; Tang et al., 2015). Therefore, there is a need to speed up the development of novel molluscicides with high efficiency, low cost, user friendliness and low toxicity to nontarget organisms; or new biological approaches for snail control, especially, environment-friendly and benefit-oriented ecological mollusciciding should be developed.

### 5.3 Theoretical research

Research on biological theory of S. japonicum and Oncomelania needs to be further strengthened to develop new strategy and technology for snail control. As studies based on morphology, anatomy, protein and DNA tests have shown, certain genetic diversity exists in Oncomelania in mainland The People's Republic of China, and Oncomelania interspecific differentiation is possible; the susceptibility of Oncomelania to schistosome infection may be under control of its genetic materials, which cause significant differences in infection rates of snails from different areas when infected with geographical strain of S. japonicum from the same or different areas (He et al., 1990; Huang et al., 2002; Jiang et al., 2007). With the development of genomics, transcriptomics and proteomics, studies on classification of infra-species, different geographical strain, even sex of Oncomelania and interspecific difference of Oncomelania need to be further pursued. Also, research on host parasite relationship between S. japonicum and Oncomelania needs to be strengthened, including genetic difference between Oncomelania and S. japonicum, molecular basis of gene co-evolution and symbiosis, biological characteristics and immunological mechanism of susceptibility difference of Oncomelania for geographical strains of S. japonicum, functional gene of Oncomelania susceptibility and transmission threshold of Oncomelania. These studies would likely contribute to the development of a schistosomiasis vaccine and chemical molluscicides.



Oncomelania hupensis, the only intermediate host of S. japonicum, shows high diversity, which makes it possible to widely distribute and explore complex landscapes in The People's Republic of China. Although routine chemical mollusciciding did lower the density of snails and thus the risk of schistosomiasis transmission, the area of snail habitats did not change as expected in the last 10 years. Along with great achievement in schistosomiasis control in humans and domestic animals, snail control should be highlighted in future control programmes. The next 10-year national plan (2016—2025) towards schistosomiasis elimination has been formulated and snail control is one of the fundamental measures (Liu et al., 2014). Facing the challenges in snail control, it is urgent to develop high-effective, low-toxic, cheap and user-friendly chemical molluscicides, to research novel, long-standing and thoroughgoing methods for snail control, and to establish sensitive and real-time monitoring and warning techniques in low schistosome-endemic areas (Tambo et al., 2014).

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