

Chapter 2

Treatment Methods of Traditional Chinese Medicines Against Intestinal Protozoan Infections

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Abstract Protozoa that parasitize the human intestine include *Entamoeba histolytica*, *Blastocystis hominis*, *Trichomonas hominis*, *Cryptosporidium parvum*, *Giardia lamblia*, *Balantidium coli*, *Isospora*, and other protozoa. Most intestinal protozoan infections can cause acute or chronic diarrhea in healthy individuals and may result in intractable, life-threatening illness in patients in immunosuppressive status. Collectively, they infect over one billion people each year. *E. histolytica* can invade the gut epithelium and subsequently other organs, where it forms abscesses. *E. histolytica* infection (amoebiasis) is the second leading cause of death from parasitic diseases. *Cryptosporidium* and *Giardia* are the most common parasitic causes of diarrheal illness worldwide, especially in the developed countries, where they were associated with waterborne outbreaks. In addition, *Cryptosporidium*, *Entamoeba*, and *Isospora* have been most frequently identified as the most commonly implicated pathogens which cause persistent diarrhea in patients with HIV, followed by *Giardia* and *Strongyloides*. Clinically, the nitroimidazoles class of agents including metronidazole, tinidazole, ornidazole, etc, is considered as the front line of defense against intestinal protozoan infections in the world. However, drug-resistant intestinal protozoa (such as *E. histolytica* and *Giardia*) have appeared in clinical and laboratory isolates regularly. Accordingly, resistant strains have been treated with longer repeat courses or higher doses of the original agent in clinic. Chinese medicine therapeutic approaches have been employed for intestinal disease caused by intestinal protozoa for a long time in China and Asian countries, with low side effects compared with western medicine. Several Chinese medicines including *Radix Pulsatillae*, *Fructus Bruceae*, *Allium sativum*, *Radix Sophorae Flavescentis*, *Fructus cnidii*, etc, are shown to be effective for anti-protozoa. This review presents the current advances in traditional Chinese medicines with a focus on effective treatment and control of intestinal protozoan infections.

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2.1 Introduction

E. histolytica is an intestinal protozoan that causes invasive amoebiasis in 40–50 million people, resulting in up to 100,000 deaths globally every year (Ali et al. 2008). *E. histolytica* infections occur worldwide but are more prevalent in the tropics. Recognized high-risk groups include travelers, immigrants from endemic areas, immunocompromised individuals, and men who have sex with men (MSM) (Weinke et al. 1990). About 90 % infections with *E. histolytica* are asymptomatic, only 10 % of infected individuals develop symptoms including amoebic colitis and amoebic liver, lung or brain abscess. In China, it was estimated that ten million persons are infected by *E. histolytica* by fecal–oral transmission, particularly distributing in Xizang, Yunnan, Xinjiang, Guizhou, Gansu Provinces according to epidemic survey data about 1988–1991 (Jiang et al. 1997) (Table 2.1).

Nitroimidazoles, particularly metronidazole, has been the mainstay of therapy for acute and chronic invasive amoebiasis since the 1960s. After treatment with metronidazole and other nitroimidazoles (such as tinidazole, ornidazole, and secnidazole), luminal amebicides such as Diloxanide furoate, paromomycin, and iodoquinol are recommended to eradicate intestinal colonization and prevent relapse (Reed 2000). Noninvasive infections may be treated with paromomycin and Diloxanide furoate as first-line and second-line agent, respectively. However, Nitroimidazoles have many side effects, such as primarily gastrointestinal, disulfuram-like intolerance reaction, and neurotoxicity (Haque et al. 2003).

In addition, some other protozoa such as *Giardia lamblia*, *Cryptosporidium parvum*, *Trichomonas hominis*, *Blastocystis hominis*, and *Balantidium coli* are also prevalent in China. Nevertheless, the infection rate of intestinal protozoa declines apparently in the last 10 years. In Xinjiang Province, which is regarded as the province with higher infection rate of intestinal protozoa in China, the rate of decline of *G. lamblia* and *B. hominis* were 85.65 and 58.60 %, respectively, in 2003 (Maimaitijiang et al. 2010). There is no report about human coccidiosis in China. Metronidazole and other nitroimidazoles have been frequently used as the mainstay of chemotherapy of these intestinal protozoa diseases in China.

In China, traditional Chinese medicines have been employed to treat parasite disease for several thousand years. Here the review presents the current advances in traditional Chinese medicines with a focus on effective treatment and control of intestinal protozoa infections.

2.2 *Radix Pulsatillae*

Radix Pulsatillae, the dried root derived from *Pulsatilla chinensis* (Bge) Reg, was recorded to treat dysentery widely in Chinese classic medicine books. As early as the 1950s, it was used in the treatment of amoebiasis in China. Lab researches

Table 2.1 The infection rate ($\pm$ SE) of *E. histolytica* in Province/Autonomous region/Municipality (Jiang et al. 1997)

Province	Infection rate ($\pm$ SE), %	No. county examined	No. positive	Positive rate
Xizang	8.12 (1.86)	13	11	84.62
Yunnan	2.54 (0.50)	28	25	89.29
Xinjiang	2.37 (0.58)	23	21	91.30
Guizhou	2.25 (0.36)	24	23	95.83
Gansu	2.04 (0.56)	19	15	78.95
Hainan	1.58 (0.43)	5	4	80.00
Neimenggu	1.80 (0.42)	21	13	61.90
Tianjin	1.73 (0.36)	5	5	100.00
Zhejiang	1.50 (0.33)	29	24	82.76
Guangxi	1.49 (0.20)	20	19	95.00
Hebei	1.47 (0.32)	31	23	74.19
Shangdong	1.10 (0.10)	35	31	88.57
Jiangsu	0.89 (0.13)	33	28	84.85
Hunan	0.89 (0.15)	30	25	83.33
Jiangxi	0.88 (0.12)	23	20	86.96
Sichuan	0.83 (0.15)	45	39	86.67
Fujian	0.69 (0.13)	26	25	96.15
Anhui	0.59 (0.10)	22	20	90.91
Henan	0.57 (0.06)	39	30	76.92
Qinghai	0.48 (0.22)	23	9	39.13
Hubei	0.44 (0.08)	31	22	70.97
Shanxi	0.40 (0.08)	25	17	68.00
Liaoning	0.36 (0.11)	29	16	55.17
Guangdong	0.34 (0.05)	31	26	83.81
Beijing	0.29 (0.05)	9	8	88.89
Heilongjiang	0.17 (0.04)	26	12	46.15
Shanxi	0.15 (0.04)	26	11	42.31
Ningxia	0.10 (0.03)	20	1	5.00
Jilin	0.04 (0.03)	25	1	4.00
Shanghai	0.01 (0.004)	10	1	10.00
Total	0.95 (0.04)	726	525	72.31

demonstrated saponin extracted from *Radix Pulsatillae* and its decoction could inhibit the growth of trophozoite of *E. histolytica* in vitro and in animal model with low toxicity (Jiang et al. 1958; Lan et al. 1996) (Fig. 2.1).

The earlier clinical studies showed that the efficacy of *Radix Pulsatillae* against amoebiasis could reach up to above 90 %, administrated with 30 g orally, 10 days as one course. The high efficacy was ascribed to the powerful eradication of *Radix Pulsatillae* against trophozoite and cyst (206 Hospital 1960). Combined prescription is a method to combine several Chinese medicines guided by the idea of therapy with syndrome differentiation. *Radix Pulsatillae* decoction based on *Radix Pulsatillae*, consisting of *Radix Pulsatillae*, *Cortex Fraxini*, *Phellodendron Amurense Rupr*, *Rhizoma Coptidis Chinensis*, *Lonicera Japonica Thunb*, *Viola Yedoensis Mak*, and *Radixet Rhizoma Rhei*, was demonstrated to be an effective



Fig. 2.1 *Radix Pulsatillae* and *Pulsatilla chinensis* (Bge) Reg. *Radix Pulsatillae* is the dried root of *Pulsatilla chinensis* (Bge) Reg. of family Ranunculaceae, with slightly bitter and astringent taste. The best quality of *Radix Pulsatillae* is thick and long in regularity, grayish yellow of superficial, white villus at the top of it. The cold property of the herb is good at removing the stagnated noxious heat in the intestine and is effective in cooling blood and detoxicating

combined prescription to treat intestinal amoebiasis by oral administration and colocolysis with 90 % efficacy (Cai 1995). Beyond that, *Radix Pulsatillae* is also active to inhibit the propagation of *B. hominis* in vitro, suggesting potential beneficial effects in *B. hominis* infections (Zhang et al. 1997).

2.3 *Fructus Bruceae*

Fructus Bruceae, the fruit of *Brucea javanica* (L.) Merr, is distributed in the south of China. It is a traditional Chinese medicine to treat dysentery and malaria. It contains many chemical compositions, such as Brucealin, Brucamarine, Ratanine, etc. Crude glycosides and Brucealin (a glycoside), extracted from *Fructus Bruceae*, are tested to be effective chemical components to inhibit the growth of trophozoite of *E. histolytica* and kill it directly in vitro (Quan 1948; Song 1949; Yang and Ou 2001) (Fig. 2.2).

The first published report of successful use of *Fructus Bruceae* against human amoebiasis was in 1937 (Liu 1937). Unique in that it is effective both in acute and chronic amebic infection, *Fructus Bruceae* has been reported to eradicate up to 54–90 % of intestinal infections (Liu 1941; Wu 1943). Meanwhile, clinical studies suggested it was more efficacious for the combination of colocolysis and oral administration with *Fructus Bruceae* than single oral administration (Feng 1957). Based on the main function of *Fructus Bruceae* against *E. histolytica*, several combined prescriptions are found to be highly effective with 95.8–100 % efficacy. This statement has support from studies 14 amoebiasis cases treated by combination with *Fructus Litseae* and 24 patients treated by Jiedushenghua Dan, which consists of *Fructus Litseae*, *Lonicera Japonica* Thunb, *Glycyrrhiza Uralensis*

Fig. 2.2 *Fructus Bruceae* and *Brucea javanica* (L.) Merr. *Fructus Bruceae* consists of the dried ripe fruits of *Brucea javanica* (L.) Merr. The bitter and cold herb has been used in clearing heat, expelling toxins, checking malaria, treating dysentery, etc.



Fisch, *Radix Notoginseng*, and *Radix Paeoniae Alba* (Jingzhou Infectious Hospital 1959; Chen 1987).

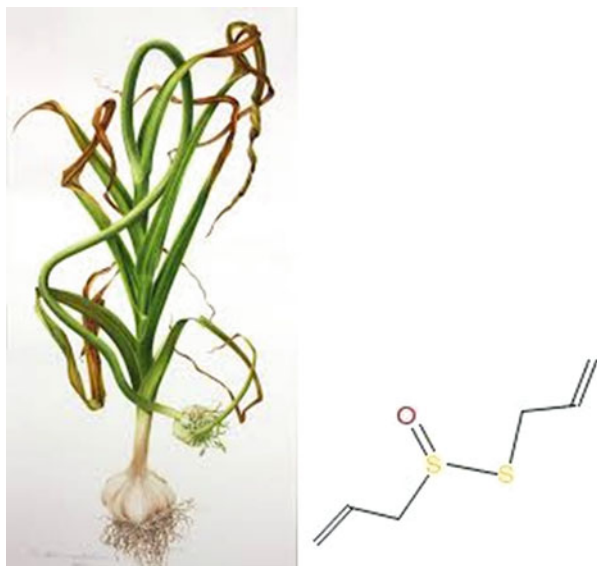
2.4 *Allium sativum* and Allicin

In addition to being a secondary ingredient for dish, *Allium sativum* (garlic) has traditionally been prescribed as a remedy for intestinal disorders (Adetumbi and Lau 1983). Precious clinical studies demonstrated that *Allium sativum* was efficacious against *E. histolytica* by oral administration and by coloclisis with fresh liquid of it. Meanwhile, it played the role in preventing amoebiasis (Ren et al. 1951; Zhang 1953). Allicin [*S*-(2-propenyl) 2-propene-1-sulfinothioate], which is one of the active principles of freshly crushed *Allium sativum* homogenates, has been shown to have a variety of antimicrobial and antitumor effects. Lab researches indicated Allicin could inhibit strongly the growth of trophozoite of *E. histolytica* by inhibiting its cysteine proteinases and alcohol dehydrogenase which are the significant contributors to amebic virulence and the enzyme required for the survival of the parasite (Mirelman et al. 1987; Ankri et al. 1997). In addition, Allicin was carried out against *Cryptosporidium* successfully in infant cases, showing its killing effect on *Cryptosporidium* and improving immune function effectively. Dosing is usually 80 mg four times a day for under 1-year-old infants, 90 mg for 1–2-year-old children, 120 mg for over 3-years-old children, 7 days as a course (Ge et al. 1991; Zheng et al. 2000) (Fig. 2.3).

2.5 *Radix Sophorae Flavescentis*

Radix Sophorae Flavescentis, a classic heat-clearing and dampness-eliminating herb, has widely been used as antimicrobial, antiparasitic, and antitumor medicine in China. It contains some chemical compositions, such as alkaloid, ethanol extract, and flavonoid which have anti-*Giardia* activity with the same efficacy in comparison with metronidazole in vitro. However, the mechanism of killing of *Giardia* is

Fig. 2.3 *Allium sativum* (garlic) and formation of Allicin. *Allium sativum*, commonly known as garlic, is a species in the onion genus, *Allium*. It has been used for both culinary and medicinal purposes. It has anticholesterol, antibacterial, antiviral, antibiotics, anti-HIV, and antifungal activities. Allicin [*S*-(2-propenyl) 2-propene-1-sulfinothioate], which is one of the active principles of freshly crushed *Allium sativum* homogenates, has been shown to have a variety of antimicrobial and antitumor effects



not thoroughly studied. It is just found that morphological changes in the trophozoites are effected by *Radix Sophorae Flavescentis*, such as separation of adhesive disk from the cell body and appearance of pit in cell membrane (Lu et al. 1993; Wu et al. 1994). Clinical studies demonstrate that *Radix Sophorae Flavescentis* could treat giardiasis with 92 % successful rate (Chen et al. 1965) (Fig. 2.4).

Previous reports also showed that combination drug therapy of *Radix Sophorae Flavescentis* is more effective in treating *Trichomonas intestinalis*. For example, it could be combined with *Fructus cnidii*, *Dictamnus Dasycarpus Turcz.*, and *Phellodendron Amurense Rupr* or combined with *Fructus cnidii*, *Cortex Fraxini*, *Radix Stemonae Sessilifoliae*, and Sijunzi decoction. The efficacy could reach as high as 95.65–100 % (Tang 1987; Qiu 2002). However, the therapeutic effect of *Radix Sophorae Flavescentis* alone on *T. hominis* is not confirmed.

Radix Sophorae Flavescentis combined with *Radix Astragali*, which is considered as an improving immune function Chinese medicine, was used to treat Cryptosporidiosis in Children. The two Chinese medicines may have repelling effect on the *Cryptosporidium* and be helpful to improve cellular immunity (Li et al. 1993). Matrine, a main active alkaloid extract from *Radix Sophorae Flavescentis*, has potential antineoplastic, antifibrotic, and anti-inflammatory activities. Lab studies clearly demonstrated that it could significantly reduce the number of *Cryptosporidium* oocysts and *Cryptosporidium*-infected cells in mouse model. Meanwhile, it could improve integrity of cell membranes and of the mucosal barrier, suggesting a potential in therapeutic applications against *Cryptosporidium* infection (Chen and Huang 2012).



Fig. 2.4 *Radix Sophorae Flavescentis* and *Sophora Flavescens* Ait. *Radix Sophorae Flavescentis* is the root of *Sophora flavescens* Ait. of family Leguminosae. It is extremely bitter in flavor and cold in nature. The cold property is capable of clearing heat. The bitter flavor could remove the dampness and purge fore downward. In addition, it could destroy intestinal parasites and induce diuresis

Kushe Decoction, consisting of *Radix Sophorae flavescentis* and *Fructus cnidii*, was successfully applied to treat human *Balantidium* infection on three cases and showed a clinical efficacy of 100 %, which was similar to metronidazole. The decoction could be used by oral and coloclysis administration (Su 1978).

2.6 *Fructus cnidii*

Fructus cnidii is the fruit of *Cnidium monnieri* (L.) Cusson. It is a classical heat-clearing and dampness-eliminating Chinese medicine with broad-spectrum activity against protozoa (*T. vaginalis*), helminthes (*Ascaris lumbricoides*), and some bacteria. It is widely used against *T. vaginalis* in China (Zhang et al. 1996) and usually combined with other Chinese medicines to treat *Trichomonas intestinalis* (Tang 1987; Qiu 2002). In vitro susceptibility testing demonstrates that *Fructus cnidii* has activity against trophozoite of *E. histolytica*, but the activity is lower than that of *Radix Pulsatillae* and *Artemisa annua* (Lan et al. 1996) (Fig. 2.5).

2.7 Other Traditional Chinese Medicines in Treatment for Intestinal Protozoan Infections

In addition to Chinese medicines discussed above, there are a few studies with other traditional Chinese herbs which also could treat intestinal protozoan infections. *Cortex Magnoliae Officinalis*, considered as a classical dampness-eliminating Chinese medicine and having antimicrobial effect, could treat intestinal amoebiasis alone successfully. Clinical trial had employed dosing two times daily (6 g/dose) for 4–9 days and shown effectiveness in 93.5 % in 46 adult patients (Sun 1960).



Fig. 2.5 *Fructus cnidii* and *Cnidium monnieri* (L.) Cuss. *Fructus cnidii* is the dried ripe fruit of *Cnidium monnieri* (L.) Cuss. of family Umbelliferae, with special aromatic smell, pungent, and cool taste. The best quality is yellowish green in color, pungent, and spicy smell under twisting and full-stacked. It could dry dampness, kill parasites, and relieve itching for external application, dispel cold, and dry dampness for internal application

Herba Calthae Membranaceae, an aquatic plant, is regarded as a delicious food and an antimicrobial Chinese medicine herb. Freshly chopped *Herba Calthae Membranaceae* is effective against *E. histolytica* (Wang and Liao 1962). It is given as 2–3 doses per day (20 g/dose) for 5 days, with obvious benefits regarding cost. *Radix et Rhizoma Thalictri*, having broad-spectrum antibacterial activity, has generally been considered as therapeutic Chinese medicine to treat dysentery. Depending on its shorter course and lower recurrence rate, it has been shown to be more potent against *E. histolytica* than emetine (Yunnan Kaiyuan Hospital 1959). *Folium Baeckeeae* is a heat-clearing, detoxifying, and diuretic herb. The oil of *Folium Baeckeeae*, distilled from the leaves of *Folium Baeckeeae*, has been demonstrated 93.9 % effective in 115 patients infected with *T. hominis* (Beihai Town Hospital 1978) (Figs. 2.6 and 2.7).

In conclusion, Chinese medicine has been used for treating intestinal protozoan infections in human populations for a long time in China and other Asian countries. However, the anti-protozoa mechanism of most of Chinese medicines is not fully elucidated, which limits the widespread use in the world. At present, clinically resistant strains have appeared with longer repeat courses or higher doses of the original western medicine. It seems Chinese medicine may be the efficacious means of eradicating the intestinal protozoan infections to avoid potential parasite resistance.



Fig. 2.6 *Cortex Magnoliae Officinalis* and *Magnoliae Officinalis*. *Cortex Magnoliae Officinalis* is the dried bark of *Magnoliae Officinalis* of family Magnoliaceae, with pungent and bitter flavor. It is a deciduous tree growing to 20 m in height. The thick and brown bark is stripped from the stems, branches, and roots. It could eliminate dampness, relieve food stagnancy, clear away phlegm, and relieve asthma



Fig. 2.7 *Radix et Rhizoma Thalictri* and *Thalictrum foliolosum* DC. *Radix et Rhizoma Thalictri* is the dried root of *Thalictrum foliolosum* DC and *T. cultratum* Wall of family Ranunculaceae. It looks like horse tail. It could clear heat, dry dampness, and remove toxicity

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