



## Review

## Chinese dark teas: Postfermentation, chemistry and biological activities



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## ABSTRACT

Chinese dark teas (CDTs) are post-fermented tea products, which are mainly produced in Southwestern China. The health benefits and chemistry of CDTs are increasing trends in the research field of teas. Deactivated leaves of *Camellia sinensis* and *Camellia assamica* are post-fermented under controlled conditions to make CDTs, the quality of which is dependent on the microorganisms like *Aspergillus*, *Penicillium* and *Eurotium* species in postfermentation process. It has been proved that CDTs have anti-obesity effects with respect to decreasing the total serum cholesterol, triglyceride, low density lipoprotein cholesterol (LDL-C) by inhibiting the lipid absorption and biosynthesis. Furthermore, CDTs possess antimicrobial, antioxidative and antimutagenic activities. Besides the health benefits, the safety of CDTs was assessed by acute and chronic toxicity evaluation. Postfermentation structurally changes the original compounds of raw CDTs, significantly decreases the contents of catechins and forms some novel catechins derivatives. In the present paper, we review the postfermentation characteristics, biological activities, chemical constituents and composition analysis of CDTs.

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## 1. Introduction

Chinese dark teas (CDTs) are post-fermented teas, the history of which should be dated back to the Ming Dynasty at 1500 A.D. In the history, tea was produced in Southwestern China, and then carried via mountain to Tibet, Xinjiang. These teas were packed and shaped into brick, mushroom, square, bowl and cake appearance so as to be convenient for transportation. After long time transportation, the color of tea became darker, and flavor was not astringent any more. Compared with other teas, the manufacture technology of CDTs is

**Abbreviations:** CDTs, Chinese dark teas; LDL-C, low density lipoprotein cholesterol; DGGE, gradient gel electrophoresis; PTP, pu-erh tea powder; SOD, superoxide dismutase; HepG2, hepatoma; TC, total cholesterol; TG, total triglycerides; GA, gallic acid; t-BHP, tert-butyl-hydroper-oxide; BTE, black pu-erh tea extract; WEPT, Water extracts of pu-erh tea.

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unique for the “piling store”, which is the postfermentation process. Raw CDTs are naturally stored for many years (usually more than five years) to make aged CDTs, the quality of which is believed to be improved correspondingly with stored years. Alternatively, raw CDTs are post-fermented under controlled conditions of high humidity and temperature to make ripened CDTs. The expression of CDT products was varied according to the interpretation preference. Actually, post-fermented, ripened, post-processed pu-erh teas are the same. Correspondingly, crude, aged and fermented pu-erh teas have the same meaning. Usually, the raw material of aged or ripened CDTs was named as raw CDTs or green tea like CDTs. To clearly understand the differences of CDTs, the manufacturing processes of various teas are profiled in Fig. 1.

In the regions inhabited by China's minority nationalities, high-calorie foods are always consumed with less dietary fiber because of their climate feature. In these places, CDTs are food ingredients and drunk simultaneously with high fat milk or red meat to balance the absorption of cholesterol and fat (Dang, Yan, Yamamoto, Wang, & Zeng, 2005). Sometimes, CDTs are cooked with ghee to make buttered tea, which is a daily beverage in Tibet.

From the studies of CDTs, the microorganisms involved in the postfermentation process have been proved to be an important factor in forming the quality and flavor of CDTs. It was reported that various CDTs including pu-erh tea (Yunnan province), fuzhuan tea (Hunan province), jinjian tea (Sichuan province), qingzhu tea (Hubei Province) and heima tea (Hunan province) contained many kinds of microorganisms (Abe et al., 2008; Van Diepeningen et al., 2004). Most of the prevailing microorganisms have been identified as *Aspergillus*, *Penicillium*, and *Saccharomyces* genus.

During the postfermentation processing, chemical analysis showed that the changes of major compounds were directly related to the fungi. In the view of chemical constituents, CDTs are highly different from other teas such as green tea (unfermented tea), black tea (fully-fermented tea) and oolong tea (semi-fermented tea) (Syu, Lin, Huang, & Lin, 2008). The contents of catechins and L-theanine in CDTs were much less than those of the unfermented and semi-fermented teas. Although the catechins levels in black tea were similar to those of CDTs, the representative catechins oxidation products in black tea, theaflavins and thearubigins were undetectable in CDTs (Xie et al., 2009). Some studies proposed that the marker constituent of pu-erh tea was theabrownine, but the exact structure of which has not yet been clarified (Tan, Peng, Gao, & Gong, 2012; Wang, Liu, et al., 2011). Furthermore, a kind of antilipemic statins, lovastatin was detected in

pu-erh tea (Yang & Hwang, 2006). All of these studies showed that the chemistry of CDTs was more complicated than others.

In this review, we survey the state of our understanding of CDTs in the field of postfermentation characteristics, pharmacological effects and chemical studies. We also highlight key experimental challenges that must be confronted to advance our understanding in this area and consider how existing knowledge might be harnessed to improve public health by CDTs.

## 2. Microorganisms during postfermentation of CDTs

Microorganisms accelerate the postfermentation process of ripened CDTs within several months to attain a similar flavor to aged CDTs stored for many years. There are many reports about the isolation and identification of these microorganisms during postfermentation of pu-erh tea. We review the microorganism characteristics in varieties of CDTs as follows.

### 2.1. Microorganisms of pu-erh tea

Pu-erh tea has become one of the most famous CDTs, and attracted consumers worldwide. The raw material of pu-erh tea is the mature leaves of *Camellia assamica*, which is also used for making black tea in India. Besides, other CDTs are made of leaves of *Camellia sinensis*. According to the classification criteria of processing, pu-erh tea should be considered as a variety of CDTs regardless of the source of raw material. After all, the common process, postfermentation by microorganism is the most important process of making CDTs.

Molecular biology methods have been employed in the microbial studies on microorganisms of pu-erh tea. Yunnan province of China is the original production place of pu-erh tea. During the traditional postfermentation, no strains were artificially introduced into the piled raw CDTs. The microorganisms found in pu-erh tea are naturally transported from air and water in situ. In these years, more and more fungi were isolated and identified from pu-erh tea. Zhao et al. isolated 385 strains, 62 of which were identified with internal transcribed spacer (ITS) region 4 and ITS5 analysis (Zhao, Tong, Zhou, Wang, & Liu, 2010). Abe et al. identified two major fungi using denaturing gradient gel electrophoresis (DGGE) (Abe et al., 2008). Other studies also reported plentiful fungi in pu-erh tea. Table 1 lists the main microorganisms in pu-erh tea.

Recently, some single fungus or mixed fungi isolated from aged pu-erh tea were industrially used to ferment raw pu-erh tea. It was

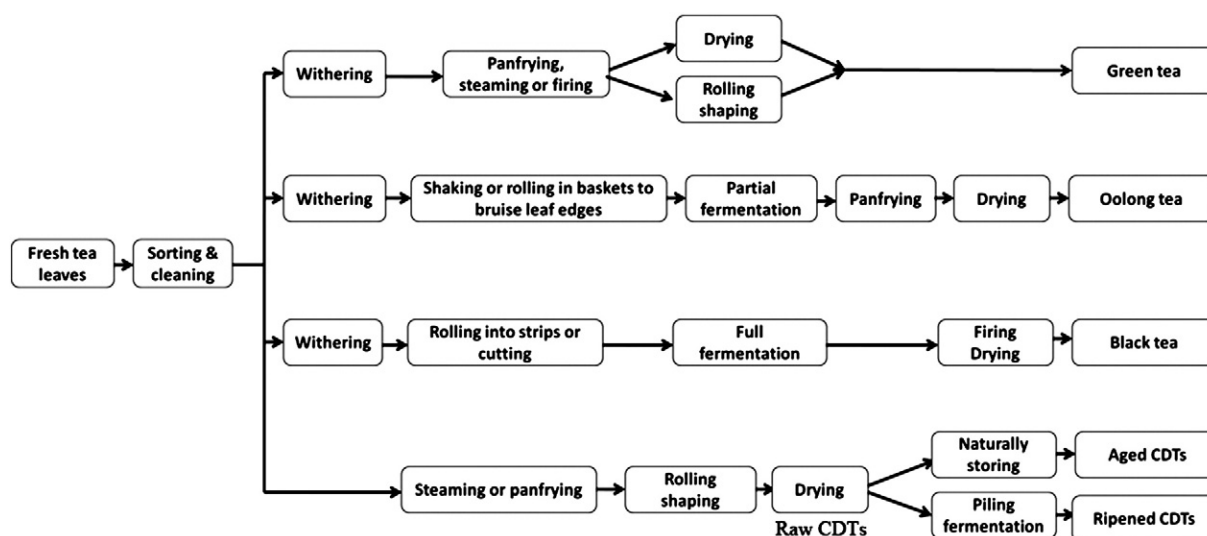


Fig. 1. Main manufacture process of various teas.

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