Postprint: Study on Components, Antioxidant Activity, and Hypoglycemic Activity of Water Extracts from Different Varieties of Raw Tea

Authors: Huo Huazhen, Cai Aihua, Guo Chunyu, Xie Yunchang, Dianpeng Li, Cai Aihua

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Abstract

This study compared the differences in active constituents and in vitro antioxidant and hypoglycemic activities of water extracts from seven different varieties of raw tea, determined the correlations between various components and activities, and provided a scientific basis for raw tea material screening and processing method selection in developing Liupao tea products with superior antioxidant and hypoglycemic activities. The contents of total polyphenols, total flavonoids, and tea polysaccharides in raw tea water extracts and their concentrates were measured. Antioxidant and hypoglycemic activities were evaluated based on DPPH· scavenging capacity, ORAC values, and inhibition of α-glucosidase and α-amylase, with Pearson correlation analysis employed. The results demonstrated: (1) Significant differences existed among the seven varieties in water extract, total polyphenol, total flavonoid, and tea polysaccharide contents, with the highest values observed for Huangjin tea (53.42% $\pm$ 0.14%), Guihong No. 4 (40.87% $\pm$ 1.09%), Yunnan Dayezhong (27.17% $\pm$ 0.26%), and Fuyun No. 6 (2.70% $\pm$ 0.02%), respectively. (2) Significant differences were also noted in DPPH· scavenging capacity and ORAC values, with Liupao population species, Guihong No. 4, and Wantian species exhibiting superior antioxidant effects in both assays. (3) Inhibitory effects on α-glucosidase and α-amylase were significantly stronger than the positive control acarbose, with Liupao population species, Guihong No. 4, and Guiqing species showing better hypoglycemic effects in both evaluation systems. (4) Both antioxidant and hypoglycemic activities exhibited strong positive correlations with total polyphenol and total flavonoid contents. In conclusion, Liupao population species, Guihong No. 4, Wantian species, and Guiqing species all demonstrated good raw tea quality. Among these, Liupao population species and Guihong No. 4 possess promising potential for developing functional foods with both antioxidant and hypoglycemic activities, while Wantian species and Guiqing species show potential for developing antioxidant and hypoglycemic functional foods, respectively. Total polyphenols and total flavonoids contributed substantially to the in vitro antioxidant and hypoglycemic activities of raw tea, and special attention should be paid to protecting these components during further processing and utilization of raw tea.

Full Text

Study on Components, Antioxidant and Hypoglycemic Activities of Water Extracts from Different Varieties of Raw Tea

HUO Huazhen[1], CAI Aihua[1]*, GUO Chunyu[2], XIE Yunchang[1], LI Dianpeng[1]

[1]Guangxi Key Laboratory of Plant Functional Phytochemicals and Sustainable Utilization, Guangxi Institute of Botany, Guangxi Zhuang Autonomous Region and Chinese Academy of Sciences, Guilin 541006, Guangxi, China
[2]Guangxi Tea Research Institute, Guilin 541004, Guangxi, China

Abstract

This study compared the differences in active components and in vitro antioxidant and hypoglycemic activities among water extracts from seven different varieties of raw tea, determined the correlations between each component and activity, and provided a scientific basis for developing Liupao tea products with superior antioxidant and hypoglycemic activities through raw material screening and processing method selection. The contents of total polyphenols, total flavonoids, and tea polysaccharides in raw tea water extracts and their extracts were determined. Antioxidant and hypoglycemic activities were evaluated using DPPH·scavenging capacity, ORAC values, and α-glucosidase and α-amylase inhibition as indicators, with Pearson correlation analysis performed to examine relationships. The results showed: (1) Significant differences existed among the seven varieties in water extract content, total polyphenols, total flavonoids, and tea polysaccharides, with the highest values observed in Golden tea (53.42%$±0.14±1.09±0.26±$0.02%), respectively. (2) Significant differences were also found in DPPH·scavenging capacity and ORAC values, with Liupao group species, Guihong No.4, and Wantian species demonstrating superior antioxidant effects in both evaluation methods. (3) All varieties showed significantly stronger inhibition of α-glucosidase and α-amylase than the positive control acarbose, with Liupao group species, Guihong No.4, and Guiqing species exhibiting better hypoglycemic effects in both assays. (4) Both antioxidant and hypoglycemic activities showed strong positive correlations with total polyphenol and total flavonoid contents. In summary, raw tea from Liupao group species, Guihong No.4, Wantian species, and Guiqing species all demonstrated good quality. Among these, Liupao group species and Guihong No.4 show promise for developing functional foods with both antioxidant and hypoglycemic properties, while Wantian species and Guiqing species have potential for developing antioxidant and hypoglycemic functional foods, respectively. Total polyphenols and total flavonoids contribute significantly to the in vitro antioxidant and hypoglycemic activities of raw tea, warranting particular attention to protecting these components during further processing and utilization of raw tea.

Keywords: different varieties, raw tea, water extract components, antioxidant activity, hypoglycemic activity

Introduction

Liupao tea, a distinctive traditional famous tea from Guangxi, belongs to the dark tea category. It not only contains abundant functional components and offers excellent health benefits but also provides a sensory experience characterized by "red, thick, aged, and mellow" qualities, making it increasingly popular among consumers (Huang et al., 2020; Ma et al., 2020). Liupao tea has become an important component of Guangxi' s "hundred-billion-yuan tea industry" that the autonomous region prioritizes for development (Li, 2019; Kong, 2020). Liupao tea is produced from fresh tea leaves through primary processing to produce raw tea, which is then further refined and aged (pile-fermented). Therefore, the quality of the intermediate raw tea material is crucial for the final quality formation of Liupao tea (Ma et al., 2020). However, preliminary market research revealed that raw tea for Liupao tea production is sourced from numerous tea cultivars with varying material quality, resulting in inconsistent quality of finished Liupao tea products and seriously hindering industry development. Consequently, there is an urgent need to evaluate quality differences in raw tea among these cultivars to help Liupao tea enterprises more accurately select intermediate raw materials and stabilize the quality of finished products.

With improving living standards and changing dietary patterns, the prevalence of diabetes is increasing with a trend toward younger onset (Yang et al., 2021). Antioxidants are substances that capture and neutralize free radicals, thereby protecting the human body from free radical damage (Zhang et al., 2017). They enhance the body' s antioxidant capacity and are commonly used for preventing and treating diabetes and its complications. As radiation exposure increases in daily life and health consciousness grows, antioxidants have become a research hotspot in food and medical fields. However, currently used antioxidants are mostly Western medicines with numerous side effects and limited effectiveness in controlling complications (Gong et al., 2012). People increasingly prefer natural antioxidants that can be consumed long-term with minimal side effects. When selecting tea products, consumers now focus not only on taste but also on health benefits related to antioxidant and hypoglycemic effects. Therefore, developing Liupao tea products with superior antioxidant and hypoglycemic activities holds broad market prospects.

Tea polyphenols (including flavanols, flavonoids, anthocyanins, and phenolic acids) and tea polysaccharides are important active components in tea. They not only exhibit health benefits such as antioxidant, hypoglycemic, and liver-protective effects but also regulate tea liquor taste, representing key material bases that determine tea quality (Liu et al., 2012; Song et al., 2018; Ma et al., 2020; Liu et al., 2022). Active components in tea are typically absorbed by the human body through brewing or boiling. The content of water extract in tea liquor reflects its thickness and taste intensity and can indicate quality to some extent (Liu et al., 2012). Therefore, determining water extract content and active component levels in raw tea water extracts is significant for quality research.

Although numerous studies on Liupao tea have been reported, most have focused on active components in finished Liupao tea (Chen et al., 2008; Lin et al., 2019; Zhang et al., 2019), aroma components (Wen et al., 2021), processing technology (Li et al., 2021), and biological activities (Ye et al., 2019; Gong et al., 2020), with relatively few studies on the intermediate raw tea material. Zhou et al. (2013) measured polyphenols, water extract, and catechin contents in fresh leaves of different seasonal Liupao tea raw materials (Liupao group species, Guiqing species, Yunnan big leaf species) and conducted sensory evaluation (appearance, liquor color, aroma, taste, and leaf base) of the raw tea, concluding that Liupao group species was more suitable for Liupao tea production. However, this study did not involve comparative analysis of active components in raw tea and used limited cultivars. Lin et al. (2012) reported active and nutritional component contents in fresh leaves of nine cultivars across different seasons, suggesting that tea cultivars with high polyphenol content were suitable for Liupao tea processing. Although this study used more cultivars, it also lacked comparative analysis of active components in raw tea. Liu et al. (2012) determined water extract, total polyphenols, total flavonoids, and total free amino acids in water extracts from seven types of Liupao tea and raw tea from a single origin (Liuzhou), finding that raw tea had higher contents than finished Liupao tea. However, the tea cultivar used for the raw tea was unspecified, and no comparative analysis between different cultivars was conducted. Huang et al. (2020) evaluated raw tea quality under different drying processes, finding that withering under a shed for 4 hours with auxiliary ventilation was optimal. Thus, existing raw tea research has focused on sensory quality and biochemical component content levels. However, from a health value perspective, studies comparing active components and antioxidant and hypoglycemic activities among different raw tea varieties are lacking, preventing scientific judgment of health values and correlations between components and activities. As the saying goes, "seven parts depend on raw material, three parts on processing," indicating that the antioxidant and hypoglycemic activities of raw tea material are closely related to the health value of Liupao tea products.

Therefore, this study selected raw tea prepared from major tea cultivars commonly used for Liupao tea production (Liupao group species, Guiqing species, Wantian species, Yunnan big leaf species) and cultivars including Guihong No.4, Golden tea, and Fuyun No.6 as subjects. We determined the content of active components and in vitro antioxidant and hypoglycemic activities in raw tea water extracts and extracts, performed Pearson correlation analysis to determine relationships between components and activities, and comprehensively evaluated raw tea quality from three aspects: active component content, in vitro antioxidant activity, and hypoglycemic activity. This research provides a scientific basis for developing Liupao tea products with superior antioxidant and hypoglycemic activities through raw material screening and processing method selection.

Materials and Methods

1.1 Materials and Reagents

Fresh tea leaves from seven six-year-old tea cultivars were collected in April 2021 from the tea garden of Guangxi Tea Research Institute, uniformly harvested according to the standard of one bud and three leaves. The cultivars were Golden tea (L1), Liupao group species (L2), Guihong No.4 (L3), Fuyun No.6 (L4), Wantian species (L5), Yunnan big leaf species (L6), and Guiqing species (L7).

Rutin (purity $ $98%, Hefei Bomei Biotechnology Co., Ltd.), gallic acid, water-soluble vitamin E (Trolox), vitamin C (VC) (purity $ $98%, Shanghai Shifeng Biotechnology Co., Ltd.), α-glucosidase, α-amylase (Sigma-Aldrich, USA), Folin-Ciocalteu (FC) reagent, sodium nitrite, aluminum nitrate nonahydrate, sodium hydroxide, 95% ethanol, potassium dihydrogen phosphate, dipotassium hydrogen phosphate, 1,1-diphenyl-2-picrylhydrazyl (DPPH), 2,2' -azobis(2-amidinopropane) dihydrochloride (ABAP), sodium fluorescein, and p-nitrophenyl-α-D-galactopyranoside (PNPG) were all analytically pure. Experimental water was laboratory-prepared ultrapure water.

1.2 Instruments and Equipment

Electronic balance (Shenyang Longteng Electronics Co., Ltd.), T6 UV-Vis spectrophotometer (Beijing Puxi General Instrument Co., Ltd.), XS205 analytical balance (Mettler-Toledo Instruments Co., Ltd.), RIOS 8 ultrapure water system (Millipore, USA), pipettes (Thermo Fisher, USA), TD5A-WS low-speed centrifuge (Hunan Xiangyi Laboratory Instrument Development Co., Ltd.), HH-S4 constant temperature water bath (Shanghai Kuangsheng Industrial Development Co., Ltd.), N1100 rotary evaporator (Shanghai Ailang Instrument Co., Ltd.), Tecan Spark multifunctional microplate reader (Shanghai Dien Trade Co., Ltd.), BCD-213D11D double-door refrigerator (Guangdong Rongsheng Electric Co., Ltd.), MJ-series mold incubator (Shanghai Yiheng Technology Co., Ltd.).

1.3 Experimental Methods

1.3.1 Preparation of Liupao Raw Tea and Its Water Extracts Referring to the raw tea processing technology of Huang et al. (2020), fresh leaves were withered under a shed for 4 hours, then subjected to drum fixation, rolling, and drying in a flavor-enhancing machine at 70°C until sufficiently dry to obtain Liupao raw tea. The raw tea was further pulverized, sealed, and stored at -20°C for later use.

Water extracts from raw tea were prepared according to GB/T 8305–2013 (Standardization Administration of China, 2013). The obtained extracts were aliquoted and stored at -20°C for later use.

1.3.2 Determination of Water Extract Content in Liupao Raw Tea The water extract content in Liupao raw tea was determined according to GB/T 8305–2013. The water extract content in tea was expressed as dry mass fraction (%).

1.3.3 Determination of Total Polyphenols in Liupao Raw Tea Water Extract The determination was performed using the Folin-Ciocalteu method from GB/T 8313–2018 (Standardization Administration of China, 2018) with slight modifications. Briefly, 0.2 mL of raw tea water extract was taken for measurement, diluted with water to 6 mL, then 0.5 mL of FC reagent was added and mixed. After 10 minutes, 1.5 mL of 20% Na_2CO_3 solution was added, mixed thoroughly, diluted with water to 25 mL, and reacted at 30°C in the dark for 30 minutes. Using water as blank control, absorbance (A) was measured at 760 nm. Gallic acid was used as standard to prepare a calibration curve. Total polyphenol content in water extract was expressed as dry mass fraction (%) and calculated using Equation (1):

$$\text{Total polyphenol content} = \frac{\text{Total polyphenols in water extract (g)}}{\text{Mass of water extract (g)}} \times 100$$

1.3.4 Determination of Total Flavonoids in Liupao Raw Tea Water Extract The determination was performed using the aluminum nitrate colorimetric method from Luo et al. (2016) with slight modifications. Briefly, 1.0 mL of raw tea water extract was taken for measurement and diluted to 10.0 mL with 50% ethanol. Then 1.0 mL of 5% sodium nitrite solution was added, mixed, and after 10 minutes, 1.0 mL of 17.6% aluminum nitrate nonahydrate solution was added, shaken, and left for 10 minutes. Next, 10.0 mL of 4% sodium hydroxide solution was added, diluted to volume with 50% ethanol, shaken, and left for 30 minutes. A reagent blank was prepared simultaneously, and absorbance (A) was measured at 510 nm. Rutin was used as standard to prepare a calibration curve. Total flavonoid content in water extract was expressed as dry mass fraction (%) and calculated using Equation (2):

$$\text{Total flavonoid content} = \frac{\text{Total flavonoids in water extract (g)}}{\text{Mass of water extract (g)}} \times 100$$

1.3.5 Determination of Tea Polysaccharides in Liupao Raw Tea Water Extract Tea polysaccharide content was determined using the phenol-sulfuric acid colorimetric method from GB/T 40632—2021 (Standardization Administration of China, 2021).

1.3.6 Determination of In Vitro Antioxidant and Hypoglycemic Activities

1.3.6.1 DPPH Radical Scavenging Capacity The method from Li et al. (2022) was referenced with slight modifications. Raw tea water extract was diluted into a series of appropriate concentrations as test samples. Then 1.0 mL of test sample was precisely mixed with 2.0 mL of 95% ethanol and 1.0 mL of 0.12 $\mathrm{mg \cdot mL^{-1}}$ DPPH solution, shaken, and reacted at room temperature in the dark for 20 minutes as the sample group. The zero adjustment group used 4.0 mL of 95% ethanol, the sample background group used the same volume of 95% ethanol instead of DPPH solution, and the blank group used 3.0 mL of 95% ethanol and 1.0 mL of DPPH solution. Absorbance was measured at 517 nm. VC was used as positive control (concentration gradient: 5, 10, 15, 20, 25 $\mathrm{\mu g \cdot mL^{-1}}$). Scavenging rate was calculated using Equation (3):

$$\text{Scavenging rate (\%)} = \left[1 - \frac{T - T_0}{C}\right] \times 100$$

where T is the absorbance of the sample group, T_0 is the absorbance of the sample background group, and C is the absorbance of the blank group.

The DPPH radical scavenging capacity of raw tea water extract was expressed as half scavenging concentration (IC_{50}), with smaller IC_{50} values indicating stronger radical scavenging capacity (Lü and Pan, 2020).

1.3.6.2 Oxygen Radical Absorbance Capacity (ORAC) The ORAC values of water extract samples were determined according to Wen et al. (2016) and expressed as μM Trolox equivalents (TE) $\cdot\, g^{-1}$ water extracts dry weight (DW). Larger ORAC values indicate stronger oxygen radical absorbance capacity and higher antioxidant activity of raw tea water extract.

1.3.6.3 α-Glucosidase Inhibition Activity α-Glucosidase inhibition activity of raw tea water extract was determined according to Pan et al. (2020). The sample concentration required to inhibit 50% of enzyme activity was expressed as IC_{50} value ($\mathrm{\mu g \cdot mL^{-1}}$), with smaller IC_{50} values indicating stronger α-glucosidase inhibition.

1.3.6.4 α-Amylase Inhibition Activity The method was based on Liu et al. (2017) with slight modifications using the DNS method. Different concentrations of sample solution (1.0 mL) were mixed with 1 $\mathrm{U \cdot mL^{-1}}$ α-amylase (1.0

mL) and incubated at 37°C for 10 minutes. Then 1.0 mL of 1% soluble starch solution (pre-boiled and gelatinized) was added and reacted at 37°C for 15 minutes. After high-temperature enzyme inactivation, 1.5 mL of DNS reagent was added and boiled for 8 minutes. The mixture was diluted to 25 mL. The zero adjustment used water instead of α-amylase solution and 1% soluble starch solution, and absorbance (A_1) was measured at 540 nm. The blank control without sample was measured as A_0. Acarbose served as positive control. Reagent background was zeroed with water, and absorbance (A_2) was measured with equal volume of water replacing DNS. Inhibition rate was calculated using Equation (4):

$$\text{Amylase inhibition rate (\%)} = \left[1 - \frac{A_1 - A_2}{A_0}\right] \times 100$$

The sample concentration required to inhibit 50% of enzyme activity was expressed as IC_{50} value ($\mu g \cdot mL^{-1}$), with smaller IC_{50} values indicating stronger α-amylase inhibition.

1.3.7 Data Processing Each experiment was performed in triplicate. Data were processed using Excel 2019 software, and results were expressed as mean ± standard deviation ($\bar{x} \pm SD$). Differences in various indicators among samples were analyzed using SPSS 19.0 software with one-way ANOVA and Duncan' s multiple range test, where $P < 0.05$ indicated significant differences and $P > 0.05$ indicated no significant differences. IC_{50} values from in vitro activity tests were calculated using Probit analysis in SPSS 19.0 software. Pearson correlation analysis was used to examine relationships between various indicators.

Results and Discussion

2.1 Analysis of Water Extract and Active Component Contents in Liupao Raw Tea from Different Cultivars

As shown in Figure 1 [Figure 1: see original paper], water extract content in raw tea from seven different cultivars ranged from (45.18%$±0.11±0.14±0.14 $36.0%) but also surpassed 45%, indicating these cultivars are rich in internal substances with high brewing tolerance (Ji et al., 2021), meeting the material basis required for Liupao tea' s characteristic of enduring multiple infusions.

Total polyphenol content in water extracts from seven raw tea cultivars ranged from (34.50%$±0.22±1.09±$1.09%), which was 6.37% higher than Guiqing species, with significant differences between them ($P < 0.05$). Both Fuyun No.6 and Guiqing species showed significantly lower total polyphenol content than other cultivars ($P < 0.05$), indicating significant differences in total polyphenol content among raw tea water extracts from different cultivars, consistent with findings from Su et al. (2018).

Total flavonoid content in water extracts from seven raw tea cultivars ranged from (16.63%$±0.32±0.26±0.26±0.04±0.02±$0.03%). Fuyun No.6 showed the highest tea polysaccharide content, 1.5 times that of Yunnan big leaf species, with significant differences among other cultivars ($P < 0.05$). These results demonstrate significant differences ($P < 0.05$) in both water extract content and active component contents among raw tea water extracts from different cultivars, likely related to genetic differences among tea cultivars (Liu et al., 2022).

Note: Different letters indicate significant differences ($P < 0.05$) in active component contents in water extracts and extracts from different tea cultivars, while the same letters indicate no significant differences ($P > 0.05$). The same applies below.

2.2 Comparison of Antioxidant Activity of Liupao Raw Tea Water Extracts from Different Cultivars

Results for DPPH radical scavenging capacity and ORAC values of raw tea water extracts from different cultivars are shown in Figures 2 [Figure 2: see original paper] and 3 [Figure 3: see original paper]. As shown in Figure 2, significant differences existed among cultivars in DPPH radical scavenging capacity (IC_{50}) ($P < 0.05$), with Liupao group species showing the strongest scavenging capacity [(16.73$±0.02)g \cdot mL^{-1}], followedbyGuihongNo.4[(17.20±0.25)g \cdot mL{-1}]andWantianspecies[(19.51±0.19)g \cdot mL^{-1}$].

As shown in Figure 3 [Figure 3: see original paper], significant differences existed among cultivars in ORAC values ($P < 0.05$). ORAC values of raw tea water extracts from seven cultivars ranged from [(5,387.41$±39.71)MTE \cdot g^{-1}$ water extracts DW] to [(6,762.63$±50.81)MTE \cdot g^{-1}$ water extracts DW], with Guihong No.4, Wantian species, and Liupao group species showing relatively high ORAC values.

The ranking of antioxidant activity among seven raw tea water extract cultivars was not completely consistent between ORAC and DPPH methods, similar to findings from Zhou et al. (2014). This discrepancy may be related to differences in free radicals and mechanisms between the two evaluation methods. DPPH exists in solution as both radical form and cationic form ($DPPH^{+}$), thus involving both single electron transfer and hydrogen transfer mechanisms (Li and Cui, 2011). ORAC is a classic hydrogen atom transfer reaction that terminates radical chain reactions with high specificity (Xu et al., 2006). Raw tea water extracts from different cultivars may contain varying amounts of macromolecular antioxidants that cannot react with DPPH radicals due to steric hindrance (Xu et al., 2006), leading to biased antioxidant results. Despite differences between ORAC and DPPH results, Guihong No.4, Wantian species, and Liupao group species showed both high ORAC values and strong DPPH radical scavenging capacity, indicating these three cultivars possess superior antioxidant activity compared to others.

2.3 Comparison of In Vitro Hypoglycemic Activity of Liupao Raw Tea Water Extracts from Different Cultivars

Inhibitory effects of Liupao raw tea water extracts from different cultivars on α-glucosidase and α-amylase are shown in Figures 4 [Figure 4: see original paper] and 5 [Figure 5: see original paper]. As shown in Figure 4, all seven cultivars showed significantly stronger inhibition of α-glucosidase than the positive control acarbose [IC_{50} = (606.21$±4.30)$g \cdot mL^{-1}], indicating these cultivars can serve as natural α-glucosidase inhibitors. Liupao group species, Guiqing species, and Guihong No.4 showed better inhibitory effects on α-glucosidase, with IC_{50} values of [(9.66$±0.11)$g \cdot mL^{\text{\{-1\}], [(10.87±0.07)} g \cdot mL}${-1}], and[(11.06±0.11)$g \cdot mL^{-1}], respectively. Both Fuyun No.6 and Wantian species showed significantly lower inhibition of α-glucosidase than Liupao group species ($P < 0.05$), while other cultivars showed no significant differences.

As shown in Figure 5 [Figure 5: see original paper], some cultivars showed significant differences in α-amylase inhibition ($P < 0.05$). Half-maximal inhibitory concentrations (IC_{50}) of raw tea water extracts from seven cultivars ranged from [(89.13$±1.07)$g \cdot mL^{\text{\{-1\}]} to [(160.15±1.88) g \cdot mL}${-1}$], all demonstrating significant α-amylase inhibition. Liupao group species, Guihong No.4, and Guiqing species showed better α-amylase inhibition than other cultivars. Both in vitro hypoglycemic evaluation methods indicated that Liupao group species, Guihong No.4, and Guiqing species possessed better in vitro hypoglycemic activity.

2.4 Correlation Analysis Between Active Components and In Vitro Antioxidant and Hypoglycemic Activities of Liupao Raw Tea Water Extracts

Since stronger activity corresponds to smaller IC_{50} values, $1/IC_{50}$ was used to represent activity strength in correlation analysis between active components and antioxidant/hypoglycemic activities. As shown in correlation analysis (Table 1), total polyphenol content showed significant positive correlation with DPPH radical scavenging and ORAC values, with correlation coefficients of 0.751 and 0.817, respectively, indicating significant correlation between total polyphenol content and in vitro antioxidant activity. Total polyphenol content showed significant positive correlation with total flavonoid content ($P < 0.05$), as flavonoids are components of polyphenols. Total polyphenol content showed good positive correlation with α-glucosidase and α-amylase inhibition, while total flavonoid content showed moderate and significant positive correlation with α-glucosidase and α-amylase inhibition, respectively, indicating that in vitro hypoglycemic activity is related to both total polyphenols and total flavonoids.

DPPH radical scavenging showed significant positive correlation with ORAC values, indicating these two antioxidant evaluation methods can corroborate each other. Similarly, α-glucosidase inhibition showed correlation coefficient of 0.929 ($P < 0.01$) with α-amylase inhibition, indicating these two in vitro hypoglycemic evaluation methods can serve as mutual references. However, tea polysaccharide content showed no positive correlation with either antioxidant or hypoglycemic activities, differing from results reported by Yu et al. (2021) and Song et al. (2018). This may be because molecular weight, chemical composition, structure, and stereoconformation of tea polysaccharides differ among cultivars (Song et al., 2018), leading to different in vitro hypoglycemic activities. Specific influencing factors require further investigation.

Significant differences ($P < 0.05$) existed among seven cultivars in total polyphenols, total flavonoids, and tea polysaccharides in water extracts and extracts, as well as in in vitro antioxidant and hypoglycemic activities. Both antioxidant evaluation methods indicated that Liupao group species, Guihong No.4, and Wantian species raw tea water extracts had better antioxidant activity, along with higher total polyphenol and total flavonoid contents. Correlation analysis showed that total polyphenol content had significant positive correlation with antioxidant activity, and total flavonoid content also had good positive correlation with antioxidant activity, indicating that total polyphenols and total flavonoids contribute significantly to raw tea antioxidant activity. Total polyphenol content showed good positive correlation with α-glucosidase and α-amylase inhibition, while total flavonoid content showed moderate and significant positive correlation with α-glucosidase and α-amylase inhibition, respectively, indicating that both total polyphenols and total flavonoids possess hypoglycemic activity and may have synergistic effects.

Both hypoglycemic evaluation methods showed that Liupao group species, Guihong No.4, and Guiqing species raw tea water extracts had better hypoglycemic activity than other cultivars. Among these, Liupao group species and Guihong No.4 had higher total polyphenol and total flavonoid contents, while Guiqing species had lower contents than other cultivars. This may be related to two factors: First, Pan et al. (2020) demonstrated that combinations of tea polyphenol components ECG with EGCG and GCG showed synergistic effects on α-glucosidase inhibition, with different ratios affecting inhibition differently. Therefore, compositional differences in ECG, EGCG, and GCG among raw tea water extracts from different cultivars may affect hypoglycemic activity. Second, monosaccharide composition, molecular weight, branched structure, and stereoconformation are important factors affecting tea polysaccharide hypoglycemic activity (Yang et al., 2021). For example, uronic acid can significantly affect polysaccharide activity, while neutral sugar and uronic acid contents show extremely significant differences among tea cultivars (Liu, 2009). Therefore, monosaccharide composition, molecular weight, branched structure, and effective structural content of tea polysaccharides in raw tea water extracts may differ among cultivars, thereby affecting hypoglycemic activity. The material basis and mechanism underlying the in vitro hypoglycemic activity of Guiqing species require further investigation.

In summary, raw tea from Liupao group species, Guihong No.4, Wantian species, and Guiqing species all showed good quality. Among these, Liupao group species and Guihong No.4 have prospects for developing functional foods with both antioxidant and hypoglycemic properties, while Wantian species and Guiqing species have potential for developing antioxidant and hypoglycemic functional foods, respectively. Total polyphenols and total flavonoids contribute significantly to the in vitro antioxidant and hypoglycemic activities of raw tea. Total polyphenols have certain thermal stability but their content decreases rapidly under high humidity, high temperature, and strong light conditions (Shen, 1993). To obtain Liupao tea products with better antioxidant and hypoglycemic activities, particular attention should be paid to protecting these components during further processing and utilization of raw tea, avoiding prolonged use of high-temperature and strong-light processing methods. During storage, raw tea should be sealed, protected from light, and placed in dry, cool, ventilated areas or refrigerated. Since total flavonoids are components of total polyphenols, their protection methods are consistent with those for total polyphenols. Additionally, to ensure quality stability of the same tea cultivar during introduction and cultivation in different regions, geographical conditions similar to the original production area should be selected in terms of region, soil, climate, and altitude (Liu et al., 2022), and unified tea picking standards should be adopted. This study provides a scientific basis for developing Liupao tea products with superior antioxidant and hypoglycemic activities through raw material screening and processing method selection.

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Note: Figure translations are in progress. See original paper for figures.