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SPECTROPHOTOMETRIC DETERMINATION OF POLYPHENOLS IN GREEN TEAS WITH 18-MOLYBDODIPHOSPHATE

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Abstract. The reactivity of 18-molybdodiphosphate heteropoly complex (18-MPC) with respect to the polyphenolic compounds contained in green teas was estimated. The one- and two-electron heteropoly blues coexist depending on the ratio of reducing agent to 18-MPC in such reactions. The results for the determination of total content of polyphenols in green teas of different brands obtained with 18-MPC and Folin-Ciocalteu reagent are close to each other. The disadvantages of the existing methods are shown.

Keywords: spectrophotometry, Dawson heteropoly complexes, Folin-Chiocalteu reagent, 18-molybdo-diphosphate, catechins, tea analysis.

1. Introduction

Tea leafs and ready-made tea drinks are multicomponent systems, in which polyphenols are essential for the proper functioning of the human body. It is well known that phenolic compounds exhibit anti-inflammatory, antihistamine, antioxidant, anti-edematous, and anti-cancer effects, stabilize cell membranes, inhibit the aging process, and positively influence the function of the cardiovascular system [1].

The difficulties encountered in the determination of polyphenols are primarily due to the fact that numerous groups of phenolic compounds contained in teas differ in structure and have different reactivity. The analysis of literature data showed that for the determination of tea polyphenols, spectrophotometric techniques are preferred, where the vanillin reagent [2] or the Folin-Chiocalteu reagent (FC reagent) [3] are used most often as reagents. It is known that vanillin forms complexes only with certain groups of phenolic compounds, such as catechins and tannins. However, at the same time, the contribution of flavonols, phenolic acids and other substances that are also contained in tea and are responsible for antioxidant activity and nutritional utility is not taken into account when using the vanillin reagent. Determination of polyphenols in tea with FC reagent is based on the reduction of the heteropoly complex at pH 11.4 with various reducing agents contained in tea, both phenolic and non-phenolic.

Therefore, improvement of the existing and/or development of the new methods that can allow to correctly estimate the content of polyphenols in tea remains a topical task. Recently, a new analytical reagent, the 18-molybdodiphosphate heteropoly complex of the Dawson structure (18-MPC), has been proposed to determine single polyphenolic compounds or their total content [4]. Compared with existing reagents, the use of 18-MPC for the determination of polyphenols in plant objects has several advantages, so it seemed important to assess its potential for determining the sum of polyphenolic compounds in tea. The main utility of the basic leaf tea and the final tea beverage is determined by the complex phenolic compounds of different nature, such as derivatives of flavan-3-ols, flavonoids (quercetin, kaempferol, myricetin), flavandioles, and phenolic acids (gallic, cinnamic, quinic, coumaric, and chlorogenic) [5-7]. It is well known from the literature [8-11] that derivatives of flavan-3-ol, or catechin predominate in green teas (30–42 % in terms of dry matter).

The four major catechins in tea leaves are epicatechin, epigallocatechin, epicatechin-3-gallate, and epigallocatechin-3-gallate (EGCG) (Fig. 1). Among all known natural antioxidants, EGCG has the greatest antioxidant effect [9]. Several methods have been proposed for the determination of catechins, including titrimetry with potassium permanganate as the titrant and indigocarmine as indicator (Leventhal's method) [12], high-performance liquid chromatography [13-16], spectrophotometry in the UV and IR region [2, 3, 17], thin-layer chromatography, and capillary electrophoresis [18].

The permanganatometric method is based on the oxidation of catechins with potassium permanganate in diluted sulfuric acid with an indigocarmine as indicator

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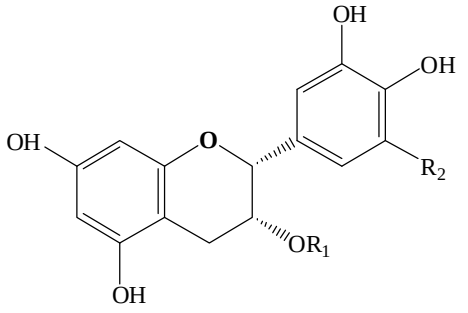
and catalyst. The method is economical and simple in execution, but not precise enough, as potassium permanganate partially oxidizes low-molecular phenolic compounds. Reactions involving potassium permanganate are possible only in strictly standardized conditions (pH, temperature, *etc*.) [12].



Catechin	R_1	R_2
Epicatechin	H	H
Epigallocatechin	gallate	H
Epicatechin-3-gallate	H	OH
Epigallocatechin-3-gallate (EGCG)	gallate	OH

Fig. 1. Structural formulas of tea catechins

The most widely used spectrophotometric method for assessment of the total content of polyphenols in plant objects is based on the oxidation of polyphenols in an alkaline medium (pH 11.4) with the FC reagent. The FC reagent is a mixture of 18-molybdotungsten heteropoly complexes of the Dawson structure, in which heteropoly anions of the formula $P_2Mo_nW_{18-n}O_{62}^{6-}$ (n = 4-5) predominate [19]. Intensely colored compounds, heteropoly blues (HPBs), are formed in the reaction of polyphenols with FC reagent.

Despite the significant advantages, the FC method has a number of shortcomings, such as low selectivity with respect to many reducing agents, high reagent consumption, insufficient reaction speed, formation of insoluble substances with the components of the analyzed sample, nonlinearity of the calibration function, and the need to work in a strongly alkaline region. Many compounds accompanying the catechins in teas are determined simultaneously with catechins by this method including phenolic acids, ascorbic acid, reducing sugars, water-soluble vitamins of group B, and some other species.

The spectrophotometric methods for the determination of catechins in tea based on their reactions with iron(III) ions, vanillin and sulfanilic acid have the highest selectivity. The spectrophotometric method with vanillin is based on the formation of its colored complexes with flavonols and dihydrocalcones, having in their structure a single bond at position 2,3 in the presence of free meta-oriented hydroxyl groups of ring-B [20]. One of the main conditions for the analysis is the need to use methanol-ethanol medium in the presence of concentrated hydrochloric acid. At the same time it is a lack of methodology. The amount of polyphenols, which is extracted, can vary greatly and depends on the composition of such a mixture. In addition, there are serious differences in the evaluation of the amount of polyphenols obtained for aqueous and organic extracts.

In the present work, 18-molybdodiphosphate heteropoly complex of the Dawson structure is proposed as a new reagent for the spectrophotometric determination of catechins in green tea [4, 19, 21-26]. The aim of the work was to develop a simple, highly reproducible, and rapid spectrophotometric procedure for determining the sum of polyphenolic compounds in green tea using 18-MPC as a reagent, which would significantly reduce the interfering effect of the reducing agents of the non-phenolic nature.

2. Experimental

2.1. Reagents, Equipment and Procedures

The following reagents were used in the work: NaOH, H_2SO_4, HCl, NH_4OH, Na_2CO_3, $Na_2B_4O_7$, $KMnO_4$, sum of tea catechins, CH_3OH, C_2H_5OH, vanillin ("Mriya", Ukraine), EGCG, indigocarmine, and oxalic acid. All the reagents were of analytical-reagent grade. Borate buffer solution (pH 9.5) was prepared according to [27]. An aqueous solution of 18-MPC with a concentration of 0.005 mol/l was prepared by dissolving 390 mg of the synthesized salt $(NH_4)_6P_2Mo_{18}O_{62}\cdot 14H_2O$ in 25 ml of distilled water. Synthesis of 18-MPC was carried out according to the procedure described in [25]. Folin-Ciocalteu reagent was prepared according to literature [3]. Both solutions of heteropoly complexes were stable for one month after preparation when preserved in refrigerator.

The preparation of working solutions for permanganatometric titration and the procedure for determining polyphenols in tea with potassium permanganate are described in [12]. Samples of unfermented teas were investigated, including "Hyleys", "Greenfield Flying

Dragon", "Qualitea", "Celestial Tea", "Curtis Bountea White Tea", and "Ahmad". To prepare 0.011 mol/l solution of EGCG, 5 mg of preparation were dissolved in 10 ml of 96% ethyl alcohol by careful heating on a water bath (313–323 K). The solution was used within one week after preparation.

Absorption spectra in the UV and visible regions were obtained with a spectrophotometer SF-26 (Russia, LOMO). The measurements of the absorbance were made in the glass cells with optical path length of 5 and 10 mm. The pH was measured using a universal ionomer EV-74 with a glass indicator electrode and a silver/silver chloride reference electrode.

Preparation of the water extract from tea. Tea leaf material was ground in a mortar. 2.5 g of tea were placed in a 250 ml flask, 200 ml of hot distilled water were added to a sample and boiled for 45 min. The resulting solution was filtered off on a Buchner funnel, and after cooling, the volume was adjusted to 200 ml with distilled water. The extraction yield of polyphenols from tea was more than 98 % [28]. A water extract from tea was used to determine polyphenols with 18-MPC, FC reagent, as well as permanganatometric method.

Preparation of the methanol extracts of polyphenols from tea. 10 ml of methanol were added to 1 g of ground raw material and boiled for 2 h under reflux. The resulting solution was filtered on a Buchner funnel. After cooling, the volume was adjusted to 10 ml with methanol. The formation of a precipitate insoluble in methanol was observed in carrying out the methanolic extraction. The precipitate was removed by centrifugation and discarded. The content of polyphenols in the methanolic extract was determined with 18-MPC and vanillin.

2.2. Procedures for the Determination of Polyphenols in Tea with Dawson Heteropoly Complexes

The content of polyphenols in tea was determined with 18-MPC by the following procedure. An aliquot of the sample solution was transferred in a 25 ml volumetric flask, then 0.8 ml of 18-MPC (0.005 mol/l) and 2 ml of borate buffer solution with pH 9.5 were added. The flask was then filled with water to the mark. Absorbance was measured after 15 min at 820 nm against water in a 10 mm glass cell.

The determination of the polyphenols with FC reagent was carried as follows: an aliquot of the sample solution was transferred in a 25 ml volumetric flask, then 0.3 ml of FC reagent and 3 ml of 20 % Na_2CO_3 were added. Finally, the flask was filled with water to the mark. The absorbance was measured after 15 min at 720 nm in a 10 mm glass cell.

2.3. The Procedure for the Determination of Catechins using a Vanillin Reagent

Our experience and literature data indicate that when using the vanillin reagent, strict adherence to a number of conditions is required [20]. It is shown that it is better to use a methanol extract, since for ethanolic extraction in certain cases no colored complex compounds are formed. Acidification is best carried out with hydrochloric acid to obtain more stable results and achieve the maximum absorbance.

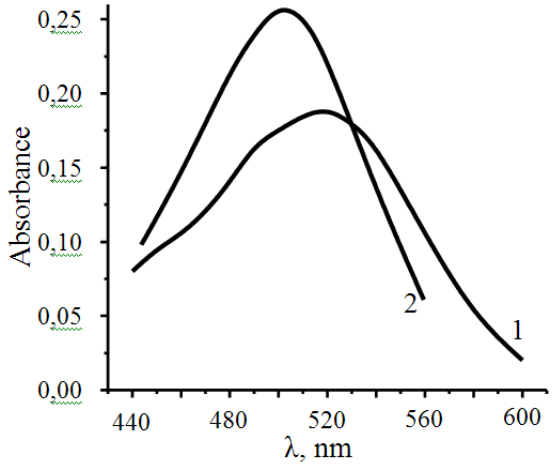


Fig. 1. Absorption spectra of complex compounds obtained in the reaction of vanillin with EGCG (1) and galotannin (2). *C*(vanillin) = 40 mg/ml, *C*(polyphenol) = 10^{-5} mol/l, *L* = 1 cm

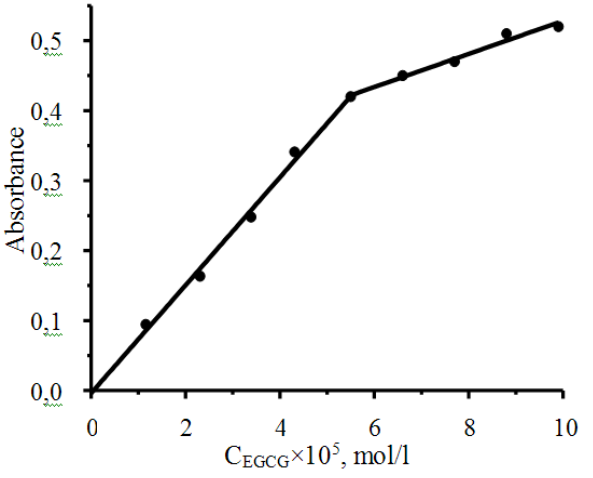


Fig. 2. Calibration graph for the determination of EGCG with vanillin. *C*(vanillin) = 40 mg/ml, λ = 500 nm, *L* = 1 cm

Determination of the polyphenols content in tea with a vanillin reagent. 0.1 ml of tea (methanolic extract) were mixed with 2 ml of ethanolic vanillin solution. Then, a solution containing ethanol and HCl (1:1 v/v) was added to the mixture to a total volume of 10 ml. After 15 min, the absorbance of the solution was measured at 520 nm. For the preparation of the vanillin reagent, 2 g of vanillin were dissolved in 10 ml of ethanol by heating on a water bath at 323–333 K. The absorption spectra of complexes obtained by the reaction of vanillin with EGCG and gallotannin are shown in Fig. 1. The spectrum for vanillin complex with EGCG has an absorption maximum at 500 nm, and the one with gallotannin – at 520 nm. As can be seen from Fig. 2, the calibration curve obtained for the determination of catechins with vanillin reagent is non-linear. However, the dependence can be approximated by two linear segments. In the concentration range from $4.4 \cdot 10^{-6}$ to $5.4 \cdot 10^{-5}$ mol/l, the equation of the calibration graph is described by the equation $A = (0.042 \pm 0.035) + (7.42 \pm 0.09) \cdot 10^{3} \cdot C_{EGGC}$ ($R^2 = 0.997$), the detection limit is of 10^{-6} mol/l ($L = 1$ cm).

3. Results and Discussion

3.1. A Comparative Study of the Features of the EGCG Reactions with 18-MPC and FC Reagent

It is known that pH 11.4 is recommended for the determination of polyphenols with a FC reagent [3]. The use of 18-MPC at this pH value is difficult, since it is much more destructable in such a medium in comparison with molybdotungstophosphate heteropoly anions. In a strongly alkaline medium, partial destruction of both the oxidized and the reduced forms of 18-MPC occurs [19]. It was found that 18-MPC can be reduced by polyphenols at pH > 5. However, a sufficiently complete formation of HPB during the reaction with catechins is possible only at pH > 9 (Fig. 3). Therefore, pH 9.5 was chosen as optimal, at which, on the one hand, the rate of the oxidation of EGCG with 18-MFC is rather high, but on the other hand, the heteropoly complex is still relatively stable.

For the optimal pH values, the kinetic curves of the reactions of epicatechin gallate with 18-MPC (Fig. 4, *curve* 1) and with the FC reagent (Fig. 4, *curve* 2) were constructed. It has been established that, for both systems, the absorbance of HPB continues to increase even after 60 min of reaction time. This is explained by the fact that the mechanism of interaction of polyphenols and, in particular, catechins, with oxidants is a complex multi-stage process and includes several stages of electron transfer, as well as the stages associated with structural changes in polyphenols [29]. In an alkaline medium, catechins are oxidized to form new phenolic compounds, which in turn can react with the heteropoly complex. Then, the oxidized and reduced forms of the polyphenols present in the solution polymerize to form condensed tannins, which are also capable of being oxidized by 18-MPC. It was found that after 15 min from the beginning of the reaction the achieved absorbance for the reaction of EGCG with 18-MPC is ~85 % of the maximum possible and then increases very slowly. This time interval was used later in the analysis of plant objects.

Fig. 4 shows the kinetic curves for the reactions of polyphenols with 18-MPC (Fig. 4, *curve* 3) and the FC reagent (Fig. 4, *curve* 4) in green tea “Celestial Tea”. As can be seen from the figure, the reaction rates of oxidation of tea polyphenols and EGCG with heteropoly complexes are approximately the same. Therefore, taking into account that EGCG in the best degree reflects the reactivity of tea polyphenols, and also dominates in the composition of green tea, it can be used as a standard in the analysis of various varieties of tea.

A break on the molar ratio plot for the reaction of 18-MPC with EGCG corresponds to the ratio of C(18-MPC):C(EGCG) = 4:1 (Fig. 5). The first product of the 18-MPC reduction is the two-electron HPB. Thus, EGCG, being oxidized, takes 8 electrons in this reaction. Formally, according to the known rule [3], this indicates the oxidation of the 8 hydroxyl groups present in the EGCG structure, but, as we have already discussed, the actual oxidation mechanism is probably much more complex.

The formation of two-electron HPB is also confirmed by the fact that the maximum in the absorption spectrum of HPB is situated at 820 nm (Fig. 6a) [21]. The half-reaction of the formation of a two-electron HPB can be written as follows:

$$P_2Mo^{VI}_{18}O_{62}^{6-} + 2\bar{e} \rightarrow P_2Mo^{VI}_{16}Mo^{V}_{2}O_{62}^{8-}$$

However, as can be seen from Fig. 6a, in the absorption spectra of HPBs obtained for different ratios of EGCG to 18-MPC, a shoulder appears in the long-wave region. We believe that this phenomenon is explained by a shift in the equilibrium towards the formation of a one-electron HPB in a large excess of the oxidized form of 18-MPC according to the equation: $P_2Mo^{VI}_{18}O_{62}^{6-} + P_2Mo^{VI}_{16}Mo^{V}_{2}O_{62}^{8-} \leftrightarrow 2P_2Mo^{VI}_{17}Mo^{V}O_{62}^{7-}$. The isobestic point at approximately 920 nm is present in the spectra plotted as a dependence of molar absorptivity of HPB versus wavelength (Fig. 6a), that proves the existence of the equilibrium between two forms of HPBs.

The calibration graphs (Fig. 7) for the determination of EGCG by using 18-MPC and the FC reagent are linear in the concentration range from $2.2 \cdot 10^{-6}$ to $4.4 \cdot 10^{-5}$ mol/l. For 18-MPC, calibration graph is

described by the equation A^{820} = (0.008±0.021) + + (3.73±0.08)·10^4·C_{EGCG} (R^2 = 0.997), and the detection limit is calculated as 5·10^{-7} mol/l (L = 1 cm). For FC reagent, the calibration equation is the following: A^{720} = = –(0.008±0.013) + (2.80±0.06)·10^4·C_{EGCG} (R^2 = 0.996), and the detection limit is 5·10^{-6} mol/l (L = 1 cm). It can be seen that the slope of the calibration curve for the 18-MPC is about 1.4 times higher than for the FC reagent.

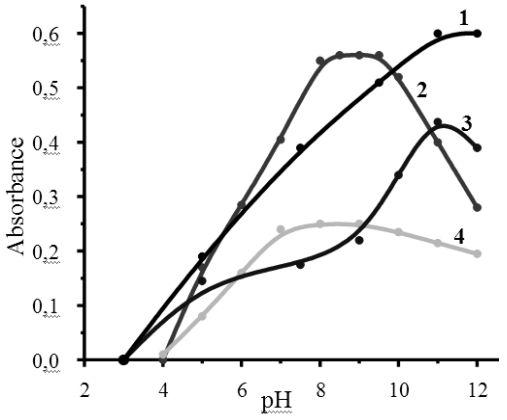


Fig. 3. Effect of solution pH on the formation of HPB in the reaction of 18-MPC with EGCG (1), quercetin (2), chlorogenic (3), and gallic (4) acids.
C(18-MPC) = 1.6·10^{-4} mol/l, C(polyphenol) = 2·10^{-5} mol/l, λ = 820 nm, t = 15 min, L = 0.5 cm

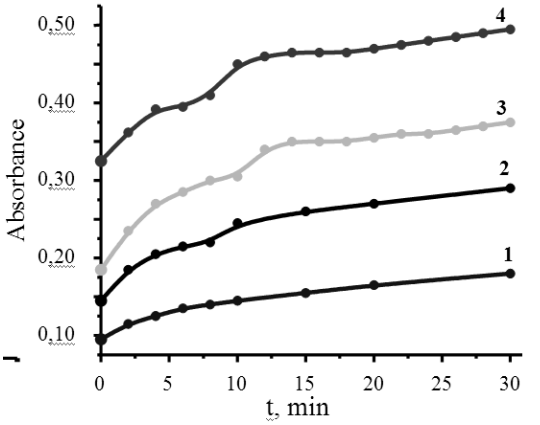


Fig. 4. Dependence of the absorbance on the reaction time for the reactions of EGCG (1, 2) and polyphenols (3, 4) of green tea with 18-MPC (2, 3) and with FC reagent (1, 4).
C(EGCG) = 1.1·10^{-5} mol/l, C(18-MPC) = 1.6·10^{-4} mol/l; pH = 9.5, λ = 820 nm, C(FC reagent) ≈ 2·10^{-4} mol/l, pH = 11.4, λ = 720 nm, L = 0.5 cm

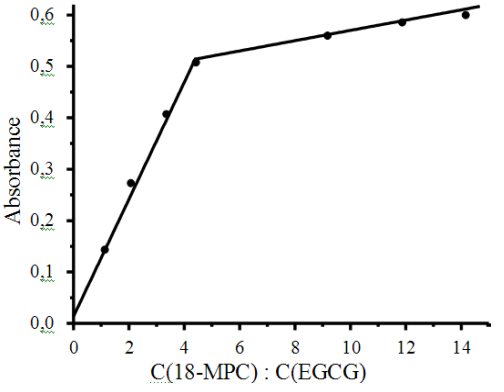


Fig. 5. Molar ratio plot for the reaction between EGCG and 18-MPC. C(EGCG) = 1.1·10^{-5} mol/l, pH = 9.5, λ = 820 nm, L = 0.5 cm

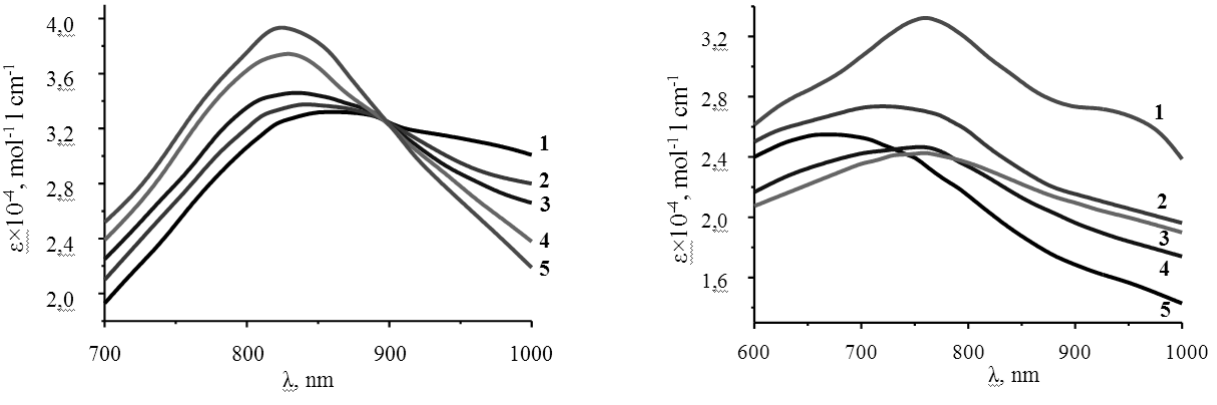


Fig. 6. Absorption spectra of HPBs obtained by reduction of 18-MPC (a) and FC reagent (b) with EGCG.
For (a) C(18-MPC) = 1.6·10^{-4} mol/l, pH = 9.5, C(EGCG), μmol/l: 4.4 (1); 8.8 (2); 13 (3); 26 (4) and 31 (5).
For (b) C(FC reagent) = 2.4·10^{-4} mol/l, pH = 11.4, C(EGCG), μmol/l: 2.2 (1); 8.8 (2); 18 (3); 26 (4) and 35 (5).
L = 1 cm, t = 15 min

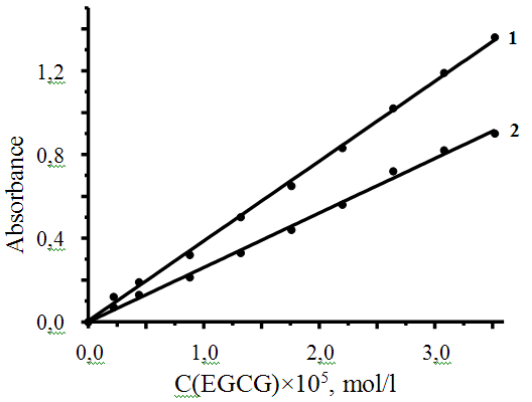


Fig. 7. Calibration graphs for the determination of EGCG with 18-MPC (1) and FC reagent (2). For (1) C(18-MPC) = $1.6 \cdot 10^{-4}$ mol/l, λ = 820 nm, pH = 9.5, L = 1 cm For (2) C(FC reagent) ≈ $2 \cdot 10^{-4}$ mol/l, λ = 720 nm, pH = 11.4, L = 1 cm

3.2. Application and Assessment of the Proposed Procedure

The spectrophotometric method for the determination of catechins with 18-MPC proposed in this study was applied to the analysis of green teas of various brands. To assess the accuracy of the developed method, we chose the methods generally accepted for the determination of catechins: permanganatometric titration in the presence of indigosulfonic acid (Leventhal's method) and spectrophotometric methods using the Folin-Chiocalteu reagent and a vanillin reagent.

The content of polyphenols in green teas found by the permanganatometric titration (Table, Fig. 8a) quite well correlates with the results obtained by using 18-MPC (correlation coefficient is 0.952). However, in all the cases the obtained content of catechins is significantly higher for the first method. Obviously, the reaction of potassium permanganate is not selective enough with respect to polyphenols. The permanganate is much stronger oxidant than 18-MPC, hence it can react with bigger number of species and oxidize the polyphenols more deeply. In addition, the oxidation of polyphenols with 18-MPC proceeds under milder conditions. We have previously shown that for other plant samples, the reaction with potassium permanganate often results in overestimated results, since permanganate to a large extent reacts with reducing sugars, group B vitamins, simple phenols, proteins, etc. The results of the analysis obtained using 18-MPC better reflect the content of polyphenolic compounds, and the evaluation of nutritional value and antioxidant activity is more objective.

As can be seen from the table, the content of polyphenols in green tea in terms of EGCG content, determined using a vanillin reagent, constitutes ~20–45 % of the sum of polyphenols found with 18-MPC and the FC reagent. These results do not contradict the literary data. The table shows that the content of polyphenols in green tea determined with vanillin reagent gives distorted results and therefore it cannot be recommended for evaluation of the content of polyphenols, antioxidant activity and nutritional value.

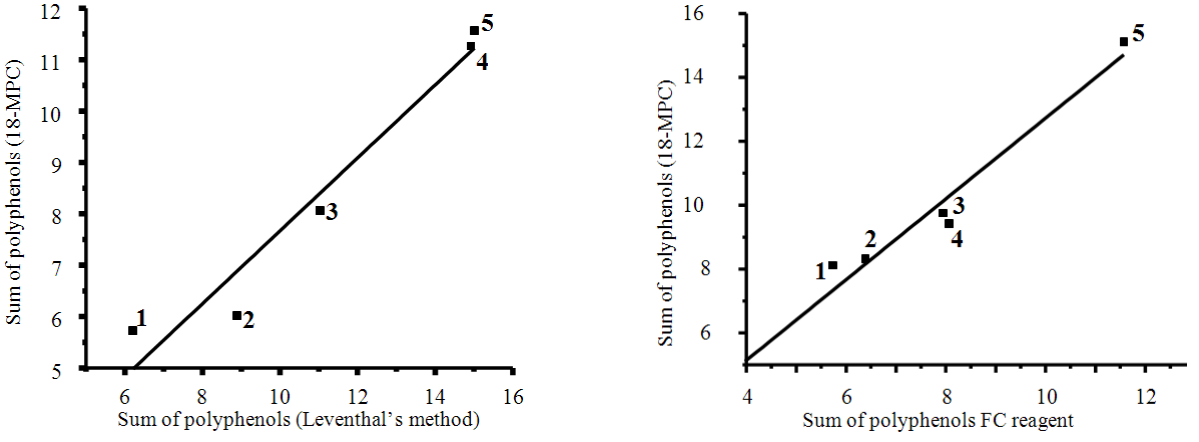


Fig. 8. Correlation between the results of the determination of total content of polyphenols found in green teas using 18-MPC and potassium permanganate (a); 18-MPC and FC reagent (b). The examined brands of tea: Qualitea (1), Hyleys (2), Curtis Bountea White Tea (3), Greenfield Flying Dragon (4) and Celestial Tea (5)

Table

The total content of polyphenols in green teas found with the 18-MPC, FC reagent (n = 6, P = 0.95), permanganate and vanillin reagent (n = 3, P = 0.95)

Brand of tea	Enzymatic type of tea	The content of polyphenols in tea, g of EGCG per 100 g of tea ± confidence interval (relative standard deviation)			
		18-MPC	FC reagent	Leventhal's method	Vanillin
Qualitea	Green	10.79 ± 0.29 (0.027)	11.73 ± 0.73 (0.062)	11.31 ± 0.20 (0.062)	–
Hyleys	Green	12.17 ± 0.19 (0.015)	11.73 ± 0.73 (0.062)	15.17 ± 0.39 (0.052)	–
Curtis Bountea White Tea	White	15.21 ± 0.16 (0.011)	14.91 ± 0.37 (0.025)	18.03 ± 0.71 (0.075)	–
Ahmad	Green	12.96 ± 0.61 (0.047)	13.3 ± 1.8 (0.14)	18.37 ± 0.40 (0.050)	10.52 ± 0.44 (0.061)
Greenfield Flying Dragon	Green	11.38 ± 0.37 (0.032)	11.19 ± 0.28 (0.022)	14.21 ± 0.91 (0.046)	6.03 ± 0.21 (0.034)
Heavenly Tea	Green	11.69 ± 0.28 (0.025)	10.91 ± 0.27 (0.025)	15.01 ± 0.41 (0.047)	3.68 ± 0.10 (0.050)

The contents of polyphenols in green teas determined using 18-MPC and the FC reagent correlate very well with each other, the correlation coefficient is ~ 0.987 (Table, Fig. 8b) and with the data given in the literature. For green tea "Curtis Bountea White Tea", "Ahmad" leaf green tea, "Greenfield Flying Dragon" the absolute values of polyphenols contents are also close to each other. It can be suggested that the mechanisms of the EGCG oxidation with 18-MPC and the FC reagent are approximately the same, the values of the apparent molar absorptivity coefficients for EGCG are also close, which means that FC reagent can be in most cases replaced by 18-MPC. Nevertheless, the selectivity of the 18-MPC reaction with polyphenolic compounds is significantly better than that for FC reagent that forces us to recommend it for the determination of the above indices in green teas. The reaction of 18-MPC with polyphenols is generally more rapid and the measured absorbance is more stable and less dependent on the variation in the conditions of the analysis.

4. Conclusions

A simple, accurate and highly sensitive spectrophotometric procedure has been developed for the determination of the sum of polyphenols in green teas by using of 18-molybdo-2-phosphate Wells-Dawson heteropoly complex. It was established that optimal pH for the reaction of 18-MPC with polyphenols is situated at pH 9.5. Under these conditions, reaction of 18-MPC is significantly more selective than that with FC reagent. The spectrum of HPB formed in the reaction of FC reagent or 18-MPC with reducing agents depends on the ratio of analyte to reagent. This phenomenon is caused by the formation of different reduced forms of heteropoly blues coexisting in such solutions. Only for 18-MPC, the measurement of the absorbance at the wavelength corresponding to the isobestic point allows to obtain strictly linear calibration graphs and to avoid systematic errors by the determination of individual reducing agents or their mixtures.

The proposed method was applied for the determination of polyphenols in green teas of different brands. The accuracy of the obtained results with the 18-MPC was confirmed by a good correlation with the data of the permanganatometric titration method and excellent proximity to the results obtained with the Folin-Chocalteu reagent. Permanganatometric titration and use of a vanillin reagent cannot be recommended for the determination of total content of polyphenols in green teas. Determination of polyphenols in tea with 18-MPC, unlike the standard methods, does not depend on the method of sample preparation (aqueous or water-alcoholic extracts), does not require a large excess of reagent, occurs at room temperature, and is more rapid and selective with respect to phenolic compounds.

References

[1] Yashin Ya., Yashin A.: Khimicheskiy Sostav Chaya i ego Vliyanie na Zdorovie Cheloveka. Translit, Moskva 2010.
[2] Ferreira E., Nogueira A., Ferreira E.: Talanta, 2000, **51**, 1. https://doi.org/10.1016/S0039-9140(99)00222-2
[3] Singleton V., Orthofer R., Lamuela-Raventos R.: Meth. Enzymol., 1999, **299**, 152. https://doi.org/10.1016/S0076-6879(99)99017-1
[4] Denisenko T., Tsiganok L., Vishnikin A.: Analitika i Kontrol, 2015, **19**, 373. https://doi.org/10.15826/analitika.2015.19.4.012

[5] Zaveri N.: Life Sci., 2006, **78**, 2073. https://doi.org/10.1016/j.lfs.2005.12.006
[6] Price K., Rhodes M., Barnes K.: J. Agric. Food Chem., 1998, **46**, 4898. https://doi.org/10.1021/jf980687v
[7] Hertog M., Hollman P., Putte B.: J. Agric. Food Chem., 1993, **41**, 1242. https://doi.org/10.1021/jf00032a015
[8] Balentine D., Wiseman S., Bouwens L.: Crit. Rev. Food Sci. Nutr., 1997, **37**, 693. https://doi.org/10.1080/10408399709527797
[9] Harbowy M., Balentine D.: Crit. Rev. Plant Sci., 1997, **16**, 415. https://doi.org/10.1080/07352689709701956
[10] Higdon J., Frei B.: Crit. Rev. Food Sci. Nutr., 2003, **43**, 89. https://doi.org/10.1080/10408690390826464
[11] Juneja L., Kapoor M., Okubo T. *et al*. (Eds.): Green Tea Polyphenols. Nutraceuticals of Modern Life. CRC Press, London, N.Y. 2013.
[12] http://www.gosthelp.ru/gost/gost16980.html
[13] http://www.internet-law.ru/gosts/gost/62348/
[14] Zuo Y., Chen H., Deng Y.: Talanta, 2002, **57**, 307. https://doi.org/10.1016/S0039-9140(02)00030-9
[15] Narumi K., Sonoda J.-I., Shiotani K. *et al.*: J. Chromatogr. B, 2014, **945-946**, 147. https://doi.org/10.1016/j.jchromb.2013.11.007
[16] Lee M.-J., Prabhu S., Meng X. *et al.*: Anal. Biochem., 2000, **279**, 164. https://doi.org/10.1006/abio.2000.4487
[17] Apak R., Güçlü K., Demirata B. *et al.*: Molecules, 2007, **12**, 1496. https://doi.org/10.3390/12071496
[18] Arce L., Rios A., Valcarcel M.: J. Chromatogr. A, 1998, **827**, 113. https://doi.org/10.1016/S0021-9673(98)00737-7
[19] Denisenko T., Tsiganok L., Vishnikin A.: Analitika i Kontrol, 2015, **19**, 242. https://doi.org/10.15826/analitika.2015.19.3.001
[20] Desphande S., Cheryan M., Salunkhe D. *et al.*: Crit. Rev. Food Sci. Nutr., 1986, **24**, 401. https://doi.org/10.1080/10408398609527441
[21] Denisenko T., Tsiganok L., Vishnikin A.: Methods Objects Chem. Anal., 2014, **9**, 130.
[22] Bulatov A., Petrova A., Vishnikin A. *et al.*: Microchem. J., 2013, **108**, 213. https://doi.org/10.1016/j.microc.2013.04.020
[23] Petrova A., Bulatov A., Vishnikin A. *et al.*: Anal. Sci., 2015, **31**, 529. https://doi.org/10.2116/analsci.31.529
[24] Vishnikin A., Al-Shwaiyat M., Petrushina G. *et al.*: Talanta, 2012, **96**, 230. https://doi.org/10.1016/j.talanta.2012.02.049
[25] Bulatov A., Petrova A., Vishnikin A. *et al.*: Talanta, 2012, **96**, 62. https://doi.org/10.1016/j.talanta.2012.03.059
[26] Vishnikin A., Sklenařova H., Solich P. *et al.*: Anal. Lett., 2011, **44**, 514. https://doi.org/10.1080/00032719.2010.500789
[27] Lurye Yu.: Spravochnik po Analiticheskoy Khimii. Khimiya, Moskva 1979.
[28] Mariya J., Vijayan D., Raj Kumar R. *et al.*: Asian J. Plant Sci., 2006, **5**, 123. https://doi.org/10.3923/ajps.2006.123.126
[29] Timbola A., de Souza C., Giacomelli C. *et al.*: J. Braz. Chem. Soc., 2006, **17**, 139. https://doi.org/10.1590/S0103-50532006000100020

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СПЕКТРОФОТОМЕТРИЧНЕ ВИЗНАЧЕННЯ ПОЛІФЕНОЛІВ В ЗЕЛЕНИХ ЧАЯХ З 18-МОЛІБДОДИФОСФАТОМ

Анотація. *Оцінено реакційну здатність 18-молібдодифосфатного гетерополікомплексу (18-МФК) щодо поліфенольних сполук, які містяться в зелених чаях. В таких реакціях в залежності від співвідношення відновника до 18-МФК співіснують одно- та двоелектронні гетерополісині. Результати визначення загального вмісту поліфенолів у зелених чаях різних марок, отриманих з 18-МФК та реагентом Фоліна-Чокальтеу, близькі один до одного. Показані недоліки існуючих методів.*

Ключові слова: *спектрофотометрія, гетерополікомплекси структури Доусона, реактив Фоліна-Чокальтеу, 18-молібдодифосфат, катехіни.*