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Rapid colorimetric determination of ascorbic acid by solid phase extraction of iodine into a polymethacrylate matrix

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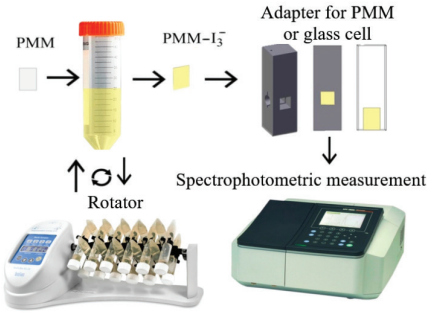
The proposed iodometric solid-phase spectrophotometry method is based on interaction of ascorbic acid with molecular iodine in a solution with further extraction of the unreacted iodine into a polymethacrylate matrix followed by its spectrophotometric measurement in the solid phase. The amount of iodine extracted by the matrix is in inverse linear dependence on the concentration of ascorbic acid in the solution. The method can be employed for the range of ascorbic acid concentrations 1.0–9.0 mg dm^{-3}, while its detection limit is 0.8 mg dm^{-3}.



Keywords: ascorbic acid, polymethacrylate matrix, solid phase spectrophotometry, iodometric method.

Ascorbic acid (vitamin C, food additive E300) is widely used in pharmaceutical, perfume, cosmetics and food industries to increase acidity of products and prolong their shelf life.[1,2] Numerous analytical techniques are available for the determination of ascorbic acid in different matrices. The instrumental methods such as electrochemical[3–8] and chromatographic ones[9–11] are paid a lot of attention. As well, a significant number of spectrophotometric techniques for the analysis of ascorbic acid,[12–16] including solid-phase ones with the use of reagents immobilized into a solid support,[17–20] have been developed.

For the officially accepted titrimetric determination of ascorbic acid,[21] 2,6-dichloroindophenol {2,6-dichloro-4-[(4-hydroxyphenyl)imino]-2,5-cyclohexadien-1-one}[22–24] is recommended. As well, ferroin [tris(1,10-phenanthroline)-iron(II)][25,26] and Cu^{II} with neocuproine[27] are used as reagents immobilized on solid carriers for the quantitative analysis of ascorbic acid. The immobilization allows one to carry out the analysis in a variety of samples including juices with pulp, turbid and intensely coloured media avoiding preliminarily filtration or centrifugation to the required transparency. Moreover, this technique also simplifies the digital image colorimetry for rapid determination of ascorbic acid in natural fruit juices at the sites of sample acquisition.[28,29]

In addition to the above mentioned advantages of the immobilized reagents, development of a solid-phase analytical system for direct determination of ascorbic acid skipping the reagent immobilization stage, *i.e.*, without preliminary preparation of the solid phase, is of definite interest. This could significantly simplify the analysis and increase its performance. Such an approach can be implemented using transformation of the classical iodometric method of ascorbic acid determination into a spectrophotometric one to increase the sensitivity.[30,31] An example is the analysis of ascorbic acid in food based on its redox reaction with triiodide ion (I_3^-) followed by extraction of the excess I_3^- into CCl_4,[32] where iodine forms an associate with the polymethine dye. In this work, we investigated feasibility of the thechnique combining solid-phase spectrophotometry and iodometric method for determination of ascorbic acid through its oxidation by iodine with further solid-phase extraction of the unreacted iodine left after the reaction into polymethacrylate matrix (PMM) and detection of its absorbance in the solid phase. Solid-phase methods in analytical chemistry correspond to the green chemistry principles as a key trend in this area of science.

PMM was used as a transparent analytical medium thanks to the hydrophilic elements in the PEG 400 chain and the hydrophobic polymer scaffold of polymethylmethacrylate. The optical analytical medium had the form of plates obtained by block polymerization of methacrylic monomers in the presence of PEG 400 and calcium methacrylate at 60 °C for 3 h initiated by benzoyl peroxide. The obtained PMM samples represented transparent colorless plates of 4 × 6 × 0.5 mm size. Combining a hydrophobic scaffold with a hydrophilic filling to build a transparent hybrid matrix is a currently relevant and actively developing approach to promising as ready-to-use analytical sensors for solid-phase spectrophotometric determinations, where accumulation of an analyte within the matrix and the change in the absorption intensity take place through the analyte extraction from a sample solution.

Determination of ascorbic acid is based on its interaction with iodine in a solution followed by extraction of the unreacted iodine into PMM, which results in the change of its color to yellowish brown in a linear dependence inversely to the concentration of ascorbic acid in the sample solution. The color of PMM remains unchanged for a few weeks, with no loss of analytical response and optical characteristics. Two absorption maxima at 290 and 365 nm can be seen (Figure 1) in the spectra

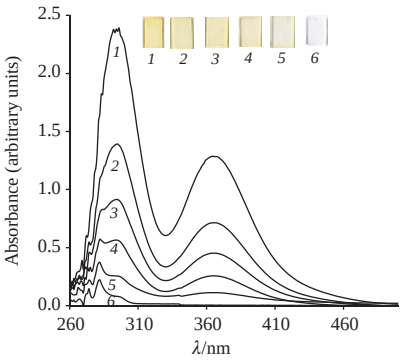


Figure 1 Absorption spectra of iodine extracted into PMM after its contact with a solution containing (*1*) 0, (*2*) 1, (*3*) 3, (*4*) 5, (*5*) 7 and (*6*) 9 mg dm^{-3} ascorbic acid. Inset: photos of PMM with extracted iodine.

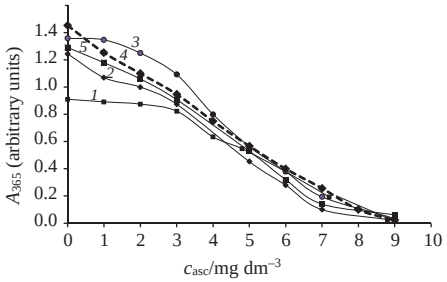


Figure 2 Calibration curves for determination of ascorbic acid at different HCl concentrations in the sample solution (in 10^{-3} mol dm^{-3}): (*1*) 0, (*2*) 2, (*3*) 4, (*4*) 6 and (*5*) 8. The curve (*4*) was used further (see the text).

of iodine extracted into PMM after the contact of the matrices with ascorbic acid solutions of various concentrations in the presence of iodine. We used the absorption maximum of 365 nm to obtain more repeatable results.

The influence of iodine addition on absorbance at 365 nm was evaluated from calibration curves resulting in their analytical performance as well as the estimated values of regression variance S^2_{ad} and the error mean square S^2_y (Table 1). Repeatable results could not be obtained with the iodine concentrations in the sample solution below 5.0×10^{-5} mol dm^{-3}, while the concentrations $\geqslant 1.0 \times 10^{-4}$ mol dm^{-3} significantly increased the blank absorbance. We concluded that the optimal iodine concentration in a sample solution for determination of ascorbic acid was 5.0×10^{-5} mol dm^{-3}, which corresponded to the lowest regression variance and error mean square of a calibration curve as well as the minimal determined concentration of ascorbic acid.

To test the influence of pH, we explored how 0 to 8×10^{-3} mol dm^{-3} of hydrochloric acid in a sample solution influenced the analytical signal with the fixed analyte concentration of 5.0 mg dm^{-3} and the iodine one of 5×10^{-5} mol dm^{-3}. Preliminary tests revealed that the analytical signal was recorded at $(1.0–8.0) \times 10^{-3}$ mol dm^{-3} HCl, the corresponding calibration curves A_{365} *vs.* c_{asc} were collected in Figure 2.

Further experiments were carried out with the sample solution containing 6×10^{-3} mol dm^{-3} HCl, the corresponding calibration curve $A_{365} = 1.423 – 0.161c_{asc}$ ($r = 0.998$, see Figure 2, curve *4*) provided for the widest linear response range with an increase in the sensitivity.

Interfering influence of cations and anions on the determination of ascorbic acid was evaluated as relative deviation of the analytical signal at c_{asc} = 3.0 mg dm^{-3} and 1 : 1 or 1 : 10 ratios of the analyte to ions. In the presence of NO_3^-, NO_2^-, Pb^{2+} or Cu^{2+} the relative deviation of the analytical signal exceeded 10%. Addition of EDTA to the sample solution as a masking agent eliminated the influence of the metal cations.

Then the integrated iodometric solid-phase spectrophotometry method developed for the determination of ascorbic acid[†] was tested on various juice samples. The results are collected in Table 2 in comparison with the known reference indophenol titration method[33] together with the significance of discrepancy in the concentration obtained by the two methods. For three repetitive determinations of ascorbic acid following the proposed and the reference methods the statistical Student's *t*- and *F*-tests revealed that there were no significant differences between the mean values and the variances for the two populations at the

Table 1 Influence of iodine concentration in the sample solution on parameters of calibration curve in the determination of ascorbic acid. For parameter explanation see the text.

c_{iodine}/ mol dm^{-3}	Regression equation	Linear range/ mg dm^{-3}	S^2_{ad}	S^2_y
5.0×10^{-5}	$A_{365} = 1.2 – 0.14c_{asc}$	3.0–9.0	0.004	0.002
7.5×10^{-5}	$A_{365} = 2.24 – 0.16c_{asc}$	6.0–14.0	0.049	0.017
1.0×10^{-4}	$A_{365} = 3.1 – 0.17c_{asc}$	8.0–16.0	0.022	0.014
1.5×10^{-4}	$A_{365} = 4.2 – 0.16c_{asc}$	15.0–27.0	0.221	0.177

Table 2 The results of ascorbic acid determination in real samples (n = 3, P = 0.95, F_{tabl} = 19, t_{tabl} = 2.8).

Juice	c_{asc} as indicated on the package/mg dm^{-3}	c_{asc} as determined by solid-phase spectrophotometry with PMM		c_{asc} as determined by titrimetry[33]		F	t
		Found/mg dm^{-3}	s_r (%)	Found/mg dm^{-3}	s_r (%)		
Lemon pulp	–	440 ± 50	4.6	450 ± 20	1.8	6.2	0.7
Orange no. 1	200	230 ± 30	5.3	210 ± 10	1.9	9.3	2.7
Orange no. 2	210	250 ± 30	4.8	260 ± 10	1.5	9.5	2.7
Carrot	300	340 ± 30	3.6	340 ± 10	1.2	9.0	0.4
Pineapple no. 1	800	930 ± 60	2.6	910 ± 60	2.6	1.0	1.8
Pineapple no. 2	800	980 ± 70	2.9	970 ± 70	2.9	1.0	1.6

[†] *Procedure for determination of ascorbic acid.* Ascorbic acid (0.05 g, Dynemic Products, India) was dissolved in double distilled water (50 cm^3), the resulting 1 g dm^{-3} stock solution was consequently diluted with distilled water to form working solutions, all the solutions were prepared on the day of the experiment. The content of iodine ampoules (Gosstandard, Russia) was diluted with distilled water in a 1000 cm^3 measuring flask resulting in 0.025 mol dm^{-3} solution, the concentration was measured by titration using the sodium thiosulfate standard solution. Sodium 2,6-dichloroindophenolate (0.05 mg, Sigma-Aldrich) was dissolved in hot distilled water (150 ml), cooled to room temperature, diluted to 200 ml and filtered into an amber-colored bottle. Hydrochloric acid (Sigma-Aldrich) was employed for the pH adjustment. All the chemical reagents were used without purification. PMM plates were placed into the test tubes with the solutions, the content was stirred for 5 min using a Multi Bio RS-24 programmable rotator (Biosan, Latvia) at 50 rpm. Then the plates were taken out and their absorption was measured at 365 nm relative to the matrix source sample employing an Evolution 201 UV-VIS spectrophotometer (Thermo Fisher Scientific, USA). The content of ascorbic acid was determined using the calibration curve built under similar conditions.

95% confidence level. Thus, the proposed method was considered to be as accurate and precise as the reference one.

In summary, we demonstrated the feasibility of using PMM for iodometric solid-phase spectrophotometric determination of ascorbic acid in a 1.0–9.0 mg dm^{-3} concentration range with the limit of detection 0.8 mg dm^{-3} as calculated by the 3*s* criterion. The method suggested is easy to implement and requires only standard spectrophotometric equipment. Compared to the reference titration method, our approach offers the following advantages: the rapidity and possibility of using it without any preliminary sample processing for the intensely natural and packaged pulpy juices.

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References

1 V. Spínola, E. J. Llorent-Martínez and P. C. Castilho, *J. Chromatogr. A*, 2014, **1369**, 2.
2 A. Barberis, Y. Spissu, A. Fadda, E. Azara, G. Bazzu, S. Marceddu, A. Angioni, D. Sanna, M. Schirra and P. A. Serra, *Biosens. Bioelectron.*, 2015, **67**, 214.
3 M. S. Mohammadnia, E. M. Khosrowshahi, E. Naghian, A. H. Keihan, E. Sohouli, M. E. Plonska-Brzezinska, A. Sobhani-Nasab, M. Rahimi-Nasrabadi and F. Ahmadi, *Microchem. J.*, 2020, **159**, 105470.
4 M. D. Vedenyapina, M. M. Kazakova and A. M. Skundin, *Russ. J. Phys. Chem. A*, 2019, **93**, 1178 (*Zh. Fiz. Khim.*, 2019, **93**, 938).
5 K. Karaboduk, *ChemistrySelect*, 2019, **4**, 6361.
6 J. Raveendran, R. G. Krishnan, B. G. Nair and T. G. S. Babu, *Microchim. Acta*, 2017, **184**, 3573.
7 P. Krzyczmonik, E. Socha and G. Andrijewski, *Anal. Lett.*, 2017, **50**, 806.
8 M. Khairy and B. G. Mahmoud, *Electroanalysis*, 2016, **28**, 2606.
9 M. E. K. Wahba, *J. Liq. Chromatogr. Relat. Technol.*, 2015, **38**, 54.
10 O. V. Trineeva, E. F. Safonova and A. I. Slivkin, *Pharm. Chem. J.*, 2018, **51**, 938 [*Khim.-Farm. Zh.*, 2017, **51** (10), 58].
11 C. D. Zappielo, A. E. Nicácio, L. P. Manin, L. Maldaner and J. V. Visentainer, *J. Braz. Chem. Soc.*, 2019, **30**, 1130.
12 M. Salkić, *J. Anal. Chem.*, 2016, **71**, 153.
13 M. Salkić and A. Selimović, *Croat. Chem. Acta*, 2015, **88**, 73.
14 B. Szpikowska-Sroka and J. Połedniok, *J. Anal. Chem.*, 2011, **66**, 941.
15 R. Monošík, V. B. dos Santos and L. Angnes, *Anal. Methods*, 2015, **7**, 8177.
16 Y. Yasunobu, M. Yasuyuki and K. Tomozo, *Bunseki Kagaku*, 1990, **39**, 567.
17 H. Filik, D. Aksu, D. Giray and R. Apak, *Drug Test. Anal.*, 2012, **4**, 493.
18 S. Kathiravan, E. Sundaram, B. A. Paulraj, P. M. Johnson, S.-T. Huang, V. Mani and V. S. Vasantha, *Food Chem.*, 2020, **332**, 127150.
19 P. Lv, Y. Xu, Z. Liu, G. Li and B. Ye, *Microchem. J.*, 2020, **152**, 104255.
20 D. C. M. Ferreira, G. F. Giordano, C. C. dos S. P. Soares, J. F. A. de Oliveira, R. K. Mendes, M. H. Piazzetta, A. L. Gobbi and M. B. Cardoso, *Talanta*, 2015, **141**, 188.
21 S. S. Nielsen, *Food Analysis Laboratory Manual (Food Science Text Series)*, 3rd edn., Springer, Cham, 2017.
22 N. A. Gavrilenko, A. V. Sukhanov and O. V. Mokhova, *J. Anal. Chem.*, 2010, **65**, 17 (*Zh. Anal. Khim.*, 2010, **65**, 20).
23 G. Goodlet and R. Narayanaswamy, *Meas. Sci. Technol.*, 1994, **5**, 667.
24 M. Siu, V. A. Yaylayan, J. M. R. Bélanger and J. R. J. Paré, *Tetrahedron Lett.*, 2005, **46**, 5543.
25 D. T. Newcombe, T. J. Cardwell, R. W. Cattrall and S. D. Kolev, *Lab. Rob. Autom.*, 2000, **12**, 200.
26 N. A. Gavrilenko, G. M. Mokrousov and O. V. Dzhiganskaya, *J. Anal. Chem.*, 2004, **59**, 871 (*Zh. Anal. Khim.*, 2004, **59**, 967).
27 G. Absalan, M. Arabi and J. Tashkhourian, *Anal. Sci.*, 2012, **28**, 1225.
28 I. S. A. Porto, J. H. S. Neto, L. O. dos Santos, A. A. Gomes and S. L. C. Ferreira, *Microchem. J.*, 2019, **149**, 104031.
29 V. B. dos Santos, E. K. N. da Silva, L. M. A. de Oliveira and W. T. Suarez, *Food Chem.*, 2019, **285**, 340.
30 L. Suntornsuk, W. Gritsanapun, S. Nilkamhank and A. Paochom, *J. Pharm. Biomed. Anal.*, 2002, **28**, 849.
31 M. Ismail, S. Ali and M. Hussain, *Int. J. Pharm. Qual. Assur.*, 2014, **5**, 22.
32 Y. Bazel, T. Riabukhina and J. Tirpák, *Microchem. J.*, 2018, **143**, 160.
33 T. L. da Silva, E. Aguiar-Oliveira, M. R. Mazalli, E. S. Kamimura and R. R. Maldonado, *Food Chem.*, 2017, **224**, 92.

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