International Journal of Pharmacy Teaching & Practices 2014, Vol.5, Issue 4, 1571-1573.

Rapid Resolution Method Determination of Parabens in Pharmaceutical Cream

Larisa Alagić-Džambić*, Midhat Vehabović, Nina Avdić, Selma Kaljun, Nihad Kokorović, Mirsad Džambić
Development Department, Bosnalijek d.d. Sarajevo, Jukićeva 53, 71000 Sarajevo, Bosnia and Herzegovina

Research Article

Please cite this paper as Larisa Alagić-Džambić*, Midhat Vehabović, Nina Avdić, Selma Kaljun, Nihad Kokorović, Mirsad Džambić. Rapid Resolution Method Determination of Parabens in Pharmaceutical Cream. IJPTP, 2014, 5(4), 1571-1573.

Corresponding Author:
Larisa Alagić-Džambić
Development Department,
Bosnalijek d.d. Sarajevo, Jukićeva 53,
71000 Sarajevo, Bosnia and Herzegovina
E-mail: larisatravnik@gmail.com
Phone No: +38733254556

Abstract

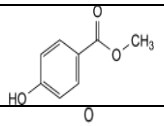
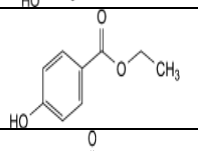
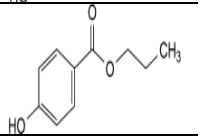
Parabens are p-hydroxybenzoic acid esters. They are widely used preservatives in cosmetic preparations. Because of theirs endocrine effect, use of paraben is limited. In Europe, limit of concentration of parabens individually is not more then 0.4%, and in mixture not more then 0.8%. In United States concentration levels are from 0.01% to 0.3%. The aim of this work is to have good separation of three parabens used in pharmaceutical cream, using Rapid Resolution System. The optimum results were obtained by the use of column Poroshel 120 SB C-18; 3 mm x 100 mm; 2.7 µm, kept at 40°C. The mobile phase was 60% V/V methanol. Flow rate was 0.4 ml/min. UV detection was performed at 254 nm. This method was validated following ICH guidelines. All parabens were separated within 5 minutes.

Keywords: *Methyl parahydroxybenzoate; Ethyl parahydroxybenzoate, Propyl parahydroxybenzoate, Rapid Resolution Method; Cream*

Introduction

Metyl, ethyl and propyl parahydroxybenzoate are synthetic parabens and they were developed from benzoic acid. It is estimated that over 90% all cosmetic preparations have some form of parabens. The structures of parabens are in Table 1. All of them have phenyl ring, and because of that they are detectable in extremely low concentration. Also, they are lipophilic and slightly soluble in water. Solubility in water decreasing as the ester chain length increases (methyl, ethyl, propyl etc..). Many reversed-phase separation of parabens are published in the chromatograpy literature. The objective of our study was to develop and validated Rapid Resolution Liquid Chromatography method for determination three parabens with good separation in short time. The system provides faster analyses and higher resolution then conventional liquid chromatography [1, 2, 3, 4, 5, 6].

Table 1: Chemical structures of the assayed compounds

Name	Molecular form	Molecular weight	Structural form
Methyl parahydroxybenzoate	$C_8H_8O_3$	152.1	
Ethyl parahydroxybenzoate	$C_9H_{10}O_3$	166.2	
Propyl parahydroxybenzoate	$C_{10}H_{12}O_3$	180.2	

Material and Method

Standards of methyl parahydroxybenzoate is 99.5% purity, ethyl parahydroxybenzoate is 98.8% purity and propyl parahydroxybenzoate is 99.8% purity. Chemicals, methanol and ethanol were from J. T. Baker, gradient grade, and other chemicals, hydrochloric acid 37%, sodium hydroxide pellets and hydrogen peroxide 30% were from Merck, pro analysi grade. Water was purified with a Milli-Q system from Millipore. The HPLC system was Waters Acquity UPLC H-class. All excipients used in our

formulation are tested according their monograph and they meet the specifications and quality.

Apparatus and chromatographic conditions

The optimum results were obtained by the use of column Poroshel 120 SB C-18; 3 mm x 100 mm; 2,7 µm, kept at 40°C. The mobile phase was 60% V/V methanol. Flow rate was 0.4 ml/min. UV detection was performed at 254 nm. Injection volume was 1 µl. Solvent solution for standards and sample solution was 50% V/V ethanol.

Standards and sample preparation

Standard solution was prepared as mixture of methyl parahydroxybenzoate in concentration 0.0195 mg/ml and ethyl and propyl parahydroxybenzoate were prepared in concentration 0.0305 mg/ml. Request was to have this System suitability:

1. Relative standard deviations (RSD) for methyl parahydroxybenzoate, ethyl parahydroxybenzoate and propyl parahydroxybenzoate peaks for six replicate injections in standard solution are not more than 2.0%.
2. Tailing factors (TF) for methyl parahydroxybenzoate, ethyl parahydroxybenzoate and propyl parahydroxybenzoate peaks in standard solution are not more than 2.0.
3. Theoretical plates (TP) for methyl parahydroxybenzoate, ethyl parahydroxybenzoate and propyl parahydroxybenzoate peaks in standard solution are not less than 1500.

Test solution were prepared by weigh about 1.0 g of cream into a 250 ml Erlenmayer flask, add 100.0 ml of 50% V/V ethanol and shut with stopper. Mix solution in an ultrasonic bath until the cream has melted on a temperature of 50°C with occasional shaking. Centrifuge solution at 4000 rpm for 15 minutes and filter through a nylon filter 0.20 µm.

Validation

The selectivity was tested by runing solution caontaining all excipents cream in the same concentrations and conditoins like samples. Retention time for Methyl parahydroxybenzoate is about 1.7 minutes, for Ethyl parahydroxybenzoate is about 2.1 minutes and for Propyl parahydroxybenzoate is about 2.9 minutes.

We also did forced degradation of standard solutions. Forsed degradation was carried out by oxidation of the solution (with 3% hydrogen peroxide, kept 2 hours at 90°C), alkaline hydrolysis of the solution (with 1 mol/l sodium hydroxide solution kept 2 hours at room condition at about 25°C), acid hydrolysis of the solution (with 1 mol/l hydrochloric acid solution, kept 2 hours at 90°C), thermal decomposition of the solution (with solvent solution, kept 2 hours at 90°C) and photolysis of the solution (solution exposed to daylight for 72 hours).

The linearity was tested by prepering standard solutions of parabens from 50 to 120% of the target analyte concentration. The accuracy of the method was tested by applaying mixture of parabens and excipients by triplicate in three levels (80, 100 and 120%). The precision was tested like system repeatibility and method repeatibility. In system repeatibility we tested by runing 6 repelication of test solution in target concentration. In method repeatibility we tested 6 different sample solutions in target concentration. We also tested reproducibility on two different batches of cream, analyzing from two different analyst. The robustness of the method was tested by changing the UV detection, flow rate, column temperature and mobile phase ratio.

Results and Discussion

In Figure 1, we can see very good separation between parabens in cream. We also meets requirements of System Suitability. Main validations parameters are shown in Tables 1, 2, 3 and 4.

Table 1: Precision, Linearity and Recovery

	Methyl parahydroxybenzoate	Ethyl parahydroxybenzoate	Propyl parahydroxybenzoate
System Suitability			
% RSD	0.50	0.46	0.38
TF	1.2	1.2	1.2
TP	6557	8847	12269
Linearity (R^2)	0.99998	0.99997	0.99996
Accuracy (% RSD) n=9	99-101	99-101	98-101
Response precision (System) (% RSD) n=6	0.14	0.12	0.14
Response precision (Method)			
(% RSD) n=6	0.88	0.97	0.47
(% C.L.)	0.93	1.02	0.50
Reproducibility			
(% RSD)- Batch A	0.9	1.0	1.2
(% RSD)- Batch B	0.9	0.8	1.5
Robustness (Mean Recovery in % and % RSD)			
UV detection			
Flow rate	102	102	102
Column temperature	RSD=0.02 to 1.34	RSD=0.13 to 2.27	RSD=0.05 to 0.47
Mobile phase ration			

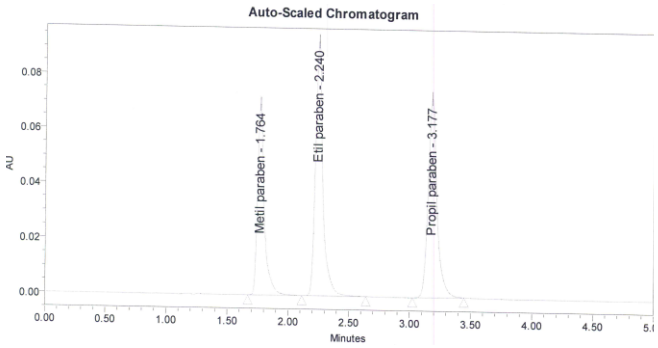


Fig. 1 Chromatogram of the standard solution for parabens

Table 2: Peak purity for standard solution of Methyl parahydroxybenzoate

Forsed degradation conditions	Purity angle	Purity threshold
Oxidation of the solution	0.315	0.336
Acid hydrolysis of the solution	0.305	0.346
Alkaline hydrolysis of the solution	0.489	0.813
Thermal decomposition of the solution	0.320	0.344
Photolysis of the solution	0.240	0.321

Table 3: Peak purity for standard solution of Ethyl parahydroxybenzoate

Forsed degradation conditions	Purity angle	Purity threshold
Oxidation of the solution	0.101	0.304
Acid hydrolysis of the solution	0.098	0.293
Alkaline hydrolysis of the solution	0.125	0.319
Thermal decomposition of the solution	0.226	0.303
Photolysis of the solution	0.101	0.293

Table 4: Peak purity for standard solution of Propyl parahydroxybenzoate

Forsed degradation conditions	Purity angle	Purity threshold
Oxidation of the solution	0.123	0.319
Acid hydrolysis of the solution	0.123	0.325
Alkaline hydrolysis of the solution	0.148	0.349
Thermal decomposition of the solution	0.119	0.316
Photolysis of the solution	0.121	0.320

Conclusion

The application of Rapid Resolution LC method to analysis parabens in cream at first reduce chromatographic time. As a reminder, run time was 5 minutes. The method was shown linear response for all parabens. Using of 50% V/V ethanol as solvent was well selected because of theirs low solubility. Paraben preservatives are quickley analyzed using rapid resolution column Poroshel 120 SB C-18; 3 mm x 100 mm; 2,7 µm. It can be conlcuded that proposed method provides an alternative procedure for quality control of methyl parahydroxybenzoate, ethyl parahydroxybenzoate and propyl parahydroxybenzoate in pharmaceutical and cosmetic products, in this case cream.

References

1. Robert R. High Speed Separation of Parabens. Agilent Technologies publication. Available at http://www.agilent.com/chem.
2. M. Borremans, J. Van-Loco, P. Roos and L. Goeyenes. Chromatographia 59., 2004: 47-53.
3. U.D. Uysal, T. Guray. Determination of Parabens in Pharmaceutical and Cosmetic Products by Capillary Electrophoresis. Journal of Analytical Chemistry.63., 2008: 982-986.
4. ICH, Validation of Analytical Procedure: Methodology (Q2B), International Conference on Harmonization, Geneva 1996.
5. International Conference on harmonization of technical requirements for registration of pharmaceuticals for human use. Validation of analytical procedures: text and methodology Q2 (R1). Current Step 4 Version, 2005.
6. European pharmacopoeia, Council of Europe, Strasbourg, 8th ed., 2015.

AUTHORS' CONTRIBUTIONS

Authors contributed equally to all aspects of the study.

PEER REVIEW

Not commissioned; externally peer reviewed.

CONFLICTS OF INTEREST

The authors declare that they have no competing interests.