*p*H -Sensitive Clays as Drug Delivery Carriers for Controlled Release of Hydrocortisone

ADI GHEBAUR[1]*, BRINDUSA BALANUCA[1], SORINA ALEXANDRA GAREA[1], HORIA IOVU[1,2]

[1]Advanced Polymer Materials Group University Politehnica of Bucharest, 1-7 Gheorghe Polizu Str., 011061, Bucharest, Romania

[2]Academy of Romanian Scientists, 54 Splaiul Independentei, 50085, Bucharest, Romania

The main goal of this study was to modify halloysite with polyethylene glycol methacrylate and polyethylene glycol dimethacrylate in order to realize some pH sensitive controlled drug delivery systems. The obtained samples were characterized by spectral techniques (FTIR, XPS) and thermal techniques (TGA, DSC). There was also studied the influence of clay type and initial clay amount onto the drug adsorption process within halloysite. The hydrocortisone release profile was also investigated in simulated gastric fluid and simulated intestinal fluid.

Keywords: halloysite, modification, drug encapsulation, methacrylate groups, hydrocortisone

In drug delivery field clays [1-3] are the most used materials because they are non-toxic and chemically inert. Also they have a high surface area, high cationic capacity and high adsorption capacity. They can be used as excipients in pharmaceutical industry or as drug carriers at the site of action [4-6]. There is a wide range of natural and synthetic clays, but recently halloysite started to be intensely studied [7-9] because it is an aluminosilicate with a morphology similar with carbon nanotubes. It presents a hallow tubular structure and the walls are porous and can be loaded with drugs or other active substances [10]. On the outer surface of halloysite are present Si-OH groups that are negatively charged while on the inner surface can be found positively charged Al-OH groups [11].

The *p*H sensitive materials are usually used in drug delivery field because pH values vary in human body (ex: stomach, intestine, liver, tumors) [12]. The hybrid materials synthesized for this type of drug delivery systems are based on biodegradable polymers with methacrylate groups like poly(methacrylic acid) (PMMA), poly[2-(*N,N*-diethylamino)ethyl methacrylate] (PDMAEMA), poly(hydroxyethyl methacrylate) (PHEMA) etc [13-15]. Some researchers tried to modify halloysite in order to use it for oral administration in cancer treatment [16].

Hydrocortisone is a corticosteroid that is used in the treatment of a wide range of diseases: skin disease, lung, adrenal insufficiency, bowel diseases due to its anti-inflammatory and immunosuppressive properties. It can be administered as tablets, injections, cream. The use of hydrocortisone in high concentration over a long time period may lead to osteoporosis and produce neurotoxic effects on central nervous system [18-21].

The aim of this study was to modify halloysite with polyethylene glycol methacrylate and polyethylene glycol dimethacrylate in order to synthesize some *p*H sensitive materials that can be used as sustained drug delivery systems of hydrocortisone. The synthesized materials were characterized by different techniques like FTIR, XPS, TGA, DSC and UV-VIS. Also the hydrocortisone release profile was monitored in two different dissolution media (simulated gastric fluid, SGF, and simulated intestinal fluid, SIF).

Experimental part

Materials and methods

Raw materials

Halloysite (HNT), polyethylene glycol methacrylate (Mw = 360 g/mol) (PEGm), polyethylene glycol dimethacrylate (Mw=720 g/mol) (PEGdm), toluene, hydrocortisone, sodium hydroxide, potassium phosphate monobasic, hydrochloric acid, sodium chloride, azoisobutyronitrile (AIBN) were purchased from Sigma Aldrich and were used as received.

Synthesis of pH sensitive drug delivery systems

HNT modification

The modification of HNT was realized with PEG m and PEGdm. A certain amount of HNT was first dispersed in toluene and maintained for 30 min at 60 °C. Then a specific amount of modifying agent (1:1 HNT:modifying agent) was added and the mixture was refluxed for 6 h under nitrogen atmosphere. The suspension was centrifuged and washed once with toluene and twice with water. The modified HNT (HNT-PEGm, HNT-PEGdm) were dried under vacuum for 24 h at 50 °C.

Hydrocortisone loading within caly

The drug encapsulation within the unmodified and modified HNT was realized at room temperature varying the initial clay amount (200 mg, 300 mg, 500 mg) for each

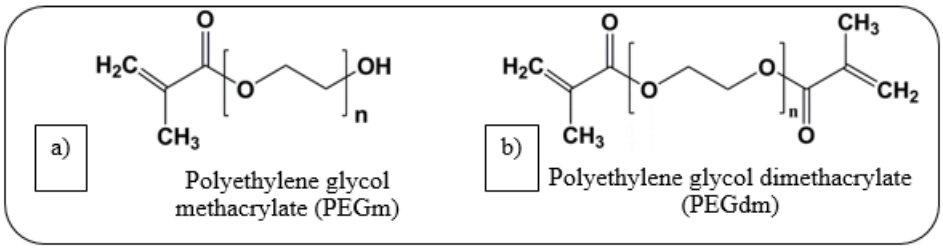


Fig. 1. Chemical structure of modifying agents: a) Polyethylene glycol methacrylate (PEGm); b) Polyethylene glycol dimethacrylate (PEGdm)

* email: ghebauradi@yahoo.com; Phone: (+40) 214022726

type of HNT. Briefly in a solution of 0.08 mg/mL concentration hydrocortisone a certain amount of clay was dispersed at room temperature and the mixture was maintained for 1 h allowing the drug molecule to be loaded within the HNT pores and lumen. The samples (HNT200, HNT300, HNT500, HNT-PEGm200, HNT-PEGm300, HNT-PEGm500, HNT-PEGdm200 HNT-PEGdm300 HNT-PEGdm500) were dried at room temperature for 48 h.

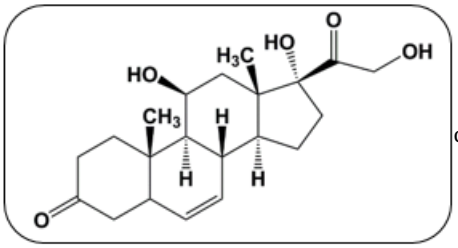


Fig. 2. Hydrochortisone chemical structure

Characterization methods

FTIR spectra were recorded on a Bruker VERTEX 70 spectrometer using 32 scans with a 4 cm^{-1} resolution.

Thermogravimetric analysis (TGA) was done on Q 500 TA instruments. The samples of 3 mg were heated from 20 to 800 °C at a scanning rate of 10°C/min under a constant nitrogen flow rate (40 mL/min).

X-Ray Photoelectron Spectroscopy (XPS) spectra were recorded on Thermo Scientific K-Alpha equipment, fully integrated, with an aluminum anode monochromatic source.

DSC curves were registered on a Netzsch DSC 204 F1 Phoenix equipment. The samples were first mixed with 2 wt. % AIBN and then were heated from 20 to 200 °C using a heating rate of 10 °C/min, under a constant nitrogen flow rate (40 mL/min).

UV-Visible absorbance of hydrocortisone solutions was measured at λ_{max} = 248 nm on a Cary 60 UV/Vis spectrophotometer equipment provided with a quartz cell having a light path of 10 mm.

In vitro drug release

The drug release profile was determined in an automated dissolution USP Apparatus 1 (708-DS Agilent) with an autocotrolled multi-chanel peristaltic pump (810 Agilent), an UV/Vis spectrophotometer (Cary 60) with 1 mm flow cell and UV-Dissolution software. The samples were introduced in a dialysis bag with 3 mL of buffer solution and were immersed in 200 mL of dissolution medium at 37 °C. . Rotational speed of 75 rpm was tested. The samples were maintained for 700 min in the dissolution medium (SGF or SIF). At specific time intervals the amount of released hydrocortisone was determined with a known concentration of the standard solutions at 248 in both dissolution media.

Results and discussions

FTIR Analysis

The modification of halloysite with PEGm and PEGdm was proved in the FTIR spectra (fig. 3). In HNT-PEGm and HNT-PEGdm FTIR spectra are present a couple of new peaks than in the neat HNT spectrum. The peak from 2950 cm^{-1} is attributed to the C-H asymmetric stretching vibration from CH_3 group while the peak from 2877 cm^{-1} is assigned to the C-H symmetric stretching vibration from the O-CH_2 groups present in the PEG chain. The peaks characteristic to the methacrylic group are present at 1715 cm_{-1} attributed to C=O stretching vibration and at 1639 cm_{-1} assigned to C-O stretching vibration [22].

XPS Analysis

Surface composition of pristine HNT and modified HNT with PEGm respectively PEGdm was investigated by XPS. In figure 4 are shown the survey spectra for the three samples while the elemental surface composition is presented in table 1. As can be observed the presence of PEGm and PEGdm on HNT surface is emphasized by the appearance of C1s peak in the spectra. Regardless of the modification agent type C1s species is present in

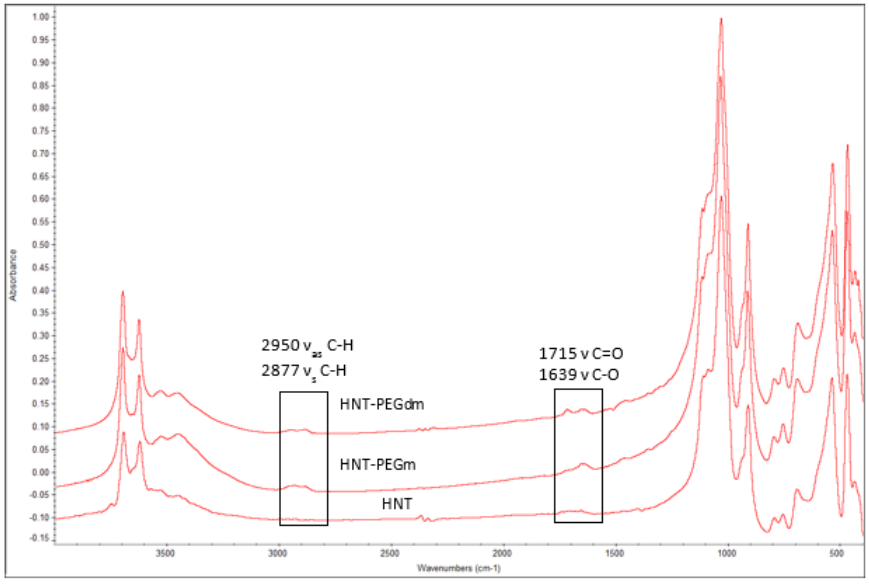


Fig. 3. FTIR spectra of HNT-PEGm, HNT-PEGdm

Sample	O1s, %	Al2p, %	Si2p, %	Na1s, %	C1s, %
HNT	63.1	17.9	17.6	1.4	
HNT-PEGm	54.7	15.3	14.8	-	15.2
HNT-PEGdm	53	15.2	15.5	-	16.3

Table 1
THE CONCENTRATION OF O1s, A12p, Si2p, Na1s SPECIES DETERMINED FROM XPS ANALYSIS

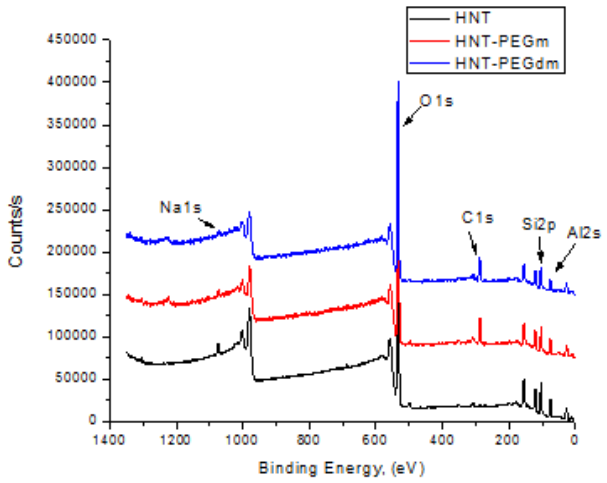


Fig. 4. XPS survey spectra of HNT, HNT-PEG and HNT-PEGdm

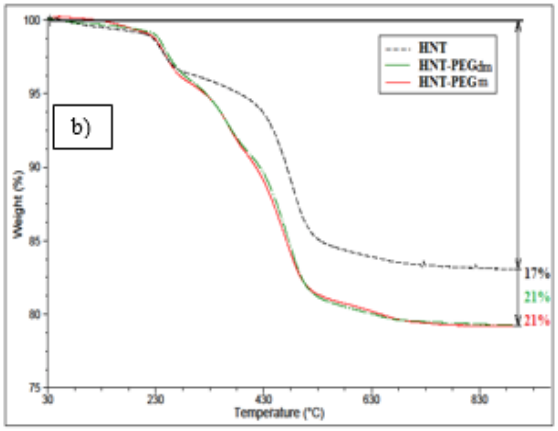
Efficiency encapsulation clay type and initial clay amount influence

The influence of initial drug concentration on the encapsulation efficiency process of hydrocortisone onto HNT was studied. The encapsulation efficiency (EE %) was calculated using equation 1.

$$EE\% = \frac{m_0 - m}{m_0} \times 100 \quad (1)$$

where m_0 is the initial hydrocortisone amount (g), m is the unloaded hydrocortisone amount (g).

In table 2 are summarized the EE% of hydrocortisone in different types and amounts of clays. It may be observed that the clay type doesn't have an important influence onto EE%. The modified HNT encapsulate with 5 % HNT-PEGm300 respectively 7 % HNT-PEGdm300 more hydrocortisone than pristine HNT probably because the modifying agents create a network where drug can be



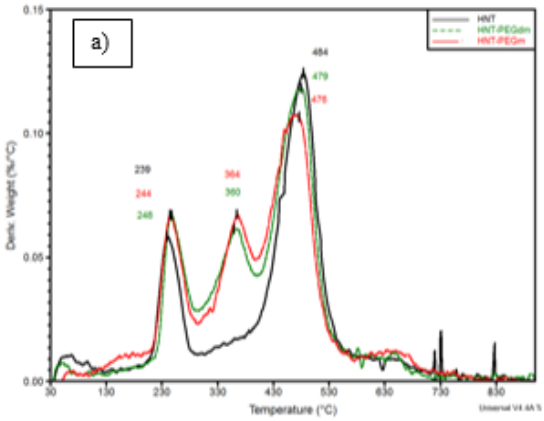


Fig. 5. Thermogravimetric alanalysis of unmodified and modified HNT: TGA curves (a); DTG curves (b)

approximately the same concentration in the two samples 15.2 % in HNT-PEGm, respectively 16.3 % in HNT-PEGdm.

Thermogravimetric analysis

The presence of the organic modifying agents onto HNT surface was also demonstrated by thermogravimetric analysis. In figure 5 are presented the TGA respectively DTG curves of the unmodified and modified HNT with organic modifying agents (PEGm, PEGdm). The overall weight loss of modified HNT (21 wt. %) is higher than for unmodified HNT (17 wt.%) due to the presence of the organic modifying agents attached by the aluminosilicate. As can be observed from the TGA curves (fig. 5a) the unmodified HNT presents two degradation steps. The first one where Tmax is at 240 °C is attributed to the dehydration of aluminosilicate and the second degradation step with a Tmax at 484 °C is assigned to the clay dehydroxylation. Both modified types of HNT present three steps of degradation. In this case the third step is attributed to the degradation of the organic component that takes place at 364 °C for HNT-PEGm respectively 360 °C for PEGdm.

DSC analysis

DSC thermograms of unmodified and modified HNT (fig. 6) offers information regarding the attaching of methacrylic groups by the aluminosilicate. As it can be observed from the DSC curves in the modified HNT an exothermic peak (120 °C) appears that corresponds to the polymerization peak of the methacrylic group. This is another proof that the modification of HNT with PEGm and PEGdm took place.

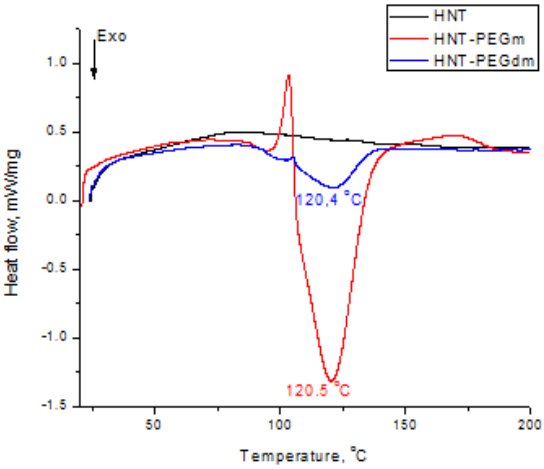


Fig.6. DSC curves of pristine HNT, HNT-PEGm, HNT-PEGdm

load. An important influence onto EE% it has the initial clay amount. The higher the initial clay amount is the higher amount of drug is encapsulated, 43% for HNT-PEGdm500, because there is a high number of hallows and pores from clay where the drug can be adsorbed.

In vitro drug release

The drug release profile of hydrocortisone from clays was monitored in SIF and SGF. There were also studied the influence of clay type (fig. 7) and initial clay amount (fig. 8). The presence of methacrylic groups onto HNT reduces the drug release in SGF and SIF. The chains of the modifying agents hinder the drug molecules movement resulting a lower release rate. The total drug amount released in SGF

Sample name	Clay amount, mg	Initial drug amount, mg	EE%
HNT300	300	40	21
HNT-PEGm300	300	40	26
HNT-PEGdm200	200	40	22
HNT-PEGdm300	300	40	28
HNT-PEGdm500	500	40	43

Table 2
THE INFLUENCE OF CLAY TYPE AND INITIAL CLAY MOUNT ONTO ENCAPSULATION EFFICIENCY

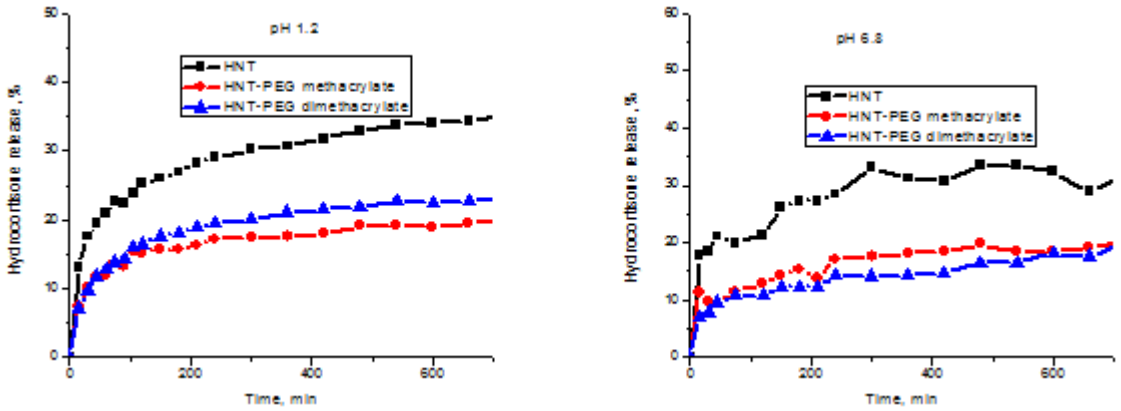


Fig.7. Hydrocortisone release profiles from different types of clays in different dissolution media

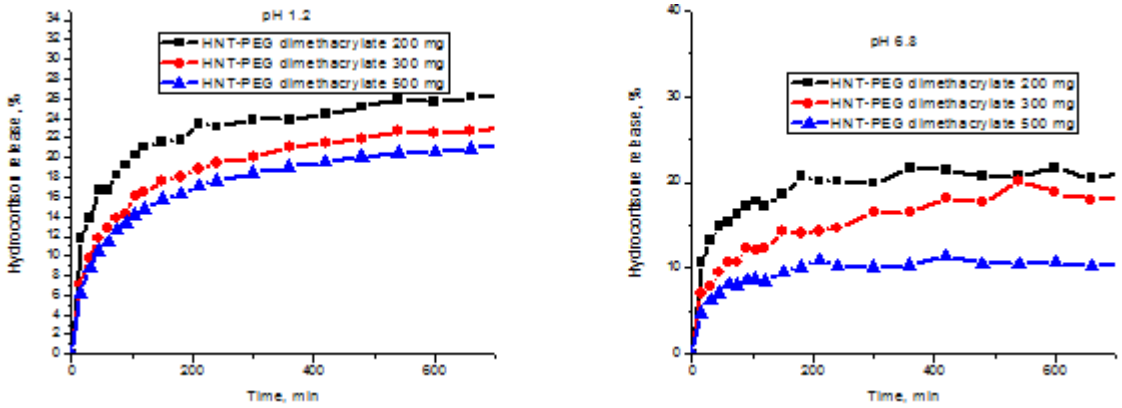


Fig.8. Hydrocortisone release profiles from HNT-PEGdm with different initial clay amounts in different dissolution media

is lower than in SIF [23]. In figure 8 are shown the hydrocortisone release profiles in SGF ad SIF from clays with different initial amounts of clays. As can be observed the sample that has the highest initial amount of clay releases the lowest amount of drug no matter the dissolution medium.

Conclusions

Some new clays that may be used as *p*H sensitive drug delivery systems were synthesized. The HNT modification with PEGm and PEGdm was proved by FTIR, XPS, TGA and DSC. The presence of the modifying agent on HNT has a small influence onto hydrocortisone adsorption. Instead an important influence onto the adsorption process it has the initial clay amount, the higher it is, a high amount of drug is encapsulated, 43% when 500 mg of HNT-PEGdm was used. The presence of methacrylic groups onto HNT surface leads to a reduced drug amount release in acid medium. The smallest drug amount released was also recorded for HNT-PEGdm in SGF.

Acknowledgement: The work has been funded by the Sectoral Operational Programme Human Resources Development 2007-2013 of the Ministry of European Funds through the Financial Agreement POSDRU/159/1.5/S/132397.

References

1.LAL, S., DATTA, M., Appl. Clay Sci., 114, 2015, p. 412
2.OLIVEIRA, M.J.A., ESTEFANIA, O.S., LUCIA, M.A.B., REGINA, M., AMATO, V.S., LUGAO, A.B. PARRA, D.F., Radiat. Phys. Chem., 94, 2014, p. 194
3.KEVADIYA, B.D., CHETTIAR, S.S., RAJKUMAR, S., BAJAJ, H.C., GOSAI, K.A., HARSHAD BRAHMBHATT, H., Colloid. Surface. B., 112, 2013, p. 400
4.CARRETERO, M.I., POZO, M., Appl. Clay Sci., 46, 1, 2009, p. 73
5.DATTA, S.M., Appl. Clay Sci., 80–81, 2013, p. 85
6.KEVADIYA, B.D., JOSHI, G.V., MODY, H.M., BAJAJ, H.C., Appl. Clay Sci., 52, 4, 2011, p. 364
7.AHMED, F.R., SHOAIB, M.H., AZHAR, M., UM, S.H., YOUSUF, R.I., HASHMI, S., DAR, A., Colloid. Surface. B., 135, 2015, p. 50

8.GHEBAUR, A., GAREA, S.A., IOVU, H., Int. J. Pharm., 436, 1–2, 2012, p. 568
9.TAN, D., YUAN, P., ANNABI-BERGAYA, F., LIU, D., WANG, L., LIU, H., HE H., Appl. Clay Sci., 96, 2014, p. 50
10.Rosas-Aburto, A., Gabaldón-Saucedo, I.A., Espinosa-Magaña, F., Ochoa-Lara, M.T., Roquero-Tejeda, P., Hernández-Luna, M., Revilla-Vázquez, J., Micropor. Mesopor. Mat., 218, 2015, p. 118
11.CAVALLARO, G., LAZZARA, G., MILIOTO, S., PARISI, F., SPARACINO, V., Polym. Degrad. Stabil., 120, 2015, p. 220
12.LIU, J., HUANG, Y., KUMAR, A., TAN, A., JIN, S., MOZHI, A., LIANG, X-J., Biotechnol. Adv., 32, 4, 2014, p. 693
13.CONSTANTIN, M., BUCATARIU, S., HARABAGIU, V., POPESCU, I., ASCENZI, P., FUNDUEANU, G., Eur. J. Pharma. Sci., 62, 2014, p. 86
14. YANG, Y.Q., ZHAO, B., LI, Z.D., LIN, W.J., ZHANG, C.Y., GUO, X.D., WANG, J.F., ZHANG, L.J., Acta Biomater., 9, 8, 2013, p. 7679
15.KIERYS, A., GROCHOWICZ, M., KOSIK, P., Micropor. Mesopor. Mat., 217, 2015, p. 133
16.RAO, K.M., NAGAPPAN, S., SEO, D.J., HA, C-S., Appl. Clay Sci., 97–98, 2014, p. 33
17.LIU, J., HUANG, Y., KUMAR, A., TAN, A., JIN, S., MOZHI, A., LIANG, X.-J., Biotechnol. Adv., 32, 4, 2014, p. 693
18.SMAJDOR, J., PIECH, R., RUMIN, M., BATOR, B.P., Electrochim. Acta., 182, 2015, p. 67
19.FU, Z.-F., LI, G.-K., HU, Y.-F., Chinese J. Anal. Chem., 43, 9, 2015, p. 1322
20.WERUMEUS BUNING, J., BRUMMELMAN, P., KOERTS, J., DULLAART, R.P.F., VAN DEN BERG, G., VAN DER KLAUW, M.M., TUCHA, O., WOLFFENBUTTEL, B.H.R., VAN BEEK A.P., Psychoneuro-endocrino., 55, 2015, p. 36
21.MILLER, R., WECKESSER, L.J., SMOLKA, M.N., KIRSCHBAUM, C., PLESSOW, F., Psychoneuroendocrino., 53, 2015, p. 148
22.BARRIENTOS-RAMIREZ, S., OCA-RAMÍREZ, G.M.D., RAMOS-FERNÁNDEZ, E.V., SEPÚLVEDA-ESCRIBANO, A., PASTOR-BLAS, M.M., GONZALEZ-MONTIEL, A., Appl. Catal. A-Gen., 406, 1–2, 2011, p. 22
23.LEVIS, S.R., DEASY, P.B., Int. J. Pharm., 253, 2003, p. 145

Manuscript received: 14.10.2015