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WORKING OUT QUALITY STANDARDS OF MODEL COMPOSITION SAMPLES OF GRANULATED DOSAGE FORM WITH GLUTATHIONE RESTORED

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Biologically active sulfur-containing compounds (BASC) exhibit pronounced antioxidant properties. Glutathione reduced (GSH) occupies a particular position among these compounds. It represents a key link in the 3 antioxidant systems of the body from the existing four. Based on the foregoing, a GSH-based dosage form with antioxidant properties was proposed. The aim of this study is to work out a model granulated dosage form based on GSH and methods of its analysis by means of pre-column derivatization with ortho-phthalic aldehyde. ***Materials and methods.*** *GSH and granulated dosage form based on GSH obtained by wet granulation were used as the object of the study. Quantitative evaluation of GSH content in the obtained granules was carried out using pre-column derivatization by the method of reversed-phase high-performance chromatography (RP HPLC). Ortho-phthalic aldehyde was used as a derivatizing agent. A diode-array detector was used to detect the resulting derivative. Ortho-phthalic aldehyde was used as a derivatizing agent. A diode-matrix detector was used to find out the resulting derivative.* ***Results.*** *In the course of the work, a model dosage form was created – granules based on GSH. By reference to the recommendations on the dosage of the drug, the concentration of the active substance was selected. Lactose was chosen as an auxiliary component. Physical and technological characteristics of a model sample of granules with GSH and lactose as a filler were studied. A method of quantitative determination of GSH in granules using pre-column derivatization with ortho-phthalic aldehyde was developed and validated by HPLC. The method of quantitative determination of GSH in granules with the use of pre-column derivatization by ortho-phthalic aldehyde by HPLC was developed and validated.* ***Conclusion.*** *The developed granulated dosage form meets the requirements given in the pharmacopoeial item "Granules" according to the analyzed indicators. Using the validation evaluation it was established, that the developed methods for the quantitative determination of GSH in granules is correct, precise and specific.*

Keywords: *reduced glutathione, ortho-phthalic aldehyde, granules, reverse phase high performance liquid chromatography, derivatization, validation*

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РАЗРАБОТКА НОРМ КАЧЕСТВА ОБРАЗЦОВ МОДЕЛЬНОГО СОСТАВА ГРАНУЛИРОВАННОЙ ЛЕКАРСТВЕННОЙ ФОРМЫ С ГЛУТАТИОНОМ ВОССТАНОВЛЕННЫМ

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Биологически активные серосодержащие соединения (БАСС) проявляют ярко выраженные антиоксидантные свойства. Особое положение из числа данных соединений занимает глутатион восстановленный (GSH). Он представляет собой ключевое звено 3-х антиоксидантных систем организма из существующих четырех. Исходя из вышесказанного, нами была предложена лекарственная форма на основе GSH, обладающая антиоксидантными свойствами. ***Целью*** *данного исследования является разработка модельной гранулированной лекарственной формы на основе GSH и методики ее анализа с помощью предколоночной дериватизации орто-фталевым альдегидом.* ***Материалы и методы.*** *В качестве объекта исследования использовали GSH и гранулированную лекарственную форму на основе GSH, полученную методом влажного гранулирования. Оценку количественного содержания GSH в полученных гранулах, проводился с помощью предколоночной дериватизации методом обращено-фазной высокоэффективной хроматографии (ОФ ВЭЖХ). В качестве дериватизурующего агента использован орто-фталевый альдегид. Для обнаружения образовавшегося деривата был применен диодно-матричный детектор.* ***Результаты.*** *В ходе работы была создана модельная лекарственная форма – гранулы на основе GSH. Исходя из рекомендаций по дозировке препарата подобрана концентрация действующего вещества. В качестве вспомогательного компонента была выбрана лактоза. Изучены физические и технологические характеристики модельного образца гранул с GSH и лактозой в качестве наполнителя. Разработана и отвалидирована методика количественного определения GSH в гранулах с использованием предколоночной дериватизации орто-фталевым альдегидом методом ВЭЖХ.* ***Заключение.*** *Разработанная гранулированная лекарственная форма по анализируемым показателям соответствует требованиям, приведенным в ОФС «Гранулы». При помощи валидационной оценки установлено, что разработанная методика количественного определения GSH в гранулах является правильной, прецизионной и специфичной.*

Ключевые слова: *глутатион восстановленный, орто-фталевый альдегид, гранулы, обращённо-фазная высокоэффективная жидкостная хроматография, дериватизация, валидация*

INTRODUCTION

Reduced glutathione (GSH) is Tripeptide containing amino acid residues of L-glutamic acid, glycine and L-cysteine. GSH is one of the non-enzymatic components involved in the antioxidant protection of living organisms. It acts as an effective antiradical agent and plays a key role in the life cycle of cells, causing their protection from free radicals, hydroperoxides and xenobiotics [1, 2]. GSH status is an indicator of cell functionality and viability [3, 4]. The exhaustion or change of its level inside the cell provokes a number of diseases such as cancer, neurodegenerative, cardiovascular [5, 6]. *In vitro* and *in vivo* studies confirm that GSH deficiency can cause cell death and mitochondrial damage due to an increase in the number of toxic forms of oxygen and an increase in the number of free radicals [7]. GSH is able to prevent the destruction of cells by conjugation with toxic substances and their metabolites. Glutathione conjugation occupies one of the central places in the mechanisms of biotransformation of a number of xenobiotics [8–10]. Nowadays, more than forty types of chemical compounds that react with GSH are known. The mating factor of such reactions is the presence of an electrophilic center capable of reacting with the SH-group of GSH [11]. Consequently, the xenobiotic detoxification system involving GSH plays a key role in the formation of the body's resistance to various influences and is one of the most important defense mechanisms of the cell. Hereby, GSH conjugates with xenobiotics are less reactive and more hydrophilic than the initial substances, so they are less toxic and eliminate from the body faster [12, 13]. GSH is also able to prevent the introduction of lipophilic

ОРИГИНАЛЬНАЯ СТАТЬЯ
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toxicants into lipid bilayer membranes [14]. GSH has a membrane stabilizing activity effect on the hepatocyte, increases the activity of enzymes and promotes detoxification and regenerative activity of the liver through the neutralization of free radicals [15]. Taking into account the spectrum of glutathione pharmacological activity, we have proposed a dosage form with GSH, which has pronounced antioxidant properties.

MATERIALS AND METHODS

As the object of the study, glutathione reduced (CAS No. 70-18-8, EC No. 2007254, Applichem, Germany) and the excipient, lactose, were used The choice of this component is due to the possibility of its application as a filler, a reagent and an agent that regulates some technological characteristics of the granules (stability, disintegration, etc.).

Advantages of lactose as an excipient [16]:

– inert material, high purity, neutral color;

– moisture resistance;

– physical and chemical stability;

– it is well subjected to grinding and sieving;

– its high degree of crystallization, low degree of amorphism.

The wetting angle of GSH with water is less than 90°, it is 45±5°, i.e. GSH is a hydrophilic substance, so purified water was used as a humidifier in the model sample of granules.

To obtain a model sample of granules, a wet granulation method was used. The procedure for the formation of granules occurs as a result of pushing the moistened mass through a perforated sieve.

The technological characteristics of the obtained granules were determined on the basis of the following techniques.

The degree of the granules flowability was established by the method of the State Pharmacopoeia of Russian Federation (SPh), XIV-th edition (pharmacopoeial item "Powder Flow") using a V-12A (VZZTO) vibrating device. The sizes of the obtained granules were determined using the sieve analysis in accordance with SPh, XIV-th edition (pharmacopoeial item "Wet sieve residue test"). The test of "disintegration" was carried out on the "Swinging basket" disintegration tester in accordance with SPh, XIV-th edition (pharmacopoeial item "Granules"). The results of the "Dissolution" test were noted on the "Impeller" dissolution tester in accordance with pharmacopoeial item 1.4.2.0014.15.

Quantitative determination of GSH in a granular dosage form was performed by method of reverse phase high-performance chromatography using pre-column derivatization of this therapeutic agent by ortho-phthalic aldehyde [17].

The analysis was performed on the chromatograph "Agilent Technologies 1200 Infinity" with automatic sampler, vacuum microdoser, gradient pump and thermostat. The gradient chromatography conditions were: mobile phase (A) – 1% aqueous solution of formic acid, (B) – ethyl alcohol 95%; column: *Ascentis express* C18 2.7 dm × 100 mm × 4.6 mm; mobile phase speed – 0.5 ml/min.; column temperature was +35±0.01°C; sample volume – 1 µl.

GSH lacks the chromophores suitable for its analysis by UV spectroscopy and HPLC with diode array detection. In this regard, we proposed the analysis of GSH by HPLC using chemical transformation with a derivatizing agent – ortho-phthalic aldehyde. As a result of the reaction, the obtained derivative of GSH and ortho-phthalic aldehyde acquires a chromophore label, which is fixed during chromatographic analysis using diode-matrix detection.

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Derivatization methods of glutathione-based granules

1 ml of a 0.1% solution of glutathione granules in a 0.05 M of aqueous solution of sodium tetraborate was placed in a 10 ml analysis bottle, 1 ml of a 0.35% solution of o-phthalaldehyde in ethanol was added, shaken thoroughly and immediately chromatographed.

Preparation of model test solutions for validation assessment methods

The characteristics of correctness and precision were investigated on model samples of the preparation with glutathione concentrations that correspond to 80%, 85%, 90%, 95%, 100%, 105%, 110%, 115% and 120% of their content relative to the nominal value. For the preparation of model samples of the drug a volumetric flasks with a capacity of 25 ml was used. The batches were taken directly from the components of the drug. The mass of the batches and the concentration of the resulting solutions of glutathione are shown in Table 2.

RESULTS AND DISCUSSION

On the base of the experimental data obtained, a model dosage form – granules based on glutathione – was created. The concentration of active substances was selected by reference to the recommendations of the

dosage of the drug (0.05–0.5 g). The composition of the granules in a single-dose package (3.0 g) is as folows: glutathione 0.1 g, lactose 2.9 g. At the next stage, organoleptic, physical and technological parameters of granules based on glutathione were determined. The results of the study are presented in Table 1.

Table 1. Pharmaceutical and technological characteristics of granulated model dosage form based on GSH

Indicator under study	Methods of determination	Values obtained experimentally	Reference values
Granule size	Pharmacopoeial item 1.1.0015.15	1.0–1.2 mm	Coarse fraction:> 1.2 mm Average fraction: 1.2 mm Fine fraction: 1.0 mm
Granule form	Pharmacopoeial item 1.2.1.0009.15	Anisodiametric (elongated)	Elongated: >3:1 Lamellar: 3:1 Equiaxed: 1:1
Tapped density	Pharmacopoeial item 1.4.2.0016.15	460.10 ±1.31 kg/m3 (Light)	Very heavy: >2000 kg/m3 Heavy: 1100–2000 kg/m3 Medium: 600–1100 kg/m3 Light: < 600 кг/м3
Flowability	Pharmacopoeial item 1.4.2.0016.15	7.14 ±0.25 g/sec (Good)	Excellent: 8.6–12.0 g/sec Good: 6.6–8.5 g/sec Satisfactory: 3.0–6.5 g/sec Permissible: 2.0–3.0 g/sec Poor: 1.0–2.0 g/sec Very poor: <1,0 g/sec
Disintegration	Pharmacopoeial item 1.4.2.0013.15	4±2 min.	Up to 15 min.
Stability	Pharmacopoeial item 1.4.2.0004.15	98.5±0.5%	Mot less than 97%
Glutathione release	Pharmacopoeial item 1.4.2.0014.15	99.9±0.50% (during 15 min.)	75% (during 45 min.))
Water solubility of granules	Pharmacopoeial item 1.2.1.0005.15	Soluble (1:30)	Highly water soluble: up to 1 ml/g Freely soluble: 1–10 ml/g Soluble: 10–30 ml/g; Sparingly soluble: 30–100 ml/g Slightly soluble: 100–1000 ml/g; Very slightly soluble: 1000–10000 ml/g Practicaly insoluble: more than 10000 ml/g
Weight loss on drying	Pharmacopoeial item 1.2.1.0010.15	1.5±1%	
Uniformity of dosing	Pharmacopoeial item 1.4.2.0008.15	In progress	
Glutathione content, g	Reverse phase highly efficient liquid chromatography 100.0±0.39%	100.09±0.39%	

According to the data of Table 1, glutathione-based granules are elongated particles of 1.0-1.2 mm. They belong to light grains and have a satisfactory flowability. According to the indicators given in the pharmacopoeial item “Granules” (disintegration, uniformity of dosage, dissolution) this dosage form meets the requirements. Glutathione-based granules can be used to fill capsules, as well as an independent dosage form. The worked out granules are soluble and highly soluble in warm water. Glutathione-based granules are an oral dosage form with preliminary dissolution in liquid.

For the purpose of quantitative determination of glutathione recovered in a granulated dosage form, a method for pre-column derivatization with ortho-phthalic aldehyde has been developed. Derivatization with the specified modifier usually occurs within 2-3 minutes. The resulting product is easily detected using a diode array or a fluorimetric detector. The wavelength of the resulting derivative is usually λ – 337 nm. The molar ratios of o-phthalaldehyde and glutathione are 3.5: 1. When chromatographing OPHA-derivative of glutathione, there is one peak with a retention time of 18.066 minutes. The UV spectrum of the derivative exhibits several absorption maxima, the most specific of which is λmax – 336 mn. At the same time, the peak of the derivatizer itself is not visible, due to a different maximum absorption of the derivative (Fig. 1).

ОРИГИНАЛЬНАЯ СТАТЬЯ
DOI: 10.19163/2307-9266-2019-7-1-13-19

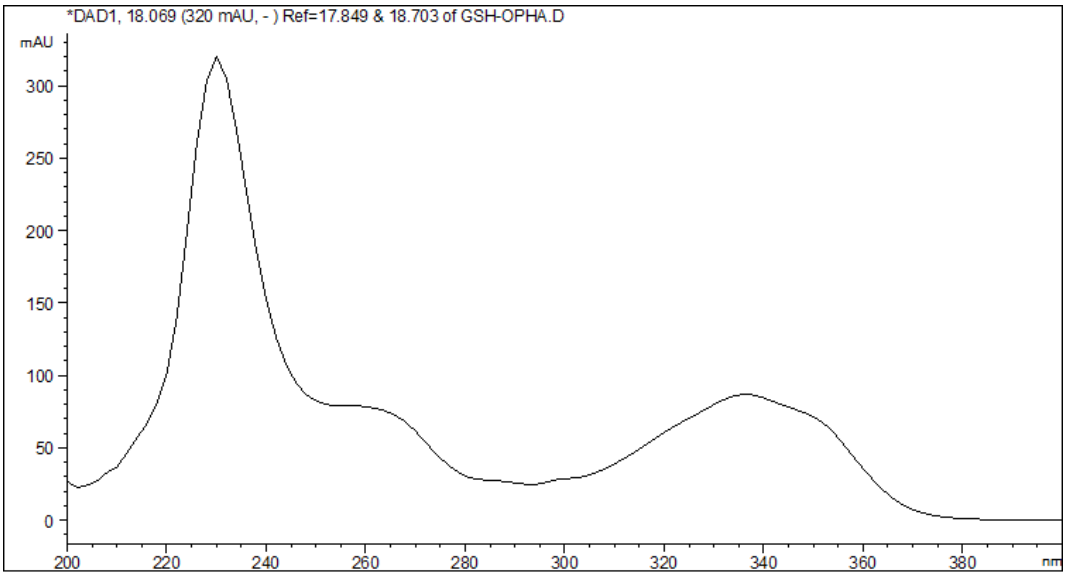

Figure 1. UV spectrum of o-phthalaldehyde derivative with GSH at the wavelength of 336 nm in a granulated dosage form

To confirm the possibility of using the proposed method of identification and quantitative determination of glutathione by the method of pre-column derivatization with ortho-phthalic aldehyde in granules, a validation assessment was carried out according to the characteristics: specificity, linearity, convergence (precision) and correctness [18].

Validation parameters of correctness, precision and linearity were studied on the preparation solutions with glutathione concentrations in the range of 80–120% of the nominal content of in granulated glutathione. This covers the whole range of concentrations and determines the minimum permissible concentration of the methods for quantitative determination [19].

The acceptance criteria were calculated for b=10%, therefore, the maximum value of the total uncertainty of the results of the methods (Δ_{As}) should not exceed the value of B×0.32=3.2% [20].

Table 2. Mass of the batches and concentrations of glutathione reduced in model granulated samples

Number of model sample	Mass of the preparation batches – granules with glutathione, g	Content of glutathione reduced in the batch, g	Concentration of glutathione reduced relative to nominal, %
1	0/6022	0.02000	80.41
2	0/6360	0.02120	85.42
3	0.6760	0.02250	90.85
4	0.7125	0.02370	95.07
5	0.7512	0.02500	100.02
6	0.7875	0.02620	105.20
7	0.8265	0.02755	110.03
8	0.8630	0.02876	114.96
9	0.9012	0.03000	119.92

The results of chromatography, the values of certain concentrations of glutathione in model samples and the calculation of the metrological characteristics of the methods are presented in Table 3.

From the information given in Table 3 it follows, that the methods of quantitative determination of glutathione does not have a statistically significant systematic error.

Thus, validation tests of the methods of quantitative determination of glutathione in granules by the method of pre-column derivatization with ortho-phthalic aldehyde showed that the validation parameters correspond to the accepted quantitative criteria for correctness, convergence in the concentration range from 80% to 120% of the nominal glutathione content in the granulated dosage form.

Table 3. Validation results of the methods for quantitative determination of glutathione in model solutions of granules

Solution number	% of Glutathione taken, compared to the concentration of the reference solution (X,%)	% of Glutathione found, compared to the concentration of the reference solution (Y,%)	Glutathione found to Glutathione taken ratio Z = 100*(Y/X)
1	80.41	80.39	99.98
2	85.42	85.49	100.01
3	90.85	90.78	99.92
4	95.07	95.23	100.17
5	100.02	100.21	100.19
6	105.20	105.01	99.82
7	110.03	110.18	100.14
8	114.96	115.15	100.17
9	119.92	120.36	100.37
Average, Z, %			100.09
Relative standard deviation, S_z, %			0.17%
Relative confidence interval, Δ, % = t (95%, 7)*S_z			0.39
Critical value for convergence of results, maxΔ_{As}, %, at B±10%			3.2
Systematic error, δ = Z_{cp} –100			0.09
Criterion of insignificance of the system error δ≤Δ/3			In progress
General conclusion about the methods			Correct

CONCLUSION

In the course of the work, a model dosage form was created – glutathione-based granules. Physical and technological characteristics of the model sample of granules with GSH and lactose as a filler have been studied. Methods of quantitative determination of GSH in granules using pre-column derivatization with ortho-phthalaldehyde by HPLC has been worked out and validated. The results of the validation assessment showed that this methods complies with all the validation parameters: it is correct, precise, specific and linear in the analytical field.

REFERENCES

1. Winterbourn CC. Superoxide as an intracellular radical sink. Free Radic. Free Radic Biol Med. 1993 Jan;14(1):85–90.
2. Cazanave S, Berson A, Haouzi D, et al. High hepatic glutathione stores alleviate Fas-induced apoptosis in mice. J Hepatol. 2007 May;46(5):858–68.
3. Go YM, Jones DP. Redox compartmentalization in eukaryotic cells. Biochim Biophys Acta. 2008 Nov;1780(11):1273–90. DOI: 10.1016/j.bbagen.2008.01.011.
4. Zimmermann AK1, Loucks FA, Schroeder EK, Bouchard RJ, et al. Glutathione binding to the Bcl-2 homology-3 domain groove: a molecular basis for Bcl-2 antioxidant function at mitochondria. J Biol Chem. 2007 Oct 5;282(40):29296–304. DOI: 10.1074/jbc.M702853200.
5. Arnér ES, Holmgren A. Physiological functions of thioredoxin and thioredoxin reductase. Eur J Biochem. 2000 Oct;267(20):6102–9. DOI: https://doi.org/10.1046/j.1432-1327.2000.01701.x.
6. Iles KE, Liu RM. Mechanisms of glutamate cysteine ligase (GCL) induction by 4-hydroxynonenal. Free Radic Biol Med. 2005 Mar 1;38(5):547-56. DOI: 10.1016/j.freeradbiomed.2004.11.012.
7. Corbucci G.G. The role of reduced glutathione during the course of acute haemolysis in glucose-6-phosphate dehydrogenase deficient patients: clinical and pharmacodynamic aspects. Int J Clin Pharmacol Res. 1990;10(5):305–10.
8. Golikov SN, Sanockij IV, Tiunov LA. Obshchie mekhanizmy toksicheskogo dejstviya [General mechanisms of toxic action]. L:Medicina.1986. 280p. Russian.
9. Eropkin MYu, Eropkina EM. Kultury kletok kak modelnaya sistema issledovaniya toksichnosti i skrininga citoprotektornyh preparatov [Cultures of cells as a model system for the study of toxicity and screening of cytoprotective drugs]. SPb:MORSAR AB. 2003. 239p. Russian.
10. Tiunov LA. Mekhanizmy estestvennoj detoksikacii I antioksidantnoj zashchity [Mechanisms of natural detoxification and antioxidant protection]. Bulletin RAMS. 1995;3:9–13. Russian.
11. Tiunov LA, Ivanova VA. Rol glutationa v processah detoksikacii [The role of glutathione in detoxification processes]. Bulletin AMS USSR. 1988;1:62–9. Russian.
12. Harman D. Free-radical theory of aging: inversing the functional life span. Ann N Y Acad Sci. 2006 May;1067:10–21. DOI: 10.1196/annals.1354.003.
13. Lőrincz T, Szarka A. The determination of hepatic

ОРИГИНАЛЬНАЯ СТАТЬЯ
DOI: 10.19163/2307-9266-2019-7-1-13-19

glutathione at tissue and subcellular level. J Pharmacol Toxicol Methods. 2017 Nov;88(Pt 1):32–39. DOI: 10.1016/j.vascn.2017.05.004.

14. Kanekal S, Kehrer JP. Metabolism of cyclophosphamide by lipoxygenases. Drug Metab Dispos. 1994 Jan-Feb;22(1):74–8.
15. Traverso N, Ricciarelli R, Marengo B, et al. Role of glutathione in cancer progression and chemoresistance // Oxidative Medicine and Cellular Longevity. Oxidative Medicine and Cellular Longevity. 2013;2013. Article ID 972913. DOI: https://doi.org/10.1155/2013/972913.
16. Polyakov NA, Astrahanova MM, Dubinskaya VA, Bykov VA. Gidrataciya i termodinamicheskie harakteristiki obrazcov alfa laktozy poluchennye s ispolzovaniem novyh tekhnologij [Hydration and thermodynamic characteristics α-lactose, received on new technologies]. Problems of Biological, Medical and Pharmaceutical Chemistry. 2011;8:7–11. Russian.
17. Lenton KJ, Therriault H, Wagner JR. Analysis of glutathione and glutathione disulfide in whole cells and mitochondria by postcolumn derivatization high-performance liquid chromatography with ortho-phthalaldehyde. Anal Biochem. 1999 Oct 1;274(1):125–30. DOI: 10.1006/abio.1999.4258.
18. Obshchaya farmakopejnaya statya OFS 1-1-0012-15 validaciya analiticheskih metodik [Pharmacopoeial item OFS.1.1.0012.15 "Validation of analytical methods"]. State Pharmacopoeia of Russian Federation XIV-th edition. Moscow;2018. Russian.
19. Rukovodstvo po validacii metodik analiza lekarstvennyh sredstv. Metodicheskie rekomendacii [Guidance on the validation of drug analysis techniques]. Moscow,2007:6–92. Russian.
20. Obshchaya farmakopejnaya statya OFS 1-1-0013-15 "Statisticheskaya obrabotka rezultatov himicheskogo ehksperimenta" [Pharmacopoeial item OFS.1.1.0013.15 "Statistical processing of the results of a chemical experiment"]. State Pharmacopoeia of Russian Federation XIV-th edition. Moscow; 2018. Russian.

Conflict of interest

The authors declare no conflict of interest.

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