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DEVELOPMENT AND METHODOLOGY FOR THE ESTIMATION OF ATENOLOL AND VALSARTAN IN PHARMACEUTICALS

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SUMMARY. According to the appropriate protocols for the treatment of hypertension are often used antihypertensive drugs of the 5 main classes – first-line drugs, which when used in equivalent doses contribute to the reduction of blood pressure and significantly reduce the risk of cardiovascular complications. Quite often, doctors prescribe two/three medicines at a time. Therefore, the creation of fixed combinations antihypertensive action in the form of solid dosage forms is an urgent task of modern pharmacy.

The aim of the study – to improve to more rapid, simple, selective, less expensive methods TLC analysis of simultaneous determination of atenolol and valsartan in pharmaceuticals.

Methods. The present study assessed mobile phases of atenolol and valsartan for TLC.

Results and Discussion. Method of simultaneous identification of atenolol and valsartan by TLC has been developed. We have established that the most optimal Rf observed using mobile phases for simultaneous determination of atenolol and valsartan: n-butanol-acetic acid-water (40:10:20). We have explored the validation characteristics – specificity and suitability of the chromatographic system that met, the eligibility criteria established by the SPU.

Conclusion. We have developed chromatographic method for simultaneous determination of atenolol and valsartan. Prospects for future research will be aimed at developing analytical methods of analysis.

KEY WORDS: atenolol; valsartan; identification; thin layer chromatography; validation.

Introduction. According to the appropriate protocols for the treatment of hypertension are often used antihypertensive drugs of the 5 main classes – first-line drugs, which when used in equivalent doses contribute to the reduction of blood pressure and significantly reduce the risk of cardiovascular complications. Quite often, doctors prescribe two/three medicines at a time. Therefore, the creation of fixed combinations antihypertensive action in the form of solid dosage forms is an urgent task of modern pharmacy. Nowadays, valsartan is one of the most effective drugs for the treatment of hypertension.

Analytical method development is increasingly being introduced into fundamental pharmaceutical research, taking into account their high sensitivity, accuracy, specificity and expressiveness. Thin layer chromatography, or TLC, is a method for analyzing mixtures by separating the compounds in the mixture. TLC can be used to help determine the number of components in a mixture, the identity of compounds, and the purity of a compound. By observing the appearance of a product or the disappearance of a reactant, it can also be used to monitor the progress of a reaction. TLC is a sensitive technique – microgram (0.000001 g) quantities can be analyzed by TLC – and it takes little time for an analysis (about 5–10 minutes). TLC is an easy-to-use, fast and highly versatile separation technique for qualitative and quantitative analysis [1, 2].

The aim of the present study was to improve to more rapid, simple, selective, less expensive methods TLC analysis of simultaneous determination of atenolol and valsartan in pharmaceuticals.

Methods. Using this technique, we have analyzed medicines Atenololum-Astrapharm 50 mg (tablets containing 50 mg of atenolol produced by Astrapharm), Valsartan (tablets containing 80 mg of valsartan produced by KRKA d.d. Novo Mesto).

All solvents were obtained from Merck pharmaceticals.

Analytical equipment

Scales AVT-120-5D, measuring vessel glass and reagents that meet the SPU requirements. TLC test was carried out using Silica gel, chromatographic plates 60 F254 Merck (Germany) and Sorbfil (Russia).

Sample preparation for investigation solution.

Investigation solution from tablets Atenololum-Astrapharm, tablets Valsartan. To sample powder tablets or powder, equivalent to 20.00 mg atenolol, 20.00 mg valsartan add 5.0 ml of *methanol R* and dilute with *methanol R* to 10.0 ml, mix and filter.

Reference solution of atenolol. 20.00 mg Pharmacopoeial standard sample SPhU of atenolol dissolved in *methanol R* and dilute with the same solvent to 10.0 ml.

Reference solution of valsartan. 20.00 mg Pharmacopoeial standard sample SPhU of valsartan dissolved in *methanol R* and dilute with the same solvent to 10.0 ml.

Mobile phase: *n-butanol-acetic acid-water (40: 10: 20).*

Samples that are applied: 10 µl, applied the test solutions and investigation solutions.

Over a path of 10 cm from the starting line.

Detection: examination in ultraviolet light at 254 nm.

Results and Discussion. The present study assessed the different solvent extracts of atenolol and valsartan for TLC. The chromatograms obtained with the test solution were detected at the main spot basic substance in the chromatograms obtained with reference solutions, corresponding in size and color. We had investigated various mobile phases in order to identify the optimal choice of atenolol and valsartan investigation by TLC. The factors of mobility in the studied of simultaneous determination of atenolol and valsartan in mobile phases, is listed in Table 1.

Table 1. Chromatographic characteristics of atenolol and valsartan in different mobile phases

Mobile phase	Stationary phase (plate) *Rf* on Sorbfil (atenolol)	Stationary phase (plate) *Rf* on Sorbfil (valsartan)	The limit of detection of atenolol, micrograms	The limit of detection of valsartan, micrograms
Chloroform-methanol (9:1)	0.05	0.07	0.4	0.4
Chloroform-ethanol (8:2)	0.08	0.12	0.4	0.4
Chloroform-methanol-ammonia (25 %) (4:4:2)	0.86	0.84	0.4	0.4
n-butanol-methanol (3:2)	0.20	0.70	0.4	0.4
Ammonia (25 %)-propanol (30:70)	0.71	0.69	0.4	0.4
Propanol-water (70:30)	0.12	0.72	0.4	0.4
n-butanol-acetic acid-water (40:10:20)	0.34	0.78	0.4	0.4
Acetone – water (3:2)	0.18	0.87	0.4	0.4

We have established that the most optimal Rf observed using mobile phases for for simultaneous determination of atenolol and valsartan: *n-butanol-acetic acid-water (40:10:20).*

The analysis considered probable, though the test requirements "Check suitability chromatographic system".

Chromatographic system is considered appropriate when:

The chromatogram obtained with reference solution is a clearly visible spot;

Rf principle spot in the chromatogram obtained with reference solution to be about 0.6.

According to the SPhU and Note for guidance on validation of analytical procedures: text and methodology (CPMP/ICH/381/95) to test the Identification must be validated, to determine such characteristics as specificity and suitability of the chromatographic system [2–4]. The maximum difference of Rf values in the same plate (for two series of plates) must not exceed the value of 0.02. Originally, plates were tested according to the requirements of SPU on chromatographic resolution. When checking for the stability of the solution at the time we started chromatography of atenolol and valsartan freshly prepared test solution sustained, over time for 30 min. Visual assessment of spots on the size and intensity of staining confirms that they clearly appear as freshly cooked and seasoned in time solutions (for plates of different series). The solutions were stable over time and new areas, had been identified [5-8].

Thus, we have explored the validation characteristics – specificity and suitability of the chromatographic system that met, the eligibility criteria established by the SPhU. Therefore, the present study provided a suitable as well as accurate method for simultaneous determination of atenolol and valsartan, which is of potential practical significance in development of analytical methods.

Conclusions. We have developed TLC method for simultaneous determination of atenolol and valsartan. We have found that the most optimal Rf observed using mobile phases for simultaneous determination of atenolol and valsartan: *n-butanol-acetic acid-water (40: 10: 20).* The validation study of the characteristics of specificity and suitability of the chromatographic system, confirmed that they meet the eligibility requirements under the SPhU. Propects for future research will be aimed at developing analytical methods of analysis.

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РАЗРАБОТКА И МЕТОДОЛОГИЯ ОПРЕДЕЛЕНИЯ АТЕНОЛОЛА И ВАЛСАРТАНА В ЛЕКАРСТВЕННЫХ СРЕДСТВАХ

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РЕЗЮМЕ. Согласно соответствующим протоколам, для лечения артериальной гипертензии чаще всего применяют антигипертензивные препараты 5 основных классов – препараты первой линии, которые при применении в эквивалентных дозах способствуют снижению АД и существенно уменьшают риск сердечно-сосудистых осложнений. Довольно часто врачи назначают два/три лекарственных средства одновременно. Поэтому, создание фиксированных комбинаций антигипертензивного действия в виде твердых дозированных лекарственных форм является актуальной задачей современной фармации.

Цель – улучшить более быстрые, простые, селективные и менее дорогостоящие методы ТСХ-анализа для одновременного определения атенолола и валсартана в лекарственных средствах.

Методы. В исследовании оцениваются подвижные фазы для одновременного определения атенолола и валсартана для тонкослойной хроматографии.

Результаты. Разработан метод одновременной идентификации атенолола и валсартана с помощью ТСХ. Установлено, что наиболее оптимальная *Rf* наблюдается с использованием подвижных фаз: *н-бутанол – кислота уксусная – вода (40:10:20)*. Мы изучили характеристики валидации – специфичность и пригодность хроматографической системы, которая соответствовала критериям отбора, установленным ГФУ.

Вывод. Разработана эффективная хроматографическая методика для одновременного определения атенолола и валсартана. Перспективы будущих исследований будут направлены на разработку аналитических методов анализа.

КЛЮЧЕВЫЕ СЛОВА: атенолол; валсартан; идентификация; тонкослойная хроматография; валидация.

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РОЗРОБКА І МЕТОДОЛОГІЯ ВИЗНАЧЕННЯ АТЕНОЛОЛУ ТА ВАЛСАРТАНУ В ЛІКАРСЬКИХ ЗАСОБАХ

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РЕЗЮМЕ. Згідно з відповідними протоколами, для лікування артеріальної гіпертензії найчастіше застосовують антигіпертензивні препарати 5 основних класів – препарати першої лінії, які при застосуванні в еквівалентних дозах сприяють зниженню АТ та суттєво зменшують ризик серцево-судинних ускладнень. Доволі часто лікарі призначають два/три лікарських засоби одночасно. Тому створення фіксованих комбінацій антигіпертензивної дії в вигляді твердих дозованих лікарських форм є актуальним завданням сучасної фармації.

Мета – удосконалення більш швидких, простих, вибіркових, менш дорогих методів аналізу тонкошарової хроматографії (ТШХ) для одночасного визначення атенололу та валсартану в лікарських засобах.

Матеріал і методи. В даному дослідженні оцінюються рухливі фази для одночасного визначення атенололу та валсартану для тонкошарової хроматографії.

Результати. Розроблена методика одночасної ідентифікації атенололу та валсартану за допомогою ТШХ. Встановлено, що найбільш оптимальна *Rf* спостерігається при використанні одночасної ідентифікації атенололу та валсартану: *н-бутанол – кислота оцтова – вода (40:10 : 20)*. Були вивчені валідаційні характеристики – специфічність та придатність хроматографічної системи, що відповідали критеріям прийнятності, встановленими ДФУ.

Висновок. Розроблена ефективна хроматографічна методика одночасного визначення атенололу та валсартану. План майбутніх досліджень буде спрямований на розробку аналітичних методів аналізу.

КЛЮЧОВІ СЛОВА: атенолол; валсартан; ідентифікація; тонкошарова хроматографія; валідація.

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