Development of Methodology for Identification of Captopril in Medicines

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Abstract

Aim: The aim of the study was to development of a thin layer chromatography (TLC) method for the estimation of captopril. The pharmaceutical industry is under increased scrutiny to constrain costs and yet consistently deliver to market safe, efficacious products that fulfill medical needs. As a part of this, drug analysis also plays an important role in its contribution to new drug development. **Materials and Methods:** Standard analytical procedure for newer drugs or formation may not be available in pharmacopoeia and it is essential to develop new analytical methods which are accurate, precise, specific, linear, simple, and rapid. The analysis of captopril is described in pharmacopeia, but the aim of our researches was to improve to more rapid, simple, selective, more accurate, precise, reliable, less expensive methods by TLC of captopril in medicines and for using this methods for analysis of their metabolites in next step of researches. **Results and Discussion:** The method of identification of captopril in medicines by TLC has been developed. Established that the most optimal Rf observed using mobile phases: Chloroform R-propanol R (9:1). The detection limits of captopril in this system are 0.4 mcg. However, those mobile phases are the most express. We explored the validation characteristics - Specificity and suitability of the chromatographic system that met, the eligibility criteria established by the segmented polyurethane. **Conclusion:** We have been developed chromatographic methods of identification of captopril in medicines. The proposed methods are rapid, economical, simple, accurate, selective, precise, and applicable to the analysis of pharmaceutical dosage forms.

Key words: Captopril, identification, thin layer chromatography, validation

INTRODUCTION

Captopril (1-[(2S)-3-mercapto-2-methylpropionyl]-1-proline), is an angiotensin-converting enzyme (ACE), has been widely used therapeutically as an antihypertensive agent,[1,2] which is also prescribed for the treatment of treatment of hypertension, congestive heart failure, and coronary artery disease.[3] The drug is considered as a drug of choice in antihypertensive therapy due to its effectiveness and low toxicity. It is mainly prescribed for patients who are chronically ill and require long-term therapeutic agents. The dose required is 37.5-75 mg to be taken three times a day in divided doses. The drug acts orally and after single oral dose ingestion, the antihypertensive action is only effective for 6-8 h.[4] Captopril competitively inhibits ACE, thereby decreasing levels of angiotensin II, increasing plasma renin activity, and decreasing aldosterone secretion.[5] This agent may also inhibit tumor angiogenesis by inhibiting endothelial cell matrix metalloproteinases and endothelial cell migration. Captopril may also exhibit antineoplastic activity independent of effects on tumor angiogenesis. Although captopril is efficacious in most forms of hypertension, its use largely has been limited because

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of safety considerations to patients with severe, treatment-resistant hypertension. Recent studies recommended that lower doses, usually in combination with a diuretic, suggest, however, that while antihypertensive efficacy is maintained, the safety profile of captopril could be reappraised.[6] The analysis of captopril in substance is described in British Pharmacopoeia (for identification-infrared absorption spectrophotometry and for assay-iodometry). For pharmaceutical dosage forms that contain captopril methods, which are described in pharmacopoeia, cannot be used, special in present another active pharmaceutical ingredient. [7-9] Therefore, the development of methods for identification and quantification of captopril in medicines is an important task of pharmaceutical analysis and standardization of medicines.

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 popular technique for the analysis of a wide variety of organic and inorganic substances, because of its distinct advantages such as minimal sample clean-up, wide choice of mobile phases, flexibility in sample distinction, high sample loading capacity and low cost. The use of chromatographic techniques for monitoring the starting materials, intermediates and the process reactions is an excellent means for controlling the purity of the final drug and thereby protecting the patient who ultimately receives it.[10] TLC is a powerful tool for screening unknown materials in bulk drugs.[11-15] It provides a relatively high degree of assurance that all possible components of the drug are separated.[16] The high specificity of TLC has been exploited to quantitative analytical purpose using spot elution followed by spectrophotometric measurement. TLC plays a key role in the early stage of drug development when information about the impurities and degradation products in drug substance and drug product is inadequate. Various impurities of pharmaceuticals have been identified and determined using TLC.[17]

The aim of this study was to improve to more rapid, simple, selective, more accurate, precise, reliable, less expensive methods TLC analysis of captopril in medicines and for using these methods for analysis of their metabolites in next step of researches.

MATERIALS AND METHODS

Using this technique, we have analyzed medicines "Captopres 12.5 - Darnitsa" (tablets containing 50 mg of captopril and 12.5 mg hydrochlorothiazide) and captopril (25 mg tablets produced by "Astrafarm").

Analytical equipment

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

Sample preparation for captopril

Investigation solutions from tablets "Captopril" and "Captopres 12.5 – Darnitsa." To sample powder tablets or powder, equivalent to 10 mg captopril, add 5.0 ml of methanol R and dilute with methanol R to 10.0 ml, mix and filter.

Reference solution: 10 mg pharmacopoeial standard sample SPU of captopril dissolved in methanol R and dilute with the same solvent to 10.0 ml.

Mobile phase: Chloroform R-methanol R (9:1).

Samples that are applied: 5 μl, applied the test solution and investigation solutions. Over a path of 10 cm from the starting line.

Detection: Examination in ultraviolet light at 254 nm.

RESULTS

This study was assessed the different solvent extracts of captopril for TLC. The chromatogram obtained with the test solution is detected at the main spot spots basic substance in the chromatogram obtained with a reference solution, corresponding in size and color. We had investigated various mobile phases (solvent system) to identify the optimal choice of captopril investigation by TLC in medicines. The factors of mobility in the studied of captopril in mobile phases are listed in Table 1.

DISCUSSION

We found that for identification by TLC using a sensitive of all investigated solvents. Established that the most optimal Rf observed using mobile phases for captopril:chloroform R-methanol R (9:1). The detection limit of captopril in these systems is 0.4 mcg. However, the most express mobile phases are chloroform R-methanol R (9:1). Similar studies were used to detect captopril in its formulation.[18,19] The analysis considered probable, though the test requirements "Check suitability chromatographic system." Checking the suitability of the chromatographic system. Chromatographic system is considered appropriate when:

Table 1: Chromatographic characteristics of captopril in different solvent systems

Mobile phase	Stationary phase (plate) Rf on (Sorbfil)	The limit of detection, micrograms	Detection in ultraviolet light at 254 nm	Detection in ultraviolet light at 365 nm
Chloroform-methanol (9:1)	0.60	0.4	Violet	Blue
Chloroform-methanol ammonia (25%) (4:4:2)	0.95	0.4	Violet	Blue
n-butanol-methanol (3:2)	0.81	0.4	Violet	Blue
Ammonia (25%)-propanol (30:70)	0.80	0.4	Violet	Blue
Formic acid-isopropanol-water (40:2:10)	-	-	-	-
Propanol-water (70:30)	0.75	0.4	Violet	Blue
n-butanol-acetic acid-water (40:10:10)	0.77	0.4	Violet	Blue
n-butanol-acetic acid-water (40:10:20)	0.75	0.4	Violet	Blue

- The chromatogram obtained with reference solution is a clearly visible spot.

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

We previously studied the behavior of placebo tablets in terms of methods of identification of captopril. It was established that the excipients are part of pills and do not affect the sensitivity and specificity of verapamil detection. The number of drugs introduced into the market is increasing every year. These drugs may be either new entities or partial structural modification of the existing one. The objective of any analytical measurement is to obtain consistent, reliable and accurate data. Validated analytical methods play an important role in achieving this goal. The results from method validation can be used to judge the quality, reliability and consistency of analytical results, which is an integral part of any good analytical practice. The validation of analytical methods is also required by most regulations and quality standards that impact laboratories.[20-23] Very often there is a time lag from the date of introduction of a drug into the market to the date of its inclusion in pharmacopoeias. This happens because of the possible uncertainties in the continuous and wider usage of these drugs, reports of new toxicities, development of patient resistance, and introduction of better drugs by competitors. Under these conditions, standards and analytical procedures for these drugs may not be available in the pharmacopoeias. There is a scope, therefore, to develop newer analytical methods for such drugs. Analytical methods development and validation play important roles in the discovery, development, and manufacture of pharmaceuticals. According to the SPU 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.[24] 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 the chromatographic resolution. When checking for the stability of the solution at the time, we started chromatography of captopril 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. Thus, we explored the validation characteristics - specificity and suitability of the chromatographic system that met, the eligibility criteria established by the SPU. The objective of any analytical measurement is to obtain consistent reliable and accurate data. The analytical method validation is a major issue in the pharmaceutical industry for controlling drug quality, development, and registration. Simply, it is used to justify the analytical method, methods submitted as a part of a new drug application, bioequivalence and bioavailability studies, and for the analysis of drugs as raw material or in their dosage forms. Therefore, this study provided a suitable as well as an accurate method for determination of captopril, which is of potential practical significance.

CONCLUSIONS

We developed TLC methods of identification of captopril in medicines. We found that captopril identification by TLC using a sensitive of all investigated solvents. Established that the most optimal Rf observed using mobile phases for captopril: Chloroform R-methanol R (9:1). The detection limit of captopril in these systems is 0.4 mcg. The validation study of the characteristics of both specificity and suitability of the chromatographic system confirmed that they meet the eligibility requirements under the SPU. Prospects for future research will be aimed at developing methods of the analysis captopril's metabolites by proposed TLC.

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