

Review on Analytical Methods for Determination of Furosemide

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Abstract

Furosemide (FRS) belongs to loop diuretics. The drug is prescribed for the treatment of hypertension and heart failure. FRS inhibits the reabsorption of electrolytes mainly in the thick ascending limb of the loop of Henle and also in the distal renal tubules. It may also have a direct effect in the proximal tubules. Using a thorough computer assisted literature survey; this review article touches upon the reported analytical methods for the quantification of FRS in raw materials, pharmaceutical dosage forms and biological fluids. Bioanalytical methods are widely used for quantitative estimation of drugs and their metabolites in biological matrices. Various analytical methods such as spectrophotometry, spectrofluorimetry, HPLC, HPTLC, and UPLC have been used in laboratories for the quantitative analysis of this drug in biological samples and pharmaceutical preparations. Therefore, the aim of this review is to give a summary of the current analytical methods used for the assay of the drug.

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1. Introduction

FRS is a potent loop diuretic with rapid action. It is used in the treatment of oedema associated with heart failure, including pulmonary oedema, with renal and hepatic disorders and may be effective in patients unresponsive to thiazide diuretics. FRS is also used in the treatment of hypertension, either alone or with other antihypertensives. FRS inhibits the reabsorption of electrolytes mainly in the thick ascending limb of the loop of Henle and also in the distal renal tubules. It may also have a direct effect in the proximal tubules. Excretion of sodium, potassium, calcium, and chloride ions is increased, and water excretion is enhanced. FRS's effects are evident within 30 minutes to 1 hour after an oral dose, peak at 1 to 2 hours, and lasts for about 6 hours; after intravenous injection its effects are evident in about 5 minutes and last for about 2 hours. It

is given orally, usually in the morning. Alternatively, it may be given intramuscularly or intravenously as sodium salt; doses are expressed in terms of FRS base (Brayfield, 2013). FRS chemically is [4-Chloro-N-furfuryl-5-sulphamoylanthranilic acid] [Brayfield, 2013] (**Fig. 1**).

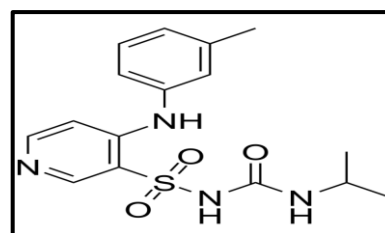


Fig. 1: Chemical structure of FRS.

2. Literature Review:

FRS has an official monograph in each of the United States Pharmacopoeia (USP) and the British Pharmacopoeia (BP). Different analytical methods for the estimation of FRS either in pharmaceutical formulations or in biological fluids have been discussed in review articles [Bosch, 2008 & Bosch, 2013]. Many other methods were then developed for its determination.

A brief description of these methods can be summarized as follows:

2.1. Spectroscopic Methods:

2.1.1. Ultraviolet and Visible Spectrophotometric Methods:

A summary of the spectrophotometric methods is presented in **Table 1**.

Table (1): Reported spectrophotometric methods for determination of FRS.

Matrix	Technique	λ_{\max}	Ref.
Pharmaceutical preparations	Absorption maxima method.	277 nm.	(Gahandule, 2016)
	Partial least squares regression.	200-350 nm	(Dikran, 2016)
	Azo coupling with resorcinol	430 nm	(Abdullah, 2021)
	Oxidative coupling reaction 1-Naphthylamine-4-sulfonic acid in the presence of potassium permanganate	465 nm	(Ahmed, 2022)
	Charge transfer reaction with pyrogallol reagent using sodium carbonate	610 nm	(Mahmoud, 2023)
	Complex formation with 2,4-Di hydroxyl Benz aldehyde	430 nm	(Okdeh, 2023)

2.1.2. Spectrofluorimetric methods:

A spectrofluorimetric method depends on measuring the quenching of fluorescence of nitrogen doped carbon quantum dots by FRS. The method was linear over the range of 0.1 to 1.0 $\mu\text{g/mL}$ with LOQ of 0.087 $\mu\text{g/mL}$. The method was further extended for determination of the drug in its pharmaceutical tablets and spiked human plasma (Abo Zaid, 2023).

Another spectrofluorimetric method was reported for the determination of FRS using acriflavine as a new reagent. The method is based on the quantitative quenching effect of FRS on the native fluorescence of acriflavine in Britton-Robinson buffer due to the formation of an ion associated complex. The decrease of acriflavine fluorescence was observed at 505 nm after excitation at 265 nm. The fluorescence – concentration plot was rectilinear over the range of 2.0–10.0 $\mu\text{g/ml}$ (Qader, 2017).

2.2. Electrochemical Methods:

A glassy carbon electrode (GCE) chemically modified with reduced graphene oxide (CRGO) was used for the determination of FRS in natural waters using batch injection analysis (BIA) as an analytical method, where CRGO-GCE is coupled to a BIA cell for amperometric measurements. Acetate buffer of pH 5.2 was used as the background electrolyte. The method provided a limit of detection of (0.7 $\mu\text{mol L}^{-1}$), and a linear range of

(1–600 $\mu\text{mol L}^{-1}$) (Vasconcelos, 2020).

An electrochemical method for the determination of FRS using tranexamic acid (Tr) derived gold nanoparticles modified glassy carbon electrode (GCE) was reported. The amperometric determination of FRS was also carried out at the Tr-AuNps modified GCE under stirred conditions using Britton Robinson buffer (BRb) of pH 5. The linear calibration plot was obtained in the range of 50 μM to 500 μM (Chandio, 2021).

Another method depending on a glassy carbon working electrode modified with poly(3,4-ethylenedioxythiophene): polystyrene sulfonate to detect FRS was reported. The electrochemical oxidation of FRS was investigated in a supporting electrolyte, 0.01M of phosphate buffer of pH 4 using differential pulse voltametry (DPV). The developed sensor displays a wide determination range of 6.0×10^{-6} to 1.0×10^{-4} M with a detection limit of 2.0×10^{-6} M (Hossain, 2023).

2.3. Capillary Electrophoresis:

A capillary electrophoretic method with photodiode array detector was reported for the determination of FRS in tablets. The separation was achieved using a fused silica capillary with (total length of 30.2 cm \times 50.0 μm id). The optimum separations conditions were: 2 mmol/L sodium tetraborate buffer solution,

pH 9.3 and 10% methanol; hydrodynamic injection 3.45 kPa/5 s; voltage and temperature set at +25 kV and 25°C, respectively, and UV detection at 273 nm. The method showed good linearity (70.0 to 130.0 µg/mL) (de Souza, 2019).

2.4. Chromatographic Methods:

Many chromatographic techniques have been reported

for the determination of FRS as follows:

2.4.1. High Performance Liquid Chromatography (HPLC):

The recent HPLC methods for determination of FRS were described in **Table 2**.

Table (2): Summary of the reported HPLC methods for the determination of FRS.

Application(s)	Mobile phase	Column	Detection	Ref.
Pure form and plasma samples.	Ethanol–deionized water (45: 55, v/v) at pH 3.5 with glacial acetic acid	C ₁₈	UV at 254 nm	(Naguib, 2018)
Pharmaceutical preparations.	Water-acetonitrile (35:65, V/V)	C ₁₈	UV at 281 nm	(Shaikh, 2018)
Spiked human urine samples	A binary gradient of 20% acetonitrile as solvent A and 80% acetonitrile as solvent B, containing 0.25% acetic acid for both solvents	C ₁₈	UV at 230 nm	(Al-Hashimi, 2018)
Pharmaceutical preparations	A mixture of 1% glacial acetic acid and acetonitrile in the ratio of 50%:50% v/v as the mobile phase	C ₁₈	UV at 272 nm	(Karunakaran, 2021)
Real patient urine and Plasma samples	A binary mobile phase consists of A (20% acetonitrile) and B (80% acetonitrile), both in 0.25% of acetic acid.	C ₁₈	UV at 230 nm	(AL-Hashimi, 2022)

2.4.2. High Performance Thin Layer Chromatography (HPTLC):

A HPTLC method for determination of FRS was reported. The separation was completed on silica gel HPTLC F₂₅₄ plates using ethyl acetate–triethylamine–acetic acid (9:0.7:0.5, by volume) as a developing system and UV detection at 254 nm. The method was used for its determination in plasma samples in the ranges of 0.2–2 µg/band ([Naguib, 2018](#)).

Another HPTLC method was reported for the estimation of FRS. The separation was carried out on TLC precoated silica gel 60 F₂₅₄ aluminum plate using chloroform: methanol: glacial acetic acid (7.5: 2:0.5 v/v) as a developing system and UV detection at 234 nm. The method was used for its determination in pharmaceutical preparations in the range of 200 – 1200 ng/band (Vanjari, 2023).

2.4.3. Ultra High-Performance Liquid Chromatography (UHPLC):

An UHPLC-MS/MS method for the quantitation of FRS in 10 µL of human urine has been applied to its

analysis of urine samples. The assay range was linear and reproducible over the calibration range of 0.100 – 50.0 µg/mL. All chromatographic separation was achieved on C₁₈ column with 0.1% formic acid in water as mobile phase A, and 0.1% formic acid in acetonitrile as mobile phase B (Vedar, 2022).

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