Constancy indication and development of UPLC stimulated benzoic, salicylic and triamcinolone acids in pharmaceutical dosage form

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Abstract

A Simple, precise, speedy and exact Ultra presentation molten chromatographic technique has been established and authenticated by will power of benzoic acid, salicylic acid, triamcinolone acetonide and ichthammol. The chromatographic separation was done on isocratic elution using pentafluorophenyl column (150x4.6mm, 3µ). Flow rate is 1mL/min and UV detection were carried out at 278nm. The mobile phase consisted by octane sulphonic acid with addition of 1ml triethyl amine and adjusts pH7.0 with ortho phosphoric acid: acetonitrile in 70:30 % V/V. The retention time for benzoic acid, salicylic acid, triamcinolone acetonide and ichthammol were found to be 2.54, 5.24, 8.62, and 7.77 minimum respectively. The detector showed rectilinear response finished the attentiveness assortment of 60-900µg/ml of benzoic acid, 30-450µg/ml of salicylic acid, 20-300µg/ml of ichthammol and 0.1-1.5µg/ml of triamcinolone acetonide for the drugs by showing a good correlation coefficient of 0.999. This proposed method is highly sensitive, precise and accurate which reduces cost of analysis, hence recommended for routine quality analysis in laboratories.

Keywords: Benzoic acid, Salicylic acid, Triamcinolone acetonide, Ichthammol, UPLC

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1 Introduction

Benzoic acid is a colorless crystalline robust and a basic fragrant carboxylic corrosive. The name infers starting with gum benzoin, which might have been to quite a while its best referred to as sourball. Corrosiveness of benzoic acid takes place alongside huge numbers plants and serves as a middle of the road in the biosynthesis (1) about numerous optional metabolites. Salts about benzoic corrosive are utilized. Similarly as nourishment preservatives (2) Furthermore benzoic corrosive is a paramount forerunner to those modern amalgamation of huge numbers other natural substances. Those salts Furthermore esters by benzoic corrosive are known as benzoates. Benzoic corrosive is a constituent of Whitfield’s ointment (3-4) which will be utilized for the medication for contagious skin diseases (5) for example tinea, ringworm, Also athlete’s foot (6). Similarly, as vital component of gum benzoin. Benzoic corrosive is also a major component of tincture (7). It can also be used as topical anesthesia antiseptics and as inhalant decongestants (8). Benzoic corrosive might have
been utilized, likewise an expectorant, analgesic (9), antiseptic (10). Salicylic corrosive may be a lipophilic mono hydroxyl benzoic acid which developed from phenolic acid (11) and a beta hydroxy corrosive. Their chemical formula is C\textsubscript{7}H\textsubscript{6}O\textsubscript{3}. This crystalline natural corrosive can be broadly utilized within natural union and works. It can also be utilized as plant hormone. It is determined from those digestion systems for salicin. It has been used as metabolite such as aspirin, which demonstrations to some degree it prodrug characteristics with salicylic acid, it best known for use as topical anesthesia anti-acne (12). The esters from similar to salicylic corrosive are known as salicylates. It is on World Health Organization (13) list of fundamental medicines. It has further uses in treating warts (14), psoriasis (15), acne, ringworm (16), dandruff (17) and ichthyosis (18).

**MATERIALS AND METHODS**

**Materials and Reagents**
Waters alliance-2695 chromatographical structure containing of quaternary propel, PDA detector-2996 and chromate graphical package Empower-2.0. Acetonitrile, Ortho Phosphoric Acid (OPA), Octane Sulphonic acid and Tri ethyl amine and water (HPLC score) were acquired from Merck (India) Ltd. Worli, Mumbai, India. All API’s of Benzoic acid, Salicylic acid, Triamcinolone acetonide and Ichthammol as reference standards were procured from Spectrum Pharma research solutions PVT ltd, Hyderabad. Octane Sulphonic Acid + Acetonitrile (70+30) are used as mobile phase. Water + Acetonitrile (50+50) are used as diluents.

**Preparation of Buffer**
Octane sulphonic acid with addition of 1ml triethyl amine and adjust pH=7.0 with OPA.

**Groundwork of Average Standard Explanation**
Precisely considered 60mg of Benzoic acid, 30mg of Salicylic acid, 20mg of Ichthammol and 0.1mg of Triamcinolone acetonide and transferred into a 10ml volumetric flask. The compounds were then dissolved in buffer, sonicate in 15min to dissolve the compounds and the volumes are completed up to the spot by solvent. Since the overhead explanation pipe out 1ml hooked on one separate 10ml volumetric flask and made up to the mark by solvent.

**Analysis of formulation**
Weigh 200mg of ointment and then occupied hooked on a 20ml volumetric flask. Add 15ml diluent, sonicate to dissolve and centrifuge for 30min. and dilute to capacity by dilutant. Filter concluded 0.45µ Nylon syringe filter (19).

**RESULTS AND DISCUSSION**

**Method Validation**
In this method structure appropriateness, segment, exactness, correctness, LOD, LOQ, strength, forced degradation and stability studies are validated for the selected drugs Benzoic acid, Salicylic acid, Ichthammol and Triamcinolone acetonide drugs. The proposed method having standard solution and sample solution chromatograms are shown in Fig.1.
Scheme appropriateness
The HPLC classification is soothed for 60min for getting steady base line. Six duplicate vaccinations of the typical solution containing 600µg/ml of Benzoic acid, 300µg/ml of Salicylic acid, 200µg/ml of Ichthammol and 1µg/ml of Triamcinolone acetonide were assessed to check the system suitability (20). The number of theoretical plate count for Benzoic acid, Salicylic acid, Ichthammol and Triamcinolone acetonide were 3038, 14504, 25887 and 22050 respectively. Tailing factor is 1.15, 1.14, 1.11 and 0.96 respectively.

Linearity
The segment of the HPLC technique is established for benzoic acid, salicylic acid, ichthammol and triamcinolone acetonide ranging from 60-900µg/ml of benzoic acid, 30-450µg/ml of Salicylic acid, 20-300µg/ml of ichthammol and 0.1-1.5µg/ml of triamcinolone acetonide (21) that is corresponding to 10% to 150% of the employee dataset. Six typical explanations at the meditations that stated series stayed organized and investigated as per the technique. The segment consequences gained are revealed in Table 1.
LOD values for Benzoic acid, Salicylic acid, and Ichthammol and Triamcinolone acetonide were 0.6µg/ml, 0.3µg/ml, and 0.2µg/ml and 0.005µg/ml. LOQ values for Benzoic acid, Salicylic acid, Ichthammol and Triamcinolone acetonide were 6µg/ml, 3µg/ml, 2µg/ml and 0.01µg/ml (22).

Accuracy
Technique precision might have been investigated by evaluating six independently prepared specimens of the same clump. Starting with these six differentiated tests result might have been injected and the crest zones got used to ascertain imply What's more rate RSD qualities. Those present method was found to be precise as % RSD of the less than 2.0%. The results are given in Table 2.

<table>
<thead>
<tr>
<th>Analyte</th>
<th>Amount present</th>
<th>%RSD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Benzoic acid</td>
<td>600</td>
<td>0.81</td>
</tr>
<tr>
<td>Salicylic acid</td>
<td>300</td>
<td>0.10</td>
</tr>
<tr>
<td>Ichthammol</td>
<td>200</td>
<td>0.28</td>
</tr>
<tr>
<td>Triamcinolone acetoneide</td>
<td>1</td>
<td>0.56</td>
</tr>
</tbody>
</table>

Table 1: Method Precision results

Accuracy
Accuracy was determined by retrieval revisions which are approved out in 3 dissimilar absorption stages (50%, 100% and 150%). APIs with absorption 300, 600 and 900µg/ml of benzoic acid, 150, 300 and 450µg/ml of Salicylic acid, 100, 200 and 300µg/ml of ichthammol and 0.5, 1 and 1.5µg/ml of triamcinolone acetonide were prepared. As per test method the test solution was injected to three preparations each spike level and the assay was performed. The percentage recovery values were found to be in the assortment of 98- 101%. The consequences are given in table 3.

Table 2: Accuracy (recovery) study results

<table>
<thead>
<tr>
<th>% of target concentration</th>
<th>Benzoic acid (% recovery)</th>
<th>Salicylic acid (% recovery)</th>
<th>Ichthammol (% recovery)</th>
<th>Triamcinolone acetonide (% recovery)</th>
</tr>
</thead>
<tbody>
<tr>
<td>50</td>
<td>100.4</td>
<td>100.25</td>
<td>99.9</td>
<td>100.45</td>
</tr>
<tr>
<td>100</td>
<td>99.8</td>
<td>100.30</td>
<td>100.2</td>
<td>100.3</td>
</tr>
<tr>
<td>150</td>
<td>100.1</td>
<td>100.55</td>
<td>100.45</td>
<td>100.7</td>
</tr>
<tr>
<td>Mean (% recovery)</td>
<td>100.1</td>
<td>100.37</td>
<td>100.18</td>
<td>100.48</td>
</tr>
</tbody>
</table>

Toughness

Toughness of the system might was also evaluated which demonstrated that chromatographic designs didn’t essentially change at different HPLC system, analyst, section. The quality by rate of RSD might have been beneath 2% exhibits that toughness of the developed method. 23.

Forced Degradation

Forced degradation conditions such as acidic, basic, peroxide, reduction and thermal stress were attempted as per ICH Q1A (R2). The effect of assay on their results is shown below table 5.

Table 3: Forced degradation results

<table>
<thead>
<tr>
<th>Degradation</th>
<th>Benzoic acid (% Degradation)</th>
<th>Salicylic acid (% Degradation)</th>
<th>Ichthammol (% Degradation)</th>
<th>Triamcinolone acetonide (% Degradation)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acid</td>
<td>16.3</td>
<td>14.9</td>
<td>14.6</td>
<td>11.6</td>
</tr>
<tr>
<td>Alkali</td>
<td>15.1</td>
<td>14.5</td>
<td>16.5</td>
<td>16.2</td>
</tr>
<tr>
<td>Peroxide</td>
<td>15.8</td>
<td>16.6</td>
<td>14.9</td>
<td>11.1</td>
</tr>
<tr>
<td>Reduction</td>
<td>13.7</td>
<td>15.3</td>
<td>13.8</td>
<td>12.7</td>
</tr>
<tr>
<td>Thermal</td>
<td>14.4</td>
<td>15.1</td>
<td>15.5</td>
<td>15.8</td>
</tr>
</tbody>
</table>

Each of these variables lead to the end that the proposed technique is straightforward, explicit, precise, exact and reproducible. Measurable investigation demonstrates that the strategy is reasonable for the examination of benzoic acid, salicylic acid, ichthammol and triamcinolone acetonide.

REFERENCES


