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New Spectrophotometric Methods for the Determination of Salmeterol Xinafoate

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Two simple and sensitive spectrophotometric methods (A and B) in the visible region have been developed for the determination of salmeterol xinafoate (SX) in bulk and in dosage forms. Method A is based on the reaction of SX with hydrochloric acid and sodium nitrite under alkaline conditions to form a stable yellow colored chromogen with absorption maximum of 430 nm and method B is based on the reaction of SX with ferric alum reagent to form a stable blue colored chromogen with the absorption maximum of 620 nm. The color obeyed Beer's law in the concentration range of 0-32 µg/ml for method A and 0-200 µg/ml for method B respectively. When pharmaceutical preparations (Laboratory prepared tablets) were analysed, the results obtained by the proposed methods are in good agreement with the labelled amounts and are comparable with the results obtained using a UV spectrophotometric method. Recovery in both the methods is 98-101%.

SALMETEROL xinafoate[1,2], chemically, (RS)-5- {1-Hydroxy-2-[6(Phenyl butoxy) hexylamino] ethyl) Salicyl alcohol 1-hydroxy-2-naphthoate, is a relatively new bronchodilator[3]. Literature cites only HPLC methods[4-7] for the determination of SX in human plasma and in pharmaceutical dosage forms. In the present work, the reaction of SX hydrochloric acid and sodium nitrite in the presence of sodium hydroxide to form a stable yellow colored chromogen with λmax of 430 nm (method A) and the reaction of SX with ferric alum reagent to form a stable blue colored chromogen with λmax of 620 nm (method B) were used for the determination of SX. These reactions have not been reported earlier for the quantitative determination of SX.

EXPERIMENTAL

A stock solution of SX (1.0 mg/ml) was prepared by dissolving 100 mg of the drug in 100 ml of methanol. Working standard solutions were obtained by appropriate dilution of the stock solution. Solutions of sodium nitrite (5.0% w/v), sodium hydroxide (2.0 N), hydrochloric acid (1.0 N) in distilled water and ferric alum reagent (1.0% w/v) in 1.0% v/v nitric acid were prepared. An ELICO UV-Visible spectrophotometer (Model: SL-150) with 1 cm matched quartz cells were used for all absorbance measurements.

Method A: The stock solution of SX was diluted with distilled water to obtain a series of dilutions containing 80,160,200,240 and 320 µg in 1 ml of the solution. To 1 ml of each dilution in a graduated text tube, 4 ml of hydrochloric acid solution was added. To each tube, 1.5 ml of sodium nitrite solution was added at room temperature, mixed and allowed to stand for 15 minutes. Then 3.5 ml of sodium hydroxide solution was added and the yellow colored chromogen formed was measured at 430 nm against a reagent blank. The results are given in Table-1.

Method B: Into a series of graduated test tubes 0.5,1.0,1.5,1.75 and 2.0 ml of stock solution of SX was pipetted and 1 ml of ferric alum reagent was added. After 5 minutes, the volume was made upto 10 ml with distilled water and the blue color developed was measured at 620 nm against a reagent blank. The results are given in Table-1.

Table -1 : Calibration Curve for the Estimation of Salmeterol Xinafoate by the Proposed methods

Method A		Method B	
Conc. (µg)	Absorbance at 430 nm	Conc. (µg).	Absorbance at 620 nm
80	0.151(1.8)	500	0.146(1.9)
160	0.302(1.6)	1000	0.292(1.7)
200	0.374(1.7)	1500	0.438(1.8)
240	0.452(1.8)	1750	0.512(1.7)
320	0.603(1.5)	2000	0.586(1.5)

Values in Parentheses are coefficient of variation (%).

Table-2 : Estimation of Salmeterol Xinafoate by the Proposed Methods*

Pharmaceutical Preparation/ excipient	Amount (mg) per Tablet	Amount found by the			Percent Recovery
		Method A (mg)	Method B (mg)	UV Method (mg)	
Tablet-I	100.00	99.97	99.89	99.99	98.0[a]
Tablet-II	100.00	99.98	99.96	99.98	99.9[a]
Talc	—	—	—	—	101.0[b]
Starch	—	—	—	—	99.7[b]
Lactose	—	—	—	—	98.5[b]

* Average of four determination
a = amount added 25 mg
b = amount added 50 mg

Estimation in Pharmaceutical dosage forms : As the tablets containing SX are not available in the market, tablets each containing 100 mg of SX were prepared by conventional wet granulation method using aqueous solution of PVP as a binding agent at 2% concentration in the formula and 20% of potato starch (Dry) as disintegrant and 2% of each of talc and magnesium stearate as lubricants. Tablet granulations were compressed into 126 mg tablets and to a hardness of 5.6 kg/Sq. Cm on a Cadmach single punch tablet machine.

For each method tablet powder equivalent to 100 mg of SX was taken in a 100 ml volumetric flask. To each flask 50 ml of methanol was added and mixed to dissolve the drug. The solutions were then filtered and the filtrates were collected into 100 ml volumetric flasks and made upto volume with distilled water. Further analyses were carried out as described under methods. The above dosage forms were also analysed by the UV spectrophotometric method by extraction of the drug into methanol and measuring the absorbance at 254 nm[7] after suitable dilution with distilled water. The results are given in Table-2.

Recovery experiments : Recovery experiments in both the methods were performed by adding known amount of drug to previously analysed pharmaceutical preparations and also to various excipients used in formulations. The results are given in Table-2.

RESULTS AND DISCUSSION

The colored solution exhibited absorbance maxima at 430 nm for method A and 620 nm for method B. The color obeyed Beer's law in the concentration range of 0-32 µg/ml and 0-200 µg/ml for method A and method B respectively. The color formed in both the method is stable over a period of six hours. The regression lines were found to be $Y=0.017 + 1.8x10^{-3} X$ and $Y=3x10^{-3}+2.9x10^{-4}X$, where X is the concentration of SX in micrograms per ml of dilution and Y is the absorbance at the corresponding λmax for method A and method B respectively. Sandell's sensitivity (µg/cm^2/0.001 absorbance unit) and molar extinction coefficient (1.mole^{-1}. cm^{-1}) were found to be 0.052 and $7.84x10^3$ for method A and 0.302 and $1.215x10^3$ for method B respectively. When the stock solution in both the methods was assayed repeatedly (n=5) the coefficient of variation (precision) was found to be in the range of 1.5-2.0% (Table-1). When pharmaceutical preparations containing SX were analysed the results obtained by the proposed methods were in good agreement with the labelled amounts and are comparable with the results of UV spectrophotometric method. Recovery in both the methods is 98-101%.

In method A, the yellow colored chromogen is formed due to the nitrosation of salmeterol under alkaline conditions. Method B is based on the reaction of phenolic hydroxy group with ferric ions to give colored complexes[8]. The blue colored chromogen formed in method B is due to the reaction of phenolic hydroxy group of salmeterol with ferric ions of ferric alum reagent.

Thus the proposed methods are simple, less time consuming and applicable for the routine determination of SX in bulk and in dosage forms. Method A is more sensitive than method B.

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