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ECO-FRIENDLY AND COST-EFFECTIVE UV SPECTROSCOPY METHOD FOR THE ESTIMATION OF PREDNISOLONE SODIUM PHOSPHATE IN BULK AND PHARMACEUTICAL DOSAGE FORM

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Keywords:

Prednisolone, Sodium Phosphate, UV Spectroscopy, Tablet Dosage Form

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ABSTRACT: Prednisolone Sodium Phosphate (PSP) is a steroidal anti-inflammatory or immune suppressive agent. It is a sodium salt of the phosphoester of the glucocorticoid prednisolone. It is freely soluble in water; soluble in methanol; slightly soluble in alcohol and in chloroform; and very slightly soluble in acetone and in dioxane. A simple spectrophotometric method was developed for the determination of Prednisolone Sodium Phosphate in Pharmaceutical tablet dosage form. PSP exhibiting λ max at 246nm in solvent media 100% water and obeyed linearity in the concentration range of 2 to 12µg and exhibited good correlation coefficient (R^2=0.999). This method was successfully applied to the determination of PSP content in four different marketed brands from local market and the results were in good agreement with the label claims. The percentage recovery was found to be 99.80±0.238. The sample solution was stable up to 10 hours. The method was validated statistically and studies for linearity, precision, repeatability, and reproducibility. Various methods for analysis of the Prednisolone Sodium Phosphate are available but are time consuming and expensive. But here developed method is new, pricise, accurate, simple, safe and stability indicating. Furthermore it was safe for operator, environmently friendly and economical in terms of cost of chemicals and waste management. Thus, this method is not only suits for routine pharmaceutical quality control analysis work, but also presents prototype for the development and utilization of greener analytical procedures in which an environmental impact is concerened. The obtained results proved the method can be employed for the routine analysis of prednisolone sodium phosphate.

INTRODUCTION: Chemically Prednisolone Sodium Phosphate is glucocorticoid and its IUPAC name is Disodium [2-[(8*S*,9*S*,10*R*,11*S*,13*S*, 14*S*, 17*R*)-11,17-dihydroxy-10,13-dimethyl - 3 - oxo - 7, 8, 9,11,12,14,15,16-octahydro - 6H – cyclopenta [*a*] phenanthren-17-yl]-2-oxoethyl] phosphate.[1]

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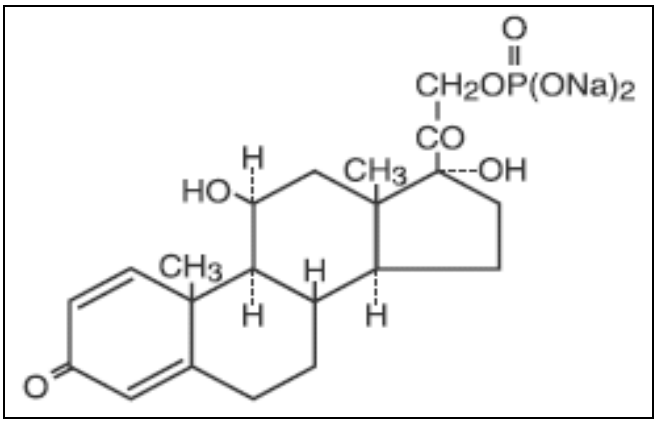
It is mainly used for the treatment of a wide range of inflammatory and auto-immune disease such as asthma multiple sclerosis, rheumatoid arthritis, autoimmune hepatitis etc [2]. Prednisolone is also known as 'disease modifying anti-arthritic drugs [3] because of its anti-inflammatory action by inhibiting gene transcription for COX-2, cytokines, cell adhesion molecules, and inducible NO synthetase [4, 5]. It is soluble in water; soluble in methanol; slightly soluble in alcohol and in chloroform; and very slightly soluble in acetone and in dioxane. In our Literature survey reveals that for Prednisolone HPLC [6, 7] Spectrophotometric [8-9] and solid phase extraction[10] and HPTLC methods

have been reported for its determination in commercial formulation. However some of these methods are costlier and time consuming.



PREDNISOLONE SODIUM PHOSPHATE
($C_{21}H_{27}Na_2O_8P$), M.Wt=484.39

At present, acetonitrile and methanol are by far the most popular modifiers because of their favorable characteristics including excellent miscibility with water, little interference in the short wavelength region in UV detection, relatively low viscosity [11], commercial availability at high purity grade and broad compatibility with the analytes. However, these solvents also have notable disadvantages. In a case of acetonitrile which is a minor by-product in the manufacture of acrylonitrile polymers and cannot be considered as a renewable resource, a downturn in plastic production has occasionally resulted in global shortages and rising cost of this solvent [12].

Furthermore, acetonitrile is ranked by the U.S. Environmental Protection Agency (U.S. EPA) as a toxic chemical as liquid or vapor and waste has to be detoxified through special chemical treatment, leading to high to very high disposal cost [13]. Later, attempts have been made to switch the media from acetonitrile/ methanol to water. However, acetonitrile/ methanol is still highly toxic to humans and causes adverse effects on aquatic life [13].

To overcome these difficulties Spectrophotometric analysis serves to be the quickest, promising and reliable method for routine analytical needs. The aim of the present study is to develop two new simple, rapid, reliable and precise UV Spectrophotometric methods for analysis of PSP from pharmaceutical formulation. To the best of our knowledge, literature search from databases has not presented the evidence of using greener and less toxic solvent for the spectroscopic analysis of Prednisolone Sodium Phosphate. Therefore, the aim of the present study was to assess the feasibility of using green solvent water as 100% solvent media for the development of an eco-friendly, operator-safe and cost-effective spectrocopic method and its validation for estimation of PSP from pharmaceutical formulation.

MATERIALS AND METHODS:

Apparatus: A Shimadzu UV–Visible Spectrophotometer (UV1800, Shimadzu Corporation, Kyoto, Japan) was used for all absorbance measurements with matched quartz cells.

Materials: All chemicals and reagents were of analytical grade. Prednisolone Sodium Phosphate in the form of powder with certificate of analysis was gifted by In agio Pharmaceutical Ltd., Pune, Maharashtra, india.

Stock solution:

Preparation of standard stock solution of Prednisolone Sodium Phosphate:

10 mg of Prednisolone Sodium Phosphate accurately weighted by electronic balance and dissolved in 80ml of double distilled water in 250ml conical flask. Content of flask was kept for stirring on magnetic stirrer for 10 min and transferred in 100ml volumetric flask. Conical flask was rinsed by 20ml of double distilled water and this water was used to make up volume 100ml of volumetric flask to give conc. of 100µg/ml.

Prepration of working standard solution of Prednisolone Sodium Phosphate:

The working solution of Prednisolone sodium phosphate was prepared by further diluting the stock solution. Then pipette out 0.2ml, 0.4ml, 0.6ml, 0.8ml, 1.0ml & 1.2ml of solution and make up to 10ml leads to 2µg/ml, 4µg/ml, 6µg/ml, 8µg/ml, 10µg/ml & 12 µg/ml concentration solution. This solution was estimated by UV spectrophotometer by using H_2O as blank at 246nm.

Fixing of wave length:
After selecting the suitable solvent, the fixing of the λmax for the proposed method is very important. This can be done by scanning the drug sample (Prednisolone Sodium Phosphate) solution in H_2O in the range of 400nm-200nm and the most repeated maximum absorbance with linearity and repeatability can be fixed as λmax for the drug. And in the proposed method for Prednisolone sodium phosphate drug shows maximum 246 nm. With more linearity, repeatability (ruggedness) and the λmax was fixed as 246 nm. Shown in the **(Figure 1)**

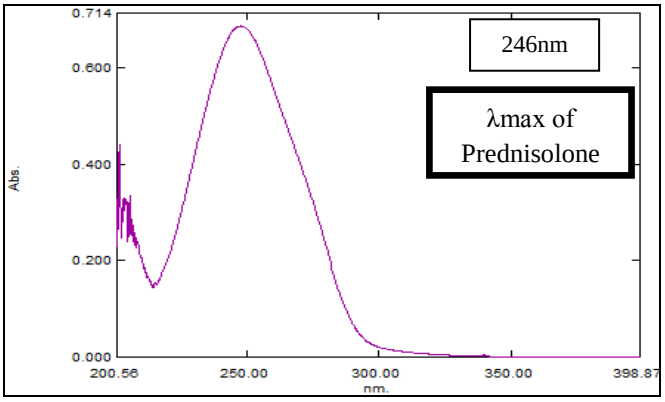


FIGURE1: FIXING OF WAVELENGTH FOR PREDNISOLONE SODIUM PHOSPHATE

Validation parameter:
Linearity and range:
For linearity study from the working standard at different concentration 2, 4, 6, 8, 10 and 12 µg/ml of drug solution were placed in 6 different 10ml volumetric flask volume was made up to the mark with water. Absorbance was measured at 246nm and results are recorded in **Table1 and Figure 2**. Then obtained data were used for the linearity calibration plot.

Intra-day precision (repeatability) and inter-day precision study (intermediate precision)
Prepared the standard stock solution of prednisolone sodium phosphate. Prepare the three concentration of (2, 6, and 12 µg/ml), by using mobile phase water. Take λ_{max} at the intraday and inter day. Calculate the % RSD

Stability study:
Samples prepared for repeatability study were preserved for 24 hour at room temperature and analyzed on the following day to test for short-term stability.

Accuracy study:
This study was carried out using the stock solution (100µg/ml). Take three concentrations 2 µg/ml, 6µg/ml, and 12µg/ml. And take six reading of these concentrations. Calculate the % RSD of the concentration.

Reproducibility:
Reproducibility is assessed by mean of an inter laboratory trial. The absorbance readings were measured at 246nm at different laboratory using another spectrophotometer and the value obtained were evaluate using t-test to verify their reproducibility data for prednisolone sodium phosphate at 246nm is recorded:

Limit of detection & limit of quantitation:
The limit of detection and quantification of drug are calculated with the standard deviation and slop.

RESULTS AND DISCUSSION:
Method validation:
Linearity and range:
The calibration curve obtained was evaluated by its correlation coefficient. The absorbance of the samples in the range of 2 to 12 mg/ml was linear with a correlation coefficient (R2) 0.999.

TABLE 1: LINEARITY AND RANGE FOR PREDNISOLONE AT 246NM

Sr. No.	Concentration (µg/ml)	Absorbance
1.	2	0.143
2.	4	0.290
3.	6	0.410
4.	8	0.540
5.	10	0.685
6.	12	0.798

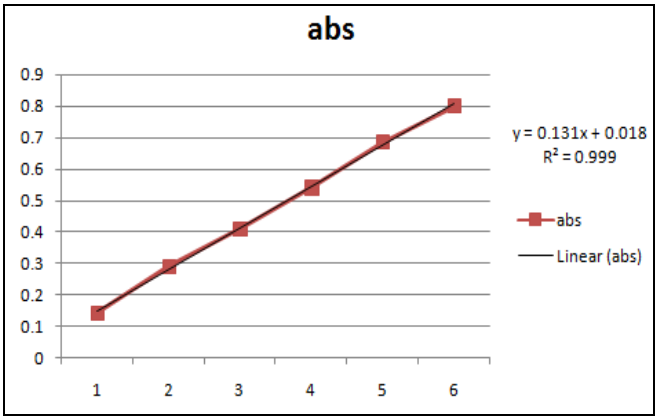


FIGURE 2: LINEARITY AND RANGE FOR PREDNISOLONE SODIUM PHOSPHATE

Parameter	Data
Range	2 µg/m to 12 µg/m
Correlation coefficient	0.999
Slope	0.131
Intercept	0.018

Intra and inter day precision:

Variation of results within the day (intra day), Variation of result between days (inter day) were analyzed. Intraday precision was analyzing Prednisolone Sodium Phosphate for three times in the same day at 246nm. Inter-day precision was determined by analyzing the drug different day for three days at 246nm. Precision data for Prednisolone Sodium Phosphate at 246nm is given **Table 2**.

TABLE 3: PRECISION DATA FOR PREDNISOLONE AT 246nm

Concentration µg/ml	Intraday (n=3)	% RSD
2	0.127 ± 0.001665	0.78745
6	0.432 ± 0.002229	0.51597
12	0.704 ± 0.002137	0.30355

Accuracy and recovery study:

Results within the range of ensure an accurate method (**Table 4**) as well as indicate non-interference with the excipients of formulation.

Concentration µg/ml	Inter day (n=3)	% RSD
2	0.126 ± 0.001251	0.7936
6	0.436 ± 0.003189	0.7314
12	0.719 ± 0.001378	0.1910

TABLE 4: ACCURACY FOR THREE DIFFERENT CONCENTRATIONS OF PSP

Concentration µg/ml	Absorbance Measured (Mean±SD)	RSD (%)
2	0.125 ± 0.000753	0.4024
6	0.449 ± 0.002338	0.4207
12	0.729 ± 0.003033	0.4160

The accuracy of the proposed methods was assessed by recovery studies at three different levels i.e. 80%, 100%, 120%. The recovery studies were carried out by adding known amount of prednisolone sodium phosphate solution of the drug to pre-analyzed tablet solutions. The resulting solutions were then reanalyzed by proposed methods; the results are reported in **Table 5**.

TABLE 5: RECOVERY DATA OF PROPOSED METHOD

Level of recovery	Amount of sample (µg/ml)	Amount of drug added (µg/ml)	Amount added %	% Mean recovery (±S.D.) (n = 3)
80%	10	8.0	80	100.49 ± 1.74
100%	10	10.0	100	99.98 ± 1.18
120%	10	12.0	120	101.35 ± 1.36

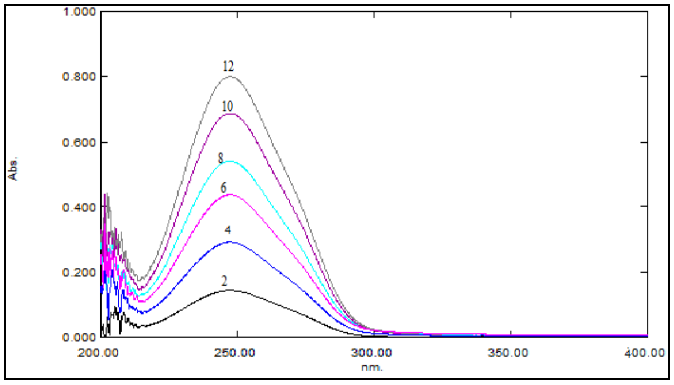


FIGURE 3: ACCURACY OVERLAY GRAPH OF PREDNISOLONE SODIUM PHOSPHATE

Reproducibility:

Reproducibility is assessed by mean of an inter laboratory trial. The absorbance readings were measured at 246nm at different laboratory using another spectrophotometer and the value obtained were evaluate using t-test to verify their reproducibility data for prednisolone sodium phosphate at 246nm is recorded in **Table 6**.

TABLE 6: REPRODUCIBILITY DATA FOR PREDNISOLONE AT 246nm

Instrument I SIMADZU	Instrument II JASCO	Result of t-test	Inference
0.295 ± 0.0005 16	0.296 ± 0.0006 16	0.99	Not significant difference

Specificity and selectivity:

Prednisolone sodium phosphate and selective as given **Table 7**.

TABLE 7: SPECIFICITY AND SELECTIVITY STUDY

Study	Prednisolone sodium phosphate
Specificity	Specific
Selectivity	Selective

Limit of Detection & Limit of Quantitation:

The limit of detection and quantification of drug are calculated with the standard deviation and slop. Its value described in **Table 8**.

$$LOD = \frac{3.3\sigma}{S}, \quad LOQ = \frac{10\sigma}{S}$$

σ =Standard deviation

S= slope of the calibration curve

LOD	LOQ
0.0538	0.1631

Assay Procedure:

Preparation of sample solution:

The proposed method was applied to analyze commercially available Prednisolone tablet. Ten tablets (5mg) were weighed and powdered. The amount of tablet powder equivalent to 50mg of was weighed accurately and transfer into the 100ml conical flask and dissolved by 40ml double distilled water. Content of flask was kept for stirring on magnetic stirrer for 10 min and transferred in 50ml volumetric flask. Conical flask was rinsed by 10ml of distilled water and this water was used to make up volume 50ml of to give conc. of 1mg/ml. Weigh accurately about 10 tablets (5mg), powder and take 50mg equivalent quantity of prednisolone and transfer into a 50ml standard flask. And dissolve the water by using magnetic staring (30min).

Then pipette out 1ml of solution and make up to 100ml leads to 10µg/ml concentration solution. This solution can be estimated in UV spectrophotometer by using water as blank at 246nm. Compare with the standard.

Assay Result of Marketed Formulation:

TABLE 8: ASSAY RESULT OF MARKETED FORMULATION

Formulation	Label claim mg/tablet	Amount found mg	% Label claim
Tablet	5mg	4.96	99.20%
Tablet	5mg	4.98	99.60%
Tablet	5mg	4.97	99.40%
Tablet	5mg	4.98	99.60%

Stability of Sample:

The sample of 4µg/ml drug solution was prepared by suitable dilution with diluents and absorbance were taken at 246nm against the blank. The stability of sample was found to be more than 10 hrs. As shown following **Table 9**.

TABLE 9: STABILITY OF SAMPLE

Sr. No.	Concentration of drug solution (µg/ml)	Time (min)	Absorbance at 246nm.
1	4	0	0.236
2	4	30	0.236
3	4	60	0.236
4	4	90	0.237
5	4	120	0.235
6	4	150	0.233
7	4	180	0.232
8	4	210	0.231
9	4	240	0.231
10	4	360	0.230
11	4	480	0.230
12	4	600	0.230

CONCLUSION: The results and the statistical parameters demonstrate that the proposed UV spectrophotometric method is simple, rapid, specific, accurate and precise. The method showed acceptable linearity and accuracy. Method was safe for operator, environmentally friendly and economical in terms of cost of chemicals and waste management. Therefore, this method can be used for the determination of prednisolone sodium phosphate either in bulk or in the dosage formulations without interference with commonly used excipients and related substances by the utilization of greener analytical procedures in which an environmental impact is concerned

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REFERENCES:

1. Tripathi KD, Essentials of medical pharmacology, 6th Edition, Jaypee Brothers Medical Publishers Ltd. New Delhi, 2008; P. 217-218.
2. GI Lambrou; S Vlahopoulos; C Papthanasiou; M Papanikolaou; M Karpusas; E Zoumakis; Prednisoloneexerts late mitoginic and biphasic effect on

resistant acute lymphoblastic leukemia cells:relation to early gene expression Leuk Res.,2009
3. S Siro; S Srinivas S. International J of Pharmaceutics. 1998,166,177.
4. Australian Medicines Handbook., Adelaide: Australian Medicines handbook Pty Ltd., 2010,497.
5. HP Rang; MM Dale; JM Ritter; PK Moore.,Hunter Pharmacology, 5th edition,2003; 413
6. Sohan S. Chitlange, Kaushalendra K. Chaturvedi, Sagar B. Wankhede, Development and Validation of Spectrophotometric and HPLC method for the Simultaneous Estimation of Salbutamol sulphate and Prednisolone in Tablet Dosage Form J Chem. Pharm. Res.; 2011; 3(1) ;304-312.
7. Yoe-Ray Ku, Yi-Chu Liu And Jer-Huei Lin, Solid-phase Extraction and High-performance Liquid Chromatographic Analysis of Prednisone Adulterated in a Foreign Herbal Medicine, J of Food and Drug Anal.Vol. 9; No. 3; 2001; 150-152.
8. Mohit Rohitas, Abhinav Agrawal, Ashish K jain, Narendra K lariya, Anil K kharya and Govind P Agrawal, Development of Simultaneous Spectrophotometric Method of Mesalazine and Prednisolone in Same Dosage Form, Int. J. of App. Pharmace. Vol 2; Issue 4; 2010.
9. Moharana AK, Banerjee M, Panda S, Muduli JN, Development and Validation of UV Spectrophotometric Method for the Determination of Mesalamine in Bulk and Tablet Formulation, Int. J Pharm. Pharm. Sci, Vol 3; issue 2; 2011; 19-21.
10. Jamakhandi CM, Javali C, Disouza JI, Chougule US, Mullani AK, Spectrophotometric Determination of Lisinopril Dosage Form By Condensation Reaction, Int. J Pharm. Pharm. Sci, Vol 3; Issue 2; 2011; 185-187.
11. Ribeiro RLV, Bottoli CBC, Collins KE, Collins CH. Reevaluation of ethanol as organic modifier for use in RP-HPLC mobile phase. J. Braz. Chem. Soc. 2004; 15: 300-6.
12. Tullo A, A solvent dries up. Chem. Eng. News. 2008; 86: 27.
13. http://www.epa.gov/ttn/atw/188.polls. html (accessed August 1, 2011).

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