# Development and Validation of HPLC Method for Estimation of Analytes in Combined Dosage Form

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***Abstract-Paracetamol,Phenylephrine HCl and Triprolidine HCl are frequently associated in pharmaceutical formulations against the common cold. A HPLC method for the simultaneous estimation ofthese compounds in pharmaceutical formulation such as tablet and syrup, including the separation of impurities and excipients has been developed and validated. The separation was achieved using Kinetex RP (C18, 150 x 4.6 mm I.D.) column. Final chromatographic conditions were a gradient elution being solvent A: Phosphate buffer 10Mm at pH 2.8 and solvent B: Methanol. At t=0, the mobile phase consisted of 88% A and 12% B for first 4 min and it changed with a linear gradient during 10 min to 85% A and 15% B and at t=10 min, it returns to the initial conditions (88% A and 12% B) during 10 min remaining at this composition until t=20 min. Photo diode array detector was used for the detection wavelength. The detection was performed at 216 nm.Validation parameters are carried out.***

***Key words: HPLC,Paracetamol, Phenylephrine HCl, Triprolidine HCl.***

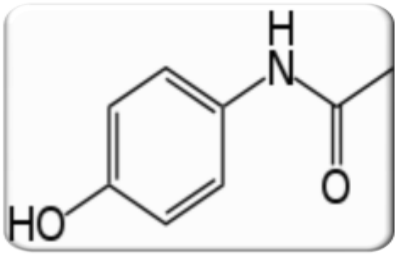
## I. INTRODUCTION

Paracetamol is an analgesic and antipyretic. Phenylephrine HCl is an sympathomimetic (decongestant) and Triprolidine HCl is an $H_1$-receptor antagonist (antihistaminic). These substances are frequently associated in pharmaceutical formulations against the common cold, but with an important imbalance between the different actives in the dosage forms. Moreover, the active compounds have very different polarity and therefore, chromatographic behaviour. The dosage forms also contain excipients, some of which may interfere with the analysis of the active ingredients. No single method is reported to determine the active ingredients quantitatively in this combination.

The aim of the present work was the development and validation, following ICH guidelines [23] of a HPLC method for the simultaneous determination of Paracetamol, Phenylephrine HCl and Triprolidine HCl in pharmaceutical formulations such as Tablets and Syrup, including the separation of impurities and excipients. The chemical structures of the assayed compounds and most of their values for the acid-base constants are shown below.

*A. Paracetamol :-*

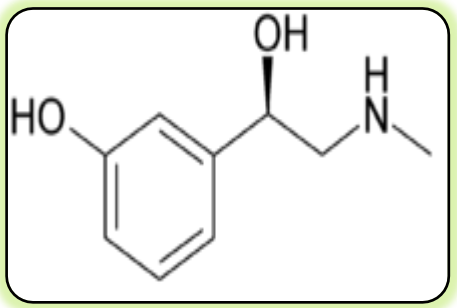
Structure :




Chemical name : *N*-acetyl-*p*-aminophenol
Molecular formula : $C_8H_9N$
pKa : 9.46, 14.17

*B. Phenylephrine HCl:*

Structure :

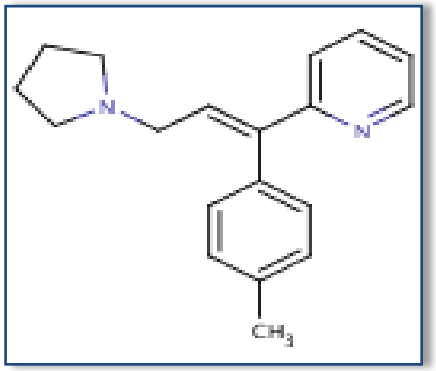



Chemical name : (*R*)-3-[-1-hydroxy-2- (methyl amino) ethyl phenol

Molecular formula :$C_9H_{13}NO_2$ pKa : 8.9

*C)Triprolidine:*

Structure:




Chemical name: 2-[(1E)-1-(4-methylphenyl)-3-(pyrrolidin-1-yl)prop-1-en-1-yl]pyridine

Molecular formula :$C_{19}H_{22}N_2$pKa : 8.64

## II. EXPERIMENTAL PROCEDURE

*Materials and methods*

*Chemicals and reagents*

All chemicals were used without further purification. Paracetamol, Phenylephrine hydrochloride andTriprolidine hydrochloride. Methanol (HPLC grade),Monobasic sodium dihydrogen phosphate (AR grade) and Ortho phosphoric acid (HPLC grade).

Apparatus

The HPLC analysis was carried out onWaters 600 quaternary gradient pump with 996 PDA Detector and 717 Autosampler. A Phenomenex Kinetex C18,(150 × 4.6mm), 5µm column was used for the analysis at ambient temperature. pHmeasurements of the mobile phase were carried out with a pH meter Hanna

*Chromatographic Procedure*

Mobile phase composed of 10 Mm phosphate buffer pH 2.8 and methanol. A three step gradient program was developed with step-1 started with 12% methanol and 88% buffer at flow rate of 1 ml for first 4 min. step-2 started with gradient changes to 85% methanol and 15% buffer for next 10 min and finally method ended with step-3 achieving initial concentration of 12% methanol and 88% buffer. During all the method flow rate was 1 ml/min. The selected wavelength was 216 nm.

*Preparation of Standard Stock Solution*

An accurately weighed quantity of Paracetamol 325 mg was transferred to the 100 ml volumetric flask and dissolved in Methanol. Finally the volume was made up to the mark with diluent to obtain the resultant concentration of 3250 ppm.

An accurately weighed quantity of Phenylephrine Hydrochloride 5 mg was transferred to the 100 ml volumetric flask and dissolved in Methanol. Finally the volume was made up to the mark with diluent to obtain the resultant concentration of 50 ppm.

An accurately weighed quantity of Triprolidine Hydrochloride 2.5 mg was transferred to the 100 ml volumetric flask and dissolved in Methanol. Finally the volume was made up to the mark with diluent to obtain the resultant concentration of 25 ppm.

*Preparation of working standard solution:*

Mix working of standard stock solution:

1ml ofParacetamol, 1mlPhenylephrine HCl and 1mlTriprolidine HCl was pipette out from above standard stock solution respectively into a 10 ml volumetric flask, and diluted up to mark by Methanol and to obtained resultant concentration of 325ppm,5ppm,and 2.5ppm of Paracetamol, Phenylephrine HCl and Triprolidine HCl respectively.

1ml of resultant solution was injected in developed chromatographic conditions.

*Mix STD*:

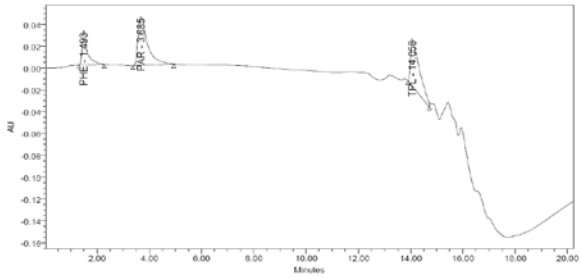



Fig:1Separation of the three drugs in selected mobile phase Showing R.T. for Phenylephrine HCl 1.493, Paracetamol 3.685, and Triprolidine HCl 14.058 min respectively.

*Pharmaceutical Preparation Procedure*

For the test solution 20 tablets (Marketed preparation (Recofast Plus) contain Paracetamol 325mg, Phenylephrine HCl 5mg, Triprolidine HCl 2.5mg)were weighed and the average weight was determined. 20 tablets were triturated and powder. Equivalent to 325mg of PAR, 5mg of PHE and 2.5mg of TPL was added into a 100 ml volumetric flask the content were mixed with diluents &sonicated for 15 min &same content were filtered through 0.45 µ membrane filter. 1 ml of resultant solution was taken in a 100 ml of volumetric flask &volume was made up to the mark with diluents.

1ml of resultant was diluted to 10ml Methanol and injected in developed chromatographic conditions.

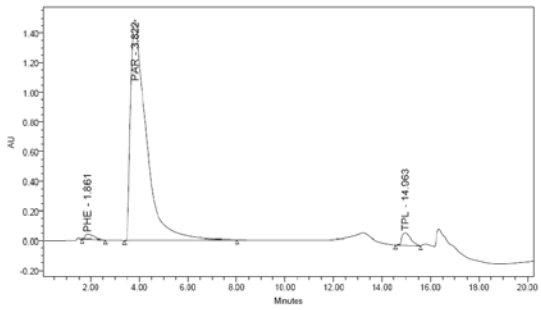



Figure:2Separation of the three drugs in selected mobile phase showing R.T. for PHE 1.861, PAR 3.822 and TPL 14.963 min respectively.

*Application Of Proposed Method For Estimation Of Par, Phe And Tpl On Marketed Formulation* (SYRUP)

For preparation of sample for analysis 5ml of syrup formulation was taken in 100ml volumetric flask and volume was made upto 100ml with Methanol. The

resultant was sonicate for 10 min on a ultra-bath sonicator and filtered through 0.45µm membrane filter.

1ml of resultant was diluted to 10ml Methanol and injected in developed chromatographic conditions.

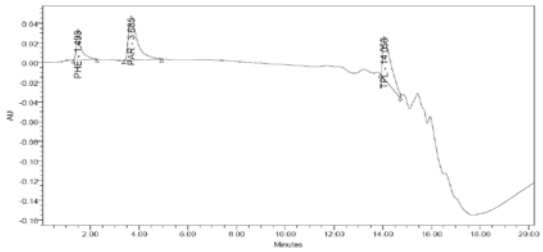



Figure. 3: Separation of the three drugs in selected mobile phase showing R.T. for PHE 1.493, PAR 3.685 and TPL 14.058 min respectively.

*Validation*

- *Linearity*

For each drug, appropriate dilutions of standard stock solutions were assayed as per the developed methods. The linearity datapresented in Table no.5.

- *Accuracy*

Accuracy of the developed method was conformed by recovery study as per ICH norms at three different concentration levels of 80 %, 100 %, 120 % by replicate analysis (n = 3). Here to a preanalysed sample solution, standard drug solutions were added and then percentage drug content was calculated. The result of accuracy study was reported in Table no.6. The recovery study indicates that the method is accurate for quantitative estimation of Paracetamol, Phenylephrine hydrochloride and Triprolidine hydrochloride in tablet dosage form as the statistical results are within the acceptance range (R.S.D. < 2.0).

*Precision*

Precision was determined by studying the repeatability and intermediate precision.

*Repeatability (System Precision)*

Repeatability result indicates the precision under the same operating conditions over a short interval of time. Repeatability was performed for three times. The results of statisticalevaluation are given in Table 2.

*Intermediate Precision (Inter-Day And Intra-Day Precision)*

An intermediate precision was carried out by intra and inter day precision study. In intra day study concentration of drugs were calculated on the same day at an interval of one hour. In inter day study the drug contents were calculated on three different days. Study expresses within laboratory variation in different days. In both intra and inter-day precision study for the methods % RSDwere not more than 1.0 indicates good intermediate precision (Table 3).

*Limit Of Detection (Lod) And Limit Of Quantitation (Loq)*

The LOD and LOQ of Paracetamol, Phenylephrine hydrochloride and Triprolidine hydrochloride by proposed methods were determined using calibration standards. LOD and LOQ were calculated as 3.3*Avg SD / S and 10*Avg.SD /S respectively, where S is the slope of the calibration curve and Avg SD is the average standard deviation of response. The results are shown in Table 4.

*Robustness*

For robustness studies some deliberate changes in chromatographic conditions was done as per ICH guidelines.

Effect of Variation in flow rate of mobile phase by ± 10%

Change in mobile phase pH by ± 0.1 units

The results are shown in Table 7.

## III. RESULT AND DISCUSSION

HPLC has gained the valuable position in the field of analysis due to ease of performance, specificity and the analysis of sample of complex nature.

This technique is commonly used for the quantitative estimation of the drugs from their formulation as well as for studying their metabolites of drugs and their estimation in their biological fluids. This method offers advantages of estimating the constituents for the multi-component system.

This technique was employed in the present investigation for estimation of Paracetamol, Phenylephrine HCl and Triprolidine HCl tablet dosage form. Careful evaluation of various parameters influencing analysis is an important aspect for the development of analytical method. In order to establish RP-HPLC method the following parameters were studied.

*HPLC column selected:*

The HPLC system consisted of waters series 600E pump quaternary gradient, waters online degasser module a 996 photo-diode array (PDA) detector, a 717 Auto injector ; data were acquired and processed by use of EMPOWER software (all equipment from Waters, Milford). The chromatographic separations were carried out on a reverse phase Kinetex-C18 column (150 mm × 4.5mm i.d.,particle size 5 µ, core shell technology).

*Mobile Phase Selected*:

Dissolve 15.6gms of monobasic Sodium mono Phosphate in 1000 ml of HPLC water, add 6.6 ml of ortho phosphoric acid in 1000 ml of HPLC water to adjusted pH 2.8, and finally made up to pH 2.8 by ortho phosphoric acid. The wavelength 216 nm was selected for the evaluation of the chromatogram of drugs. The selection of the wavelength

was based on the λmax obtained by scanning of standard solution. This system gave good resolution and optimum retention time with appropriate tailing factor (<2). The mean values of system suitability test result are depicted in Table

The following chromatographic conditions were established by trial and error and were kept constant throughout the method.

*Analysis In Tablet Formulation*:

Table no. 1: summary of system suitability of test results

| Sr. No. | Parameter | Observations | | | Limits |
|---|---|---|---|---|---|
| | | PHE | PAR | TPL | |
| 1 | The % RSD of peak area response for three replicate injections of standard | 0.37 | 0.03298672 4 | 0.86054667 | NMT 2.0 |
| 2 | Theoretical plates | 151592 | 102295 | 54520 | NLT 2000 |
| 3 | Tailing factor | 0.6 | 0.8 | 1.6 | NMT 2.0 |

*System Suitability Acceptance Criterion*:

1. Relative standard deviation of the area of Paracetamol, Phenylephrine HCl and Triprolidine HCl peaks in standard chromatograms should not be more than 2.0 %.

2. Theoretical plate of peaks in Std. chromatograms should not be less than 2000.

3. Tailing factor (Asymmetry) of peaks in standard chromatograms should be less than 2.0

*thus the results obtained for such method are given as follow*:

After establishing the chromatographic conditions, Mix standard and marketed preparation were prepared and analysed by following procedure described under experimental work. It gave accurate, reliable results and was extended for estimation of drugs in marketed tablet formulation.

Table No.2: Results of Method Precision Using Marketed Sample

Thus the above analysis, passes the limit of % L.C. as between 95 – 105%

| | PHE | | PAR | | TPL | |
|---|---|---|---|---|---|---|
| Sr.no. | Assay (mg) | Assay % of LC | Assay (mg) | Assay % of LC | Assay (mg) | Assay % of LC |
| 1 | 5.49 | 109.79 | 328.35 | 101.03 | 2.54 | 101.68 |
| 2 | 5.51 | 109.34 | 328.57 | 101.06 | 2.55 | 101.85 |
| 3 | 5.47 | 109.42 | 328.46 | 101.05 | 2.58 | 101.7 |
| Average | 5.49 | 109.516667 | 328.46 | 101.04 | 2.55 | 101.74 |
| SD | 0.02 | 0.24006943 | 0.11 | 0.01527525 | 0.02081666 | 0.09291573 |
| % RSD | 0.36429872 | 0.21920813 | 0.03348962 | 0.01511802 | 0.81633961 | 0.09132665 |

*Validation*

Validation of these methods was performed as per the USP guidelines for these following parameters

*Precision*

*System Precision*

Prepared the standard solution as per test method and injected into the HPLC system in three replicates. It was found that all system suitability parameters are well within the limits.

*Method Precision*

Replicate estimation of tablet analysed by the proposed method has yielded quite consistent result indicating repeatability of method. Study showed R.S.D. less than 2.

INTERMEDIATE PRECISION (RUGGEDNESS):

Prepared six sample solutions as per the test method. Injected into the different HPLC system (preferably with different manufacturer or same manufacturer with different configuration) by using the different column and by the different analyst at different date.

Table no. 3: data analysis between precision and intermediate precision

| | PHE | | PAR | | TPL | |
|---|---|---|---|---|---|---|
| Sr.no. | Set I | Set II | Set I | Set II | Set I | Set II |
| 1 | 109.79 | 108.51 | 101.03 | 100.6 | 101.68 | 101.82 |
| 2 | 109.34 | 108.3 | 101.06 | 100.06 | 101.85 | 101.8 |
| 3 | 109.42 | 109.42 | 101.05 | 101.5 | 101.7 | 101.81 |
| Average | 109.516667 | 108.743333 | 101.046667 | 100.72 | 101.743333 | 101.81 |
| SD | 0.24006943 | 0.59534304 | 0.01527525 | 0.72746134 | 0.09291573 | 0.01 |
| % RSD | 0.21920813 | 0.54747544 | 0.01511703 | 0.72226106 | 0.09132366 | 0.00982222 |

SET – I:Method Precision data SET – II:Intermediate Precision data

*Acceptance Criteria*:

The overall % RSD for the twelve determinations shall be NMT 2.0

Table No.4 Lod And Loq

| Sr.no. | API | LOD (µg/ml) | LOQ (µg/ml) |
|---|---|---|---|
| 1 | Phenylephrine Hcl (PHE) | 1.56 | 4.72 |
| 2 | Paracetamol (PAR) | 1.51 | 4.6 |
| 3 | Triprolidine (TPL) | 0.122 | 0.371 |

*Linearity &Range*:

The linearity of an analytical procedure is its ability (within a given range) to obtain test results, which are directly proportional to the concentration (amount) of analyte in the sample. Linearity was carried out for five levels in the range of 80% to 200%.

Linearity of method was ranging from concentration 80% to 200 %ml for Paracetamol, PhenylephrineHcl and Triprolidine Hcl.

A graph was plotted with concentration on X axis and mean peak areas on Y-axis. The R2 value was found to be 0.984, 0.990 and 0.992 for Phenylephrine HCl, Paracetamol and Triprolidine HCl are respectively (R2value should be always near to 1).The result show that an excellent correlation exist between concentration and mean peak areas within the concentration range. From the studies carried out and result obtained the proposed methods are compared in terms of statistical data, ease of application and reliability. Thus the method developed is accurate, precise, specific, &linear Hence it can be said that, RP-HPLC is the most accurate, precise and reproducible among all methods.

Table No 5: Linearity and Range Studies

| Sr. no. | Addition level range labeled claim | PHE | | | PAR | | | TPL | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | Am.t (µg/mL) (n=) | Peak area response | Amt. recovered | Amt.(µg/mL) (n=3) | Peak area response | Amt. recovered | Amt. (µg/ml) (n=3) | Peak area response | Amt. recovered |
| 1 | 80% | 4 | 520379.3 | 3.94 | 260 | 948265.7 | 248.52 | 2 | 19236.67 | 2.282 |
| 2 | 100% | 5 | 993855.7 | 4.93 | 325 | 1491082 | 314.66 | 2.5 | 37502.67 | 3.168 |
| 3 | 120% | 6 | 1317399 | 5.84 | 390 | 2632877 | 384.95 | 3 | 65516 | 4.528 |
| 4 | 150% | 7.5 | 1722462 | 7.41 | 487.5 | 3577556 | 473.75 | 3.75 | 83608.33 | 5.406 |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| 5 | 200% | 10 | 2138679 | 9.94 | 650 | 4429972 | 555.90 | 5 | 101649.7 | 6.281 |
| Correlation Coefficient | | 0.984 | | | 0.990 | | | 0.992 | | |
| Slope | | 43715 | | | 87798 | | | 20609 | | |
| Intercept | | -362497 | | | -1271857 | | | -27806.9 | | |

Fig. Graph of linearity of Phenylephrine HCl.

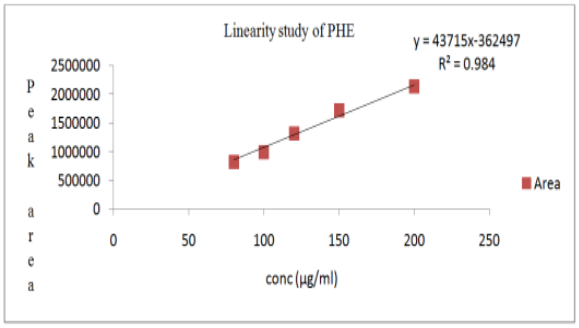



Fig. Graph of Linearity of Paracetamol

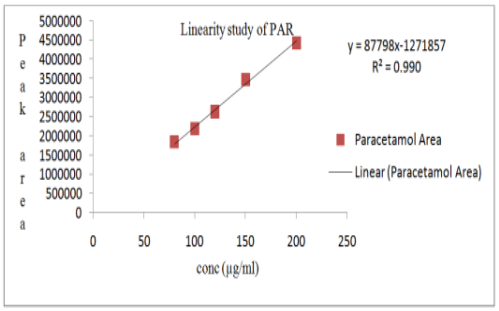
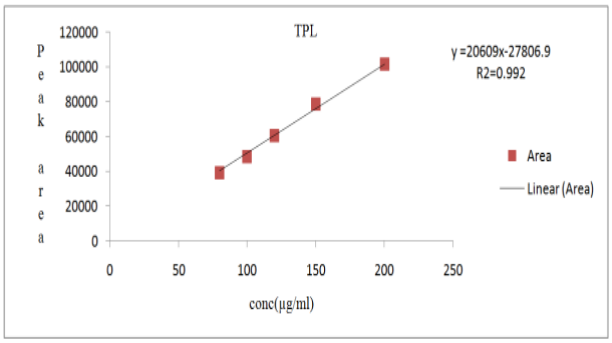



Fig. Graph of Linearity of Triprolidine HCl




*ACCURACY*:

Accuracy of the proposed method was ascertained from the recovery Studies by standard addition method. Results are shown in the Table. Recovery results were well within the range **94-102%.** Thus the method was found to be accurate.

Table no. 6: results of accuracy

| | PHE | | | PAR | | | TPL | | |
|---|---|---|---|---|---|---|---|---|---|
| | Levels | | | Levels | | | Levels | | |
| | 80% | 100% | 120% | 80% | 100% | 120% | 80% | 100% | 120% |
| Amt added (µg/ml) | 4 | 5 | 6 | 260 | 325 | 390 | 2 | 2.5 | 3 |
| | 4 | 5 | 6 | 260 | 325 | 390 | 2 | 2.5 | 3 |
| | 4 | 5 | 6 | 260 | 325 | 390 | 2 | 2.5 | 3 |
| Amt recovered (µg/ml) | 3.94 | 4.93 | 5.66 | 243.62 | 314.66 | 382.58 | 1.91 | 2.49 | 2.98 |
| | 3.91 | 4.77 | 5.84 | 248.52 | 313.52 | 375.73 | 1.9 | 2.44 | 2.92 |
| | 3.85 | 4.89 | 5.9 | 243..40 | 306.8 | 384.95 | 1.87 | 2.5 | 2.98 |
| % Recovery | 98.46 | 98.59 | 94.3 | 93.7 | 96.82 | 98.1 | 95.47 | 99.66 | 99.23 |
| | 97.87 | 95.38 | 97.39 | 95.58 | 96.47 | 96.34 | 95.23 | 97.53 | 97.45 |
| | 96.19 | 97.75 | 98.39 | 93.61 | 94.4 | 98.7 | 93.28 | 99.85 | 99.38 |
| Mean recovery | 97.51 | 97.24 | 96.69 | 94.3 | 95.9 | 97.71 | 94.66 | 99.02 | 98.69 |
| % RSD | 1.21 | 1.71 | 2 | 1.18 | 1.36 | 1.26 | 1.27 | 1.3 | 1.09 |

*Acceptance criteria:*

1) The % RSD for the triplicate at each spike level shall be NMT 2.0.

2) The overall % RSD for % recovery for all spike levels shall be NMT 2.0.

3) The % recovery at each spike level shall be NLT 94.0 and NMT 102.0 of the

added amount.

*Robustness*:

Robustness of the proposed analytical method was evaluated by making deliberate changes in the chromatographic system method parameters, the standard solution and test solutions were injected for each of the changes made to access the Robustness of proposed analytical method.

*Following Parameters Were Covered Under Robustness Parameter.*

1. Effect of Variation in flow rate of mobile phase by ± 10%:

2. Change in mobile phase pH by ± 0.1 units:

Table no. 7: results of robustness

| Sr. No. | System Suitability parameter | | Observations for flow rate | | | Limits |
|---|---|---|---|---|---|---|
| | | | Unchanged | 0.9 ml | 1.1 ml | |
| 1 | The % RSD of peak area response for five replicate injections | PHE | 1.37 | 0.19 | 0.51 | NMT 2.0 |
| | | PAR | 1.25 | 0.24 | 0.12 | |
| | | TPL | 1.71 | 0.64 | 0.64 | |
| 2 | Theoretical plates | PHE | 151592 | 117880 | 176612 | NLT 2000 |
| | | PAR | 102295 | 92604 | 12435 | |
| | | TPL | 54520 | 50824 | 57750 | |
| 3 | Tailing factor | PHE | 0.6 | 0.92 | 0.8 | NMT 2.0 |
| | | PAR | 0.8 | 0.99 | 1.0 | |
| | | TPL | 1.6 | 1.9 | 1.4 | |
| 4 | Retention Time (Min) | PHE | 1.493 | 1.675 | 1.394 | |
| | | PAR | 3.685 | 4.097 | 3.422 | |
| | | TPL | 14.058 | 15.694 | 14.193 | |

| Sr. No. | System Suitability parameter | | Observations for pH | | | Limits |
|---|---|---|---|---|---|---|
| | | | Unchanged | 2.7 | 2.9 | |
| 1 | The % RSD of peak area response for five replicate injections | PHE | 1.37 | 0.55 | 0.56 | NMT 2.0 |
| | | PAR | 1.25 | 0.40 | 0.67 | |
| | | TPL | 1.71 | 0.84 | 0.82 | |
| 2 | Theoretical plates | PHE | 151592 | 117880 | 176612 | NLT 2000 |
| | | PAR | 102295 | 92604 | 12435 | |
| | | TPL | 54520 | 50824 | 57750 | |
| 3 | Tailing factor | PHE | 0.6 | 0.92 | 0.8 | NMT 2.0 |
| | | PAR | 0.8 | 0.99 | 1.0 | |
| | | TPL | 1.6 | 1.9 | 1.4 | |
| 4 | Retention Time (Min) | PHE | 1.493 | 1.493 | 1.484 | |
| | | PAR | 3.685 | 3.685 | 3.042 | |

| | | | | | | |
|---|---|---|---|---|---|---|
| | | TPL | 14.058 | 14.058 | 14.968 | |

*Acceptance Criterion*:

1. The % RSD of peak area response for five triplicate injection level shall be NMT 2.0
2. The therotical plate shall be NMT 2000
3. The tailing factor shall be NMT 2.0
   All the system suitability parameters shall pass.

Recovery results were well within the acceptance criterion of system suitability parameters. Thus the method was found to be accurate.

*Specificity*:

Is the ability to assess unequivocally the analyte in the presence of impurities, degrades, matrix etc. It is evaluated by injecting the blank, placebo and the control sample solution prepared as per the proposed method to check for the interference if any peak at the retention time of Paracetamol, Pheneyleprine HCl and Triprolidine HCl. Thus no interference was found at the Retention of, Pheneyleprine HCl, Paracetamol and Triprolidine HCl which is **1.493 , 3.685 and 14.058** mins. respectively.

*Application of Hplc Method For Analysis Of Marketed Formulation*:

Table No.8

| Sr.no. | Formulation | % Label claim | | |
|---|---|---|---|---|
| | | PHE | PAR | TPL |
| 1 | Recofast plus (Tablet) | 109.565 | 101.04 | 101.74 |
| 2 | Recofast plus (Syrup) | 102.633 | 100.88 | 101.41 |

## IV. SUMMARY AND CONCLUSION

- Combined dose tablet formulation containing Paracetamol, Phenylephrine HCl and Triprolidine HCl is available in solid dosage form and liquid dosage form for the treatment of cold.
- Multicomponent formulations are gaining precedence over single component formulations owing to the following reasons:
  - ✓ Synergism of effects.
  - ✓ Reduction of cost of treatment
  - ✓ Increased patient compliance.
- Due to this rise in the multicomponent formulations, the challenges faced by the analytical chemist are on the rise. Estimation of drugs from a multi-component formulation requires a method capable of discriminating the more than two components. Approaches to multi component analysis can be broadly categorized into those which rely on physical separation of components prior to analysis (e.g. chromatographic methods).

The present work involved the development of accurate, precise, and simple suitable RP-HPLC method for estimation of the drugs in multicomponent tablet formulations.

## V. FUTURE SCOPE

- Paracetamol, Phenylephrine HCl and Triprolidine HCl is a new combination recommended as Anti-cold.
- Paracetamol, Phenylephrine HCl and Triprolidine HCl can be estimated by HPLC method by developing the mobile phase composition.
- Comparative estimation of different brands can be conducted.
- The developed method can be applied for estimation of the proposed drugs in bio- fluids for pharmacokinetic and studies associated with metabolites.
- This method can be used for the estimation of given drugs in different dosage forms such as capsules, micro pellets etc.
- The method can be further extended for studies such as degradation and related substances experiments.

The method can be further extended for Dissolution studies.

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