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Original Research Article

Determination of the synthesised jelly by UV method development and validation of ramipril

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ABSTRACT

Chalcone is an α,β- unsaturated ketone and central core for a variety of important biological compounds extracted from natural products and the experiment is conducted by performing identification tests to identify the chemical components in the compounds such as *prunusamygdalusbatsch shells* and *saccharum officinarum* are the main components used in this procedure. The synthesized jelly is developed and validated by ramipril and its linearity is found to be 10-50 μg/ml, precision % RSD of in intraday 35.7142 and in inter day 35.7277, Accuracy and recovery studies 80 %, 100%,120% are found to be 82,95,103,. Specificity % RSD is 29.43, Robustness wavelengths are taken as 210 nm, 211nm,212 nm found to be 3.345,26.182,21.038, LOD – 0.8μg/ml, LOQ - 2.5 μg/ml. The amount present in chalcone jelly in given sample of ramipril was 81μg/ml.

1. Introduction

Bitter almond kernel shell (Prunusamygdalusbatsch shell):

Bitter Almond shells correspond to the hard, lignified endocarp that is removed to obtain the kernels. Almond shells represent 33% of the total fresh weight of the almond fruit.[1] Almond is the general name of Prunus dulcis or *Prunusamygdalus L.* Almond fruit consists of the hull, shell, and kernel (nut).It has been estimated that the annual output in north China is up to 40 thousand tons, and the amount has tended to increase due to a strong demand for the fruit. During the harvest period, the hull and kernel are the desirable products; the almond shell consists of 70% of the dry mass of a whole almond fruit.[2]

1.1. Jaggery powder (Saccharum officinarum L.):

Gur (Jaggery) is a natural, traditional sweetener made by the concentration of sugarcane juice and is known all over the world in different local names. It is a traditional unrefined non-centrifugal sugar consumed in Asia, Africa, Latin America and the Caribbean. Containing all the minerals and vitamins present in sugarcane juice, it is known as healthiest sugar in the world.[3] In India, jaggery making is one of the ancient agro-processing industries that prepares 70% of the total jaggery produced in the world to different countries.[4]

2. Materials and Methods

2.1. Preparation of jelly

1. 6 gm of glucose syrup and 4 ml of water were mixed and heated in a brass pan add 3.5 gm gelatin solution was prepared by dissolving the gelatin in warm water.
2. Gently mix into mixture and prevent from burning,

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Figure 1:

Figure 2:

3. When temperature raised to 116, 0.8 gm of citric acid was added and mixed.
4. Straining of extract and addition of sugar.
5. Judging of endpoint.
6. Foam/ Scum is removed from mixture
7. Fill the hot into clean sterilized bottles
8. Capping is done for mixture.
9. Stored at ambient temaparature.

3. Method Validation

3.1. Preparation of solutions

Standard stock preparation

Figure 3:

First 5 ml of Ramipril is mixed with 10 ml water. Then transfer it into a separating Funnel. Shake it vigorously up to 10-15mins. Then separate the organic layer and again add 10 ml of water, shake it and separate the organic layer from and continue same for 3 times. After 3 shakings separate the organic layer. And by using the pipette take 0.05 ml of solution and mix with 10 ml of water.

3.2. Validation parameters

The method was validated according to ICH 028 guidelines.

Method validation: The proposed method was validated for different parameters like linearity, precision, accuracy, specificity, robustness, LOD, LOQ and assay.

Linearity Study: The linearity was determined by plotting concentration against corresponding absorbance. Standard stock solutions, 1000μg/mL were further diluted with the diluent to obtain 10 μg/mL-50 μg/mL solutions. The calibration curves were constructed by plotting absorbance versus concentration and the regression equations were calculated.

Table 1: Calibration curve data of Ramipril

S. No	Concentration(μg/ml)	Absorbance
1	0	0
2	10	0.2444
3	20	0.4146
4	30	0.5979
5	40	0.7603
6	50	0.9375

3.3. Precision

Intra-day precision study: Test sample was diluted further to obtain 10-50 ug/ml concentration. Six replicates were measured and the percentage RSD was calculated.

Inter-day precision study: The selected concentrations for the intra-day precision study were again analysed

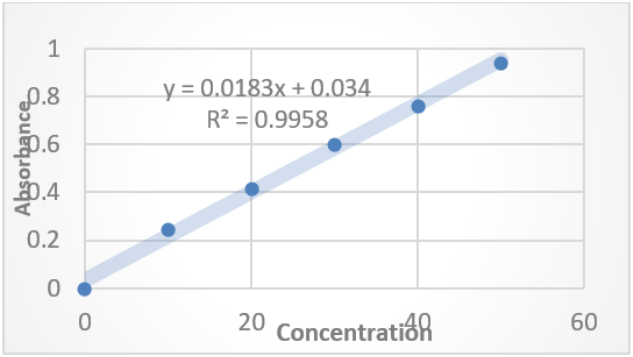

Figure 4: Calibration curve of ramipril

for consecutive three days and the percentage RSD was calculated.

Table 2: Intraday nd Inter day data of ramipri

Sample No	Intraday precision	Inter day precision
1	0.030	0.271
2	0.036	0.543
3	0.044	0.619
4	0.048	0.820
5	0.080	0.921
Mean	0.0476	0.6348
SD	0.017	0.2268
%RSD	35.7142	35.7277

Accuracy and Recovery Studies: Accuracy of the method was calculated by recovery studies at three different levels (80%, 100% and 120%) by standard addition method to study the accuracy of the method and to check the interference from excipients. The first recovery study was conducted on the excipients mixture (placebo) prepared by adding accurately weighed amount of extract to the excipient mixture and calculating the percentage recovery in each case.

Table 3: Results of recovery study

%Recovery level	% Recovery	Mean % Recovery	SD	% RSD
80%	89.47 88.44 87.69	88.53	0.729	82
100%	85.64 85.97 86.12	85.9	0.82	95
120%	89.32 87.75 89.93	89	0.918	103

Specificity in the presence of excipients: The specificity test was carried out using only excipients. Spectra for blank and sample were measured for different time intervals and compared.

Table 4: Results of specificity

Time	Standard	Sample
0	0.003	0.007
2	0.005	0.009
4	0.008	0.014
6	0.011	0.017
Mean	0.005	0.010
SD	0.002	0.002
%RSD	41.09	29.43

Robustness: The robustness of an analytical products interfered with the quantification of the drug. Procedure is the measure of its capacity to remain unaffected by small but deliberate variations in method parameters and provides an indication of its reliability during normal usage. It was determined by carrying out the analysis by at different wavelengths i.e. at 265nm, 266 nm and 267 nm. The absorbance was measured and assay was calculated for six times.

Table 5: Results of robustness

S.No	Wave lengths		
	210nm	211nm	212nm
1	2.147	1.800	1.539
2	2.008	1.053	1.004
Mean	2.0775	1.4265	1.2715
SD	0.0695	0.3735	0.2675
%RSD	3.345	26.182	21.038

4. Limit of Detection (LOD) and Limit of Quantitation (LOQ):

LOD and LOQ were calculated from the data obtained from the linearity studies. The slope of the linearity plot was determined. For each of the ten replicate determinations of same concentration, standard deviation (SD) of the responses was calculated. Limit of detection can be calculated by using the following formula:

LOD = 3.3 σ /S= 0.8μg /ml

Limit of quantitation can be calculated based on standard deviation of the response and the slope.

LOQ = 10 σ/S= 2.5μg /ml

Where σ = Standard deviation of the response; S = Slope of the calibration curve.

5. Assay of Prunus Amygdalus Batsch Shell, Saccharum Officinarum Extract:

To analyze the concentration of (prunusamygdalusbatsch shell, saccharum officinarum) in the vial, a portion of powder equivalent to 10mg of prunusamygdalusbatsch shell, saccharum officinarum extract was transferred in 100ml volumetric flask and was diluted with water This solution was further diluted with water to get final concentration of 10μg/mL of prunusamygdalusbatsch shell, saccharum officinarum The % assay of the drug was calculated. All determinations were conducted by thrice time.

Table 6: Results of assay

Drug	Declared Concentration(ug/ml)	Amount found Concentration (ug/ml)	Amount found (%)
Sample 1	10	8.1±0.08	81

6. Results and Discussion

The synthesized jelly is developed and validated by ramipril and its linearity is found to be 10-50 μg/ml, precision % RSD of in intraday 35.7142 and in inter day 35.7277, Accuracy and recovery studies 80 %, 100% ,120% are found to be 82,95,103,. Specificity % RSD is 29.43, Robustness wavelengths are taken as 210 nm, 211nm, 212 nm found to be 3.345, 26.182, 21.038, LOD – 0.8μg/ml, LOQ - 2.5 μg/ml, Assay was found to be 81%. The amount of ramipril was found to be 81μg/ml.

7. Conclusion

Determination of the synthesized product by method development and validation with ramipril is linear, precise, accurate and specific. The amount present in chalcone jelly in given sample of ramipril was 81μg/ml. And hence the synthesized jelly is novel, simple, economical, and hence proved by its interpretation and with validation method the amount was determined in Ramipril.

8. Acknowledgment

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9. Source of Funding

None.

10. Conflict of Interest

None.

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