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FORMULATION AND EVALUATION OF POLYHERBAL TABLETS FOR MANAGEMENT OF OSTEOPOROSIS

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ABSTRACT

Background: Formulation, standardization and evaluation of a polyherbal tablet for managing osteoporosis, was the goal of this study.**Methods:** Aqueous root extract powders were used to produce polyherbal tablets by wet granulation method of selected plant *Nigella sativa*, *Gingiberofficinale*, *Curcuma longa, Termania chebula* and *Kukkutantatwak bhasma with the help of* calcium phosphate dibasic, calcium carbonate, Starch, Mg stearate, Gum acacia in different concentration **Results:** The powder mixtures were found to possess good flow properties based on the angle of repose, Carr's index, and Hausners ratio. As for the uniformity in weight, hardness, drug content, and friability, the powder mixtures were found to comply with the standards as specified in IP. The PHT-4 kept for stability studies and observed that it was reproducible even on stored for three months.**Conclusion**: In order to cure bone fracture, osteoporosis and calcium deficiency related problems, this polyherbal formulation from Nigella sativa, Gingiber officinale, Curcuma longa, Termania chebula and Kukkutantatwak bhasma balances hormonal imbalance and increases the rate of ossification of bone.

Keywords: Polyherbal tablet; Osteoporosis; wet granulation; stability studies; drug content.

1. INTRODUCTION

Nowadays, the only way to survive a long and healthy life is through traditional medicine. Ayurveda is perhaps the most ideal system due to it have a few normal components to dispose of the basic reasons for the sickness, restoring health by restoring harmony and preventing further decay[1]. WHO estimates that around 80% of the world's populations actually trust conventional or Ayurvedic drugs for their healthy lives [2].There is an Indian philosophy behind Ayurveda that is dedicated to preventing unnecessary suffering of endurance when curing human illnesses, along with the fact that about 45,000 natural plant species exist, of which some 15,000 are therapeutic plants have been chronicled to dismissing diverse human ailments by consuming single or numerous spices for the entiretermination of infection [3].

The blend of different spices (polyherbal) in a specific proportion will give a positive helpful impact in light of the fact that individual plants lack the powerful phytochemical constituents to achieve their beneficial effects [4, 5]. In general, polyherbal details have been able to deliver favorable results during the administration of human illnesses containing various phytoconstituents with comparative or divergent useful potential [6, 7]. The ubiquity of the polyherbal definition is exceptional on account of their wide remedial reach i.e., powerful at a low portion and protected at high portion, however creates less side results while abused [8, 9].

Ayurvedic medical treatment uses a variety of parts, such as seeds, roots, bark, stems, gum, leaves, blossoms, organic product etc and so on of the plants [10, 11]. The purpose of the current investigation was to define and assess new polyherbal tablets containing watery root tinctures of some chosen plants with active phytoconstituents and described in experimental evidence for the treatment of Glomerulonephritis [12]. The forename of these elected plants for the review viz. Nigella sativa (Ranunculaceae), Terminalia chebula Retz (Combretaceae), Zinger (Zingiberacae), Curcuma longa L(Zingiberaceae)., KukkutandaTvak, Bhasma A medication resistance study and a harmfulness report determined the number of medications used in the detailed account.This definition uses equivalent amounts of concentrate powder, 25 mg, for preparing polyherbal dispersible tablets as active constituents [13, 14].

2. MATERIALSANDMETHODS

2.1 Collection and Authentication

Nigella sativa, *Gingiber. officinale*, *Curcuma longa, Termania chebula* were purchased from the Local market,Kala Amb, H.P. (India) and *Kukkutandatvak bhasma* were purchased from Regional & Unani Pharmacy, Dehradun, Uttarakhand. Other required components such as starch, magnesium stearate and talc were purchased from Research-Lab. Industries, India and were of analytical grade.The Department of Botany, Punjab University, Chandigarh, has authenticated fresh and shade dried roots of all selected plants.

2.2 Preparation of Extract

The polyherbal plant's roots were washed, dried in the shade, and then crushed with a mechanical grinder. The powdered roots of each plant were put into distilled water

until they were completely depleted. Separate extracts were filtered using Whatman filter paper, concentrated on a rotary evaporator at a suitable temperature (40°C), and dried in a freeze drier. The finished dry powder was kept in a closed container at a cold temperature, and the percentage yield was determined to be between 20 and 25 percent (w/w) [15]. Following that, numerous physicochemical characteristics of different powder plants were determined, such as Ash value, Acid insoluble ash, Water-soluble extractive value, Moisture Content, and Drying Loss [16, 17]. Bhasma and herbs contain calcium, magnesium, phosphate, potassium, chloride, nitrate and sulphate as inorganic constituents. Trace material or elements and analysis can be used as a standardization tool of the plant material. Heavy metals were analyzed using an atomic absorption spectrometer (Anallytic Jena) [18].

2.3 Development of Polyherbal Tablets [19, 20]

Polyherbal tablets prepared by using wet granulation technique. All the herbal ingredients were properly mixed with bhasma in a mortar. This was followed by subsequent addition of starch and calcium phosphate dibasic and calcium carbonate (Table 1). After proper mixing of all the ingredients, sufficient quantity of distilled water was added to form a lumpy mass which was then passed through sieve No. 10 to form granules.

The granules were dried in the oven at 90°C for 5 h. The dried lumps were passed through sieve no. 12 to get appropriate granules. The granules were thoroughly mixed with magnesium stearate and compressed using 10 mm punch into a 500 mg tablet contains a number of herbs and mineral constituents using a single rotary punching machine).

Table No.1 Composition of Polyherbal Tablet (mg)

S.No.	Ingredients	PHT1	PHT2	PHT3	PHT4	PHT5	PHT6
1	*Nigella sativa,*	60	60	60	60	60	60
2	*Gingiber officinale,*	60	60	60	60	60	60
3	*Curcuma longa,*	50	50	50	50	50	50
4	*Termania chebula*	60	60	60	60	60	60
5	*Kukkutandatvak bhasma*	50	50	50	50	50	50
6	calcium phosphate dibasic	70	65	75	70	70	75
7	calcium carbonate	60	65	60	60	65	60
8	Starch	80	75	75	80	75	75
9	Mg stearate	5	5	5	5	5	5
10	Gum acacia	5	5	5	5	5	5
11	Net	500	500	500	500	500	500

2.4 Pre-Compression Studies of Powder Blend

The biologically potent polyherbal extract powder with a stand forvariedphysicalpropertiesandmicromeriticproperties.Extractpowdered are heterogeneous because it was composed of individual particles of different sizes and shapes randomly interspersed with air spaces and becomes more complicated with polyherbal. The preformulation study including phytochemical analysis, Bulk density, tapped density hausner's ratio Porosity, angle of repose and Carr's compressibility index were performed [21, 22, 23]. Organoleptic properties like colour, odour and taste of drug were observed.

2.5 Evaluation of the Formulated Tablets [24, 25, 26]

Prepared polyherbal tablets were evaluated for various parameters.

- ***Physical appearance***

The general appearance of tablet was studies visually in shape, color, texture and odour.

- ***Thickness***

The tablet thickness was calculated by Vernier calipers. The tablet was put in between two jaws vertically and measured thickness and 6 tablets were used for this test and expressed in mm.

- ***Weight variation***

Randomly selected 20 tablets were weighed individually, calculating the average weight and comparing individual tablet weight to the average. The weight variation test would be a satisfactory

method of determining the drug content uniformity of tablets.

- ***Hardness***
  The tablet hardness was determined by the Monsanto hardness tester. The tablet was placed lengthwise between upper and lower plunger and force applied by turning a threaded bolt until the tablet fractures and measured hardness of tablet in kg/cm^2.
- ***Friability***
  It is determined by Roche friabilator, subjects a number of tablets to the combined effects of abrasion and shock by utilizing a plastic chamber that revolves at 25 rpm, dropping tablet from inches distance operated for 100 revolutions. The pre weighed tablets were dusted and reweighed and according to standard limit friability should be <1%. It is calculated by formula;
  % Friability= (Initial weight-final weight/initial weight)*100
- ***Disintegration time***
  Disintegration test was performed according to the Indian Pharmacopoeia specification. Six tablets were taken for the test and water was as the disintegration medium. The temperature of the medium was kept at 37°C, the beakers were filled at a volume of 800 ml and care was taken that the tablets were always below the level of the water at the highest and lowest position of basket-rack assembly. The discs were introduced over each tablet to avoid their floating of the tablets in the medium. The apparatus was operated until all the tablets were disintegrated.
- **Preparation of tablet samples for calcium analysis with the ethanol-water solvent**
  About 0.5 g of the dried finely ground tablet material (ca. 30–60 mesh) was refluxed for 15–30 min in a round bottomed flask with about 35 ml of 20% v/v ethanol-water mixture. The resulting solution of activated carbon and filtered directly into a 50 ml volumetric flask through Whatman No. 42 filter paper. The filter was washed with the solvent and the filtrate was received in the same volumetric flask. The volume of the combined filtrates was completed with the same ethanol-water mixture. With these solutions, adequate aliquots (10.0 ml) were taken and diluted to 50.0 ml to determine calcium using calibration curves [27].
- **Content uniformity for calcium**
  A stock solution of calcium standard (1000 mg/l) was prepared by dissolving 2.4973 g of dry CaCO3 in 200 ml of distilled water containing 5 ml of concentrated HCl. The solution was heated to drive out CO2 and after cooling, it was made up to 1000 ml. The stock was diluted to produce working standards of concentration range of 1–100 µg/ml. Lanthanum (0.1%) was used in these solutions, and the same procedure was used in plant samples, to avoid interference of phosphorus. 1.0 ml of the ethanol-water mixture was added to the final solutions. The AAS (Atomic Absorption Spectroscopy) was set at the operating wavelength of calcium. A Carl Zeiss FMD/PMQ3 atomic absorption spectrophotometer with a single element hollow cathode lamp, a 10 cm universal burner and acetylene air flame, was used for measurements of calcium at 422.7 nm. Each working standard was run in the emission spectrometer and the intensity for each standard was recorded [27]. The

calibration curve was constructed to determine the concentration of calcium in the tablets sample. The blanks were also determined in the same way and subtraction done where necessary.

Calculations: Ca (mg/tab) = (a-b) × V × f × 1000/1000× w

Where: a=Concentration of Ca in the sample extract, b=Concentration of element in the blank extract, *V*=volume of the extract solution, w=weight of the sample f=dilution factor.

2.5 Stability studies [28, 29]

Accelerated stability study was carried out as per ICH guidelines for polyherbal combination to check the physical, chemical and physiological property of prepared formulation in a short period. The optimized PHF was subjected to accelerated stability studies at three different conditions of temperature and relative humidity i.e., at 25 °C/60% RH, 30 °C/65% RH and 40 °C/75% RH for a period of 3 months. After each month tablet sample was analyzed for physical characteristics and disintegration time.

Results And Discussion

The present investigation was undertaken to design, formulate and evaluate a

polyherbal tablet. Polyherbal tablets were compressed each of 500 mg weight on a 10-station Mini Pres

s-I rotary tablet compression machine fitted with 12 mm punches size. None tablet manufacturing defects like capping, lamination, and chipping were observed.

3.1 Physicochemical Properties, Phytochemical Screening, Inorganic Constituents of Different Plant Powders

All the parameters regarding Physicochemical properties of different plant powders (loss on drying at 150°C, water-soluble matter, total ash and acid insoluble ash), traces of **inorganic constituents** (Aluminium, Chloride, Copper, Calcium, Carbonates, bicarbonates, Magnesium etc) shown in Table No. 2 & 4. The results of phytochemical screening of different plant extracts are shown in Table No. 3.

Table:2 Physicochemical properties of different plant powders

Powdered drug	Physicochemical Parameters				
	Total Ash (% w/w)	Water soluble ash (% w/w)	Acid insoluble ash (% w/w)	Moisture content (% w/w)	Loss on drying (% w/w)
Nigella sativa	6.12	2.4	0.9	5.12	0.42
Gingiber officinale	3.4	1.4	1.4	3.4	3.4
Curcuma longa,	4.2	1.3	0.9	2.0	1.4
Termania chebula	4.7	1.1	0.22	8.4	1.1
Kukkutandatvak bhasma	4.4	3.51	4.5	0.2	0.60

Table:3 Phytochemical Screening of Different Plant Extract

Plant constituents	**Extract**			
	Nigella sativa	***Gingiber officinale***	***Curcuma longa***	***Termania chebula***
Alkaloids	+	+	+	+
Glycosides	+	+	+	+
Phenols	+	+	++	+
Resins	-	-	+	-
Saponins	-	-	+	+
Sterioids	-	-	++	+
Tannins	-	+	+	+
Terpenoids	-	+	+	+
Water soluble extractive value	28	12	13	25
Alcohol soluble extractive value	32	16	19	30

Table 4: Qualitative test for inorganic constituents					
Elements	***Nigella sativa***	***Gingiber officinale***	***Curcuma longa***	***Termania chebula***	***Kukkutandatvak bhasma***
Aluminum	-	-	-	-	-
Chloride	-	+	-		+
Copper	-	-	-	+	-
Calcium	-	-	+	-	+
Carbonate,bicarbonate	+	-	-	+	-
Magnesium	-	-	+	-	-
Nitrate	-	-	-	-	-
Phosphate	+	+	-	-	+
Potassium	-	+	-	-	-
Sodium	-	+	-	-	-
Zinc	+	-	-	-	-

Physicochemical properties of different plant powders were checked. It was observed that total ash value was found to be very high in ***Nigella sativa*** and ***Termania chebula***. Ash values are the criteria to judge the identity and purity of crude drugs, where total, water soluble and acid insoluble ashes are considered. All the parameters were found to comply with the standards. All the components were found to be within specified limits

3.2 Characterization of Powder

The prepared granule was evaluated for angle of repose, characterizes the flow properties and is a characteristic related to inter particulate friction resistance to movement between particles. The basic characterization of powder and micromeritic properties of formulations containing polyherbal aqueous extracts powder used for preparing tablets mentioned in Table 5.

Table 5: Micromeritic parameters of polyherbal aqueous root extracts powder

Formulation Code	Bulk density (gm/ml)	Tapped density (gm/ml)	% Compressibility	Hausner Ratio	Angle of Repose (°)
PHT1	0.36±0.02	0.42 ±0.01	14.28	1.39 ± 0.02	24.12±0.13
PHT2	0.41±0.01	0.47 ±0.02	12.76	1.25 ±0.03	22.62±0.01
PHT3	0.45±0.02	0.60 ±0.03	25	1.31±0.04	26.56±0.31
PHT4	0.44±0.01	0.57 ±0.01	22.80	1.29 ±0.03	30.05±0.62
PHT5	0.42±0.01	0.54 ±0.03	22.2	1.28±0.02	26.56±0.31
PHT6	0.46±0.01	0.53 ±0.01	13	1.15 ±0.01	23.06±0.12

Table 5. Shows that the physical properties of the granules, like bulk density, tapped density angle of repose, % Compressibility and Hausner's ratio, were found to be within limits, which shows a good flowability of the granules. Tablets were evaluated for various parameters such as color, average weight, hardness, friability and disintegration time, which were found to be acceptable as per the Pharmacopoeial specifications. All the polyherbal tablets passed the weight variation test as the average percentage weight variation was within the USP limits of ±5%. The weight variation of tablet causes variation of active medicament which changes the bioavailability. The hardness of the tablets was found to be between 6.2±0.050 and 8.50±0.025 kg/cm^2. The friability of the tablet was found to be below 1%, indicating a good mechanical resistance, and the disintegration time of all the batches was found to lie in the range of 22±0.20–38±0.20 min. given in table 6.

Table 6: STANDARDIZATION PARAMETERS FOR POLYHERBAL TABLET

Evaluation Parameters	Formulation Code					
	PHT 1	PHT 2	PHT 3	PHT 4	PHT 5	PHT 6
Color	Yellowish brown	Yellowish brown	Yellowish brown	Yellowish brown	Yellowish brown	Yellowish brown
Odor	Characteristic	Characteristic	Characteristic	Characteristic	Characteristic	Characteristic
Texture	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth
pH	7.9	8.1	7.8	8.4	8.1	8.0
Diameter (mm)	10.55 mm ± 0.25 mm	10.67mm ± 0.28 mm	10.61mm ± 0.31 mm	10.78mm ± 0.25 mm	10.78mm± 0.30	10.70mm± 0.30
Thickness (mm)	6 mm±	6.4 mm±	6.7mm±	6.5 mm±	6.4mm±	6.7mm ±
Weight Variation (%)	1.77 ± 0.004	1.8 ± 0.007	1.041 ± 0.005	1.21 ± 0.004	1.04 ± 0.005	1.47 ± 0.004
Average weight (mg ±sd)	513 mg± 0.60	525mg± 0.40	512 mg± 0.70	528 mg± 0.20	502mg±0.20	505mg±0.30
Hardness (Kg/cm^2)	8.5 Kg/cm^2± 0.25	6.9 Kg/cm^2± 0.40	6.7 Kg/cm^2± 0.20	6.2 Kg/cm^2± 0.30	6.4 Kg/cm^2± 0.10	6.2 Kg/cm^2± 0.50
Friability (%)	0.140 % ± 0.20	0.158%±0 .25	0.21 % ± 0.40	0.12 % ± 0.50	0.2% ± 0.40	0.24 % ± 0.40
Disintegration Time (Minutes)	28min±0.3 0	22 min± 0.40	38 min ± 0.20	24 min ± 0.40	30min±0.50	27min±0.50

3.3 Quantitative Estimation of Calcium by Atomic Absorbance Spectrophotometry

The calibration curve for quantitative estimation of calcium by atomic

absorption spectrophotometer is shown in Fig.1. The quantitative estimation of calcium by atomic absorbance spectrophotometry was carried out using ethanol-water solvent extraction method and found 51.27±0.85, 60.0±1.18, 63.06±1.74, 69.27±0.49, 72.17±0.79, and 76.07±0.48mg/tab of elemental calcium in PHT1, PHT2, PHT3, PHT4, PHT5 and PHT6 respectively.

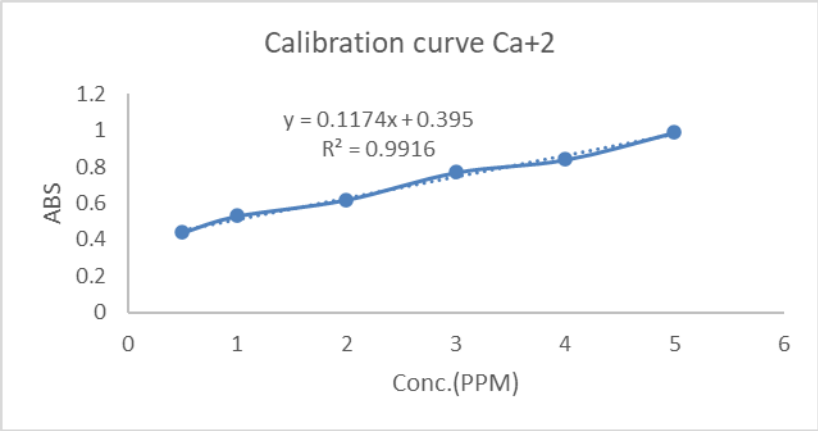

Figure 6.1: Calibration Curve Ca^{+2}

3.4 Stability Study data

The stability of herbal products depends on the various factors such as stability of herbal ingredients, manufacturing processes, the reaction between active herbal ingredients and excipients, containers, environmental conditions encountered during storage and microbe's etc. Accelerated stability testing based on the single condition of elevated temperature and humidity is more appropriate and suitable for herbal products because of their very basic nature. The optimized batch (4) was subjected to the accelerated storage condition **(25 °C 60% RH), (30°C 65% RH)** (40°C/75% RH) for 6 months according to ICH guideline. The results shows stable with no significant changes in the physio-chemical properties of the tablets

Table 7 : Stability study data for polyherbal tablet

TIME	% DRUG CONTENT AT DIFFERENT STORAGE CONDITIONS		
	25 °C 60% RH	**30°C 65% RH**	**40°C 75 % RH**
30 DAYS	99.1	99	99.5
60 DAYS	98.7	98.4	98.7
90 DAYS	97	98.1	97.2

3. CONCLUSION

From the above study, we conclude that the polyherbal tablets were prepared by the wet granulation method and gave satisfactory and acceptable results. The results from the angle of repose, Carr's index and Hausner's ratio showed that the powder mixtures possess good flow properties. The study revealed that the composition ratio of ingredients of polyherbal tablets, not affect the stability parameters. The physical properties of PHT-1 to PHT-6 were determined for the uniformity in weight, hardness, drug content and friability which have complied with the official requirements, and comply with the official limits mentioned in IP. The PHT-4 kept for stability studies and observed that it was reproducible even on stored for three months

From this study it is concluded that using traditional knowledge and the recent technologies, the medicinal plants can be prepared in the form of cost effective tablet formulations to improve their stability, consumer compliance and acceptability. However, further research is needed to study their activity clinically and to study their precise mechanism of action and efficacy with long term use as a calcium supplement to rectify the calcium deficiency.

4. REFERENCES

1. Hankey A. Ayurveda and the battle against chronic disease: anopportunity for ayurveda to go mainstream? J Ayurveda IntegMed2010;1:9–12.
2. ChaudharyA,SinghN.Contributionofworldhealthorganization in the global acceptance of ayurveda. J AyurvedaIntegMed 2011;2:179–86.
3. Parasuraman S, Thing GS, Dhanaraj SA. Polyherbal formulation:conceptofayurveda.PharmacognRev2014;8:73–80.
4. Pandey MM, Rastogi S, Rawat AKS. Indian traditional ayurvedicsystem of medicine and nutritional supplementation.J EvidenceBased Complementary Altern Med2013:1–12.http://dx.doi.org/10.1155/2013/376327
5. Garg V, Dhar VJ, Sharma A, Dutt R. Facts about standardizationofherbalmedicine:a review. Zhong Xi Yi Jie He XueBao2012;10:1077-83.
6. AwasthiH,ManiD,NathR,NischalA,UsmanK,KhattriS.Standardization, preparation and evaluation of an Ayurvedicpolyherbal formulation in a capsule dosage form suitable foruseinclinicaltrials.IndoAmJPharmRes2014;4:4093-9.
7. MathewL,BabuS.PhytotherapyinIndia:thetransitionoftraditiontotechnology.CurrBot2011;2:17–22.
8. Srivastava S, Lal VK, Pant KK. Polyherbal formulations based onIndianmedicinalplantsasantidiabeticphytotherapeutics.Phytopharmacology 2012;2:1-15.
9. KapoorVK,SinglaS.Herb-druginteractions–anupdateonsynergisticinteractions.J AltMedRes2015;1:1-11.
10. GurleyBJ.Pharmacokineticherb-

druginteractions(Part1):Origins,mechanisms ,andtheimpactofbotanicaldietarysupplem ents. PlantaMed2012;78:1478–89.

11. Rawat R, Vashistha DP. Common herbal plant in Uttarakhand,used in the popular medicinal preparation in ayurveda. Int JPharmacognPhytochemRes2011;3:64–73.
12. Kulkarni,RaghavendraG.Extractionofr ootsofquinticsbydivisionmethod. IntJMathemEduSciTech2009;40:407–10.
13. Rajab NA. Preparation and *in vitro* evaluation of lacidipine oralliquid-solid tablet as an approach of solubility and dissolutionrateenhancement.IntJApplPhar m2018;10:145-53.
14. ParfatiN,RaniKC,CharlesN,GeovannyV .Preparationandevaluationofatenolol-β-cyclodextrinorallydisintegratingtablets using co-process crospovidone-sodium starch glycolate.Int JAppl Pharm2018;10:190-94.
15. *Tirath et al.* Formulation, Standardization, and Evaluation of Polyherbal Dispersible Tablet, *Int J App Pharm, Vol 11, Issue 1, 2019, 158-167*
16. The Ayurvedic Pharmacopoeia of India, 1st ed. Part-I, Vol-3. New Delhi: Government of India, Ministry of health and family welfare, Department of Indian system of medicine and homoeopathy. 2001. p.123-6.
17. Anonymous, The Ayurvedic herbal Pharmacopoeia of India, 1st ed. Part-1, Vol-2. Ministry of Health and Family Welfare 2001. p.162-71.
18. Gupta AK. Coordinator Quality Standards of Indian Medicinal Plant. Vol-4. Published by Indian Council of Medicinal Research, Ansari nager, New Delhi. India 2006. p. 205-11.
19. *Mulchand and Seema* , Formulation Of Polyherbomineral Matrices For Treatment of Osteoporosis, *Asian J Pharm Clin Res, Vol 11, Issue 1, 2018, 217-223*
20. Kushwaha SK *et al.*, Development and Evaluation of Polyherbal Tablet from Some Hepatoprotective Herbs Sch. Acad. J. Pharm., 2014; 3(3):321-326
21. Lachman L, Lieberman HA, Kanig JL. The Theory and Practice of Industrial Pharmacy. Philadelphia, PA: Lea and Febiger; 2009. p. 182-4, 296-303.
22. Mishra US, Murthy PN, Pasa G, Mishra D. Formulation development and evaluation of herbal tablet containing methanolic extract of *Butea frondosa*. Int J Inst Pharm Life Sci 2011;1:1–15.
23. Christianah I, Ekere K, Judith O, Olayemi O, Darlington O, *et al.* Phytopharmaceutical properties of herbal teas circulating in the Nigerian market. Afr J Pharm Pharmacol 2016;10:1007–13.
24. Nagasamy VD, Jawahar N, Ganesh GNK, Suresh K R, Senthali V, Samanta MK, Sankar S, Elango K; Development and *in vitro* evaluation of sustained release matrix tablets of theophylline using hydrophilic polymer as release retardant. Int J Pharm Sci and Nanotech, 2009; 2(1):370-375.
25. Sahoo HB, Asati AK, Toppo FA, Kori ML; Evaluation of polyherbal formulation for diuretic activity in albino rats. Asian Pacific J Tropical Disease, 2012; 2(Suppl1):S442-S445.
26. Indian Pharmacopoeia. The Controller of Publication. Delhi: Vol. 2.1996. p. 735.
27. Tubino M, de Oliveira Torres JR. Determination of calcium, phosphorus and potassium in leaf tissues by extraction with ethanol-water solvent. Anal Lett 1990;23:2339-49.

28. ICH Guideline for stability study of tablets. Section Q1 a R2.
29. Blume HH, Schug BS; The biopharmaceutical classification system (BCS). Europ J Drug Metabol Pharmacokinet, 1999; 117.