

## FORMULATION OF *HAJRAL YAHUD PISHTI* AND ITS *IN VITRO* ANTIUROLITHIATIC EFFECT

CHHAYA KUMARI, DILEEP SINGH BAGHEL\*, BIMLESH KUMAR, SAURABH SINGH

School of Pharmaceutical Sciences, Lovely Professional University, Punjab, India. Email: baghel\_12@rediffmail.com

Received: 30 October 2017, Revised and Accepted: 24 November 2017

### ABSTRACT

**Objectives:** The objectives of this study were to formulate and analyze the physicochemical properties of *hajral yahud pishti* and its *in vitro* evaluation for antiurolithiatic effect.

**Methods:** *Hajral yahud pishti* was prepared using *chandan arka*, *nagarmotha arka*, and *jala* (water), respectively, as *bhawana dravya*. Physicochemical parameters were carried out using standard methods mentioned in official compendium of *Ayurveda*. *In vitro* antiurolithiatic activity of the prepared *hajral yahud* was done on artificially prepared urine.

**Results:** No significant variation was observed in the physicochemical analysis of all prepared samples in different concentrations, i.e., 100, 200, 400, 800, and 1000  $\mu$ L of the *hajral yahud pishti* showed dose-dependent antiurolithiatic activity in prepared artificial urine 1000  $\mu$ L concentration exhibited 74.47% protection.

**Conclusion:** The prepared *hajral yahud pishti* gave protection against urolithiasis.

**Keywords:** *Pishti*, *Hajral yahud*, Antiurolithiatic, *in vitro*, Urolithiasis.

© 2018 The Authors. Published by Innovare Academic Sciences Pvt Ltd. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>) DOI: <http://dx.doi.org/10.22159/ajpcr.2018.v11i2.23385>

### INTRODUCTION

*Hajral yahud* was considered as another miracle of Allah's creations by the *Unani* writers [1,2], these stones are tiny ovular and resemble like the shape of olive fruits and their name is more than 1,000 years old, *Sahib-ul-Qanoon* wrote about them that triturated *Hajar-al-Yahud* is widely used in *Unani* medicine for dissolving kidney stones. *Mutrakrichha* (dysuria) means retention of urine, accompanied by painful micturition. It was considered to be the one of the important causes of all type of kidney stones [3-7]. Urolithiasis (*Mutra ashmari*) commonly known as kidney stones. It is basically urinary tract disorder which is characterized by the formation of stone in the kidneys. It occurs due to decreased volume of the urine or accumulation of stone-forming components such as calcium oxalates, cysteine, and xanthine which we used to take in our day-to-day meal [8-12]. These crystals get deposited in the form of precipitates in the basement of the loop of Henley, leading to the formation of renal calculi. These stones are responsible cause for the blood in urine associated with intense pain in the abdomen [13-15]. For the treatment of urolithiasis, many treatment modalities have been adopted using traditional as well as allopathic system of medicines, but the pathogenesis behind the stones is not always avoidable. Ayurvedic system of medicine exemplifies *ahara dravyas* as well as *aushadh dravyas* for the treatment of urolithiasis [16,17]. This present investigation was aimed to formulate the herbomineral formulation of *Hajral yahud* in *pishti* form and its *in vitro* evaluation for antiurolithiasis against 0.01M sodium oxalate in artificial urine (AU).

### METHODS

#### Procurement of materials

The drug *hajral yahud* was purchased from local market of Jalandhar, Punjab, and was authenticated by chemist in-charge, Herbal Health Research Consortium, Amritsar, under reference broacher number 01/2016/MIS/004 dated 10/02/2016. The plant samples (*chandan* and *nagarmotha*) were collected also from local market of Jalandhar, Punjab, and was identified and authenticated by Dr. Satiwinderjeet Kaur, Head, Department of Botanical and Environmental Sciences,

Guru Nanak Dev University Amritsar, Punjab, with reference broacher number 1452, dated 08.01.2016.

#### Morphology [18]

The morphological characters of herbal drug were studied using dissecting microscope and observed characters were tabulated (Table 1).

#### Preparation of *chandan* and *nagarmotha arka* by distillation [19-21]

One part each of powder of *nagarmotha* and *chandan* was taken in two different round bottom flask (RBF), and further, eight parts of water were added in both RBF. The distillation assembly was assembled to prepare its *arka*.

#### Preparation of *hajral yahud pishti* [22-24]

*Hajral yahud* washed with hot water (5% salt solution) followed by washing with cold water 4-5 times until the removal of salt, further dried under sun rays. The purified stones were crushed in iron mortar and pestle, then sieved it through 100 mesh size to get powder. The powder was triturated with *chandan arka*, *nagarmotha arka*, and distilled water to prepare 9 different samples of *hajral yahud pishti*. The prepared *pishti* was packed in a well-closed container.

#### Physicochemical evaluation [25-30]

Different physicochemical investigations of *pishti* and its raw materials were carried out using standard pharmacopeial methods including organoleptic parameters, determination foreign matter, loss on drying, total ash, acid insoluble ash, water-soluble extractive, alcohol-soluble extractive, and pH determinations as represented in Tables 2-4.

#### Determination of physical characteristics of powder [31]

Physical characteristics such as bulk density, tapped density, Carr's compressibility index, and angle of repose were determined for the prepared *pishti* powder as represented in Table 5.

#### Infrared spectroscopy (Shimadzu) [32,33]

Infrared spectroscopy was performed and obtained spectra with values were represented in Table 6 and Figs. 1-9.

**In vitro antiurolithiatic study [34-37]****Preparation of AU**

The AU was prepared according to the method Burns and Finlayson and had the following composition: Sodium chloride 105.5 mmol/l, sodium phosphate 32.3 mmol/l, sodium citrate 3.21 mmol/l, magnesium sulfate 3.85 mmol/l, sodium sulfate 16.95 mmol/l, potassium chloride 63.7 mmol/l, calcium chloride 4.5 mmol/l, sodium oxalate 0.32 mmol/l, ammonium hydroxide 17.9 mmol/l, and ammonium chloride 0.0028 mmol/l. The AU was prepared fresh each time and pH adjusted to 6.0.

**Study without inhibitor**

A volume of 1.0 ml of AU was transferred into the cell and 0.5 ml of distilled water added to it and blank reading was taken. The 0.5 ml of 0.01M sodium oxalate was added, to the previous volume, and the measurement is immediately started for 10 min. For each experiment, six replicates were taken.

**Study with inhibitor**

The drug was dissolved in distilled water and filtered through membrane filter, and the concentration of 50, 100, 150, 200, and 250 µg/ml was obtained. A mixture of 1 ml of AU and 0.5 ml of drug

solution was versed in the cell. A blank reading was taken, and then, 0.5 ml of 0.01 M sodium oxalate solution was added and immediately the absorbance was measured for 10 min at 620 nm. For each experiment, six replicates were taken. The percentage of inhibition was calculated using the following formula:

$$\% \text{ inhibition} = \{1 - [\text{Si}/\text{Sc}]\} \times 100$$

Where

Si: Slope of graph in the presence of inhibitor (extract),

Sc: Slope of graph without inhibitor (control)

The obtained data are represented in Table 7.

**Microscopic study [20,38,39]**

The crystals of calcium oxalate (with and without inhibitors) were observed using a light microscope (Labmed) equipped with a digital camera. The photographs of CaOx were taken using the objective of 40× and eye piece of 10×.

**Acid neutralizing capacity [40]**

The neutralization capacity of the compound is determined in terms of requirement of 0.5 N sodium hydroxide to neutralize 1 g of *Hajral yahud pishti*. Accurately weighed 100 mg *Hajral yahud pishti* was taken and dissolved in 10 ml of 3 N HCL. The solution was titrated with 0.5 N NAOH using phenolphthalein as indicator. A blank was also performed by titrating 10 ml of 3N HCL (solution used for dissolving the sample) with 0.5 N NAOH. The difference between the two readings gives the amount of 0.5 N NAOH required for neutralizing 100 mg *Hajral yahud pishti* (Table 8).

**Complexometric titration**

These type of titrations are those in which a complexing agent is used to estimate polyvalent ions. Take weighed amount of drug in conical flask and add 45 ml distilled water, ammonia solution, drop of concentrated sulfuric acid and adjust pH 10, add solochrome black as indicator. Then, titrate with ethylenediaminetetraacetic acid starting point is red wine color then end point turned into green (Table 9).

**Table 1: Morphological characters of chandan and nagarmotha**

Characters	Chandan (heart wood)	Nagarmotha (rhizomes)
Color	Creamish brown	Dark brown
Odor	Aromatic	Aromatic
Taste	Sweetish	Bitter
Shape	Cylindrical	Unorganized
Size	4.5-5 cm	4-5.2 cm
Surface	Smooth	Rough
Fracture	Hard	Soft and fibrous

**Table 2: Physicochemical evaluation of raw materials of hajral yahud pishti**

Parameters	Chandan		Nagarmotha	
	Standard value (%)	Observed value (%)	Standard value (%)	Observed value (%)
FM	N.M.T 1	0.3	N.M.T 2	0.5
LOD	-	11.8	-	8
TA	N.M.T 1	0.9	N.M.T 8	6.9
AIA	N.M.T 0.2	0.1	N.M.T 4	3.2
WSE	N.L.T 1	12	N.L.T 11	15.6
ASE	N.L.T 8	10.4	N.L.T 5	12.4

FM: Foreign matter, LOD: Loss on drying at 105°C, TA: Total ash, AIA: Acid insoluble ash, WSE: Water-soluble extractive, ASE: Alcohol-soluble extractive

**RESULTS****Standardization of raw material**

Morphological characters and physicochemical parameters were performed to check the quality of raw drugs used in formulation and compared with the values given in the Ayurvedic Pharmacopeia of India. The obtained results are represented in Tables 1 and 2.

**Standardization of formulation**

The *bhawana* process was done with different liquid media and time consumed are tabulated in Table 3. Physicochemical characterization of prepared *hajral yahud pishti* was performed which showed slight

**Table 3: Time consumed for each Bhawana**

Batch	Bhawna (Time in h)	Bhawna with Chandan arka	Bhawna with Nagarmotha arka	Bhawna with water
1	62	1	1	1
2	73	1	1	1
3	76	1	1	1

**Table 4: Physicochemical evaluation of formulation of hajral yahud pishti**

Parameters	C1	C2	C3	Mean	N1	N2	N3	Mean	W1	W2	W3	Mean
LOD	0.49	0.47	0.45	0.47	0.39	0.72	0.82	0.64	1.52	1.14	0.79	1.15
TA	59.3	57.6	61.4	59.43	58.4	68.76	64.35	63.84	58.84	56.66	55.43	56.98
AIA	1.29	0.57	1.31	1.06	0.71	1.43	1.28	1.14	1.91	1.83	1.47	1.74
ASE	2.03	1.64	1.98	1.88	1.15	4.46	4.74	3.45	6.02	8.20	1.93	5.38
WSE	1.36	1.58	2.40	1.78	1.92	9.67	6.16	5.92	3.85	3.42	1.98	3.08
pH	8.8	9.1	9.3	9.07	9.4	10.2	10.6	10.07	9.3	9.7	8.4	9.13

LOD: Loss on drying at 105°C, TA: Total ash, AIA: Acid insoluble ash, ASE: Alcohol-soluble extractive, WSE: Water-soluble extractive, IR: Infrared, C1-C3: Chandan Arka, N1-N3: Nagarmotha Arka, W1-W3: Water

Table 5: Flow properties prepared *hajral yahud pishti*

Parameters	C1	C2	C3	Mean	N1	N2	N3	Mean	W1	W2	W3	Mean
Bulk density	0.3	0.4	0.3	0.33	0.4	0.4	0.3	0.36	0.6	0.5	0.3	0.46
Tapped density	1.2	1.1	1	1.1	1.4	1.3	1.2	1.3	1	1.1	1.4	1.16
Carr's compressibility index	0.72	0.63	0.7	0.68	0.71	0.69	0.75	0.72	0.4	0.54	0.78	0.57
Angle of repose	32.3	31.45	30.4	31.38	34.60	33.21	34.12	33.98	28.81	30.12	31.4	30.11

C1-C3: Chandan Arka, N1-N3: Nagarmotha Arka, W1-W3: Water

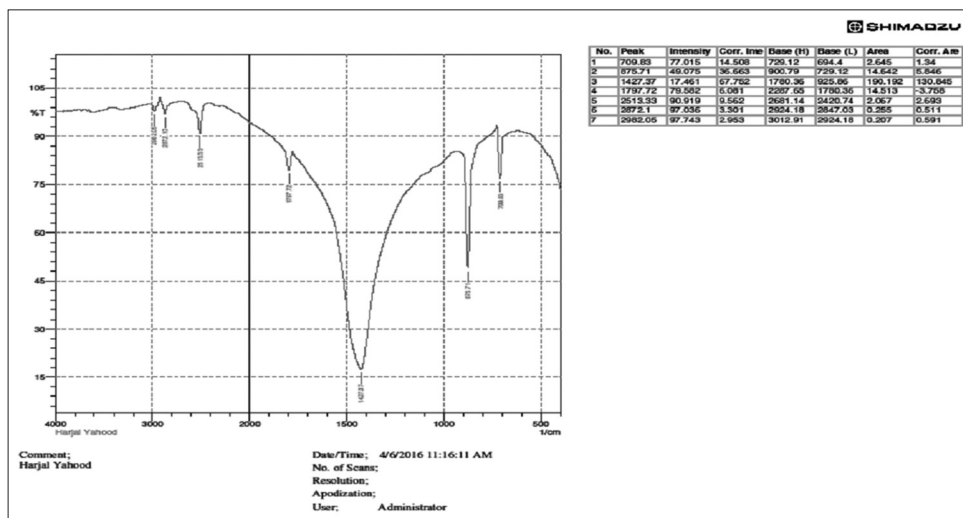


Fig. 1: Infrared *hajral yahud* (Chandan 1)

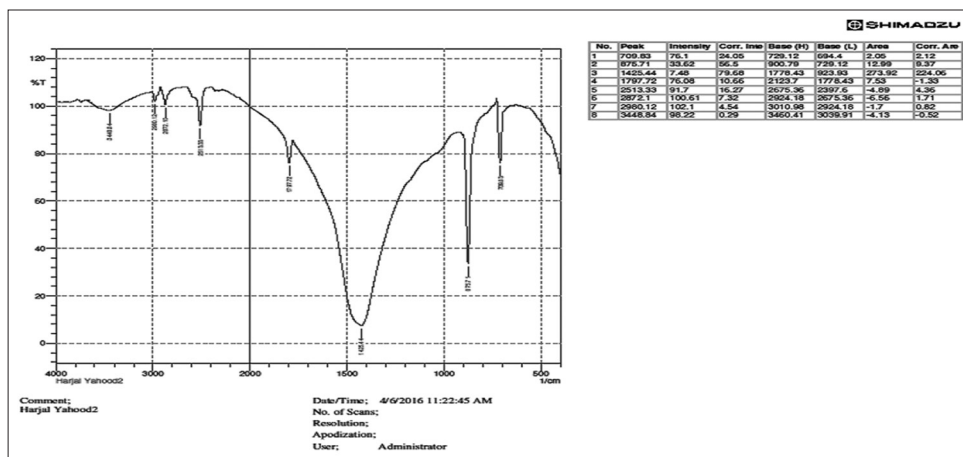


Fig. 2: Infrared *hajral yahud* (Chandan 2)

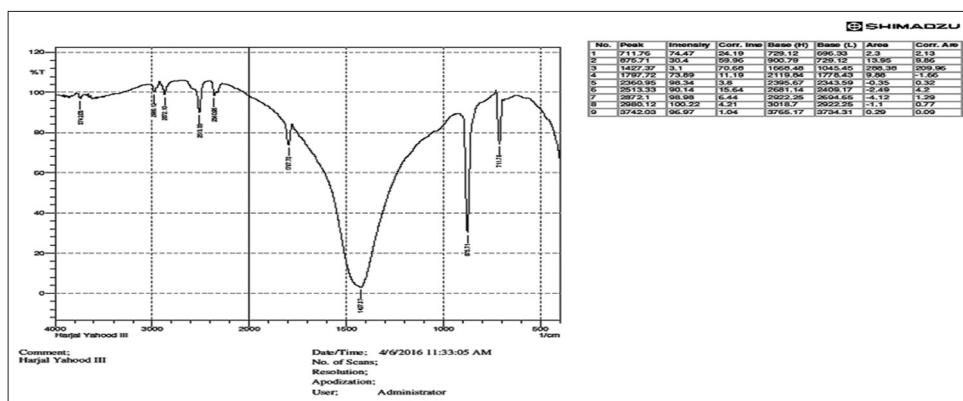


Fig. 3: Infrared *hajral yahud* (Chandan 3)

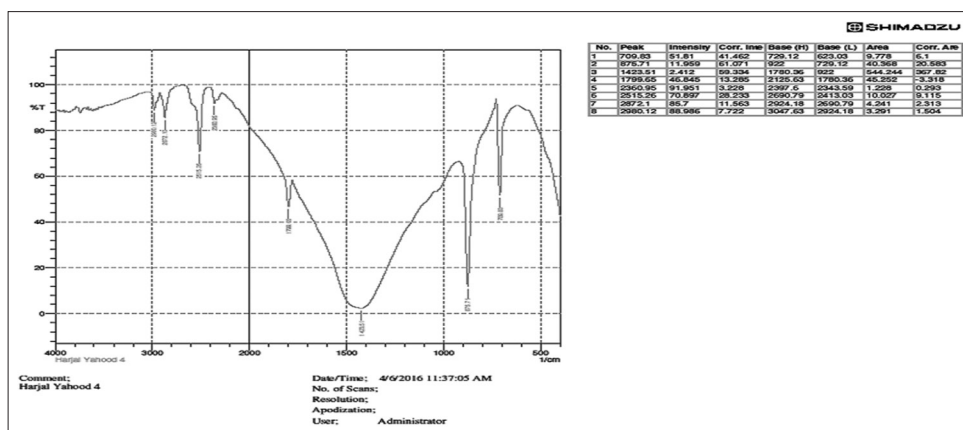


Fig. 4: Infrared hajral yahud (Nagarmotha 1)

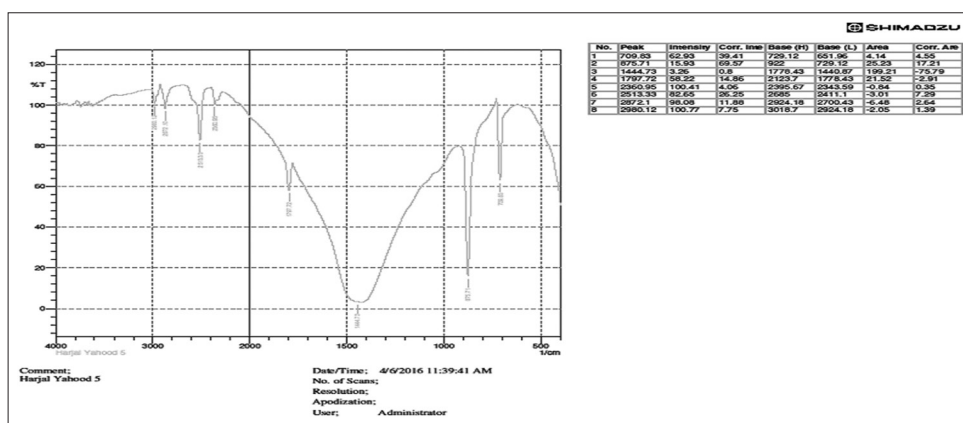


Fig. 5: Infrared hajral yahud (Nagarmotha 2)

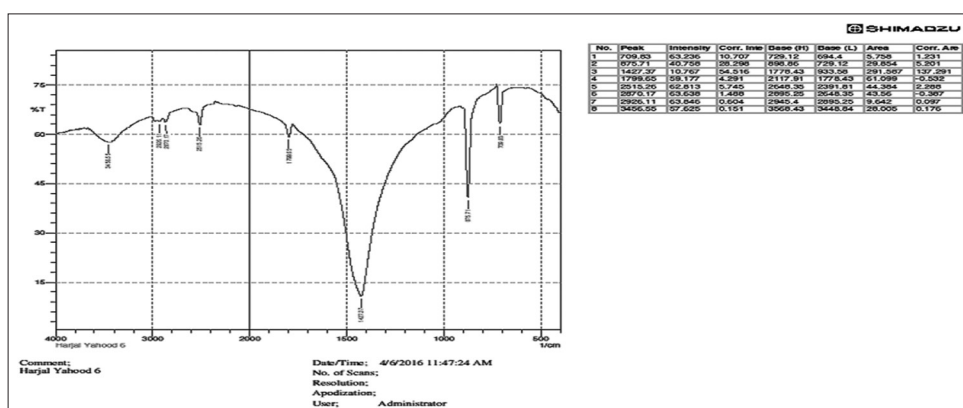


Fig. 6: Infrared hajral yahud (Nagarmotha 3)

difference in the readings of prepared batches of *hajral yahud pishti*, which suggests that the liquid mediums does not affect the formulation physically, and therefore, the method preparation could be planned using any of the liquid mediums according to the availability. The obtained value is represented in Table 4.

**Infrared spectroscopy (Shimadzu)**

Infrared spectroscopy was performed and observed major peaks are in the same range which confirm the presence of calcium carbonate, but the liquid mediums used were not differentiated by the IR, may be due to the evaporation of added liquid mediums during levigation process. The obtained peaks and intensity are represented in Table 6 and spectra is represented in Figs. 1-9.

**Flow properties prepared hajral yahud pishti**

Bulk density, tapped density, Carr’s compressibility index, and angle of repose were found in limits which implies that the formulation *hajral yahud pishti* has good flow properties and will not affect the formulation during mixing, packaging, and transportation. The data related to flow properties are represented in Table 5.

**Determination of particle size**

Two methods were used for determining the particle size of prepared *hajral yahud pishti* batches. Both the methods were performed manually. From the microscopy, the particle size was found to be in the range of 0.3–0.5 mm, while from the sieving method, maximum particles were passed through from mesh size 120 which indicates

Table 6: Infrared spectroscopy

Sample	No.	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
C 1	1	709.83	77.015	14.508	729.12	694.4	2.645	1.34
	2	875.71	49.075	36.663	900.79	729.12	14.642	5.846
	3	1427.37	17.461	67.752	1780.36	925.86	190.192	130.845
	4	1797.72	79.582	6.081	2287.65	1780.36	14.513	-3.758
	5	2513.33	90.919	9.562	2681.14	2420.74	2.067	2.693
	6	2872.1	97.036	3.301	2924.18	2847.03	0.255	0.511
	7	2982.05	97.743	2.953	3012.91	2924.18	0.207	0.591
C 2	1	709.83	76.1	24.05	729.12	694.4	2.05	2.12
	2	875.71	33.62	56.5	900.79	729.12	12.99	9.37
	3	1425.44	7.48	79.68	1778.43	923.93	273.92	224.06
	4	1797.72	76.08	10.66	2123.7	1778.43	7.53	-1.33
	5	2513.33	91.7	16.27	2675.36	2397.6	-4.89	4.36
	6	2872.1	100.61	7.32	2924.18	2675.36	-6.56	1.71
	7	2980.12	102.1	4.54	3010.98	2924.18	-1.7	0.82
	8	3448.84	98.22	0.29	3460.41	3039.91	-4.13	-0.52
C 3	1	711.76	74.47	24.19	729.12	696.33	2.3	2.13
	2	875.71	30.4	59.96	900.79	729.12	13.95	9.86
	3	1427.37	3.1	70.68	1668.48	1045.45	288.38	209.96
	4	1797.72	73.89	11.19	2119.84	1778.43	9.88	-1.66
	5	2360.95	98.34	3.8	2395.67	2343.59	-0.35	0.32
	6	2513.33	90.14	15.64	2681.14	2409.17	-2.49	4.2
	7	2872.1	98.98	6.44	2922.25	2694.65	-4.12	1.29
	8	2980.12	100.22	4.21	3018.7	2922.25	-1.1	0.77
	9	3742.03	96.97	1.04	3765.17	3734.31	0.29	0.09
N 1	1	709.83	51.81	41.462	729.12	623.03	9.778	6.1
	2	875.71	11.959	61.071	922	729.12	40.368	20.583
	3	1423.51	2.412	59.334	1780.36	922	544.244	367.82
	4	1799.65	46.845	13.285	2125.63	1780.36	45.252	-3.318
	5	2360.95	91.951	3.228	2397.6	2343.59	1.228	0.293
	6	2515.26	70.897	28.233	2690.79	2413.03	10.027	9.115
	7	2872.1	85.7	11.563	2924.18	2690.79	4.241	2.313
	8	2980.12	88.986	7.722	3047.63	2924.18	3.291	1.504
N 2	1	709.83	62.93	39.41	729.12	651.96	4.14	4.55
	2	875.71	15.93	69.57	922	729.12	25.23	17.21
	3	1444.73	3.26	0.8	1778.43	1440.87	199.21	-75.79
	4	1797.72	58.22	14.86	2123.7	1778.43	21.52	-2.91
	5	2360.95	100.41	4.06	2395.67	2343.59	-0.84	0.35
	6	2513.33	82.65	26.25	2685	2411.1	-3.01	7.29
	7	2872.1	98.08	11.88	2924.18	2700.43	-6.48	2.64
	8	2980.12	100.77	7.75	3018.7	2924.18	-2.05	1.39
N 3	1	709.83	63.236	10.707	729.12	694.4	5.758	1.231
	2	875.71	40.758	28.298	898.86	729.12	29.854	5.201
	3	1427.37	10.767	54.516	1778.43	933.58	291.587	137.291
	4	1799.65	59.177	4.291	2117.91	1778.43	61.099	-0.532
	5	2515.26	62.813	5.745	2648.35	2391.81	44.384	2.288
	6	2870.17	63.638	1.488	2895.25	2548.35	43.56	-0.387
	7	2926.11	63.846	0.604	2945.4	2895.25	9.642	0.097
	8	3456.55	57.625	0.151	3568.43	3448.84	28.005	0.176
W 1	1	399.28	54.128	0	565.16	399.28	33.277	-2.669
	2	711.76	53.591	14.851	729.12	694.4	7.599	1.878
	3	875.71	28.932	33.062	900.79	729.12	39.419	7.362
	4	1427.37	14.925	42.173	1778.43	922	364.772	159.754
	5	1799.65	48.757	5.984	2117.91	1778.43	78.648	-0.963
	6	2515.26	54.864	8.346	2650.28	2395.67	54.483	3.707
	7	2872.1	57.446	2.136	2897.18	2650.28	52.661	-0.445
	8	2980.12	58.205	1.294	3009.05	2943.47	15.09	0.26
	9	3458.48	53.675	0.026	3462.34	3448.84	3.647	0.002
W 2	1	709.83	50.57	12.218	729.12	624.96	23.611	2.36
	2	875.71	30.717	25.288	900.79	729.12	45.978	6.314
	3	1425.44	9.142	43.235	1780.36	920.08	390.891	151.953
	4	1799.65	46.638	4.072	2119.64	1780.36	91.126	-0.831
	5	2364.81	56.416	0.907	2395.67	2345.52	12.198	0.132
	6	2515.26	51.525	6.136	2650.28	2395.67	63.964	3.023
	7	2870.17	52.581	1.579	2895.25	2650.28	62.139	-0.567
	8	2924.18	52.585	0.915	2947.33	2895.25	14.314	0.181
	9	3456.55	48.566	0.114	3583.86	3448.54	41.553	0.24
W 3	1	709.83	37.943	25.482	729.12	644.25	21.785	4.732
	2	875.71	8.819	43.6	920.08	729.12	65.818	17.584
	3	1442.8	2.066	0.222	1778.43	1440.87	287.933	-63.468

(Contd...)

Table 6: (Continued)

Sample	No.	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
	4	1799.65	32.112	8.808	2119.84	1778.43	109.382	-2.491
	5	2515.26	42.767	14.519	2654.14	2395.67	70.263	7.62
	6	2872.1	47.321	3.573	2895.25	2654.14	64.704	-1.112
	7	2924.18	48.45	1.41	2945.4	2895.25	15.419	0.286
	8	2980.12	49.062	1.231	3026.41	2964.69	18.03	0.01
	9	3443.05	43.628	0.135	3448.84	3026.41	133.068	-1.624

IR: Infrared spectroscopy, C1-C3: Chandan Arka, N1-N3: Nagarmotha Arka, W1-W3: Water

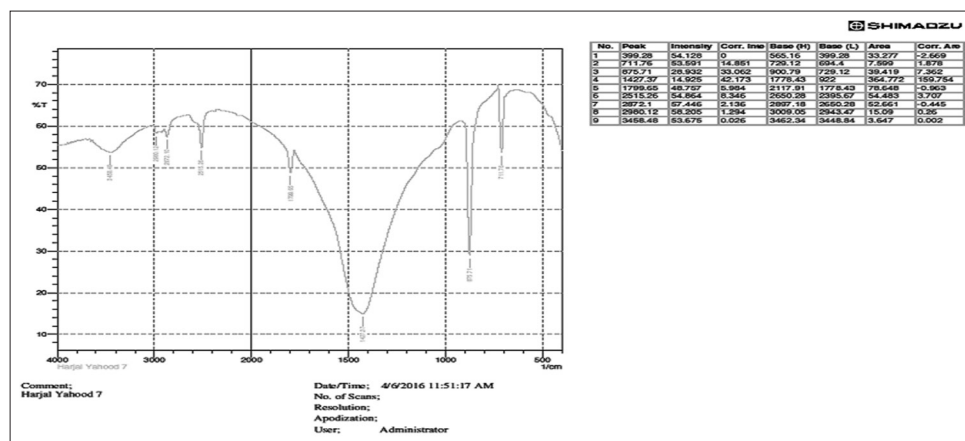


Fig. 7: Infrared hajral yahud (Water 1)

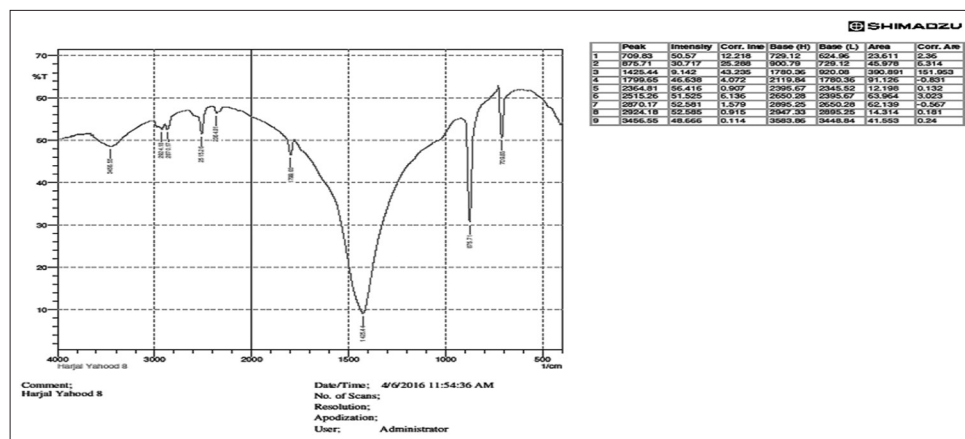


Fig. 8: Infrared hajral yahud (Water 2)

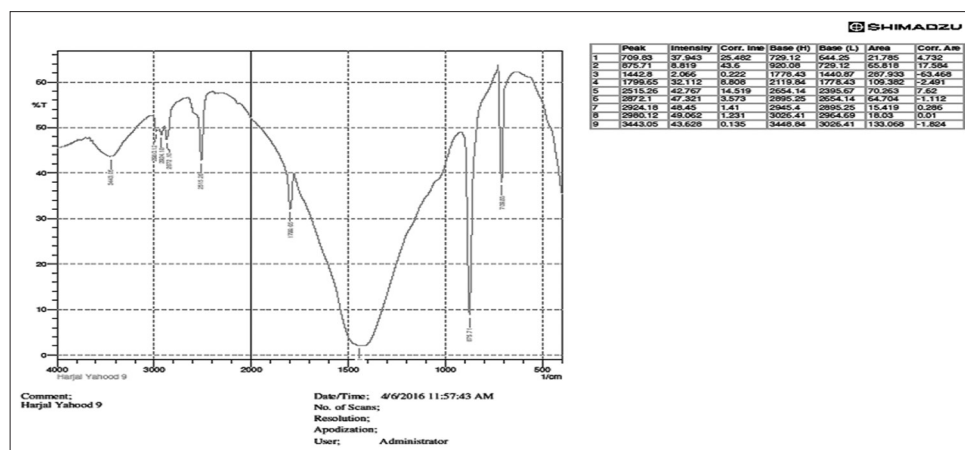


Fig. 9: Infrared hajral yahud (water 3)

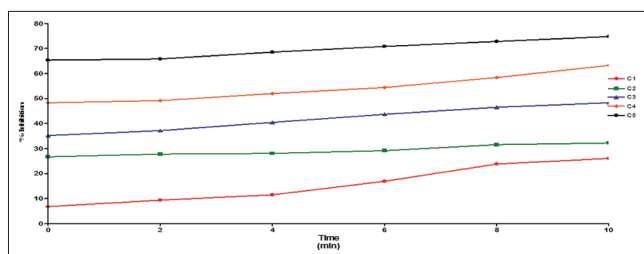


Fig. 10: Percentage inhibition with respect to time

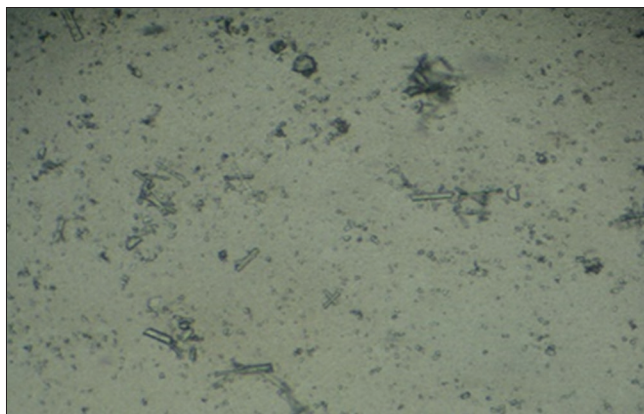


Fig. 11: Crystal growth in AU preparation

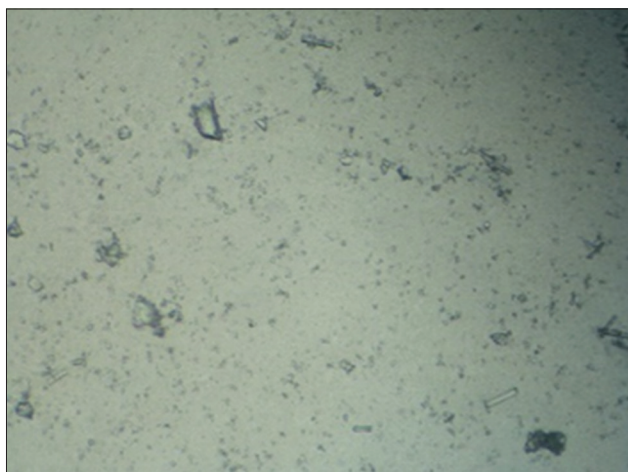


Fig. 12: Crystal inhibition in 100 µg/ml drug sample

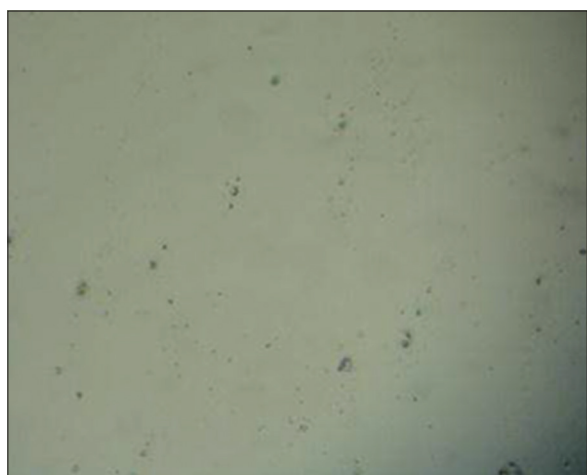


Fig. 13: Crystal inhibition in 100 µg/ml drug sample

Table 7: % inhibition of Hajral Yahud Pishti with respect to time

Time in minutes	100 µg/ml	200 µg/ml	400 µg/ml	800 µg/ml	1000 µg/ml
0	6.8182	26.705	35.23	48.3	65.34
2	9.4017	27.778	37.18	49.15	65.81
4	11.517	28.09	40.45	51.97	68.54
6	16.926	29.188	43.7	54.4	70.81
8	23.84	31.572	46.52	58.38	72.81
10	26.037	32.258	48.27	63.25	74.47

that the particle size of the prepared formulation was fine enough to produce therapeutic efficacy.

#### Acid neutralizing capacity for prepared hajral yahud pishti

An average of 0.26 mg NaOH was required to neutralize 100 mg of hajral yahud pishti. The obtained data are represented in Table 8.

#### Complexometric titrations for prepared hajral yahud pishti

An average of 0.24 g of calcium was found in 1 g of the Hajral yahud Pishti. The obtained data are represented in Table 9.

#### In vitro study

In all the treatments, concentration-dependent initial steep rising in turbidity (nucleation) followed by decrease turbidity (aggregation) was seen and with the time % inhibition also increases. The last two concentrations showed the maximum percentage of inhibition which implies that the drug is showing positive response toward antiurolithiatic activity in the *in vitro* studies. The data of inhibition in percentage are represented in Table 7 and the graph of inhibition in percentage is represented in Fig. 10. The microscopic images of crystal growth in AU and their inhibition in construction of 100 µg/ml and 1000 µg/ml are shown in Figs. 11-13.

#### DISCUSSION

In the present investigation, Hajral yahud pishti was evaluated for its physicochemical parameters and antiurolithiatic activity. Hajral yahud pishti was prepared using different liquid mediums. The physicochemical parameters were performed for the prepared formulation for three different batches treated with different liquid mediums, i.e., Chandan Arka, Nagarmotha Arka, and water for the 9 samples, respectively, but no significant variation was observed which indicates that there is not much effect of the liquid medium physically. In the same manner, the analytical parameters such as total ash, acid insoluble ash, alcohol-soluble extractive, water-soluble extractives, and IR were also performed which also not showed remarkable variations in functional groups. During the *in vitro* studies, the crystals were observed in the prepared sample of urine when mixed with sodium oxalate when observed under microscope. These crystals were found decreasing due to the addition of drug which shows that the drug is helpful in breaking crystals. *In vitro* studies were observed under ultraviolet spectrophotometer (Shimadzu 1800) when viewed under 620 nm with different dilutions without and with the drug having 100, 200, 400, 800, and 1000 µl, respectively, with time gap of 0, 2, 4, 6, 8, and 10 min showed maximum 74.77% drug inhibition in the highest concentration.

#### CONCLUSION

The physicochemical parameters was performed and no significant variation was observed in all the prepared batches which was prepared using different rations of herb and concentration of water. IR study was also carried out to find functional groups in the prepared pishti samples, all the prepared batches showed the presence of calcium. Spectral analysis showed that there is no alteration in primary composition of hajral yahud. *In vitro* antiurolithiatic activity

Table 8: Readings of acid neutralizing capacity for prepared *hajral yahud pishti*

C1	C2	C3	Mean	N1	N2	N3	Mean	W1	W2	W3	Mean
14.5	14.1	15.1	14.57	13.5	11.6	13.8	12.97	15.1	13.4	14.7	14.4
15.5	13.8	15.2	14.93	13.6	11.6	12.6	12.6	15.4	13.2	14.5	14.37
13.4	14.1	14.9	14.13	14.1	11.4	13.5	13	15.8	13.1	13.6	14.17

C1-C3: Chandan Arka, N1-N3: Nagarmotha Arka, W1-W3: Water

Table 9: Complexometric titrations for prepared *hajral yahud pishti*

C1	C2	C3	Mean	N1	N2	N3	Mean	W1	W2	W3	Mean
5.8	6.1	6.4	6.1	5.4	5.1	5.4	5.3	4.7	4.9	5.2	4.93
5.3	5.6	5.8	5.57	5.6	4.8	6.2	5.53	4.4	5.3	4.6	4.77
6.8	5.5	6.1	6.13	4.8	5.7	5.3	5.27	4.1	4.7	4.5	4.43

C1-C3: Chandan Arka, N1-N3: Nagarmotha Arka, W1-W3: Water

was performed for the all prepared batches using AU, the crystal inhibition capacity was observed through optical microscopy in all the batches, so it is concluded that all the prepared batches of *hajral yahud pishti* at different concentrations, i.e., 100, 200, 400, 800, and 1000 µL showed dose-dependent antiurolithiatic activity in prepared AU and 1000 µL concentration exhibited maximum 74.47% protection.

#### ACKNOWLEDGMENT

The authors wish to express their sincere thanks to Professor (Dr.) Monica Gulati, Sr. Dean, School of Pharmaceutical Sciences, Lovely Professional University, Phagwara, Punjab, for guidance and support.

#### AUTHORS CONTRIBUTION

Chhaya Kumari: Principal investigator, performed preparation and analysis on all samples, interpreted data, write manuscript. Dileep Singh Baghel: Guide, supervised development of work, helped in data interpretation, manuscript evaluation and acted as corresponding author. Bimlesh Kumar: Helped to evaluate and edit the manuscript. Saurabh Singh: Helped to evaluate and edit the manuscript.

#### CONFLICTS OF INTERESTS

None Declared.

#### REFERENCES

- Ahmed S, Hasan M. Muslim heritage in medicine: A concise review on Greco-Arabic contribution. *J Pharmacogn Phytochem* 2016;5:273-83.
- Rajesh. Study of hasate Kulya (Renal Calculus) with therapeutic evaluation of unani formulation (Sufoof Hajrul Yahud) in its management. *Int J Sci Res* 2016;5:268-71.
- Shastri S. Agnivesha Charaka Samhita. Chikitsa Sthana 26/599. Varanasi: Chaukhamba Bharti Academy; 2006.
- Sushruta. Sushruta Samhita. Sutra Sthana 33/126. Varanasi: Chaukhamba Sanskrit Sansthan; 2005.
- Sodimbaku V, Pujari L. Urolithiasis-an updated review over genetics, pathophysiology and its clinical management. *Int J Pharm Pharm Sci* 2014;6:23-31.
- Moe OW. Kidney stones: Pathophysiology and medical management. *Lancet* 2006;367:333-44.
- Evan A, Lingeman J, Coe FL, Worcester E. Randall's plaque: Pathogenesis and role in calcium oxalate nephrolithiasis. *Kidney Int* 2006;69:1313-8.
- Stamatelou KK, Francis ME, Jones CA, Nyberg LM, Curhan GC. Time trends in reported prevalence of kidney stones in the United States: 1976-1994. *Kidney Int* 2003;63:1817-23.
- Mohanty NK, Nayak RL, Patki PS. Safety and efficacy of an ayurvedic formulation cystone in management of ureteric calculi: A prospective randomized placebo controlled study. *Am J Pharm Toxicol* 2010;5:58-64.
- Shanmugapriya J, Kumar SS. A prospective study on quality of life in patients with urinary calculi. *Asian J Pharm Clin Res* 2017;10:191-3.
- Coe FL, Evan A, Worcester E. Kidney stone disease. *J Clin Invest* 2005;115:2598-608.
- Prasad K, Sujatha D, Bharathi K. Herbal drugs in urolithiasis - A review. *Phcog Rev* 2007;1:175-9.
- Parks JH, Coward M, Coe FL. Correspondence between stone composition and urine super saturation in nephrolithiasis. *Kidney Int* 1997;51:894-900.
- Khan SR. Calcium phosphate/calcium oxalate crystal association in urinary stones: Implications for heterogeneous nucleation of calcium oxalate. *J Urol* 1997;157:376-83.
- Mustapha B, Hanane B, Idrissi M, Benramdane Larbi OB, Mustapha D. *In vitro* effect of acetylsalicylic acid on calcium oxalate crystallization: An approach to antilithiasis. *Int J Pharm Pharm Sci* 2015;7:329-31.
- Aggarwal D, Sharma M, Singla SK. The role of natural antioxidants as potential therapeutic agent in nephrolithiasis. *Asian J Pharm Clin Res* 2013;6:48-53.
- Sayana SB, Khanwelkar CC, Nimmagadda VR, Chavan VR. Antiurolithic activity of aqueous extract of roots of *Cissampelos pareira* in albino rats. *Asian J Pharm Clin Res* 2014;7:49-53.
- Wahab S. Authentication and quality evaluation of an important ayurvedic drug *Averrhoa carambola* Linn Leaves. *Asian J Pharm Clin Res* 2013;6:52-6.
- Ravana L. Arkaprakasha. 3<sup>rd</sup> ed. Varanasi, India: Chaukhamba Krishnadas Academy; 2011.
- Rahul U, Hussain G, Ganti BY. Pharmaceutical review of Arka Kalpana. *Int Ayurvedic Med J* 2014;2:1113-20.
- Gaud SD, Nighantu S. Reprinted edition Varanasi: Chaukhamba Vidyabhanan; 2002. p. 431, 141, 87.
- Trikamji Acharya VY. Sidhiyog Sangrah. Ch. 18. Nagpur, India: Sh. Baidyanath Ayurveda Bhawan Limited 2003. p. 99.
- Krishnanad S. Rasatantra Sara Va Siddha Prayoga Samraha 16<sup>th</sup> ed. Ajmer, India: Krishan Gopal Ayurveda Bhawan; 2003. p. 219.
- Zhang J, Wider B, Shang H, Li X, Ernst E. Quality of herbal medicines: Challenges and solutions. *Complement Ther Med* 2012;20:100-6.
- Patra KC, Kumar KJ, Suresh P. Standardization of a polyherbal Siddha formulation, Amukkara Chooranam. *Indian J Tradit Knowl* 2009;8:449-e52.
- Lohar DR. Protocol for testing of Ayurvedic, Siddha and Unani medicines. Ghaziabad: Pharmacopoeial Laboratory for Indian Medicine, AYUSH. Ministry of Health and Family Welfare. Government of India; 2011. p. 25.
- Satheesh NV, Upadhyaya K, Bisht A. Phytochemical screening and standardization of polyherbal formulation for dyslipidemia. *Int J Pharm Pharm Sci* 2011;3:235e8, 17,18.
- Thakkar R, Baghel DS, Singh S. Physicochemical screening of Godanti Bhasma: An effort to pharmaceutical standardisation. *J Pharm Res* 2017;11:887-94.
- Sharma S, Baghel DS, Singh S, Singh SK. Dosage form development and preliminary physicochemical characterization of Trikantakadi Kvatha. *Asian J Pharm Clin Res* 2017;2017:52-6.
- Sharma R, Amin H, Galib, Prajapati PK. Validation of standard manufacturing procedure of Guduchi satva (aqueous extract of *Tinospora cordifolia* (Willd.) Miers) and its tablets. *Anc Sci Life* 2013;33:27-e34.
- Jain V, Swarnlata S, Saraf S. Spectrophotometric determination of piperinein trikatu churna: An ayurvedic formulation. *Asian J Chem* 2007;19:5331-5.



32. Almeida RM, Pantano CG. Structural investigation of silica gel films by infrared spectroscopy. *J Appl Phys* 1990;68:4225-32.
33. Ahmed S, Hasan MM, Mahmood ZA. *In vitro* urolithiasis models: An evaluation of prophylactic management against kidney stones. *J Pharmacogn Phytochem* 2016;5:28-35.
34. Rajeshwari P, Rajeswari G, Jabbirulla S, Vardhan IV. Evaluation of *in vitro* anti-urolithiasis activity of *Convolvulus arvensis*. *Int J Pharm Pharm Sci* 2013;5:599-601.
35. Agarwal K, Varma R. *In-vitro* Calcium oxalate crystallization inhibition by *Achyranthes aspera* L. and *Bryophyllum pinnatum* Lam. *Br J Pharm Res* 2015;5:146-52.
36. Rupam VP, Khan ND, Khan ZH, Mular SM. Study on *in vitro* antiurolithiatic activity of *Bryophyllum pinnatum* and *Ocimum gratissimum* leaves. *Biosci Discov* 2017;8:290-4.
37. Priya K, Gazala H, Reshma SM, Govindan ND. Pharmaceutico analytical study of Vacha arka. *J Biol Sci Opin* 2013;1:314-6.
38. Atmani F, Khan SR. Effects of an extract from *Herniaria hirsuta* on calcium oxalate crystallization *in vitro*. *BJU Int* 2000;85:621.
39. Kok DJ, Papapoulos SE, Blomen LJ, Bijvoet OL. Modulation of calcium oxalate monohydrate crystallization kinetics *in vitro*. *Kidney Int* 1988;34:346-50.
40. Rao GD. *Practical Pharmaceutical Analysis*. 1<sup>st</sup> ed. Delhi: Publisher Birla publications (Regd) Pvt. Ltd.; 2007. p. 137-8.