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Case Study

Scientific studies on *Lycium shawii*

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Lycium (Solanaceae) comprises of ca. 90 species of thorny shrubs distributed throughout the tropical regions of the world. *Lycium shawii*, desert thorn, or Arabian boxthorn, is a species of thorny shrub adapted to desert environments and can be found throughout the Arabian peninsula, and some places in Africa [1]. The thin-leaved, rigid bush grows up to 3 meters (9.8 feet) high, with a lot of branches and alternating spines that vary in size, and grow along the branches and on their tips. The leaves narrow towards their base. It produces small whitish-pink or purple flowers from September until April and red pea-sized seedy berries that are edible. Habitats include gravel plains and foothills up to 4,000 ft (1,200m), as well as wadis. Plants often growing nearby include *Acacia tortilis* and *Prosopis cineraria*.

Figure 1

The stems, leaves, and berries are used in traditional medicine. In Yemen, the pounded leaves of this shrub have been used as a cure for eye ailments. The berries have a laxative effect and were used in traditional medicine to relieve constipation and as a diuretic. Livestock eats new growth on the plant [2].

Phytochemical investigation of *Lycium shawii* Roem. and Schult provided fourteen compounds, including lyciumate, dehydrocostus lactone, costunolide, catechin, lyciumaside, emodin, emodin8-O-β-D-glucoside, aloe-emodin, aloe-emodin-8-O-β-D-glucoside, aloe emodin-11-Orhamnoside, chrysophanol-8-O-β-D-glucoside, nonacosane-10-ol,betulinic acid, and β- sitosterol glucopyranoside [3]. The compounds may be classified as three sesquiterpene lactones), two phenolic compounds, six anthraquinones, one long chain alcohol), one lupane-type triterpenoid and a steroid. Compounds were reported for the first time as isolated from any species of *Lycium* as well as from the Solanaceae family while three compounds were reported to be found for the first time in the genus *Lycium* [3].

Pharmacognosy and phytochemistry

Microscopical description

Sporting bright red berried, and often reaching a height of 12 feet, the Lycum plant can be found throughout much of China and Tibet. *Lycium* has played a major role in Chinese medicine since at least the 1st Century AD when it was extolled in the Divine Husbandman.

Organoleptic characteristics

Appearance: Solid Powder

Colour: Grayish green

Odour: No Specific Smell

Taste: Tending Sweet

UV- Absorbance values of *Lycium shawii* absolute alcohol extract.

Wavelength Vs absorbance

Wavelength in Nm	Absorbance
200	0.924
210	1.791
220	1.304
230	1.079
240	0.743
250	0.533
260	0.451
270	0.485
280	0.552
290	0.527
300	0.455
310	0.442
320	0.452
330	0.416
340	0.323
350	0.219
360	0.145
370	0.101
380	0.072
390	0.052
400	0.040
410	0.032
420	0.026
430	0.020
440	0.015
450	0.011
460	0.009
470	0.008
480	0.007
490	0.006
500	0.005

Table 1

* The concentration of sample solution: 100 microgram/mLμμ

Figure 2

Physicochemical constants

Loss in weight on drying at 105^{0}C (%): 9.40

Solubilities (%)

Alcohol solubility: 1.60

Water solubility: 8.00

% ethanolic extractive: -

Ash values (%)

Total ash: 5.40

Water soluble ash: 2.00

Acid-insoluble ash: 0.30

Successive extractive (%)

Petroleum ether (60-80^0): 1.30

Chloroform: 0.80

Absolute alcohol: 4.20

Distilled water: 8.00

pH values

pH of 1% solution: 6.9

pH of 10% solution: 6.2

Chemical constituents [1,2]

The plant contains alkaloids, sterols and terpenes.

A market sample of *Lycium* Plus Tablets were checked for different parameters and toxicity of the sample:

Product Name: Forever *Lycium* Plus Tablets

Manufact. date: 03/2017

Expiration date: 03/2020

Manufacturer: Forever

Batch Number: Bo60317H.

Chromatographic Studies: Different mobile phases were used to develop the thin layer chromatograms using petrol/acetone, methanol extracts. The following plants and compounds are identified in the 'Forever *Lycium* Plus 'taken in above solvents and on comparison with reference standards on chromatographic and GCMS analysis.

Lycium: Pentadecanoic acid

Palmitic acid: Heptadecanoic acid

Margaric acid: Tridecylic acid

Valeraldehyde: Butyl propyl ether

Succinaldehyde: 2-Deuterio-2-Methylpropane

Mesifurane: 1,3-Cyclohexane-1,3-d2-diamine

Glycyrrhiza glabra: Oleic acid methyl ester

Total Ash Value (%): 1.6735

Toxic elements (ppm): Arsenic: 0.110

Cadmium: 0.160

Mercury: 0.0850

Microbial Contamination: Lead 4.830

Discussion: *NMT 100 CFU/g

Recommendation: *Pathogenic organisms were not isolated.

The sample does not contain any toxic compound.

Based on above studies 'Forever *Lycium* Plus' seems to be safe to use.

Pharmacological studies

Different pharmacological studies have been carried out on rats and it was concluded that the plant is not toxic and have medicinal properties as concluded in below experiments, besides a market sample of *Lycium* Plus Tablets were checked for different parameters and toxicity of the sample:

The diuretic study of *Lycium* on rats

Group	Dosage and Routine	B.W (g)	Output of Urine In 24 hrs (ml)	Food consumption In 24 hrs (g)
Control	10 ml. Kg^{-1}, p.o, once	298.75 ± 38.14	10.55 ± 0.08	23.00 ± 1.83
Lycium	0.8 $g.kg^{-1}$, p.o. once	305.75 ± 34.42	14.06 ± 2.09	19.62 ± 0.72

Table 2: The diuretic effects of Lycium on rats (Mean ± SE, n = 8 in each group).

Animals

Wistar rats, female and male were used.

Extract

Lycium (water extract, Code:).

Dosage and routine

0.8 g.Kg^{-1} was given orally, once.

Result

Body weight (B.W), food consumption and output of urine were checked 24 after extract was given. The difference of Body weight, output of urine and water consumption in treated and control group were calculated. Experimental results showed output of urine increased slightly 24 hrs after Cymbopogon 0.4—0.8 g.Kg^{-1} was given orally once. but the difference did not have significance.

The effects of *Lycium* on gastrointestinal motility activity of Rats

Animals: SD rats, both of male and female were used.

Extract: *Lycium* (Water-extract),

Dosage and Routine: 1.0g.Kg^{-1}, p.o. once.

Animal Model: SD rats were fasted for 18 hrs. Each animal was administered orally with 1 ml of charcoal meal (5% deactivated charcoal in 20% aqucous Arab gum), 1 hrs before extract was given p.o. 30 minutes later, animal was sacrificed with ethel and the intestinal distance moved by the charcoal meal from the pylorus to the beginning of plug (D1), length of plug (D2), from the end of plug to cacacum (D3) and the distance from the pylorus to cacacum (TD) was measured, and the percentage was also calculated.

		Control	Lycium (1.0 g.kg^{-1})
BW (g)		222.90 ± 17.00	265.70 ± 29.10
TD (cm)		80.29 ± 2.75	99.00 ± 1.53**
D1 (cm)	Distance	39.14 ± 1.83	46.43 ± 1.35*
	D1/TD(%)	48.73 ± 1.38	47.02 ± 1.62
D2 (cm)	Distance	20.29 ± 1.34	24.14 ± 3.16
	D2/TD(%)	25.40 ± 1.79	24.17 ± 2.52
D3 (cm)	Distance	20.86 ± 1.60	28.43 ± 1.25**
	D3/TD(%)	25.87 ± 1.45	48.81 ± 1.36

Table 3

(n = 7, Mean ± SE)

*P < 0.05, ** P < 0.01, VS Control Group.

Conclusion

There is no effects to gastrointestinal mobility moved by the charcoal meal when *Lycium* 1.0 g.Kg^{-1} was given orally once.

Effect of *Lycium* on CHOL and Liver ccl4 hepatotoxicity induced in rats

Figure 3

			Normal	CCl4 model	Lycium
Dosage and Routine			Water, 10 ml/kg po for 5 days	Water, 10 ml/kg po for 5 days	0.5-0.8 g.kg^{-1}, p.o
B.W	Initial reading		248.50 ± 11.79	255.00 ± 14.39	254.00 ± 10.35
	Pre-CCl4	Reading	259.50 ± 13.83	269.50 ± 17.57	271.00 ± 12.22
		% of initail	104.55 ± 1.45	105.23 ± 1.38	106.59 ± 1.28
	Post-CCl4	Reading	257.00 ± 14.55	257.00 ± 16.67	265.00 ± 13.27
		% of initail	103.76 ± 1.61	100.35 ± 1.58	104.08 ± 1.86
TBIL (mg.dL)			0.21 ± 0.07	0.38 ± 0.04#	0.33 ± 0.03
CHOL (mg.dL)			69.40 ± 3.48	50.4 ± 5.98#	50.80 ± 3.36
LDH (U/L)			726.50 ± 90.50	1630.00 ± 165.10###	1015.00 ± 109.40**
AST (U/l)			143.80 ± 13.01	1052.00 ± 92.77##	925.20 ± 157.20
ALT (U/l)			57.80 ± 3.76	586.00 ± 92.77#	636.60 ± 92.18
Weight of Liver/B.W (mg.g^{-1})			37.39 ± 0.84	40.46 ± 0.96#	39.68 ± 0.80

Table 4

#P < 0.05**, ##** P < 0.01, ###P < 0.001 vs. Normal group; *P < 0.05 vs. CC4 group.

The hepatoprotective activity of *Lycium* (70% Alcohol extracts) on rats (n = 10, X ± SE)

Animals: Wisatr rats, both of male and female were used.

Extracts: *Lycium* (70% alcohol extracts), Code: LYS-A/7/2000

Dosage and Routine: 0.8 g.Kg^{-1} of *Lycium* was given orally, for 3 day and then 0.5 g.Kg^{-1} for 2 days.

Animal model: Carbon tetrachloride (CCl4) in Sesmon oil (1:3).

Procedure: 30 rats 250-300 g were divided into 3 group, G1 served as normal control and received dilation water, G2 is vehicle control and received dilation water, G3 was given *Lycium* 0.8 g.Kg^{-1}, all animals was given daily treatment orally for 5 days and then treated with CCl4 2.0 (1:3 in Sesmon oil) ml.Kg^{-1} p.o except G1 was given Sesmon oil 2.0 ml. Kg^{-1} p.o. All animals were weight and blood sample were taken from heart 24 hr after CCl4 administration with over dosage of ether. The serum was used for checking TBIL, CHOL, LDH, AST and ALT with biochemical analyzer. The liver weight was also calculated according the body weight.

Conclusion: CCl4 induced the hepotoxicity. It showed TBIL, LDH, AST, ALT and liver weight was increased significantly, while the level of CHOL in serum was decreased. It is showed it can inhibit the abnormal LDH level when 0.8 g.Kg^{-1} of *Lycium* (70% alcohol extracts) was given orally.

Time (Min)	Control Group			Lycium (400 mg/kg)			Lycium (800 mg/kg)		
	SBP	Fine V.	Percentage	SBP	Fine V.	Percentage	SBP	Fine V.	Percentage
Initial	126.67 ± 10.21			122.50 ± 9.64			125.00 ± 14.76		
5 min	121.70 ± 9.09	-5.00 ± 5.00	-3.33 ± 3.33	132.50 ± 11.38	10.00 ± 6.83	8.62 ± 5.55	127.90 ± 13.71	2.86 ± 5.10	3.43 ± 4.65
10 min	123.30 ± 8.03	-3.33 ± 5.58	-1.48 ± 4.12	110.80 ± 15.19	5.00 ± 3.42	4.94 ± 3.66	129.30 ± 14.41	4.29 ± 5.39	4.32 ± 4.73

15 min	128.30 ± 8.33	1.67 ± 5.43	2.41 ± 4.26	125.80 ± 7.79	3.33 ± 3.33	3.70 ± 3.70	134.30 ± 13.82	9.27 ± 4.68	9.64 ± 4.95
30 min	125.00 ± 8.06	-1.67 ± 4.01	-0.37 ± 3.16	127.50 ± 8.34	5.00 ± 3.42	4.93 ± 3.66	134.30 ± 13.82	9.29 ± 4.55	9.64 ± 4.95
60 min	130.00 ± 7.30	3.33 ± 6.15	4.26 ± 5.29	127.50 ± 8.34	5.00 ± 3.42	4.93 ± 3.66	147.10 ± 14.09	22.14 ± 14.05	25.14 ± 15.48

Table 5

The effects of *Lycium* on blood pressure and heart rate in rats

Animals: Wistar rats, both of male and female were used.

Extract: *Lycium* (water-extract), provided by DPS, ZCHRTM

Dosage and Routine: 400-800 mg.Kg^{-1}, ip.

Results are tabulated in the table.

Systolic blood pressure: mm Hg (MEAN ± SE)

120 min	126.70 ± 7.15	0.00 ± 5.16	1.48 ± 4.68	113.00 ± 10.17	12.00 ± 27.75	13.00 ± 12.53	123.60 ± 17.68	-1.43 ± 19.08	3.14 ± 17.01
180 min	126.70 ± 7.15	0.00 ± 5.16	1.48 ± 4.68	113.00 ± 12.38	-7.00 ± 14.55	-2.95 ± 11.89	137.50 ± 17.01	15.00 ± 23.60	19.17 ± 21.54
n	6	6	6	6	6	6	7	7	7

Table 6

Diastolic Blood Pressure (mm Hg)

Time (Min)	Control Group			Lycium (400 mg/kg)			Lycium (800 mg/kg)		
	DBP	Fine V.	Percentage	DBP	Fine V.	Percentage	DBP	Fine V.	Percentage
Initial	80.00 ± 20.43			66.67 ± 5.43			72.85 ± 10.46		
5 min	76.67 ± 6.67	-3.33 ± 3.33	-3.33 ± 3.33	72.50 ± 9.47	5.83 ± 7.79	9.21 ± 13.62	74.29 ± 9.09	1.43 ± 5.53	3.90 ± 6.82
10 min	76.67 ± 6.67	-3.33 ± 3.33	-3.33 ± 3.33	65.83 ± 9.69	-0.83 ± 5.54	-3.21 ± 8.48	75.71 ± 8.41	2.86 ± 5.55	6.79 ± 6.80
15 min	81.67 ± 7.93	1.67 ± 1.67	2.08 ± 2.08	67.50 ± 8.92	0.83 ± 5.23	0.12 ± 7.78	82.14 ± 8.79	9.29 ± 4.56	16.19 ± 6.67
30 min	80.00 ± 7.74	0.00 ± 0.00	0.00 ± 0.00	74.17 ± 8.41	5.83 ± 7.35	9.22 ± 12.54	82.14 ± 8.79	9.29 ± 4.56*	16.19 ± 6.67
60 min	86.67 ± 9.19	66.67 ± 14.94	8.33 ± 6.18	78.33 ± 6.91	10.00 ± 6.71	16.16 ± 11.56	92.86 ± 8.30	20.00 ± 7.15	30.48 ± 14.72

120 min	88.33 ± 9.46	8.33 ± 4.01	10.00 ± 5.00	83.00 ± 9.53	14.00 ± 7.96	20.83 ± 12.38	80..00 ± 9.45	7.14 ± 10.23	14.61 ± 14.08
180 min	90.00 ± 8.16	10.00 ± 3.65	13.33 ± 4.77	78.00 ± 11.33	9.00 ± 10.65	14.17 ± 15.76	80.00 ± 8.17	10.00 ± 6.17	13.20 ± 6.17
n	6	6	6	6	6	6	7	7	7

Table 7

*P < 0.05 VS Control group.

(70% ethanolic extract)

Activity	Results
Effect on Sleeping time-Pentobarbitone sleeping time	Extract significantly increased latency of sleep time
Gross behavioral studies - Tremor/Twitches	No toxic effect
Gross behavioral studies - Writhing	No toxic effect
Gross behavioral studies - Diarrhea, Urination	No diarrhea
Mortality	No death
Motor co-ordination (String and Platform test)	Not affected
Acute toxicity studies	Not toxic
LD_{50} evaluation	-

Table 8

Summary of Results

The plant extract showed antidiarrheal activity that is not free of any toxic symptoms according to the results of the acute toxicity assessment test. The plant extract has failed to produce visible anti-inflammatory, analgesic or n-analgesic effects.

The results of present investigation clearly indicate that *Lycium shawii* exhibited an important antibacterial activity. Thus, the study ascertains the value of fruit which could be of considerable interest to the development of new drugs.

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