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PRELIMINARY PHYTOCHEMICAL SCREENING OF *Hylocereus polyrhizus* PEEL (DRAGON FRUIT)

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Abstract:

Dragon fruit (Hylocereus polyrhizus) or pitaya is an exotic tropical plant that belongs to the family of Cactaceae. Dragon fruit is gaining popularity in the market for consumption of fresh fruit due to its highly attractive fruit colour with nutritional and immense bioactive potential that reduces blood sugar levels, cholesterol, prevention of liver injuries, cancer, etc. It became an interesting subject for many researchers mainly due to its unique taste, shape and the flesh colour. Dragon fruit own a range of beneficial biological activities against pathogenic microbes including bacteria, fungi and viruses, and diseases like diabetes, obesity, hyperlipidaemia and cancer. The red variety of dragon fruit has many pharmacological uses like anticancer, antioxidant, antiulcer, etc. The peel has various pharmacological activities reported but no phytochemicals are reported. Due to lack of this we uptaken to perform the preliminary phytochemical screening tests using 3 solvents maceration extraction procedures (water, water+alcohol-1:1, ethanol). Results have been reported and it is found that Ethanol is the suitable solvent for maximum extraction of pytoconstituents.

Key words: *Hylocereus polyrhizus, tropical fruit, peel, phytochemistry, antioxidant, pharmacological studies, phytoconstituents.*

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INTRODUCTION:

Phytochemicals (Greek: phyton=plant) are chemicals compounds naturally present in the plants attributing to positive or negative health effects. Some of the important phytochemicals include alkaloids, flavonoids, phenolics, tannis, saponnis, steroids, glycosides, terpenes etc. They play a vital role in survival by medication of ecological interactions with competitors, protect them from diseases, pollution, stress, UV rays and also contribute for colour, aroma, and flavor with respect to the plant. The metabolites produced by the plants to protect themselves against biotic and abiotic stresses have turned into medicines that people can use to treat various diseases. (13)

Dragon fruit (*Hylocereus polyrhizus*) or red pitaya is one of the awe-inspiring tropical fruits that belongs to the family of Cactaceae. Pitaya is native to the tropical areas of Mexico, north central and south America, it is now cultivated worldwide due to its commercial interest. (1)

Dragon fruits have been classified based on the color of flesh and peel, *Hylocereus undatus* variety has white flesh with pink skin known as white dragon fruit, flesh of *Hylocereus polyrhizus* was red in color with pink skin known as red pitaya. And *Hylocereus megalanthus* with white pulp and yellow skin know as yellow pitaya. Especially red pitaya *(Hylocereus polyrhizus*) cultivated in Malaysia, Thailand, Vietnam, Australia, Taiwan, and some other parts of world. (1)

The fruit of red pitaya (*Hylocereus polyrhizus*) is oval shape, large in size, weighing about 135-350gm, 10-12cm in size (1,3). The fruit has delicate and sweet flesh with intense red purple color of the flesh and peel. It has a lot of small black seeds which are rich in essential fatty acids. The fruits often consumed fresh or by making into juices, cordial, jams and ice cream by processing it (1). The pigment that is responsible for the red color of the fruit is Betacyanin. (6)

Dragon fruit peel (DFP) which accounts for more than 20% by weight of the whole fresh fruit, is usually discarded as waste during processing. (7)

It is also rich in nutrients and minerals, including vitamin c, B1, B3, dietary fibers, making addition to a health-conscious diet. (6,3)

The peel contains betasianin, flavonoids, and phenol. In addition, dragon fruit skin also contains vitamin C, vitamin E, vitamin A, terpenoids, flavonoids, thiamine, niacin, pyridoxine, cobalamin, phenolic, carotene, and phytoalbumin which are thought to have antioxidant benefits and can also be potential for antimicrobial activities. (12)

In addition, the colorful of plants have become attention of man throughout history for art as well as for food coloring such as *anthocyanin's, betalains, carotenoids, and chlorophylls*. (12)

The peels are mostly waste materials resulting from the dragon fruit juice processing industry and are normally discarded. Thus, in addition to being fed to animals, the peels can be used in the production of pectin, which would then increase the potential return for the dragon fruit juice processing industry. (11)

Pectin is widely used in the food industry as a thickener, emulsifier, texturizer and stabilizer. Pectin is usually added in jams and jellies as a gelling agent. It has also been used as a fat substitute in spreads, ice-cream and salad dressings. (9)

The 3 varieties of pitaya:

1. White-fleshed pitaya with yellow peel (*Selenicereus megalathus*),

2. White-fleshed pitaya with red peel (*Hylocereus undatus*)

3. Red-fleshed pitaya with red peel (*Hylocereus polyrhizus*)

PLANT PROFILE: (3)

Synonym: Selenicereus monacanthus, red pitaya, pitahaya.

Biological source: *Hylocereus polyrhizus* (*weber*) is native to Mexico belongs to *Cactaceae* family.

Geographical source: *Hylocereus polyrhizus* is an exotic fruit grown in tropical and subtropical region of Mexico, but now cultivated worldwide which includes Vietnam, Taiwan, southern China, Israel, and recently in Thailand, Australia, united states, and Malaysia.

In India, dragon fruit is cultivated in Karnataka, Kerala, Tamil Nadu, Maharashtra, Gujarat, Orissa, west Bengal, Andhra Pradesh, and Andaman and Nicobar Islands. (5,6)

BOTANICAL/SCIENTIFIC CLASSIFICATION: (3)

Kingdom: Plantae

Division: Magnoliophyte (Flowering plant)

Class: Liliopsida (Monocotyledons)

Sub-order: Caryophyllidae

Order: Caryophyllales

Family: *Cact*aceae

Genus: *Hylocereus*

Sub- family: Cactoideae

Species: *polyrhizus*

CHEMICAL CONSTITUENTS: (3,4)

- *Fruit contains various phytochemicals such as flavonoids, betacyanin (responsible for red or purple colour), polyphenols, carotenoids and vitamin C. (4)*
- *β-Amyrin (15.87%), α-amyrin (13.90%), octacosane (12.2%), γ-sitosterol (9.35%), octadecane (6.27%), 1-tetracosonal (5.19%), stigmast-4-en-3-one (4.65%), and campestrol (4.16%). (3)*

TRADITIONAL USES: (2)

Pitaya has been used as a traditional medication and consumption purposes in central America, where it is also common for pitayas to be grown in family

gardens. The leaves and flowers of pitaya were used by the ancient Mayas for the medicinal use as a diuretic and healing agent. Mayas also utilizes the pitaya fruits as a diuretic, hypoglycaemic, against heart disease, wound disinfectant, and tumour dissolution, and as a cure for dysentery. In addition, the flowers can be consumed as it is or by drinking it as a tea, the seeds possess a laxative effect, the fruit has shown an effect on gastritis, and the stalk can also be used for kidney problems.

EXTRACTION AND PREPARATION OF EXTRACT:

Separation and drying of peel

The fruit parts are separated into

1.Peel including spines

2. Flesh and they are air dried in shade

Carefully remove peel from the dragon fruit. Place the peel in a well-ventilated area with good air circulation. Allow the peel to dry naturally for 20 days turning it occasionally to ensure even drying.

1. After 20 days of drying the peel of the dragon fruit can be grind separately by taking the dried dragon fruit peel and break it into a smaller piece if necessary.
2. Place the pieces in a grinder.
3. Grind the dried peel until it turns into a fine powder and weighed.
4. Transfer the powdered peal to an airtight container for storage.

Maceration extraction:

- ✓ Maceration involved soaking plant materials (coarse or powdered form) in a stoppered container along with a suitable solvent and let it be stand at room temperature for a period of minimum 3 days with frequent agitation or shaking.
- ✓ This process helps to soften and break the plant's cell wall to release the soluble phytochemicals which are present inside the plant cells. After 3 days, the mixture is pressed or strained by filtration.
- ✓ Three extraction solvents are proposed like water, water: alcohol (50:50), alcohol for the maceration of the DF peel.

Preliminary phytochemical test (13)

- ✓ Various preliminary phytochemical screening tests proposed for/ to identify various classes of phytoconstituents.
- ✓ Like:
- ✓ Alkaloids, Carbohydrates, Detection of reducing sugars, Cardiac glycosides, Proteins and Amino acids, Flavonoids, Phenolic compounds, Tannins, Phlobatannins, Phytosterols, Cholesterol, Terpinoids, Triterpenoids, Diterpinoids, Lignins, Carotenoids, Quinines, Anthraquinones, Anthocyanin's, Leuconthocyanins, Carboxylic aid, Coumarins, Emodins, Gums and Mucilages, Resins, Fixed oils and fat, Volatile oils.

Preparation for extraction:

- The collected peel is initially weighed and dried.
- The initial weight of peel is 202.91gm.
- The dried weight of peel is about 17.98 gm.
- Loss of water content is reported as 91.13%.
- The powder is divided into equal amount for 3 parts then kept for maceration with 3 solvents.

1. 5.99 gm in 50 ml of water (water is added eventually due to absorption of water by peel
2. 5.99 gm in 50 ml alcohol (preferably ethanol)
3. 5.99 gm in 1:1 ratio of water and alcohol

PRELIMINARY PHYTOCHEMICAL SCREENING OF VARIOUS EXTRACTS (13)

Detection of alkaloids

TEST	PROCEDURE	OBSERVATION
Hager's test	Few ml of filterate+1-2 ml of Hager's reagent	A creamy white precipitate
Mayer's/ Bertrand's/ Valser's test	Few ml of filterate+1-2 drops of Mayer's reagent (along the walls of test tube)	A creamy white or yellow precipitate
Wagner's test	Few ml of filterate+1-2 drops of Wagner's reagent (along the walls of test tube)	A brown or red dish precipitate

Detection of carbohydrates

TEST	PROCEDURE	OBSERVATION
Barfoed's test	1ml filtrate+1ml Barfoed's reagent+ heated for 2 mins	A red precipitate (monosaccharides)

Detection of reducing sugars

TEST	PROCEDURE	OBSERVATON
Fehling's test	1ml each of Fehling's solution A & B +1ml filtrate+ boiled for 2 mins.	A red precipitate

Detection of phenolic compounds

TEST	PROCEDURE	OBSERVATION
Lead acetate test	Plant extract+3ml of 10% lead acetate solution	A white precipitate

Detection of Glycosides

TEST	PROCEDURE	OBSERVATION
Aq. NaOH test	Alcoholic extract +dissolved in 1 ml of water+ few drops of aq. NaOH solution	A yellow color

Detection of cardiac glycosides

TEST	PROCEDURE	OBSERVATION
Baljet test	2 ml extract+ a drop of baljet's reagent	A yellow- orange color

Detection of proteins and amino acids

TEST	PROCEDURE	OBSERVATION
Xanthoproteic test	Plant extract + few drops of conc. Nitric acid	A yellow colored solution.

Detection of flavonoids

TEST	PROCEDURE	OBSERVATION
Alkaline reagent test	1ml extract +2ml of 2%NaOH sol+ (few drops of dil. HCL)	An intense yellow color becomes colorless on addition of dil. acid
Conc. H2SO4 test	Plant extract+ Conc. H2SO4	An orange color

Detection of phlobatannins

TEST	PROCEDURE	OBSERVATION
HCL test	2ml aq. Extract +2ml 1%HCL (boiled)	A red precipitate

Detection of saponins

TEST	PROCEDURE	OBSERVATION
NaHCO3 test	plant extract +few ml of NaHCO3 sol.+ distilled water (vigorously shaken)	Stable honey comb like froth

Detection of carotenoids

TEST	PROCEDURE	OBSERVATION
Test for carotenoids	Extract + few drops of chloroform (vigorously shaken and filtered). Filtrate+ conc. H2SO4	A blue color at the interface.

Detection of tannins

TEST	PROCEDURE	OBSERVATION
10%NaOH test	Plant extract +4ml 10%NaOH + shaken well	Formation of emulsion (Hydrolysable tannins)
Bromine water test	Bromine water + plant extract	Decoloration of Bromine

Detection of triterpinoids

TEST	PROCEDURE	OBSERVATION
Salkowski's test	Filtrate + Few drops of Conc.H2SO4 (Shaken well & allowed to stand)	Golden yellow layer (At the bottom)

Detection of phytosterols

TEST	PROCEDURE	OBSERVATION
Salkowski's test	Filtrate + Few drops of Conc.H2SO4 (Shaken well & allowed to stand)	Red color in lower layer
Hesse's Response	Aq. Extract+2ml chloroform + 2mlConc. H2SO4	Pink ring/Red color (in lower chloroform layer)

Detection of cholesterol

TEST	PROCEDURE	OBSERVATION
	2ml extract + 2ml chloroform + 10 drops of acetic anhydride+2-3 drops of Conc. H2SO4	A red- rose color

Detection of terpinoids

TEST	PROCEDURE	OBSERVATION
	2ml chloroform + plant extract (evaporated on water bath) +3ml Conc.H2SO4(boiled on water bath)	A grey colored solution

Detection of lignin's

TEST	PROCEDURE	OBSERVATION
Labat test	Extract solution + Gallic acid	An olive green color

Detection of diterpenes

TEST	PROCEDURE	OBSERVATION
Copper acetate test	Plant extract+ 3-4 drops of copper acetate solution.	Emerald green color

Detection of Quinone's

TEST	PROCEDURE	OBSERVATION
Conc. HCL test	Plant extract +Conc. HCL	A green color

Detection of Anthraquinones

TEST	PROCEDURE	OBSERVATION
Borntrager's test	10ml 10%ammonia sol.+ few ml of filtrate (shaken vigorously for 30 sec)	A pink, violet, or red colored solution

Detection of fats and fixed oils

TEST	PROCEDURE	OBSERVATION
	Extract solution is applied on filter paper	A transparent appearance (oils &resins)

Detection of leuconthocyanins

TEST	PROCEDURE	OBSERVATION
Isoamyl alcohol test	5ml plant extract+5ml isoamyl alcohol	Upper layer appears red

Detection of carboxylic acids

TEST	PROCEDURE	OBSERVATION
Effervescence test	1ml plant extract +1ml sodium bicarbonate solution	Appearance of Effervescence

Detection of coumarins

TEST	PROCEDURE	OBSERVATION
NaOH test	Plant extract+10% NaOH +Chloroform	A yellow color

Detection of emodins

TEST	PROCEDURE	OBSERVATION
	Plant extract+2mlNH4OH+3ml benzene	A red color

Detection of gums and mucilage's

TEST	PROCEDURE	OBSERVATION
Alcohol test	Plant extract+ few ml of alcohol (constant stirring)	White or cloudy precipitate

Detection of resins

TEST	PROCEDURE	OBSERVATION
Acetic anhydride test	Plant extract+ Acetic anhydride solution+1ml Conc. H2SO4	Orange to yellow

RESULTS:

The solvents of water and mixture of water –alcohol (1:1) and alcohol has been tested. The following data gives the information about the presence of phytoconstituents in the peel of dragon fruit (*Hylocereus polyrhizus*) in the respective extracts.

Phytoconstituents	**Test**	**Water solvent**	**Water + Ethanol (1:1)**	**Ethanol Solvent**
ALKALOIDS	Hager's test	+ (Present)	+ (Present)	- (absent)
	Mayer's test	+ (Present)	+ (Present)	+ (Present)
	Wagner's test	+ (Present)	+ (Present)	+ (Present)
TANNINS	10%NaOH test	- (absent)	- (absent)	- (absent)
	Bromine water test	+ (Present)	- (absent)	- (absent)
REDUCING SUGAR	Fehling's test	- (absent)	- (absent)	- (absent)
GLYCOSIDES	Aq. NaOH test	- (absent)	+ (Present)	+ (Present)
CARDIAC GLYCOSIDES	Baljet test	+ (Present)	+ (Present)	+ (Present)
FLAVONOIDS	Alkaline reagent test	+ (Present)	+ (Present)	- (absent)
	Conc. H2SO4 test	+ (Present)	+ (Present)	+ (Present)
PHENOLIC COMPOUNDS	Lead acetate test	+ (Present)	+ (Present)	+ (Present)
TERPINOIDS		- (absent)	- (absent)	+ (Present)
TRITERPINOIDS	Salkowski's test	- (absent)	- (absent)	- (absent)
DITERPENES	Copper acetate test	+ (Present)	+ (Present)	+ (Present)

LIGNINS	Labat test	- (absent)	- (absent)	- (absent)
QUINONES	Conc. HCL test	- (absent)	- (absent)	- (absent)
ANTHRAQUINONES	Borntrager's test	- (absent)	- (absent)	- (absent)
CARBOHYDRATES	Barfoed's test	- (absent)	- (absent)	- (absent)
CARBOXYLIC ACID	Effervescence test	+ (Present)	+ (Present)	- (absent)
EMODINS		- (absent)	- (absent)	- (absent)
GUMS &MUCILAGES	Alcoholic test	+ (Present)	+ (Present)	+ (Present)
RESINS	Acetic anhydride test	+ (Present)	+ (Present)	+ (Present)
FIXED OILS & FATS		- (absent)	- (absent)	- (absent)
LEUCONTHOCYANINS	Isoamyl alcohol test	- (absent)	- (absent)	----------
COUMARINS	NaOH test	- (absent)	+ (Present)	+ (Present)
PHYTOSTEROLS	Salkowski's test	- (absent)	- (absent)	+ (Present)
	Hesse's response	- (absent)	- (absent)	- (absent)
CHOLESTROL		- (absent)	- (absent)	- (absent)
CAROTENIODS	Test for carotenoids	- (absent)	- (absent)	+ (Present)
PHLOBATANNINS	HCL test	- (absent)	- (absent)	- (absent)
PROTIENS &AMINO ACIDS	Xanthoproteic test	- (absent)	- (absent)	----------
SAPONINS	NaHCO3 Test	+ (Present)	+ (Present)	+ (Present)

DISCUSSION:

The Extraction carried out using different solvents for Dragon fruit peel (red pitaya).

- It is found that the extract range of phytoconstituents by using water as solvent. The phytoconstituents are- Alkaloids, Tannins, Cardiac glycosides, Flavonoids, Phenolic compounds, Saponins, Diterpenes, Carboxylic acids, Gums & mucilage, Resins.
- It is also found that the extract range of phytoconstituents by using water + alcohol(ethanol) as solvent. The phytoconstituents are- Alkaloids, Cardiac glycosides, Flavonoids, Phenolic compounds, Saponins, Diterpenes, Coumarins, Carboxylic acids, Gums & mucilage, Resins.
- It is found that the extract range of phytoconstituents by using Ethanol as a solvent. The phytoconstituents are -Alkaloids, Glycosides, Cardiac glycosides, Flavonoids, Phenolic compounds, Saponins, Phytosterols, Terpinoids, Diterpenes, Carotenoids, Coumarins, Gums &Mucilage, Resins.
- This work helps in selection of suitable solvents for maximum extraction of phytoconstituents.

CONCLUSION:

This work reveals the presence of specific class of phytoconstituents in common of 3 solvents like- Alkaloids, Cardiac glycosides, Flavonoids, Phenolic compounds, Saponins, Diterpenes, Carboxylic acids, Gums & mucilage, Resins.

Respective solvents found a specific class of compounds, they are

Water as solvent-tannins

Water +alcohol-coumarins

Alcohol-terpinoids, phytosterols, glycosides, carotenoids.

As there are many pharmacological activities reported. This work supports the correlation of chemical compounds to pharmacological activities.

Through this work we made an account to give complete phytochemical profile for the class of chemical compounds present in the peel.

It is found that ethanol is the best solvent for maximum extraction of phytoconstituents.

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