



Received on 19 November, 2017; received in revised form, 11 January, 2018; accepted, 13 February, 2018; published 01 April, 2018

A SHORT REVIEW ON CARNIVOROUS PLANTS AND RECENT DEVELOPMENTS IN THE FIELD OF CANCER RESEARCH

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Keywords:

Drosera, Dionea,
Metabolic Profiling, *In vitro*
culture, Insectivorous plants

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ABSTRACT: Nature has been providing many essential sources which are being utilized in every form of our lives. There are many novel techniques which are being developed from natural resources most importantly plants. By this review we tried to prove the efficiency of Carnivorous plants like *Dionaea muscipula*, *Drosera indica* in preventing varied types of cancers. We also tried to include various metabolites which are extracted through *in vitro* culture techniques. Some of the plants related to this type such as *Dionaea muscipula*, *Drosera indica*, have the capacity to cure cancer which is one of the most dangerous disease and due to which millions of people are dying every year. Some of the plants of this variety such as *Dionea* and *Drosera species* have essential metabolites which are extracted through *in vitro* culture techniques. In this review we also tried to compare various metabolites which are extracted from different carnivorous plants. We also tried to discuss some of the recent research developments made in the area of anticancer drugs.

INTRODUCTION: This review article is said to be prepared on the purpose of evaluation of metabolites and anti cancer properties present in carnivorous plants. There are various secretory glands such as alluring glands, mucilage glands and digestive glands which are helpful in trapping insects and other microbes which are essential for their survival. Carnivorous plants are those type of plants which derive most of their nutrients from animals primarily insects or protozoan. They are said to be one of those group of plants which are not self dependent. Carnivorous plants grow mostly in tropical areas of the world.

They grow mostly in the areas where soil is thin layered or poor in nutrients. There are different types of trapping mechanisms such as pitfall traps, flypaper traps, snap traps *etc.*, which decide the type of secretion of these plants used for trapping insects or any other animal for consumption. These traps would be active or passive, depending on whether movement aids the capture of prey.

Carnivorous plants are considered to be herbs and their traps are said to be produced by primary growth. In *Drosera species*, the stalked glands or tentacles secrete both mucilage and enzymes and function also in absorption of the digestive products. Multi cellular alluring glands of epidermal origin are present on the under surface of the lids of the pitcher plant which secrete nectar. These glands help in attracting the insects. The bladderwort which is also known as *utricularia* contains very much specialised types of trichomes.

<p>QUICK RESPONSE CODE</p> 	<p>DOI: 10.13040/IJPSR.0975-8232.IJP.5(4).205-12</p> <p>Article can be accessed online on: www.ijpjournals.com</p>
<p>DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.IJP.5(4).205-12</p>	

On the inner side of the trap are present four-armed (quadrifid) and two-armed (bifid) hairs. In the outside part of the plant there are dome shaped external glands and closely arranged epithelial cells lining the threshold of the doorway. The evolution of carnivorous plants is obscured by the paucity of

their fossil record. Most of the carnivorous plants live in waterlogged conditions and low nitrogen and phosphorous soil content conditions. Here are some of those valuable insectivorous plants which are rich in metabolites which help in cure of various diseases:



FIG. 1: DROSERA



FIG. 2: DIONAEA



FIG. 3: DARLINGTONIA



FIG. 4: SARRACINEA

Drosera Plant Species:

Bio Inspired Hydrogel for Catching and Killing Cancer Cells: This modern world has highly developed materials with multiple functions that can be performed synergistically or sequentially. The recent study on Drosera performed by Shihui Li¹, a carnivorous plant was found to be functionally similar to the bioinspired hydrogel that can interact with cancer cells (catching and killing prey)¹. This hydrogel is bilayered in which one layer consists of oligonucleotides and the other with double stranded DNA.

The study concluded that the top layer was able to trap the target cells and the bottom layer could sequester Doxorubicin (DOX) that sustains drug release¹. This released DOX plays a vital role in killing about 90% of cells after the residence on cells of hydrogel. Therefore, the study

demonstrated the potential of bioinspired hydrogel in attracting, catching and killing diseased cells or invading microorganisms like tumor cancer cells and bacteria¹. In Drosera the tentacles of leaves have adhesive secretions that helps to trap the prey. Along with this, the plant also releases few digestive enzymes such as esterase, peroxidase, proteases on its leaves which helps to capture, kill and digest the prey¹. Hence, when there are circulating diseased cells or microorganisms like cancer cells and bacteria in the human body the development of novel hydrogel with target-catching and drug-releasing functions is helpful.

As discussed the top layer of hydrogel has nucleic acids and the bottom layer has double stranded DNA which is an affinity site for sequestration of small toxic drugs. Thus, this hydrogel can be locally released in a sustainable manner. This

bifunctional hydrogel is synthesized by a two step free radical polymerization. During the polymerization, the immobilized DNA (ID₁, ID₂) are incorporated in the top and bottom layers of hydrogel¹. This chemical incorporation of ID's into hydrogel and intermolecular hybridizations between them are illustrated by treating hydrogel with FAM-CD₁ and Cy5.5-CD₂¹, washed and examined under fluorescence microscope, The hydrogels prepared with acrydite shows negligence fluorescence signals of FAM / Cy 5.5 whereas the hydrogels prepared with acrydite exhibited strong fluorophore signals.

***Drosera indica* Linn.:**

Potential effect on liver enzyme, lipid profile and hormone change in Dalton's lymphoma ascites (DLA) bearing mice:

This study was mainly carried out to observe the effect of ethanol and aqueous extracts of *Drosera indica* L. in mice using Dalton's lymphoma ascites cells (DLA) which is performed by Raju Asirvatham *et al.*,³. Cancer induced liver enzyme, lipid profile and hormonal variations were also studied along with the preparations of ethanol and aqueous extracts. Firstly animals were divided into seven groups and each group was designated as the normal control, DLA control, standard (5FU) and the ethanol and aqueous extracts (250 and 500 mg/kg each) of *D. indica* L. + DLA (four groups)³ were given the respective treatments 24 h after tumor cell inoculation, for 14 days. The doses of ethanol and aqueous extracts of *D. indica* at 250 and 500 mg/kg showed significant effects on elevated liver enzyme, lipid profile and hormonal variations.

Cure for cancer has always been done by utilization of metabolic syndrome which often leads to several side effects such as elevated triglycerides, reduced high density lipoprotein (HDL), low testosterone levels and overall connection with sex hormones³. Thus in order to prevent such type of problems Ayurvedic therapy has been a solution which involves only plant extracts to cure any type of disease. Alcoholic extract was prepared firstly by weighing a quantity of air powdered drug which was extracted with ethanol in Soxhlet apparatus. Then after extraction, the extract was concentrated in a rotary flash evaporator at a temperature not exceeding 50 °C. The ethanol extract was suspended in distilled water for experimental

purposes. Similarly for preparing the aqueous extract, the marc from the ethanol extract was made to undergo maceration process with chloroform-water for 24 h to obtain the aqueous extract.

Aqueous extract was concentrated under vacuum and was dissolved in distilled water for experimental studies. The ethanol (EEDI) and aqueous (AEDI) extracts of *D. indica* L. were then stored in air tight container, The Swiss Albino mice was used as a host in inducing cancer through DLA cells³. In order to know the hormonal levels in female mice blood, RIA method was used in this experiment. Cholesterol, HDL cholesterol, Triglyceride levels were recorded and studied in this experiment. Serum enzymes like aspartate amino transferase (AST), alanine amino transferase (ALT), alkaline phosphatase (ALP) and lactate dehydrogenase (LDH) were also evaluated in this study³.

When the alcoholic and aqueous extracts were administered into the body at high levels of concentration there was no sign of any toxicity manifestation which proved that it was safe and healthy to use which was an unique factor which differentiates this from other metabolic drugs. The wider fluctuation of serum hormonal levels of female mice was normalized after addition of ethanol and aqueous extracts at required doses. Thus this study on alcoholic and aqueous extracts of *D. indica* proved to be a novel solution for the treatment of cancer without any involvement of side effects.

***Dionaea* Species:**

Anti - Cancerous Secondary metabolites present in Venus flytrap (*Dionaea muscipula* Solander ex Ellis):

In this study on evaluation of the secondary metabolites of Venus fly trap performed by François Gaascht *et al.*,⁴ discusses about the importance of chemoprevention process which utilizes synthetic or natural molecules without toxic effects to block infectious diseases most importantly cancer from spreading in our body. Most of the natural molecules used in chemoprevention help by modulating mitotic signals involved in cell survival, apoptosis, cell cycle regulation, angiogenesis, or on processes involved in the development of metastasis which occur naturally, especially in fruits, vegetables and

also in non-comestible plants. The secondary metabolites of this plant are very much essential for the survival in an indirect manner. Various metabolites like naphthoquinones (plumbagin and its derivatives), phenolic acids (ellagic acid, gallic acid, vanillin, protocatechuic acid, caffeic acid, chlorogenic acid, ferulic acid and salicylic acids) and flavonoids (quercetin, myricetin and kaempferol) are present in this plant⁴. Plumbagin which is also known as yellow naphthoquinone is said to be having antibacterial, antifungal, anti-inflammatory, and anticancer properties. In this plant plumbagin has a protective action from predators and parasites. This particular naphthoquinone acts as an inhibitor in the case of activated NF-kB (Nuclear Factor-kappa B) signaling pathway induced by carcinogens which proves its anticancer causing nature. In the case of H460 lung cancer cells,⁴ plumbagin increases the expression of p53 and p21, which leads to cell cycle arrest in G2/M and triggers death by apoptosis which leads to cure of lung cancer in the effected patient⁴.

Several *in-vivo* experiments on mice have proved that plumbagin helps in preventing growth of tumor in humans by inhibiting the expression of several markers like MMP-9, 2, and VEGF in ovarian and prostate-cancer cells. Ellagic acid of phenolic acids acts as a chemo preventive agent as it reduces cell proliferation and inhibits NF-kB by interfering with the binding of this transcription factor to DNA, which leads to apoptosis of pancreatic cancer cells by cytochrome c release and activation of caspase - 3⁴. Ellagic acid also helps to reduce prostate carcinoma by growth of PC3 cells in a dose dependent manner which triggers apoptosis⁴. Gallic acid is a widely used Anti cancer agent which has several cellular targets. Vanillin which is the most widely used flavonoid at its non toxic concentrations inhibits growth of mammary adeno carcinoma cell line 4T1 but also decreases MMP-9 activity and thus reduces cell migration and invasion. At concentrations of 400 and 1000 mg/L, vanillin induces apoptosis in HT-29 human colorectal cancer cell line and NIH/3T3 normal cell lines.

Caffeic acid is a well known anti-inflammatory agent which decreases expression of IL-8 and NF-kB pathway activity by triggering TNF-alpha-induced IκB degradation that further leads to a

reduction of NF-kB target genes expression which are regularly involved in carcinogenesis⁴. Chlorogenic acid induces apoptosis by inducing ROS generation which leads to reduction of the mitochondrial membrane potential in U937 human leukemia cells. Chlorogenic acid has the capability of destroying A549 lung cancer cells which clearly signifies the anti cancerous property of this phenolic acid. *In vivo* studies made on mammary carcinogens induced Sprague- Dawley rats which were fed with ferulic acid showed immense drop in tumor develop in 80% of animals on which the experiment was performed. Aspirin when taken regularly in a dose of 75 mg per day for several years may have the capacity of preventing colorectal cancer from attacking our body.

Quercetin one of the flavonoids produced from this plant is widely used as antibacterial agent. Quercetin is also known to induce cell cycle arrest in G2/M and to induce cell death in human HeLa cervical cancer cells by mitochondrial apoptosis through a p53-dependent mechanism which proves its Anti- cancerous nature. Myricetin which is widely known for its antibacterial and anticancer properties as it helps in inhibition mutagenesis of organic compounds such as benzo(a)pyrene. This flavonoid also leads to apoptosis of pancreatic cancer cells by activating caspase-3 and caspase-9⁴. Lower concentrations of kaempferol which is another type of flavonoid present in this plant which helps in inhibiting the proliferation of oral cancer cell lines such as SCC-1483, SCC-25, and SCC-QLL1.

Metabolic Profiling of *Darlingtonia* and *Sarracenia Carnivorous Plants*: Sarraceniaceae, a new carnivorous plant family mainly comprising of three genera namely, *Darlingtonia* Torr, *Heliophora* Benth, *Sarracenia* L.². The sarraceniaceae members are characterized by the presence of large number of diverse metabolites [about 600 metabolites] in lids as well as pitchers. Coniine, a metabolite, was recently found in seven sarracenia species. Integrating the polygenetic information of sarraceniaceae, the study done by Hannu Hotti¹ *et al.*, concluded that the metabolic composition of the plant can be demonstrated by the phylogeny which explains the absence and presence of the compounds².

The metabolic profiles lids and pitcher and lids are analysed separately, concluding that each plant lid and the pitcher contains about 48 compounds each in total. The biosynthesis of coniine enhances the insect attraction and retention.

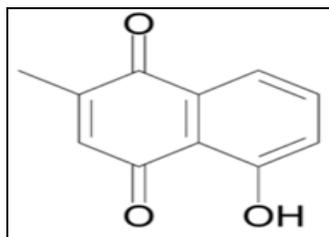
Recent Developments in the Area of Cancer Research:

Cancer is caused to abnormal growth of cells which spreads to all parts of the body in no time. There are around 100 types of cancer diseases out of which one fourth of cancer deaths are due to intake of tobacco by humans. Infection caused in the body may also become one of the symptoms of cancer. Large amount of consumption of alcohol may also lead to cancer. Lung cancer, cervical cancer and colorectal cancer are one of the most common types of cancers which are affecting humans. Around 15% of deaths out of total cancer patients are occurring every year. Radiation therapy, chemotherapy are helping only one section of people who can afford financially and others who cannot have no other option but death. This is the present scenario of cancer patients which we are encountering in the World. Immunotherapy has been developing to a larger extent from past decade. New study identifies that cells are being detected with weak immune response before the initiation of treatment for cancer cells with the help

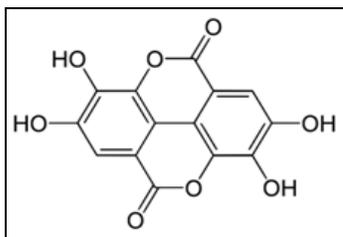
of biomarkers. This study helped inhibitors to activate weak immune cells which would detect cancer cells as foreign bodies and would try to eliminate them. Cytometry by time of flight (CyTOF) cell analysis method helps in analyzing 50 different proteins in each cell at a time and thus this helps in identifying activation status of every cell of our body. Immunotherapy analysis helps in early treatment for patients suffering with melanoma and lung cancer.

Pan beta blockers which are least prescribed and helps in treatment of heart attacks and also in prevention of lowering blood pressure in patients when given during immunotherapy helps in effectively eradicating melanoma cancer cells.

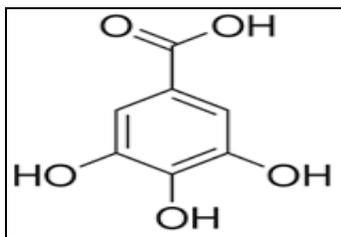
This study states that patients who were treated with immunotherapy along with beta blockers lived longer than those patients who were treated only with immunotherapy. Radiation therapy when given to the patients in different doses help in reducing 20% of side effects caused due to this therapy which could not be reduced by conventional radiation therapy. Fractionation is the principle which are involved in reducing side effects of radiation therapy which are helpful in preventing cancer cells.



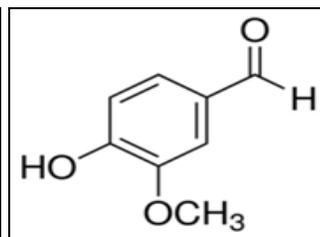
(A) PLUMBAGIN



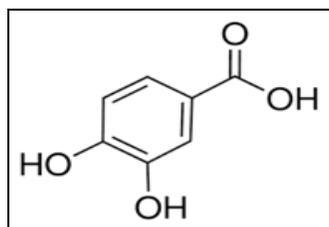
(B) ELLAGIC ACID



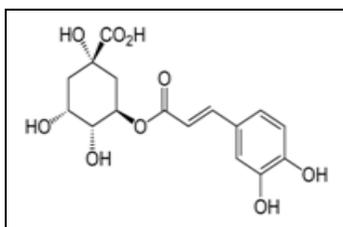
(C) GALLIC ACID



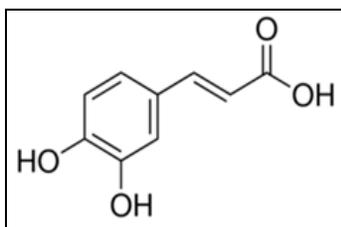
(D) VANILLIN



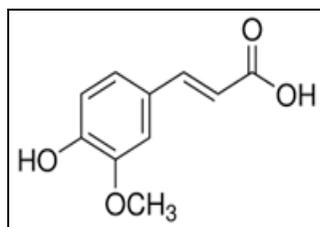
(E) PROTO CATECHUIC ACID



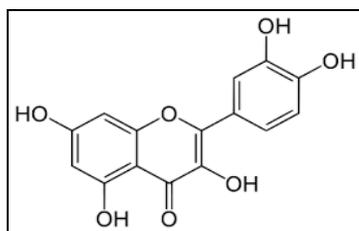
(F) CHLOROGENIC ACID



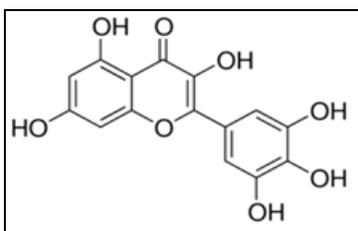
(G) CAFFEIC ACID



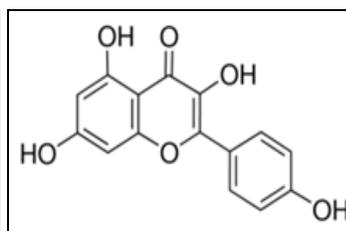
(H) FERULIC ACID



(I) QUERCETIN



(J) MYRICETIN



(K) KAEMPFEROL

CONCLUSION: From this review article we have found that insectivorous plants have been found to be rich in secondary metabolites which are very much useful in treatment of cancer. There are many secondary metabolites identified in plants such as *Drosera indica*, *Dionaea muscipula*, *Darlingtonia* and *Sarracenia* which possess anti cancer property. Metabolites like naphthoquinones, phenolic acids, flavonoids are present in these insectivorous plants. Quercetin helps in cell cycle arrest and induce cell death in HeLa cervical cancer cells by mitochondrial apoptosis. Chlorogenic acid has the property of killing lung cancer cells. Vanillin helps in apoptosis of cervical cancer cells. Caffeic acid acts as an anti inflammatory agent. Ellagic acid shows apoptosis of pancreatic cancer cells due to release of cytochrome c and activation of caspase3. Quercetin can also be used as an antibacterial agent. Both alcoholic and aqueous extracts of *D. indica* have been proved to be most safe and best solution to kill cancer cells as they do not show any side effects. From this review we also tried to brief about some of the recent developments of Cancer research mainly on biomarkers, pan beta blockers and radiation immunotherapy. Doxorubicin which is released from bottom layer of hydrogel which is functionally similar to *Drosera species* has the capability in killing 90% of cancer cells. Thus by all the results obtained from these carnivorous plants prove these to be as a valuable source to treat various threatening diseases like cancer and it is very much important in future for pharmaceutical field.

ACKNOWLEDGEMENT: Nil

CONFLICT OF INTEREST: Nil

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How to cite this article:

Rahaman ST and Sai PR: A short review on carnivorous plants and recent developments in the field of cancer research. *Int J Pharmacognosy* 2018; 5(4): 205-12. doi link: [http://dx.doi.org/10.13040/IJPSR.0975-8232.IJP.5\(4\).205-12](http://dx.doi.org/10.13040/IJPSR.0975-8232.IJP.5(4).205-12).

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