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Medicinal Properties of Phytochemicals and Their Production

Aanchal Bansal and Chinmayee Priyadarsini

Abstract

Phytochemicals are produced by plants as a defence mechanism against pathogens. They are used to treat various metabolic, immunological and neurological disorders in humans in various parts of the world as a part of traditional medicine. The use of indigenous plants in commercial medicine is rising with increasing population. The antimicrobial properties of plant extracts led to increased demands. Plant tissue culture on the other hand, has proved to be a reliable alternative for the production of bioactive compounds from plants. Artificial plant culture can enhance the production of phytochemicals in medicinal plants. This review focuses on the medicinal properties of phytochemicals and their in-vitro production.

Keywords: plants, health, phytochemicals, antimicrobial, metabolic, in-vitro

1. Introduction

In the modern era of medicine, plants are still used as traditional mode of healthcare against certain disorders [1]. Plants can protect themselves from pathogenic microorganisms, harmful insects and adverse environmental changes by producing certain chemicals or secondary metabolites which are non-nutritive [2], but useful in defence mechanism. These are known as Phytochemicals, and somewhat essential oils. It can not only protect plants, but also humans and animals against certain diseases which are either caused by microorganisms or toxins produced by the microorganism. This is due to its antimicrobial property [3]. In future, phytochemicals can be used as chemo-preventive agents [4]. Till date, a number of phytochemicals have been discovered based on difference in chemical structure and have been classified as major groups [5]. The major groups of phytochemicals are phytosterols, flavonoids, terpenoids, saponins, alkaloids, carotenoids, aromatic acid, organic acid, essential oils and protease inhibitors [6]. Due to certain properties like antimicrobial, anti-inflammatory, anthelmintic, anticarcinogenic, antigenotoxic, antiproliferative, antimutagenic and antioxidative, the metabolites can provide direct or indirect defensive mechanism against pathogens or harmful ailments [7] (**Figure 1**).

Figure 1.
Medicinal properties of phytochemicals.

2. Antimicrobial properties

2.1 Antibacterial

Helicobacter pylori colonises on the epithelial layer of gastric mucosa and cause peptic ulcers and adenocarcinoma of distal stomach. Successful treatment of *H. pylori* infection has been observed by combination treatment of a proton pump inhibitor with two antibiotics. The bacterium has evolved and become resistant to the antibiotics [8]. Thus, there is a need to find alternative to current antibiotics. Recently, certain plant extracts and substances have been isolated such as alkaloids, polysaccharides and flavonoids which have shown effective cure against *H. pylori* infection. *Daucus carota* (carrot) seed oil has been found most effective against *H. pylori* in vitro [9].

Mycoplasmas are microorganisms lacking a rigid cell wall. Generally, their physiological habitats are plants and animals, but they can cause infection to humans. Furneri et al. [10] in his recent experiment has exposed 25 clinically isolated strains to TTO (Tea Tree Oil). They used broth microdilution assay to determine the MIC values.

Diseases like pneumonia, sinusitis, bronchitis, tonsillitis and viral infections such as common cold develop due to bacterial infection in the respiratory tract and the microorganisms commonly related to this infection are *Streptococcus pneumoniae*, *Streptococcus pyogenes*, *Haemophilus influenzae* and *Moraxella catarrhalis*. Traditionally, essential oils have been in use for the treatment of respiratory tract infections. They are either inhaled by steam, administered orally or applied by rubbing on the chest due to its secretolytic and secretomotoric properties. Except *Streptococcus pyogenes*, all other respiratory tract infecting microorganisms are susceptible to essential oils extracted (*in vitro*) from lemon grass (*Cymbopogon citratus*), lemon balm (*Melissa officinalis*), cinnamon bark (*Cinnamomum verum*) and thyme (*Thymus vulgaris*). Essential oils from peppermint (*Mentha piperita*) and eucalyptus (*Eucalyptus globulus*) can also be used but it shows low activity. Most of the active essential oils showed antibacterial effect at a concentration ranging from 1.56 to 6.25 µg/ml in its gaseous phase [11]. Thus, these can be used to treat the infection by inhalation. Tea tree oil (TTO) is used worldwide in sectors including skin care cosmetics, nursing, and for successful treatment of bacterial

and fungal infections [12]. It showed high antibacterial activity against *S. aureus* both *in vitro* and *in vivo* [13].

2.2 Antiviral

Natural products in the form of pure compound or as a plant extract can be used as antiviral drugs due to unmatched availability of chemicals [14]. Besides certain chemicals, natural products can be used as novel therapeutic agents for treatment of various diseases including viral infections. Viral infections have resisted prophylaxis as compared to than other microorganisms which is of major concern worldwide. Currently, very few antiviral medicines are available and there is a need to find new substances showing both extracellular and intracellular antiviral properties. The parameters commonly taken into consideration during evaluation of antiviral property of a substance (may be natural or synthetic) are reduction in the virus yield, inhibition of cytopathic effects, reduction or inhibition of plaque formation, and other viral functions in selected host cell cultures.

Scientific evidences have shown human viral infections can be treated by plant-derived phyto-antiviral agents produced *in vitro* [8]. To investigate the antiviral activity of essential oils, they were tested against enveloped and non-enveloped RNA and DNA viruses. Most of the tests were performed against the former such as herpes simplex virus type 1 and type 2 (DNA viruses), dengue virus type 2 (RNA virus), influenza virus (RNA virus) and Junin virus (RNA virus). Only a few essential oils such as oregano (*Origanum vulgare*) oil and clove (*Syzygium aromaticum*) oil were tested against non-enveloped viruses such as adenovirus type 3 (DNA virus), poliovirus (RNA virus), and coxsackievirus B1 (RNA virus).

Herpes simplex virus type 1 (HSV-1) causes infections such as herpetic encephalitis, herpetic keratitis, mucocutaneous herpes infections and neonatal herpes. For the treatment of this infection Acyclovir is used which is a nucleoside analogue and a selective anti-herpetic agent. It inhibits the replication of viral DNA through viral thymidine kinase, inhibition its synthesis. But, resistant HSV strains against this drug have been isolate from immunosuppressed hosts, such as patients suffering from AIDs and malignancy, and patients undergoing bone marrow or organ transplants [15]. Recent researches have demonstrated the antiviral effect of certain essential oils against these resistant HSV strains [16].

Currently, Coronavirus disease of 2019 (COVID-19) is a global threat. Unfortunately, very limited drugs have shown effectiveness against SARs-CoV-2 virus and its inflammatory complications [17]. Combinations of certain medicines are used such as antiviral (remdesivir), dexamethasone, antimalarials (chloroquine/hydroxychloroquine), and IL-6 receptor blocking monoclonal antibodies (tocilizumab), for the treatment of the disease. Studies have proposed essential oils showing activity against the SARs-CoV-2 virus due to various properties such as antiviral, anti-inflammatory, broncho dilatory, immuno-modulatory. Since essential oils have lipophilic nature, it can easily penetrate the viral membrane causing its membrane disruption. Essential oils also contain multiple active phytochemicals which can act on multiple viral replication stages inducing positive effect on the host's respiratory system including lysis of mucus and bronchodilation. Thus, a combination of chemo-herbal essential oils could be feasible and effective to combat the pandemic virus.

2.3 Antifungal

Phytochemicals are antifungal as they can induce cytotoxicity in fungi by disrupting the membrane permeability of cell; inhibiting enzymes involved in

Effect	Microbial species	Plant extract	References
Antibacterial	<i>Helicobacter pylori</i>	<i>Daucus carota</i> (carrot) seed oil	Bergonzelli et al. [9]
	<i>Mycoplasmas</i>	Tea tree oil	Furneri et al. [10]
	<i>Streptococcus pneumoniae</i> , <i>Streptococcus pyogenes</i> , <i>Haemophilus influenzae</i> and <i>Moraxella catarrhalis</i>	Essential oils	Inouye et al. [1]
Antiviral	Herpes simplex virus type 1 (HSV-1)	Essential oils	Schnitzler et al. [16]
	Adenovirus, poliovirus	Oregano oil, clove oil	Reichling et al. [8]
	Coronavirus	Essential oils	Asif et al. [17]
Antifungal	<i>Aspergillus</i> , <i>Fusarium</i>	Wild stevia extract Curcumin and ellagic acid	Abdel-Fattah et al. [19] Gowda et al. [20]
	Zearalenone producing species	Lycopene	Aydin et al. [21]

Table 1.
Antimicrobial properties of phytochemicals.

mitochondria, cytoplasm and cell wall synthesis; and altering the cell compartment, and redox and osmotic balance. Some herbal plants showed antifungal and anti-mycotoxigenic activities along with antioxidant activity against phytotoxic fungal strains such as *Fusarium verticillioides*, *spergillus*. *Ochraceous* and *Aspergillus flavus*. The results reported, use of selected medicinal plants as bio fungicides may prevent food spoilage due to oxidation [18]. Study by Abdel-Fattah et al. reported, the extract of wild stevia shows potential antifungal, anti-mycotoxigenic and antioxidant activities against *Aspergillus flavus*, *Aspergillus ochraceus*, *Aspergillus niger*, and *Fusarium moniliforme* [19]. Besides, essential oils have the ability to regulate the growth of mycotoxigenic fungi including *Aspergillus favus*, *Aspergillus oryzae*, *Aspergillus niger*, *Alternaria alternata*, *Fusarium moniliforme*, *Fusarium graminearum*, *Penicillium citrinum* and *Penicillium viridicatum* and many more to list.

Curcumin and ellagic acid are some phytochemicals used in feed and food supplements [20]. These prevent the Aflatoxin B1 (AFB1) metabolism and increases glutathione-S-transferase activity which are involved in xenobiotic detoxification.

Red fruits and vegetables such as papaya and tomato have a compound named Lycopene. This compound has shown defensive mechanism against reproductive, hormonal damage and ZEN oxidation in mice [21]. It also prevents oxidative stress induced by T-2 toxin and maintains GSH cellular level in vivo. The antibacterial, antiviral and antifungal properties have been described in **Table 1**.

3. Metabolic disorders

3.1 Obesity

Obesity occurs due to high intake of dense energy foods (carbohydrates) along with less physical activity which is required to burn the food [22]. Overweight is related to a number of comorbidities such as type 2 diabetes mellitus, cardiovascular diseases (stroke and heart) and certain cancers (breast, prostate, kidney, colon) [23]. There are various factors which affects our weight and to reduce or maintain weight one must have a healthy lifestyle, physical activity, reduce consumption

of saturated fats, consume less sugars and salts and increase the intake of dietary vegetables and whole grains, as well as pharmacological therapies and surgical interventions [24]. Still obesity treatment is a challenging as only 5–10% of individuals are able to maintain their weight loss over years [25]. The weight loss is reversed when an individual abandons his/her healthy lifestyle or ceases pharmacotherapy [26]; also use of some synthetic drugs have side effects [27]. The pharmacological drugs can be replaced by herbal supplements which are not only efficient but also less expensive and most importantly safe.

3.2 Diabetes mellitus

Diabetes mellitus is a metabolic disorder and has increased rapidly in the past 20 years. Some medicinal plant species (*Tarchonanthus camphoratus*, *Strychnos henningsii*, *Elaeodendron transvaalense*, *Euclea undulata*, *Hypoxis argentea*, *Schkuria pinnata* and *Cissampelo campensis*) have the ability to increase glucose uptake in cultured cells such as hepatic cells, muscle cells and preadipocytes and thus, might show hypoglycemic activity by increasing peripheral glucose uptake [28]. *Cucurbita pepo*, *Senna alexandri*, *Nuxia floribunda* and *Cymbopogon citratus* are some medicinal plants containing α -glucosidase and α -amylase inhibitors which might help in the reduction of post-prandial hyperglycemia [29]. *H. argentea* and *Carica papaya* are example of antidiabetic plants which has the ability to preserve and increase the regeneration of pancreatic β -cells resulting in increased insulin release [30]. In South Africa, diabetes treatment is done by some medicinal plants such as *Hypoxis hemerocallidea*, *Catharanthus roseus*, *Vernonia amygdalina*, *Sutherlandia frutescens* and *Mimusops zeyheri*. Although traditional practitioners have cited some medicinal plants showing antidiabetic properties, but still very few pharmacological data is available to confirm their efficiency. Also, the interaction of these medicinal plants with modern antidiabetic pharmacological drugs, its effective doses and toxicity levels are still unknown.

3.3 Cardiovascular disease (CVD)

CVD associated complications can be prevented by using anti-hypersensitive regimes to lower high blood pressure. Some traditionally used hyper-sensitive pharmaceutical drugs used are β -blockers, angiotensin receptor blockers, thiazide diuretics, calcium channel antagonists and vasodilators [31]. Several plant extracts have been identified possessing potential to treat CVDs including hypertension, congestive heart failure, ischemic heart disease and atherosclerosis [32].

For hypertension treatment, some healers of South Africa have used orally administered tincture of *Helichrysum ceres*. The hypotensive effect of the extract is due to the presence of diuretic and natriuretic bioactive phytochemical compounds [33]. In vivo studies have shown that *H. ceres* leaf ethanolic extract lowers blood pressure [34]. The extract acts on the vascular smooth muscles resulting in vasodilation which leads to total peripheral resistance (TPR) reduction. The ethanolic extract of *Ekebergia capensis* leaf prevents hypertension development in murine models. This hypotensive effect is due to the modulatory effect on TPR of vascular smooth muscles. Studies have shown that crude leaf extract of *Opuntia megacantha* can overturn the inability of kidney to excrete sodium in a streptozotocin-induced (STZ) diabetic rat model [35]. This indicates the beneficial effects of plant extracts in hypertension management by influencing the ability of kidney to regulate blood volume. *Allium sativum* (phenols and flavonoids) [36], *Sclerocarya birrea* (flavonoids and triterpenes) [37], *Ficus thonningii* (anthraquinones, flavonoids, and saponins) [38], and *Olea europea* (triterpenes, flavonoids, and glycosides) [39] are

some medicinal plants used popularly in South Africa for hypertension management due to their cardioprotective, vasorelaxant and bradycardic effects. Isolated phytochemicals from wild African olive leaves (*Olea europea*) collected from Cape Town showed anti-hypersensitive, diuretic and anti-atherosclerotic effects [40]. When insulin-resistant rat model was treated with *O. europea* extracts for six-weeks, development of hypertension and atherosclerosis was prevented displaying its potential in hypertension management in Africans.

Renin-angiotensin-aldosterone system (RAAS) is a signalling pathway in blood pressure regulation which is targeted by the phytochemical constituents of medical plants showing anti-hypertensive property. During hypertension development, angiotensin I is converted to angiotensin II by an enzyme known as angiotensin-converting enzyme (ACE) and the enzymatic inhibition of ACE is analysed while

Metabolic disorder	Medicinal plant	Mode of action	References
Diabetes mellitus	<i>Tarchonanthus camphoratus</i> , <i>Strychnos henningsii</i> , <i>Elaeodendron transvaalense</i> , <i>Euclea undulata</i> , <i>Hypoxis argentea</i> , <i>Schkuria pinnata</i> and <i>Cissampelo campensis</i>)	Increase glucose uptake	Oyedemi et al. [28]
	<i>Cucurbita pepo</i> , <i>Senna alexandri</i> , <i>Nuxia floribunda</i> and <i>Cymbopogon citratus</i>	Inhibit α -glucosidase and α -amylase and reduces hyperglycemia	Boaduo et al. [29]
	<i>H. argentea</i> and <i>Carica papaya</i>	regeneration of pancreatic β -cells	Akinrinde et al. [30]
	<i>Helichrysum ceres</i>	Lowers blood pressure	Musabayane et al. [38]
Cardiovascular Disease (CVD)	<i>Ekebergia capensis</i>	Prevents hypertension	KAMADyAAPA et al. [34]
	<i>Opuntia megacantha</i>	Influence kidney to regulate blood volume	Bwititi et al. [35]
	<i>Allium sativum</i> , <i>Sclerocarya birrea</i> , <i>Ficus thonningii</i> , and <i>Olea europea</i>	Hypertension management	Al-Qattan et al. [36]; Braca et al. [37]; Musabayane et al. [38]; Bennani-Kabchi et al. [39]
	<i>Tulbaghia violacea</i>	Inhibition of angiotensin-converting enzyme (ACE)	Ramesar et al. [42]
Non-alcoholic Fatty Liver Disease	<i>Hoodia gordonii</i>	Suppresses the appetite	Smith et al. [44]
	<i>S. frutescens</i>	Modifies lipid metabolism in 3 T3 adipocytes	MacKenzie et al. [45]
	<i>Aloe vera</i>	Hepatoprotective activity, reduces lipid accumulation	Bhalla et al. [46]; Misawa et al. [47]

Table 2.
Mode of action of plant extracts against metabolic disorders.

screening for anti-hypertensive medicines [41]. In vitro studies have been done on South African medicinal plants to evaluate their inhibition potential against ACE. Ethanolic and aqueous extract of some medicinal plants have demonstrated inhibition activity against ACE, and they are *Tulbaghia violacea*, *Amaranthus ybridus*, *Amaranthus dubius*, *Galinsoga parviflora*, *Stangeria eriopus*, *Oxygonum sinuatum*, *Physalis viscosa*, *Justicia flava* and *Oxygonum sinuatum*. Among these medicinal plants, *T. violacea* exhibited highest activity [42]. The inhibition activity observed, is due to the presence of tannins in most of these plants as tannins interferes with the activity of ACE [43].

3.4 Non-alcoholic fatty liver disease

Non-alcoholic fatty liver disease (NAFLD) is a major cause of morbidity and mortality. The disease is generally related with obesity, but recent studies show it can also develop independent of metabolic syndrome. *Hoodia gordonii*, a succulent plant was used as an appetite suppressant and due to its appetite-suppressing property, the succulent plant can be potentially used for NAFLD management. Recent studies have shown, *Hoodia gordonii* extracts have reduced the body mass of obese rats along with reduction in muscle mass and adipocyte size [44].

S. frutescens, a legume has various medicinal properties. It not only exhibits anti-diabetic properties but recent studies have shown, it also has the ability to modify lipid metabolism in 3 T3 adipocytes as well as in insulin-resistant rats [45]. Studies have also shown, the aqueous extract of the plant can reverse fructose induced hepatic steatosis in vivo.

Aloe vera is well known for its medicinal properties against hepatic steatosis and it has also been demonstrated that its extract improves this condition in rats. Kaempferol is a bioactive compound in *A. vera* which exhibits hepatoprotective activity [46]. Lophenol and cycloartenol are some other *A. vera* phytosterols which when administered to Zucker diabetic fatty rats shows significant decrease of lipogenic gene expression and reduced hepatic lipid accumulation [47]. The mechanism of effectiveness of plant extracts in metabolic disorders is given in **Table 2**.

4. Cancer

Cancer involves uncontrolled cell growth which can be initiated by various factors. Chemoprevention is a treatment makes use of natural, biological or synthetic agents to suppress, prevent or reverse carcinogenesis in its initial phase or prevent the invasion of premalignant cells [48]. Carcinogenesis occurs in three steps, initiation, promotion and progression. At molecular level, chemoprevention has been distinguished by altering these three pathways [49]. FDA has recently approved ten new agents for the treatment of precancerous lesions, reducing the risk of cancer [50]. Clinically, chemoprevention has be grouped as primary, secondary and tertiary. The primary chemoprevention is for people with no cancer, as well as for those who have the risk of developing cancer in future. The secondary chemoprevention is suitable for patients with pre-malignant lesions which in future may lead to invasive cancer. The tertiary prevention is to cure or prevent recurrence of cancer [49].

Capsaicin (trans-8-methyl-N-vanillyl-6-nonenamide), an active and pungent alkaloid found in *Capsicum* [51]. It has been reported that capsaicin has been used as an anticancer, tumour suppressing, chemopreventive and radio sensitising agent in various cancer models [52]. Capsaicin exhibited its ability to reduce pain and effective against osteoarthritis when applied topically [53]. It has been used as an

alternative for oral non-steroidal anti-inflammatory drugs which had side effects. Capsaicin can be used as cancer treatment due to its properties such as carcinogens activity inhibition and inducing apoptosis in several cancer cell lines in vitro and in rodents [54].

Catechins are found in various beverages such as green tea [55]. These are naturally occurring dietary phytochemical and polyphenols. Very few studies have been reported showing association of cancer with consumption of dietary phytochemicals [56]. Major components of green tea are Catechin (C), epicatechin (EC), epigallocatechin (EGC) and epigallocatechin-3-gallate (EGCG) [57]. It has been reported that EGCG could enhance the activity of several anticancer drugs such as retinoids [58]. A synthetic retinoid, AM80, has been clinically used for relapsed and intractable acute promyelocytic leukaemia patients. A study demonstrated the use of AM80 and EGCG in combinations, induced apoptosis and upregulated the expression of inducible gene of damaged DNA including death receptor 5 (DR5), GADD153 and p21waf1 in lung cancer. The combination also showed downregulation of deacetylase 4, -5, and -6, inducing apoptosis in lung cancer by Am80 and EGCG.

Lycopene is an antioxidant and thus shows protective effect against various diseases including cancer, hypertension, osteoporosis, cardiovascular diseases and neurodegenerative diseases [59]. Studies have been reported that lycopene accumulates in prostate tissue as compared to other tissues and this might be responsible for its anti-prostate cancer activity [60]. A study showed lycopene exhibits anti-angiogenic activity in in vitro as well as in vivo, suggesting the mechanism may involve modulation of PI3K-Akt and ERK/p38 signalling pathways [61]. Several studies have demonstrated use of lycopene and melatonin in combination showed strong chemopreventive activity through antioxidant and anti-inflammatory activities [62].

Isoflavones are isoflavonoids present in plants of leguminosae family [63]. It is extensively found in lentil, chickpeas, beans, soy and have importance in mammals as phytoestrogens. Isoflavones have several health benefits and are used for treating hormone-dependent medical conditions such cancer, cardiovascular disease, menopause and osteoporosis. Isoflavones extracted from soy, such as genistein, have been developed to have significant anticancer effects against as lymphoma, leukaemia, breast, prostate gastric and non-small cell lung cancer [64]. Studies have reported genistein showing anticancer effects in various cancer models such as breast cancer, lung cancer, ovarian cancer, prostate cancer, bladder cancer, renal cancer, cervical cancer, liver cancer, and head and neck squamous cell carcinoma [65].

Chemotherapy is an approach for cancer treatment but has several undesired side effects, including chemotherapy-induced peripheral neuropathy (CIPN) [66]. Recent studies have reviewed the preclinical and clinical studies on the efficiency of herbal medicines in CIPN. Cinnamon (*Cinnamomum cassia* (L.) D. Don), chamomile (*Matricaria chamomilla* L.), sage (*Salvia officinalis* L.), and sweet flag (*Acorus calamus* L.) are some medicinal plants, and curcumin, thiocetic acid and matrine are some phytochemicals which have shown effective properties in CIPN animal models.

5. Oral health

Oral health reflects the physical and social well-being of an individual. The food consumed affects the oral health as they are naturally bioactive and is composed of minerals, vitamins and antioxidants [67]. Aromatic vegetables and spices used in it are not only appetising and savoury but also has therapeutic and preservative properties. Foods we consume have a number of benefits such as antibiotic,

anti-inflammatory, anticarcinogenic and immunogenic properties. A study shows, vegetables (more than 440 g/day), fruits and spices rich diet can prevent 20% of all cancers [68]. According to a WHO report, there is 15% chance of suffering from oropharyngeal cancers due to dietary imbalance or deficiencies [69]. The people of Asia, USA and Europe suffer from oral squamous cell carcinoma due to low antioxidant and fibre intake. Many studies have proved that antioxidants and fibres exhibit chemotherapeutic and chemo-preventive properties.

In Mexico, various herbal therapies are used for the treatment of oral disorders such as mouth infection, teeth discoloration, gingivitis and periodontitis [70]. Even though, very less research has been performed demonstrating the antiplaque, antimicrobial and antibacterial effects of Mexican herbs, they can still be used for treating several periodontal diseases or as anticarcinogenic agent [70].

6. Wound healing

Wounds are injuries caused physically due to skin rupture which may lead to anatomical or functional disorders. Wound healing is a complex and dynamic process leading to reformation of tissue integrity and homeostasis [71]. The process involves inflammation, tissue formation, neovascularization, reepithelization, extracellular matrix remodelling and wound contraction. The process is coordinated by various signalling mechanisms involving numerous growth factors, chemokines and cytokines. During the process, cell proliferation is necessary for tissue repair and its regeneration [72].

For more than 500 years, “Ayurveda” has been practiced in India to prevent and cure diseases. The process utilises plants for disease prevention and cure. Traditional Chinese medicine system has been in use all over eastern Asia for over 3000 years and it uses numerous medicinal plants [73]. Modern science has analysed the traditional medicinal plant species to identify bioactive constituents present in it and as many as 12 medicinal plants have undergone clinical trials with regard to their wound healing property.

7. Production

Plants immediately activate their defence mechanism when they are attacked. This also includes the biosynthesis of phytochemicals which occurs rapidly resulting in reduction of nutrients and amino acids. But the optimization of mass production of phytochemical is still unknown.

An efficient way for phytochemical production is creation of metabolic highways through protein complexes known as metabolons. Three decades ago, the existence of metabolons was first proposed. But its in vivo protein–protein interaction and structures are still challenging. Metabolons are involved in metabolic pathways, mostly primary and secondary including lignin, Krebs cycle and flavonoid pathways [74]. For the biosynthesis of toxic cyanogenic glucoside dhurrin which highly gets accumulated in sorghum, metabolons are essential (*Sorghum bicolor*) [75]. Metabolons can efficiently produce inducible phytochemicals. The biosynthesis efficiency can be increased by assembling the sequential enzymes of a pathway into a single protein complex and would also limit release of harmful or reactive intermediates. In these protein complexes, the phytochemical intermediates are released only when the metabolon is disassembled. Th 2019, Mucha et al. studied if a metabolon channels the biosynthesis of an essential defence metabolite in *Arabidopsis* (*Arabidopsis thaliana*), known as camalexin [76]. The biosynthesis of camalexin is an

enzyme catalysed multi-step reaction and from tryptophan (Trp) various intermediates are generated which have been detected in knockout genotypes [77].

In vitro plant production is a solution which is favoured by biotechnologists. The in vitro technology allows to produce plants uniformly by controlled manipulation of environmental condition, growth regulators, and strategies that can enhance production as well as overall yield of phytochemicals. The naturally occurring instable chemical composition in plants can be avoided by growing them on media prepared according to strict recipes for plant's nutritional necessities under controlled environmental conditions including temperature, light duration and intensity [78].

The most efficient method for in vitro secondary metabolite production is plant micropropagation. This is a technique that uses clonal propagation to produce plants which are identical genetically as well as free from pathogens and contaminants, and this process requires very less space, time and supplies. A study demonstrated in vitro culture of tansy (*Tanacetum vulgare*) from seeds collected from natural population for production of secondary metabolites such as essential oils and methanol extracts [79]. Studies have reported, the use of plant growth regulators exogenously (auxin and cytokinin) might hinder genetical stability leading to somaclonal variation, which is undesirable for in vitro plant production when used for isolation of secondary metabolites [80].

8. Conclusion and future perspectives

The non-nutritive part of the plants that is, phytochemicals have antimetabolic, anti-cancer, anti-neurological and wound healing properties. They also help in maintaining oral health. The antimicrobial nature of phytochemicals has led to its increased demand. To meet the requirements of modern medicine, plants and their extracts are cultured in vitro. The use of huge bioreactors and mass propagation has led to the establishment of inexpensive and efficient method for phytochemical production. The regulation of in-vitro conditions to multiplication can be a promising technique for medicine.

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