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A REVIEW ON HERBAL ANTIOXIDANTS

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INTRODUCTION

Antioxidants or inhibitors of oxidation are compounds which retard or prevent the oxidation and in general prolong the life of the oxidizable matter. (1) Free radicals are fundamentals to any biochemical process and represent an essential part of aerobic life and metabolism. Majority of the diseases / disorders are mainly linked to oxidative stress due to free radicals. (2) The oxidants / free radicals are species with very short half life, high reactivity and damaging activity towards macromolecules like proteins, DNA and lipids. These species may be either Oxygen derived (ROS) or Nitrogen derived (RNS). The most common reactive oxygen species include superoxide anion (O_2), hydrogen peroxide (H_2O_2), peroxyl radicals (ROO) and reactive hydroxyl radicals (OH). The nitrogen derived free radicals are nitric oxide (NO), peroxy nitrite anion (ONOO), Nitrogen dioxide (NO_2) and Dinitrogen trioxide (N_2O_3).

In general, the reactive oxygen species circulating in the body tend to react with the electron of other molecules in the body and these also effect various enzyme systems and cause damage which may further contribute to conditions such as cancer, ischemia, aging, adult respiratory distress syndromes, rheumatoid arthritis etc.(3) The exogenous sources of ROS include electromagnetic radiation, cosmic radiation, UV-light, ozone, cigarette smoke and low wavelength electromagnetic radiations and endogenous sources are mitochondrial electron transport chain, β-oxidation of fat. Chemical compounds and reaction capable of generating potential toxic oxygen species / free radicals are referred to as 'pro-oxidants'. They attack macromolecules including protein, DNA and lipid causing to cellular / tissue damage on the other hand, compounds and reactions disposing off these species, scavenging them suppressing their formation or opposing their actions are called antioxidants.

In a normal cell there is an appropriate pro-oxidant: antioxidant balance. However, this balance can be shifted towards the pro-oxidant when production of oxygen species is increased or when levels of anti-oxidants are diminished. This state is called 'oxidative stress' and can result in serious cell damage if the stress is massive or prolonged.(4)

Classification of anti-oxidants:- It is of two types

1. Based on solubility:

(a). Hydrophilic antioxidants:- They are soluble in water. Water soluble antioxidants react with oxidants react with oxidants in the cell cytoplasm and blood plasma.

(b). Hydrophobic antioxidants:- They are soluble in lipids. Lipid soluble antioxidants protect cell membranes from lipid peroxidation.

2. Based on line of defense:

(a). First line defense (preventive antioxidant):- These are enzymes like superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GT_X), glutathione reductase and some minerals like Se, Mn, Cu etc.

SOD mainly acts by quenching of superoxide (O_2), catalase by catalyzing the decomposition of hydrogen peroxide (H_2O_2) to water and oxygen. GT_X catalyses the reduction of H_2O_2 and lipid peroxide generated during lipid peroxidation to water using reduced glutathione as substrate.

(b). Second line defense (Radical scavenging antioxidant):- These are glutathione, Vit C, uric acid, albumin, biliribin, vit E, carotenoids, flavonoid etc. β-

carotene is an excellent scavenger of singlet oxygen. Vit C interacts directly with radicals like O_2, OH. GSH is a good scavenger of many free radicals like O_2, OH and various lipid hydroperoxides and may help to detoxify many inhaled oxidizing air pollutants like ozone.

(c). Third line defense (Repair and de-novo enzymes):- These are a complex group of enzymes for repair of damaged DNA, protein, oxidized lipids and peroxides and also to stop chain propagation of peroxyl lipid radical. These enzymes repair the damage to biomolecules and reconstitute the damaged cell membrane.(4)

This includes study of following drugs:-

1. *Aspergillus candidus*:-
Family: Aspergillaceae.
Aspergillus candidus broth filtrate, extract was studied using different antioxidant models. It's ethyl extract scavenge the stable radical diphenyl picryl hydrazyl (DPPH) and radical cation 2,2-azinobis-(3-ethylbenzothiazoline-6-sulphonate) and thus, it shows antioxidant property due to presence of phinolic compounds.(5)

Aspergillus candidus

2. Quercetin:-
It belongs to an extensive class of poly phenolic flavonoid compounds. In vitro anyioxidant activity was tested for DPPH free radical, superoxide anions, hydrogen peroxide and hydroxyl radical. It scavenges oxygen radicals, inhibits xanthine oxidase, protects against lipid peroxidation, chelates metal ions and forms inert complexes that can't take part in the conversion of superoxide radicals and hydrogen peroxide into hydroxyl radicals.(6)

Quercetin

3. *Sphaeranthus indicus* Linn.:- It is popularly known as 'Gorakmundi'
Family: Asteraceae.
The ethanolic extract scavenges radical cation, DPPH, SOD and NO. Constituents are flavonoids, carbohydrates, alkaloids, gums and mucilage. (7)

Sphaeranthus indicus Linn

4. *Rhizophora mangle* bark:-
Family: Rhizophoraceae.
Deoxyrebose assay was used. The total extract and its fraction showed scavenging activity of hydroxyl radicals and ability to chelate iron ions. Chemical constituents are polyphenols, carbohydrates, fatty acids and sterols. (8)

Rhizophora mangle

5. *Punica granatum* fruits:-
Family: Punicaceae
Activity was evaluated using DPPH test, 5-lipoxygenase assay and luminal / xanthine oxidase system (Chemiluminescence assay). Chemical constituents are tannins, alkaloids, glycosides. (9)

Punica granatum fruits

6. *Origanum dictamnus*:-
Family: Labiatae.
The aqueous extract scavenges free radicals generated by the fenton reaction and reducing oxygen consumption of a methyl linoleate emulsion. The active components of herb are phenolic compounds, mainly flavonoids and phenolic acids. (10)

Origanum dictamnus

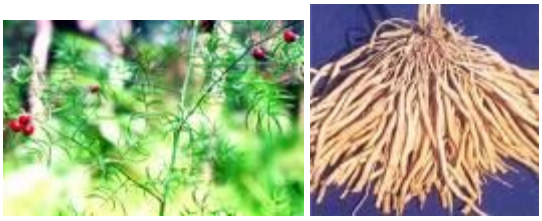
7. *Rhus oxycantha* root cortex:-
Family: Anacardiaceae.
Antioxidant activity has been examined ascorbic acid oxidation and inhibition of toxicity induced by an organochlorine pesticide, dichloro diphenyl-trichloro ethane (DDT) in rat thymocytes. Chemical constituents are (+) epicatechin-3-0-gallate and proanthrocyanidins oligomers and polymers. (+) and (-) epicatechin show hydroxyl radical scavenging activity and proanthrocyanidins are efficient free radical scavenger. (11)

Rhus oxycantha

8. *Diospyros malabarica kostel* bark:- It is popular as "Gab or Tinduk",
Family: - Ebenaceae.
Different *in vitro*, like DPPH, nitric acid, superoxide, hydroxyl radical and lipid peroxide radical model were used in the study. Oxygen reacts with the excess nitric oxide to generate nitrite and peroxynitrite anions, which act as free radicals. The extract competes with oxygen to react with nitric oxide and thus, inhibits the generation of anions. Chemical constituents are phenolic compounds. Its stem bark is used for the treatment of intermittent fever and fruit juices for healing of wound ulcer. (12)

Diospyros malabarica kostel

9. *Asparagus racemosus*:-
Family: - Liliaceae.
It shows antioxidant activity through the free radical scavenging, superoxide anion radical scavenging, hydrogen peroxide scavenging, nitric oxide scavenging, metal chelation, reduction power and inhibition of lipid peroxidation in rats. Its chemical constituents are saponins (Shatavarin I-V), alkaloids, polyphenols, flavonoids, vit.C. (2)



Asparagus racemosus

10. *Glycyrrhiza glabra*: - It is popular as licorice 'yastimadhu'.
Family: - Leguminosae.
Its extract was tested by studying the inhibition of radiation induced lipid peroxidation in rat liver microsomes. Chemical constituents are glycyrrhizin, flavones, coumarins. It shows its activity through free radical scavenging property. Its other actions are diuretic, demulcent, tonic etc. (13)

Glycyrrhiza glabra

11. *Boerhavia diffusa* leaves:-
Family: - Nictaginaceae.
Alloxan induced diabetic rats were used in the study. Chemical constituents are rich in alkaloids and sterols including ursolic acid, hypoxanthine 9-L arabinofuranoside, punarnavine 1 and 2, myricyl alcohol and myristic acid. It decreases the level of thiobarbituric acid reactive substances (TBARS) and increases the activity of glutathione peroxidase (GP_X) and glutathione-S- transferase (GST). (14)

Boerhavia diffusa

12. *Auricularia auricula*: - It is popular as 'tree ear or wood ear'.
Family: - Auriculaceae.
It has potent hydroxyl radical scavenging and lipid peroxidation inhibition activities. Chemical constituents are flavonoids. (15)

Auricularia auricular

13. *Annona squamosa*: - It is popular as 'Custard apple or Sitaphal'.

Family: - Annonaceae.

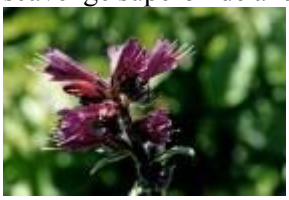
Streptozotocin induced diabetic rats were used. It reduces the lipid peroxidation and increases the activity of antioxidant enzymes and strong super oxide radicals and singlet oxygen quenchers. Chemical constituents are flavonoids. (16)

Annona squamosa

14. *Echium amoenum fisch* and C.A. Mey Flower:-

Family: - Annonaceae.

Chemical constituents are rosmarinic acid and flavonoids. Flavonoids highly scavenge most types of oxidizing molecules including singlet oxygen and various free radicals and rosmarinic acid scavenge superoxide and hydroxyl radicals. (17)



Echium amoenum fisch

15. *Eucalyptus globules*: - It is popular as "Karpura maram".

Family: - Myrtaceae.

The antioxidant activity of eucalyptus oil was estimated by two in vitro assays namely diphenyl picryl hydrazyl radical scavenging activity and inhibition of Fe-ADP-ascorbate induced lipid peroxidation method. (18)

Eucalyptus globules:

16. Pepticare: - It is a herbomineral formulation it was administered orally to rats to investigate its effect on isoproterenol induced myocardial infraction and cisplatin induced renal damage. It increases the levels of SOD, CAT and reduces GSH; membrane bound enzymes like Ca^{2+}, Mg^{2+} and $Na^{+}K^{+}$ ATPase and decreases lipid peroxidation (MDA) in heart and kidney. Thus, it protects the heart and kidney from damage caused by isoproterenol and cisplatin. (19)

17. *Acacia arabica* bark:-

Family: - Mimosae.

There are in vivo and in vitro experimental models. In vitro, lipid peroxidation was carried out by tertiary butyl hydroperoxide (TBH) induced lipid peroxidation. In vivo, experiments were carried out in CCl_4-induced hepatotoxicity in rats. The bark contains (+) catechin, (-) epicatechin, quercetin and gallic acid. The polyphenol rich active fraction pf acacia Arabica is a potent free radical scavenger and protects TBH induced lipid peroxidation and CCl_4-induced hepatic damage. It is used in the treatment of asthma, bronchitis, diabetes, dysentery and skin diseases. (20)

Acacia Arabica

18. Arthritin (Apolyherbal formulation):- It consisting of extracts of *Acacia Arabica, Withania somnifera, Juniperus communis, Asparagus racemosus, Tinospora cardifolia, Tribulus terrestris, Anethum sowa, Curcuma zerumber* and *Zingiber officinalis.* Phenols, flavonoids, terpenoids, alkaloids, glycosides are present in the various constituents of the polyherbal formulation act as natural free radical scavengers. It causes decreases in serum lipid peroxidase and increase in SOD & GT_X. It possess a significant anti-inflammatory and free radical scavenging activity and also responsible for antiarthritic activity. (21)

Withania somnifera

Juniperus communis

Tinospora cardifolia,

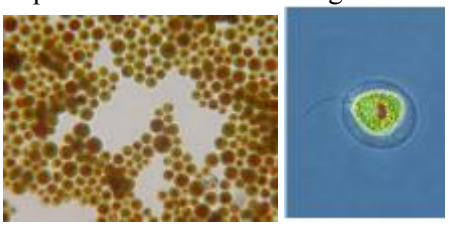
Tribulus terrestris

Anethum sowa

19. *Acacia catechu:* - It is popular as 'Khadira' (black catechu).
Family: - Leguminosae.
Its extract was tested by studying the inhibition of radiation induced lipid peroxidation in rat liver microsomes. Its chemical constituents are catechin, tannic acid, quercetin, red tannin. It's a powerful astringent. (13)

Acacia catechu

20. Astaxanthin: - It is the member of the carotenoid family. It is obtained from *haematococcus pluvialis*. It can serve as a potent free radical scavenger. It provides many essential biological functions, including protection against lipid membrane peroxidation of essential polyunsaturated fatty acids and proteins, DNA damage and UV light effects. It also plays an important role in immunological defense. (20)



haematococcus pluvialis

21. *Ligustrum vulgare* and *L. delavayanum* leaves:-
Family: - Oleaceae.
Activity was evaluated using DPPH test. Its chemical constituents are flavonoids, iridoids, coumarins, phenyl propanes and essential oil. Flavonoid aglycones are responsible for the activity. It shows free radical scavenging activity. (22)

Ligustrum vulgare and *L. delavayanum*

21. Triphala: - It is a traditional ayurvedic herbal formulation consisting of the dried fruits of three medicinal plants. *Terminalia chebula, Terminalia belerica* and *Phyllanthus emblica,* also called as 'three myrobalans'. Activity was evaluated using DPPH test. (23)

Terminalia belerica

22. *Terminalia chebula*: - It is known as '*Myrobalanus chebula* or Harde'.
Family: - Combretaceae.
Its chemical constituents are tannins, chebulinic, ellegic and gallic acids. Its extract was tested by studying the inhibition of radiation induced lipid peroxidation in rat liver microsomes. It shows free radical scavenging activity due to presence of tannins. It inhibits the development of duodenal ulcer and appeared to extract a cytoprotective effect on the gastric mucosa. (23)

Terminalia chebula

23. *Phyllanthus emblica*: - It is known as '*Emblica officinalis* or Amla.
Family: - Euphorbiaceae.
Its chemical constituents are tannins and other phenolic compounds and flavonoid quercetin. It is a rich source of Vit. C. It interacts directly with radicals and scavenges them and hence, shows antioxidants activity. It is used for the treatment of common cold, scurvey, cancer and heart disease. (23)

Phyllanthus emblica

24. *Lobelia nicotianaefolia*:-
Family: - Campanulaceae.
Its chemical constituents are alkaloids important of them are lobeline. Also contains volatile oil, resin, gum and fixed oil. It is mainly used in the treatment of asthma and as respiratory stimulant. (24)

Lobelia nicotianaefolia

25. *Arnica Montana Linn* - It is commonly called as 'Arnica'.
Family: - Compositae.
Its chemical constituents are volatile oil, flavonoids and terpenoids, sesquiterpene lactones from helenanolide group like helenalin and other ester derivatives reported to possess antioxidant, antihyperlipidemic and antitumor activity. It shows free radical scavenging activity due to presence of flavonoids. (25)

Arnica Montana Linn

26. *Zingiber officinale*: - It is commonly called as 'ginger'.
Family: - Zingiberaceae.
Its chemical constituents are volatile oil, starch, acrid resinous matter, shagoals, zingerone, peradols etc. (26)

Zingiber officinale

27. *Rosmarinus officinalis*:-
Family: - Labiatae.
The flowers contain volatile oil (rosmary oil), resin and ursolic acid. Volatile oil contains mainly borneol. It is also used as a spice in tea to help ward off cancers, heart diseases etc. Antioxidant activity is due to borneol. (27)

Rosmarinus officinalis

28. *Mentha arvensis*:-
Family: - Labiatae.
The antioxidant activity of menthe oil was estimated by two in vitro assays, DPPH radical scavenging activity and inhibition of Fe-ADP-Ascorbate induced lipid peroxidation (LPO) method. It contains 80% L-menthol and due to this, it shows antioxidant property. It shows antioxidant activity by decreasing lipid peroxidation. (28)

Mentha arvensis

29. *Citrus lemon*:-
Family: - Rutaceae.
The antioxidant activity was estimated by two in vitro assays, DPPH radical scavenging activity and inhibition of Fe-ADP-Ascorbate induced lipid peroxidation (LPO) method. It contains mainly citral and limonene. The antioxidant property is shown due to presence of citral. (3)

Citrus lemon

30. *Decalepis hamiltonil*:-

Family: - Asclapiadaceae.

Various model systems like DPPH, β-carotene linoliate and hydroxyl radical scavenging activity were used in the study. This is act by easing the level of endogenous defenses by upregulating the expression of genes encoding the enzymes such as superoxide dismutase (SOD), catalase (CAT) or glutathione peroxidase (GT_X). The main constituent by which activity is shown is 2-hydroxyl-4-methoxy benzaldehyde. (29)

Decalepis hamiltonil

CONCLUSION

Oxidative stress has been implicated in the pathology of many diseases and condition including diabetes, cardiovascular diseases, inflammatory conditions, cancer and aging.

Antioxidant may offer resistance against the oxidative stress by scavenging free radicals, inhibiting lipid peroxidation and by many other mechanisms and thus prevent disease, and today widely used as free radicals inhibitors in food for maintaining the freshness, flavor and odor for a longer period.

Current research reveals the different potential application of antioxidant / free radical manipulations in prevention or control of diseases. Natural products from dietary components such as Indian species and medicinal plants are known to possess antioxidant activity.

Future approach include gene therapy to produce more antioxidant in the body, genetically engineered plant products with higher level of antioxidant, synthetic antioxidant enzymes (SOD mimics), novel biomolecules and the use of functional foods enriched with antioxidant.

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