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Research Article

Anti Oxidant Potential of "Cureit"- A Novel Bio Available Curcumin Formulation

Sreeraj Gopi*a, Robin Georgea, V. T. Sriraamb

ABSTRACT

The major impediment towards the use of Curcumin for health benefits is its poor availability in the blood and tissues. This problem can be trounced by our innovative Curcumin preparation. The formulation treats curcumin as it occur inside the turmeric rhizome, where curcumin is more available in the blood. A human study conducted, where the innovative curcumin found to absorb ~15 times more as that of the normal curcumin. This Bio available curcumin was branded as "cureit" and its anti oxidant potential established. The study proves that "cureit" could be good source of natural antioxidant.

Key-words: curcumin, bio availability, anti oxidant, turmeric rhizome

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*Email Id- Sreeraj.gopi@plantlipids.com

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Corresponding Author:

Principal Scientist

- A Plantlipids company,

Cochin 682 311, Kerala,

R&D Centre, Aurea Biolabs (P) Ltd

Sreeraj Gopi*

India

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a- R&D Centre, Aurea Biolabs (P) Ltd – A Plantlipids company, Cochin

b- Aurous HealthCare Research and Development India Private Limited, Chennai

Introduction

Turmeric, is commonly used as a spice in curries, food additive and also, as a dietary pigment. It has been used to treat various illnesses in the Indian subcontinent from the ancient times. Turmeric finds its use in one form or the other in the textile and pharmaceutical industries ⁽²⁾ Turmeric has been used as a nontoxic drug in Ayurveda for centuries to treat a wide variety of disorders including rheumatism, bodyache, skin diseases, intestinal worms, diarrhea, intermittent, fevers, hepatic disorders, biliousness, urinary discharges, dyspepsia, inflammations, constipation, leukoderma, amenorrhea, and colic ⁽³⁾. Turmeric has been considered as an emmenagogue, diuretic, and carminativewhen taken orally, whereas topical application is commonly used to treat bruises, pains, sprains, boils, swellings, sinusitis, and various skin disorders ⁽⁴⁾. Turmeric is used to treat angina pectoris, stomachache, postpartum

abdominal pain, and gallstones in the Chinese system of medicine (5). It seems to relieve menstrual pain (6). The poultices prepared from turmeric are topically applied to relieve pain and inflammation (7). The major chemical principles of turmeric are curcuminoids, which impart characteristic yellow color to it. The curcuminoids can be separated from turmeric by ethanol extraction and it usually contains 0.3-5.4% curcumin (one of the major curcuminoids) depending on the season of its harvest (7).

FREE RADICALS AND ANTIOXIDANTS

It has been nearly 50 years since Denham Harman6 suggested that free radicals produced during aerobic respiration cause cumulative oxygen damage, resulting in aging and death. Oxygen is an essential molecule for all aerobic forms; however, oxygen plays univalent roles. Although oxygen is indispensable for all cells for chemical energy production (ATP), it is also often transformed into highly reactive forms: reactive oxygen species (ROS), which are often very toxic to the cells.

Approximately 2% of the oxygen reduced by the mitochondria then forms superoxide (0-2) or the dismutation product H2O2. Superoxide and peroxide reacts with metal ions (Heiber-Weiss and Fenton's reactions) to promote additional radical generation, particularly with the generation of hydroxyl radicals. The hydroxyl radical reacts with all components of the cell, including lipid membrane, DNA and proteins. Nitric oxide (NO) has an unpaired electron and is therefore a free-radical species. It is a short-lived, lipophilic molecule generated from l-arginine by NO synthase (NOS). NO is involved physiologically in vasorelaxation, neurotransmission, inhibition of platelet aggregation, immune defense, and intracellular signaling. However, NO reacts with 0-2 to form peroxynitrite (0N00-), which is a powerful oxidant. NO bioactivity is related to the production of many reactive intermediates, but many of these reactive nitrogen species (RNS) are capable of damaging DNA or hindering DNA repair. It is now beyond doubt that oxidants are generated *in vivo* and can cause significant damage to cells. When an imbalance occurs between oxidants and defense systems, in favor of oxidants, oxidative stress occurs. This oxidative stress in cells results in severe metabolic dysfunctions, including loss of cell integrity, enzyme function, genomic stability, and so forth, which ultimately lead to pathogenesis of many human diseases (e.g., inflammation, ischemia, atherosclerosis, arthritis, cancer, Parkinson's disease, Alzheimer's disease, and so forth).

CURCUMIN AND ANTIOXIDANTS

Curcumin is known to protect biomembranes against peroxidative damage. Peroxidation of lipids is known to be a free-radical-mediated chain reaction, leading to the damage of the cell membranes, and the inhibition of peroxidation by Curcumin is mainly attributed to the scavenging of the reactive free

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radicals involved in the peroxidation. Most of the antioxidants have either a phenolic functional group or a beta-diketone group. Curcumin is an unique antioxidant, which contains a variety of functional groups, including the B-diketo group, carbon-carbon double bonds, and phenyl rings containing varying amounts of hydroxyl and methoxy substituents. The central argument is whether the phenolic or the central methylenic hydrogen in the heptadienone moiety is responsible for its antioxidant activity. Jovanovic and Collaborators concluded that curcumin is a superb H-atom donor by donating the H-atom from the central methylenic group rather than from the phenolic group in acidic and neutral aqueous and acetonitrile solutions. On the other hand, Barclay et al. proposed that curcumin is a classical phenolic chain-breaking antioxidant, donating H-atoms from the phenolic group. Priyadarsini et al. have also claimed that the phenolic group is essential for the free-radical-scavenging activity and that the presence of the methoxy group further increased the activity. Theoretical calculations by the density functional theory (DFT) demonstrated that the enol form of curcumin is significantly more stable than the diketo form and that the bond dissociation enthalpy (BDE) of the phenolic O:H bond is significantly lower than the BDE of the central O:H bond, suggesting that the hydrogen atom abstraction takes place in the phenolic group. It was also pointed out that the relative contribution of the phenolic group and the central methylenic group on the antioxidant activity depends on the activity of attacking radical and the reaction medium.(12)

The major impediment towards the use of Curcumin for health benefits is its poor availability in the blood and tissues. This problem can be trounced by our innovative Curcumin preparation. The formulation treats curcumin as it occur inside the turmeric rhizome, where curcumin is more available in the blood. A human study conducted, where the innovative curcumin found to absorb \sim 15 times more as that of the normal curcumin. This Bio available curcumin was branded as "cureit" and its anti oxidant potential established.

DPPH FREE RADICAL SCAVENGING ASSAY

DPPH is a common abbreviation for an organic chemical compound 2,2 diphenyl -1- picrylhydrazyl. It is a dark colored crystalline powder composed of stable free radical molecules.

DPPH has two major applications, one is to monitor the chemical reactions involving radicals, most notably it is a common antioxidant and another is a standard of the position and intensity of electron paramagnetic resonance signals.

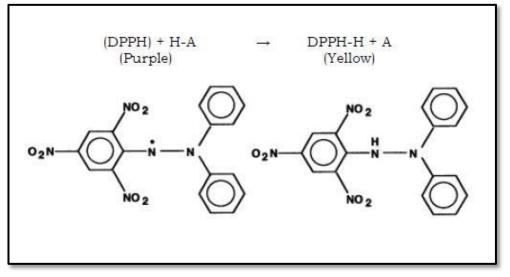


Figure1: DPPH Free Radical Scavenging Assay

- Test Compound Curcumin
- DPPH
- Methanol
- Sodium Ascorbate

METHODOLOGY

- 1. 10mg of DPPH is dissolved in 10 ml of methanol and used as master stock.
- 2. This is stored at -20°C
- 3. From the above solution 1 ml is taken and diluted with methanol and the volume is adjusted to get an absorbance in the range of 0.2 to 0.5 at 515nm.
- 4. Sodium Ascorbate **(SA)** is used as positive control. Sodium Ascorbate solution has to be prepared freshly for every experiment.
- 5. The DPPH assay method is based on the reduction of DPPH, a stable free radical. The free radical DPPH .with an odd electron gives a maximum absorption at 517 nm (purple colour). When Antioxidants react with DPPH, which is a stable free radical becomes paired off in the presence of a hydrogen donor (e.g., a free radical-scavenging antioxidant) and is reduced to the DPPHH and as consequence the absorbance's decreased from the DPPH. Radical to the DPPH-H form results in decolorization (yellow colour) with respect to the number of electrons captured. More the decolorization more is the reducing ability. This test has been the most accepted model for evaluating the free radical scavenging activity of any new drug.

RESULTS AND DISCUSSION.

The following are the results of the assay performed.

Sodium Ascorbate	OD at 515nm	С-Т	C-T/C	% inhibition
Control	0.2531			
10µg/ml	0.2011	0.012	0.05631	5.631159
25µg/ml	0.1556	0.0575	0.26983	26.98264
50µg/ml	0.1059	0.1072	0.50305	50.30502
100µg/ml	0.0352	0.1779	0.83482	83.48193

 Table 1: DPPH Free Radical Scavenging Assay – CONTROL

Curcumin	OD at 515nm	C-T	C-T/C	% inhibition
Control	0.205			
10ug/ml	0.1922	0.0128	0.06244	6.243902
25ug/ml	0.1839	0.0211	0.10293	10.29268
50ug/ml	0.1237	0.0813	0.39659	39.65854
100ug/ml	0.054	0.151	0.73659	73.65854

 Table 2: DPPH Free Radical Scavenging Assay – Consolidated Data

 Control ID
 Image: Consolidated Data

OD: Optical Density C:Control Sample T:Test Sample

Conclusion

From the data, it has been inferred that the test compound shows a concentration dependent DPPH free radical scavenging activity indicating its antioxidant potential equivalent to ascorbic acid. Based on the result in the study, it was concluded that the bio available curcumin-"cureit" could be a good source of natural antioxidant.

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