Therapeutic Potential of Polyphenols in Parkinson’s Disease

Rajeswara Babu Mythri¹,*, G. Harish¹,*, N. Raghunath¹ and M.M. Srinivas Bharath¹,**

National Institute of Mental Health and Neurosciences (NIMHANS), Bangalore, Karnataka India

1. Introduction

Increased human life expectancy has resulted in larger geriatric population throughout the world. Consequently, there is increased prevalence of age-associated neurodegenerative diseases including Parkinson’s disease (PD) with serious impact on healthcare. This has intensified the scientific research on the pathology and therapy of the central nervous system (CNS). Although modern approach in biomedical research has made major breakthroughs in understanding the pathological basis of PD, the knowledge about therapy is limited. Consequently, PD does not have a permanent cure.

During recent years, there have been several options for PD therapy including both pharmacological and neurosurgical approaches. These strategies are primarily aimed at improving the motor symptoms without any major side effects ultimately improving the quality of the life of the patient. Drugs such as L-dihydroxyphenyl alanine (levodopa or L-DOPA) replenish the lost dopamine in the brain in early PD and provide symptomatic relief. Apart from L-dopa therapy, other drugs including dopamine receptor agonists, MAO-B inhibitors and anti-cholinergic drugs have been utilized in the pharmacotherapy of PD (Almeida & Hyson, 2008). But, each class of drugs is limited by potential side effects and motor complications with chronic treatment. Further, most PD medications do not effectively tackle tremor, postural instability and cognitive deficits. Moreover, most of these drugs are not completely effective against degeneration of the remaining dopaminergic neurons (Almeida & Hyson, 2008). Due to these serious lacunae, there is a tremendous momentum to develop newer treatments involving disease modifying, restorative, possibly curative drugs with lesser side effects. Most importantly, these drugs should protect the dying neurons thus preventing further neuronal loss. In this direction there have been novel strategies of PD pharmacotherapy applicable either as independent therapies or as a supplement with the existing therapies. These have been tested in experimental models and although many of these molecules show promise, their toxicology, bioavailability and clinical efficacy in human subjects needs to be examined thoroughly before application to PD.

* Equal contribution,
** Corresponding author

www.intechopen.com
Neurodegeneration in PD involves multiple pathways such as oxidative stress, mitochondrial damage, protein aggregation, neuroinflammation etc. Modern medicine utilizes a well-defined chemical molecule(s) for pharmacotherapy which might not be effective against different disease pathways. Hence, the challenge is to come up with novel molecules that could simultaneously target multiple disease pathways without significant side-effects, be non-toxic at higher concentrations and have the ability to cross the blood-brain barrier indicating a paradigm shift from monotherapy to multi-therapy based on various targets. Towards this, natural phytochemicals which possess medicinal properties are being exploited (Kumar, 2006) which has increased the interest in the use of herbal products (Tsao, 2010). Consequently, there is increasing support for combination of modern drugs with medicine to evolve better therapies. It is interesting to note that although phytochemicals normally function as toxins and protect the plant source against damage due to pests and harmful organisms, at the lower concentrations consumed by humans, these compounds activate adaptive cellular stress responses \textit{in vivo} (“hormetic mechanisms”) thereby providing cytoprotection against exogenous toxins. Such hormetic mechanisms relevant to neuronal survival and improved brain function ultimately elevate the levels of antioxidant enzymes, protein chaperones and neurotrophic factors (Mattson et al., 2007). Polyphenols from various plant sources form a major group of phytochemicals with potential therapeutic and curative properties.

2. Polyphenols: Structure and biological properties

Polyphenols are the most widely distributed natural compounds in the plant kingdom. They are the secondary metabolites which were originally synthesized to protect the plants against microbial attack, pests and ultraviolet radiation. Polyphenols are present in fruits, vegetables, oils etc. and provide plants with brilliant colours and fragrance. Polyphenols might possess many biologically significant functions with implications for human degenerative diseases (Han et al., 2007).

2.1 Chemistry of Polyphenols

Polyphenols in general have a phenolic structure with several hydroxyl groups. Natural polyphenols vary from simple molecules (viz., phenolic acids) to complex polymeric forms (viz., condensed tannins) (Tsao, 2010). Flavonoids are the largest group of polyphenols and have been studied extensively. Currently more than 8000 phenolic structures are known and among them over 4000 flavonoids have been identified (Bravo, 1998, Cheynier, 2005, Harborne & Williams, 2000). They are classified based on the number of phenolic rings and the structural elements that link these rings (Butterfield et al., 2002, Ramassamy, 2006) (Figures 1 and 2) as follows:

a. **Phenolic acids**: These are non-flavonoid polyphenols that constitute less than 1/3 of the phenolic content in our diet and are represented mainly by benzoic acids and cinnamic acid derivatives.

![Fig. 1. Classification of polyphenols.](www.intechopen.com)
Fig. 2. Chemical structures of common polyphenols.
b. **Flavonoids**: These represent approximately 2/3 of the polyphenolic content in our diet. They have the C6-C3-C6 structural backbone, of which two are phenolic rings named Ring A and Ring B, and one chromane ring (Tsao 2010). Flavonoids are further classified based on the oxidation state of the chromane ring into Anthocyanins and anthoxanthins. Anthoxanthins are further classified into Flavones, Flavanones, Flavanols, Isoflavones, Flavanols (D’Archivio et al., 2007).

c. **Stilbenes**: Stilbenes are characterized by the presence of 1,2-diphenylethylene nucleus with hydroxyl groups substituted on the aromatic rings (Han et al., 2007). The best known compound among Stilbenes is trans-resveratrol.

d. **Tannins**: Tannins are water soluble polyphenols and are classified into condensed and hydrolysable tannins.

e. **Diferuloylmethanes**: Diferuloylmethanes are characterized by two aromatic rings substituted with hydroxyls, and linked by aliphatic chain containing carbonyl groups. Curcumin is one of the best known polyphenol among diferuloylmethanes.

Most polyphenols are chemically modified by acetylation, hydroxylation, methoxylation. These modifications could significantly influence the physicochemical properties and \textit{in vivo} absorption/degradation of the polyphenols. For e.g., acetylated flavonoids (epicatechin, epigallocatechin) are absorbed without deconjugation and hydrolysis (Rice-Evans & Miller 1996, Scalbert & Williamson 2000). Similarly, polyphenols with most antioxidant potential possess 3-6 hydroxyl groups. However, hydroxylation in the C3 position significantly inhibits the antioxidant activity and chelation of metals (Huguet et al., 1990, van Acker et al., 1996). Most polyphenols also exist as glycosides with the addition of sugar and acylated sugars at different positions of the polyphenolic structure (one to three moieties per polyphenol) (Tsao, 2010). The attached sugar could be either glucose or rhamnose and each glycosylation influences the physicochemical properties and intestinal absorption of the polyphenol (Rice-Evans & Miller, 1996, Scalbert & Williamson, 2000). Accordingly, if polyphenols are glycosylated with glucose/galactose, they will be absorbed through the small intestine by specific carriers (Scalbert & Williamson, 2000). Those containing rhamnose cannot be absorbed through the small intestine and are degraded by rhamnosidases produced by the colonic microflora. Further, glycosylation of polyphenols might decrease their antioxidant properties (Fukumoto & Mazza, 2000).

### 2.2 Biological functions of polyphenols

Polyphenols might ameliorate neurodegeneration mainly via (i) intrinsic antioxidant properties and/or by (ii) modulation of cell signalling pathways that control cell survival, death and differentiation (Chen et al., 2000, Molina et al., 2003, Shen et al., 2007, Spencer, 2007).

#### 2.2.1 Antioxidant properties

It has been suggested that consumption of foods or beverages which are rich in polyphenols can increase the antioxidant levels \textit{in vivo} (Frankel et al., 1993). Polyphenols can protect against cellular oxidative stress by directly scavenging free radicals and by chelation of divalent metal ions. The phenolic hydroxyl groups are potent nucleophiles that neutralize free radicals and form arylxyl radicals and this is central to the antioxidant properties of polyphenols. The antioxidant potential is enhanced by the vicinal hydroxyl groups depending on their number and orientation relative to the electron withdrawing groups.
such as COOH, CH$_2$COOH, or (CH)$_2$CO$_2$CH (Rice-Evans et al., 1996). The radical scavenging activity of polyphenols is measured by Trolox (a water soluble $\alpha$-tocopherol analog)-equivalent antioxidant capacity (TEAC) assay and expressed as the millimolar concentration of Trolox (TEAC=1) equivalent to the activity of a 1 mM solution of the given compound (Rice-Evans et al. 1996). Most of the common polyphenols have higher TEAC value compared to the endogenous antioxidant glutathione (GSH), whose TEAC is 1.0. Since free divalent metal ions such as iron generate free radicals by fenton reaction, chelation of free iron by polyphenols protects neurons from oxidative stress (Li et al., 2010) with implications for PD and Alzheimer’s disease (AD) (Sahu & Gray 1997, Sugihara et al., 1999). Flavonoids protect against oxidative damage either by forming complexes with iron or copper or by direct detoxification due to inherent structural characteristics that enhance the antioxidant potential (Bors et al., 1990).

### 2.2.2 Modulation of cell signalling pathways

Beyond antioxidant activities, polyphenols such as curcumin, resveratrol, quercetin and other flavonoids modulate cellular signalling pathways, that could affect the gene expression and interfere with cell death mechanisms (Williams et al., 2004). Polyphenols exert modulatory effects on neurons by selectively interacting with protein kinase and lipid kinase signalling cascades such as phosphatidylinositol 3-kinase (PI3 kinase or PI3K)/akt, tyrosine kinase, protein kinase C (PKC) and Mitogen activated protein kinase (MAPK) pathways (Agullo et al., 1997, Gamet-Payrastre et al., 1999, Matter et al., 1992, Schroeter et al., 2002, Spencer et al., 2003, Vlahos et al., 1994). Inhibitory or stimulatory actions at these pathways immensely affect the cellular functions by altering the phosphorylation states of target molecules and/or by modulating gene expression (Spencer, 2007). Many Polyphenols bind to ATP binding sites of proteins (Conseil et al., 1998) like PKC (Gamet-Payrastre et al. 1999, Kantengwa & Polla, 1991), protein kinase A (PKA) (Revuelta et al., 1997), calcium plasma membrane ATPase, mitochondrial ATPase and topoisomerase (Boege et al., 1996). For example, the stilbene resveratrol and the citrus flavones, naringenin and hesperetin have been shown to inhibit the activity of a number of protein kinases (Spencer 2007). Flavonoids and their metabolites can selectively interact within MAPK signalling pathways (Kobuchi et al., 1999, Kong et al., 2000).

**a. Extracellular signal-regulated kinase (ERK) pathway:** Some flavonoids are found to have inhibitory effect on the ERK pathway and some others activate this pathway. Flavonoids have close structural homology to specific inhibitors of ERK pathway, such as PD98059 (2‘-amino-3’-methoxyflavone), which is a selective non-competitive inhibitor of the mitogen activated kinase, MEK 1 (Alessi et al., 1995). Flavonoids and their metabolites may also act on this pathway in a similar manner. Certain flavonoids such as Quercetin and its O-methylated metabolites are shown to induce neuronal apoptosis by inhibition of ERK pathway (Spencer 2003). The MEK inhibitor PD98059 has been shown to effectively block inducible nitric oxide synthase (iNOS) expression and generation of nitric oxide (NO•) (Bhat et al., 1998), suggesting that flavonoids may also be capable of exerting anti-inflammatory actions via inhibitory actions on MEK1 within the ERK signalling pathway. Studies have indicated that flavanols (Huang et al., 2005, Li et al., 2004), flavones (Chen et al., 2004, Kim et al., 2001, Lee et al., 2003, Shen et al., 2002, Woo et al., 2006), and flavonols (Chen et al., 2005) are all capable of inhibiting the release of NO• by activated microglia via the down-regulation of iNOS gene
expression. Epicatechin (EC), and one of its metabolites, 3'-O–methyl-(−)- epicatechin (mEC), have been shown to stimulate phosphorylation of ERK1/2 and the downstream transcription factor cAMP response element binding protein (CREB) at physiological concentrations (Schroeter et al., 2007). Some polyphenols may act on the ERK pathway via acting through steroid-like receptors in neurons to modulate ERK and CREB-mediated gene expression. For example, resveratrol rapidly activates ERK signalling through alpha and beta estrogen receptors (Klinge et al., 2005).

b. c-Jun-N-terminal kinase (JNK): There are a number of potential sites where flavonoids may interact in the JNK pathway. EC and mEC have been shown to protect neurons against oxidative damage via a mechanism involving the suppression of JNK, and downstream partners, c-jun and pro-caspase-3 (Schroeter et al., 2001). The flavone, baicalein, significantly inhibits 6-hydroxydopamine (6-OHDA) induced JNK activation and neuronal cell death and quercetin suppresses JNK activity and apoptosis induced by hydrogen peroxide (Ishikawa & Kitamura 2000), 4-hydroxy-2-nonenal (4HNE) (Uchida et al., 1999) and tumour necrosis factor-alpha (TNF-α) (Kobuchi et al. 1999). Flavonoids may inhibit the activation of JNK pathway upstream by decreasing oxidative stress and maintaining calcium homeostasis (Davis 1999). They may also inhibit JNK activation by modulation of the apoptosis signal-regulating kinase 1 (ASK1) phosphorylation state, and its association with 14-3-3 protein, which is essential for suppression of cellular apoptosis (Zhang, L. et al., 1999). Investigations have indicated that flavonoids might have anti-apoptotic property by blocking oxidative stress induced activation of caspase-3 in neurons (Schroeter et al. 2001, Schroeter et al., 2000). Flavonoid o-quinones, formed from the intracellular oxidation of flavonoids also inhibit JNK and other MAPKs via the nucleophilic addition to the cysteine residues of these proteins (Spencer et al., 2004).

c. PI3 kinase pathway: Flavonoids can modulate signalling through the serine/threonine kinase, Akt/PKB, one of the main downstream effectors of PI3K in neuronal survival (Coffer et al., 1998). Experimental studies suggest that flavonoids inhibit PI3K via direct interactions with its ATP binding site. Indeed, one of the most selective PI3K inhibitors available, LY294002, was modelled based on the structure of quercetin (Matter et al. 1992).

In the current review, we have discussed the properties and therapeutic potential in PD of three important polyphenols: curcumin, resveratrol and tea polyphenols.

3. Curcumin

Curcumin, the yellow curry spice from the rhizome of turmeric is a polyphenolic compound with several beneficiary properties. Chemically characterized to be differuloyl methane (C_{21}H_{20}O_{6}) or 1,6-heptadiene-3,5-dione-1,7-bis(4-hydroxy-3-methoxyphenyl), curcumin exists as a group of compounds called curcuminoids.

3.1 Absorption

Although curcumin has several beneficial properties in vivo, a major limiting factor is its poor bioavailability. Its poor bioavailability is due to insolubility in aqueous media, poor absorption, rapid metabolism and fast systemic elimination. Following oral administration of curcumin (1.0 g / kg) in mice, 0.13 μg / ml was detected in the plasma after 15 min and peaked at 0.22 μg / ml at 1 hr. Plasma concentration of curcumin however declined to 5 ng /
ml at 6 h. Following intraperitoneal administration of curcumin (0.1 g/kg), only 2.25 μg / ml appeared in the plasma in the first 15 min. Upto 99% of curcumin and 85% of tetrahydrocurcumin was conjugated with glucuronide thus limiting their bioavailability. It has also been suggested that oral administration of curcumin results in very low absorption into the blood. At 1 h time point, the intestine, spleen, liver and kidney had 177.04, 26.06, 26.90, and 7.51 μg / g curcumin respectively, while the brain had as less as 0.41 μg / g. Mass spectrometry analysis suggested that curcumin was first biotransformed to dihydrocurcumin and then tetrahydrocurcumin which were subsequently converted to monoglucuronide conjugates (Pan et al., 1999). In a more recent study, curcumin was administered orally to rats at 500 mg / kg and the peak curcumin concentration of ~36 mg / whole tissue, in the intestine was reached at 1 h time point, while in the blood, liver and kidney, peak concentrations were reached at 6 h (Suresh & Srinivasan 2010). In a pilot human trial on cancer patients, the concentration of curcumin in the liver following oral administration (450 – 3600 mg/ day for 1 week prior to surgery) was measured. The results indicated that only trace levels of curcumin and its metabolites were present in the liver and in circulation and the order of magnitude was too less to exert pharmacological effects (Garcea et al., 2004). As a result of this constraint, research is now directed towards enhancing the bioavailability of curcumin. Animal and human studies suggest that administration of curcumin along with piperine / turmeric oil enhances its bioavailability (Bishnoi et al., 2010, Suresh & Srinivasan 2010). Administration of curcumin along with piperine enables curcumin to stay for longer in the tissues. When administered with piperine, curcumin was detected in the brain at 24, 48 and 96 h and at 48 h the levels of curcumin in the brain exceeded that found in the kidney (5.87 μg vs 1.16 μg) (Suresh & Srinivasan 2010). More recently, curcumin nanoparticles which are entirely water soluble have been suggested to enhance its bioavailability (Ray et al., 2011). Curcumin complexation with phosphatidyl choline (PC) and liposomal curcumin PC complex enhanced its bioavailability in terms of better absorption and improved pharmacokinetics (Gupta & Dixit 2011a, b).

3.2 Antioxidant properties of curcumin

The antioxidant activity of curcumin has been attributed to the various functional groups of curcumin. Jovanovic et al. suggest that the central methylenic group rather than the phenolic group is the H-atom donor, while others attribute its radical scavenging property to the phenolic group (Barclay et al., 2000, Priyadarsini et al., 2003) and the methoxy group further enhances this activity (Priyadarsini et al. 2003). The interactions between curcumin and biological relevant free radicals have been thoroughly investigated. Iwunze and McEwan demonstrated by biophysical techniques that curcumin directly detoxified the reactive nitrogen species peroxynitrite (PN) (Iwunze & McEwan 2004). Our group demonstrated that curcumin can protect neuronal mitochondria against PN induced nitrination and nitrosylation of brain mitochondrial proteins (Mythri et al., 2007). It was also shown that curcumin protected against PN induced protein nitrination and inhibition of enzyme activity of mitochondrial complex I with implications for PD (Mythri et al. 2007). Recently, we demonstrated that curcumin protects against PN mediated loss of mitochondrial membrane potential and mitochondrial integrity and curcumin derivatives offered improved protection compared to curcumin (Mythri et al. 2011). Mishra et al demonstrated that at low superoxide concentrations, curcumin effectively caused superoxide dismutation without itself undergoing any chemical change, but at higher
concentrations of superoxide, curcumin inhibited superoxide activity by reacting with it (Mishra et al., 2004). The enol structure with the intramolecular hydrogen bond of curcumin strongly enhances the free radical scavenging activity (Ohara 2005, Toniolo et al., 2002). In vivo, the antioxidant activity of curcumin could be mediated through antioxidant enzymes such as superoxide dismutase (SOD), catalase, and glutathione peroxidase (GPx). Curcumin has been shown to serve as a Michael acceptor, reacting with GSH and thioredoxin (Adams et al., 2005). Curcumin has been found to be at least ten times more active as an antioxidant than even vitamin E (Khopde et al., 2000). The phenolic and the methoxy group on the phenyl ring and the 1,3-diketone system seem to be important structural features of curcumin that can contribute to these effects. The antioxidant activity of curcumin increases when the phenolic group has a methoxy group at the ortho position (Motterlini et al., 2000).

3.3 Intracellular targets

The important molecular targets of curcumin include growth factors, growth factor receptors, transcription factors, transcription factor receptors, cytokines, enzymes and genes which regulate apoptosis. Curcumin activates the NF-E2-related factor 2/ Antioxidant Response Element (Nrf2/ARE) pathway which in turn induces synthesis of phase II antioxidant (Jeyapaul & Jaiswal, 2000, Wild et al., 1999), glutathione-S-transferase (GST) (Chanas et al., 2002, Ye et al., 2007), NADH quinone oxidoreductase 1 (NQO1) (Ye et al., 2007) and hemeoxygenase-1 (HO-1). Curcumin suppresses activation of the pro-survival transcription factor, nuclear factor kappa B (NFkB) by inhibition of IxB kinase (Jobin et al., 1999, Plummer et al., 1999). Suppression of NFkB leads to down regulation of cell cycle regulatory protein cyclin D1, cyclooxygenase 2 (COX2) and matrix metalloproteinase 9 (MMP-9) (Shishodia et al., 2005). Curcumin further, suppresses interleukin 6 (IL-6) mediated signal transducers and activators of transcription (STAT-3) phosphorylation and its subsequent nuclear translocation (Bharti et al., 2003). Curcumin, it has been reported, activates the peroxisome proliferator activated receptor γ (PPAR γ) and hence mediates expression of cyclin D1 and epidermal growth factor receptor (EGFR) (Shen & Xu, 2005). Curcumin down regulates the transcription factor, AP-1, by direct interaction with its DNA binding motif, (Bierhaus et al., 1997, Huang, T. S. et al., 1991) and also inhibits interleukin 1 alpha (IL-1α) and TNF-induced AP-1 activation (Xu, Y. X. et al., 1997). Moreover, curcumin targets the JNK pathway preventing JNK 1/2 and c Jun phosphorylation and caspase 3 activation (Yu et al., 2010). Curcumin probably interferes with the signalling molecules at the same level or proximally upstream of MAPKKK level, hence indirectly affects the JNK pathway (Chen, Y. R. & Tan 1998). Furthermore, curcumin induces anti apoptotic genes such as Bcl-2 and inhibits iNOS resulting in reduction of ROS, abrogates oxidative stress mediated cytochrome c release or caspase 3 activation, significantly increases levels of anti apoptotic proteins Bcl-2 and Bcl-xL and decreases levels of pro-apoptotic proteins Bax and Bad (Chen, J. et al., 2006, Chen, Y. R. & Tan 1998). Curcumin treatment causes a 3-7 fold increase in the modifier subunit of the GSH synthesizing enzyme, gamma-glutamyl cysteine ligase (γ-GCL), in both the neurons and astrocytes (Dale & Russell 1956, Lavoie, S. et al., 2009) and enhances GSH levels (Jagatha et al., 2008) probably by enhancing astrocytic efflux of GSH (Stridh et al., 2010). In addition, curcumin regulates iron metabolism by activation of iron regulatory protein and repression of ferretin, suggesting a role as an iron chelator (Jiao et al., 2006, Jiao et al., 2009).

Curcumin has been used as an antioxidant, antiseptic, anti-inflammatory, antibacterial and antitumor agent and has been used against various disorders such as arthritis,
cardiovascular diseases, lung fibrosis, gall stone formation, cardiotoxicity, diabetes, wound healing, AD and many more (Aggarwal 2006, Sharma et al., 2006). Recent studies have also shown curcumin to be protective against cancer (Duvoix et al., 2005), diabetes (Cheng et al., 2009, Peeyush et al., 2009) and for the reduction of blood cholesterol (Alwi et al., 2008, Ejaz et al., 2009, Jang et al., 2008). Curcumin protects against ischemia induced elevation in malondialdehyde (MDA) and lactate dehydrogenase (LDH) release in animal models (Dikshit et al., 1995). Faster restoration of muscle architecture upon curcumin treatment in injury models is probably mediated via NFkB activation (Thaloor et al., 1999).

3.4 Effects of curcumin in models of PD

More recently, curcumin has found its role as a therapeutic agent in models of neurological disorders such as stroke and AD (Lim et al., 2001, Thiyagarajan & Sharma 2004, Yang et al., 2005). Lim et al. have shown that dietary curcumin reduces amyloid pathology, oxidative stress and pro-inflammatory markers in transgenic mice, in addition curcumin reduces the levels of GFAP, a marker of astrocytic proliferation (Lim et al. 2001). Further, curcumin not only prevented formation of abeta aggregates but could also disintegrate preformed aggregates by directly binding to plaques (Yang et al. 2005). Curcumin can cross the blood brain barrier and is found to be least toxic in human subjects at high doses (Lao et al., 2006, Qureshi et al., 1992, Shankar et al., 1980, Shoba et al., 1998). It can detoxify ROS, prevent protein aggregation and induce neurogenesis in vivo (Kim, S. J. et al., 2008, Priyadarsini et al. 2003, Xu, Y. et al., 2007).

Since PD is a neurological condition involving oxidative and nitrosative stress pathways, curcumin can serve as a potential therapeutic molecule for PD. Although thiol reducing agents such as GSH have been evaluated for their anti-PD activity, their efficiency is limited due to poor ability to cross the blood brain barrier (Bharath et al., 2002). Curcumin on the other hand can cross the blood brain barrier and can be administered at higher doses without the risk of toxicity. Curcumin treatment protects substantia nigra (SN) neurons, improves striatal dopamine levels and chelates Fe^{2+}, following administration of 6-hydroxy dopamine (6-OHDA) in rats (Zbarsky et al., 2005). Curcumin treatment abrogates dopamine induced striatal neuron cell death (Luo et al., 1999). Curcumin treatment could further increase the density of dopaminergic neurons in the SN (Vajragupta et al., 2003). In all these studies, protection was afforded only by pretreatment with curcumin prior to administration of the toxin. A recent study in our laboratory indicated that chronic dietary consumption of turmeric caused an increase in the tyrosine hydroxylase (TH) positive neurons in the SN (Mythri et al. 2011). This is consistent with earlier reports suggesting that curcumin contributes to neurogenesis (Kim, S. J. et al. 2008, Xu, Y. et al. 2007). Similarly, our laboratory also demonstrated that curcumin derivatives with improved bioavailability offers better defense against 1-methyl-4-phenylpyridinium (MPP^{+}) in dopaminergic neurons (Mythri et al. 2011).

Curcumin treatment attenuates 3-nitropropionic acid mediated oxidative stress in vivo (Kumar, A. et al., 2007). PN is a highly labile reactive nitrogen species which inflicts severe cellular damage by nitrosative protein modifications. However, PN mediated nitrosative stress and mitochondrial dysfunction were attenuated by pretreatment with curcumin in vitro (Mythri et al. 2011). Curcumin mediated protection from PN toxicity is probably due to its direct interaction with and inactivation of PN in vitro (Iwunze & McEwan 2004, Mythri et al. 2007) or indirectly by enhancing GSH levels in vivo ((Jagatha et al. 2008, Mythri et al.
Curcumin down regulates iNOS which is probably mediated by the cJun/AP-1 pathway (Brouet & Ohshima 1995, Chan et al., 1998, Chen, J. et al. 2006). It has been suggested that curcumin binding to the AP-1 binding site on the promoter region of iNOS, results in its inactivation (Brouet & Ohshima 1995). Pan et al. have further reported that curcumin reduces iNOS mRNA levels and blocks induction of NFκB (Pan, M. H. et al., 2000). Rajeswari and colleagues have reported an increase in striatal dopamine and dihydroxy phenyl acetic acid (DOPAC) levels following curcumin injection in 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-injected mice (Rajeswari & Sabesan 2008). In addition curcumin restores mitochondrial membrane potential, causes elevation in Cu-Zn SOD and modulates NFκB nuclear translocation (Wang, J. et al., 2009a) by inhibition of IL6 and TNFα (Wang, S. L. et al., 2009b). Curcumin treatment inhibits AP-1 pathway by prevention of c-Jun phosphorylation (Luo et al. 1999) in addition to inhibition of JNK phosphorylation (Sawada et al., 2002, Yu et al. 2010) and caspase 3 activation (Yu et al. 2010). It has been reported that curcumin induces anti apoptotic genes such as Bcl-2 and inhibits iNOS resulting in reduction of ROS, abrogates oxidative stress mediated cytochrome c release or caspase 3 activation, significantly increases levels of anti apoptotic proteins Bcl-2 and Bcl-xL and decreases levels of pro-apoptotic proteins Bax and Bad (Chen, J. et al. 2006, Chen, Y. R. & Tan 1998). Curcumin induced cytoprotection was probably mediated by the Bcl-2 – mitochondria – reactive oxygen species (ROS) – iNOS pathway.

Oxidative damage of lipids results in the formation of aldehydes such as 4HNE and MDA, which can integrate into lipid membranes resulting in disruption of membrane integrity, energy crisis and eventually cell death. Increased levels of 4HNE adducts have been reported in PD brains (Yoritaka et al., 1996). Lipid peroxidation mediated mitochondrial damage and apoptosis were abrogated by curcumin treatment and this was probably by checking cytochrome c release and caspase and PARP activation (Zhu et al., 2004). Curcumin was further able to protect from 4HNE mediated alterations in mitochondrial respiratory function and redox metabolism, cytochrome c release, DNA fragmentation and PARP activation in PC12 cells suggesting a protective function against lipid peroxidation mediated damage (Raza et al., 2008).

Interestingly, not only is curcumin an antioxidant molecule but can also increase the levels of GSH, an endogenous antioxidant molecule, probably by enhancing astrocytic efflux of GSH (Stridh et al. 2010). Results from our laboratory and others have shown that curcumin enhances GSH levels in vitro and in vivo (Jagatha et al. 2008, Rajeswari 2006), in addition to enhancing the antioxidant enzyme activities of SOD and catalase in the striatum and mid brain of MPTP injected mice (Rajeswari 2006). In addition curcumin treatment results in a 3-7 fold increase in the modifier subunit of the GSH synthesizing enzyme, y-GCL, in both the neurons and astrocytes (Dickinson et al., 2003, LaVoie, M. J. et al., 2005). Curcumin mediated increase in the transcription of GCL genes is probably via regulation of the binding of transcription factors to the 12-tetradecanoate 13-acetate (TPA)-responsive elements (TRE) and electrophilic response element (EpRE) elements (Dickinson et al. 2003). It further activates the Nrf2/ARE pathway which in turn induces synthesis of phase II antioxidant enzymes such as GCL (Jeyapaul & Jaiswal 2000, Wild et al. 1999), GST (Chanas et al. 2002, Ye et al. 2007), NQO1 (Ye et al. 2007) and HO-1. We have used diester derivatives of curcumin (di-piperoyl, di-valinoyl and di-glutamoyl) with a resultant enhanced protection against GSH depletion mediated oxidative stress and toxin induced cell death in a dopaminergic neuronal cell line (Harish et al., 2010, Mythri et al. 2011). Particularly the di-glutamoyl derivative showed maximum protection due to its deesterification following
absorption thus providing glutamate as a precursor for GSH synthesis. Thus one could use such pro drugs with improved uptake and better radical scavenging properties to combat oxidative and nitrosative stress in various neurodegenerative disorders such as PD.

Curcumin further inhibits aggregation of α-synuclein in vitro (Pandey et al., 2008, Wang, M. S. et al., 2010) and aggregation of A53T mutant α-synuclein in SH-SY5Y cells in a dose dependent manner. Curcumin, thus enhances α-synuclein solubility rendering them nontoxic (Pandey et al. 2008). It also exhibits neuro-restorative properties as shown by its ability to disintegrate preformed α-synuclein fibrils (Ono & Yamada 2006). Curcumin pretreatment resulted in a reduction in aggregation of synphilin-1, a component of Lewy neuritis seen during PD, as a function of rotenone induced nitrosative stress, in SH-SY5Y dopaminergic cells suggesting its efficacy in ameliorating nitrosative stress induced damage (Pal et al. 2011).

Curcumin has been suggested to regulate proteins involved in iron metabolism. Increased brain iron content is a major risk factor for PD pathogenesis. It has been reported that the SN of PD brains has increased iron content (Sofic et al., 1988). Not only is iron responsible for the production of OH radical, but also promotes dopamine autooxidation in the SN neurons resulting in the release of H₂O₂ (Ben-Shachar et al., 1995). It has been reported that curcumin induces activation of iron regulatory protein and repression of ferretin and a reduction in levels of hepcidin, suggesting a role as an iron chelator (Jiao et al. 2006, Jiao et al. 2009).

Epidemiological studies have suggested that consumption of curcumin by the Asian Indians is probably the reason for the low incidence of AD / PD in India when compared to the Caucasians (Ganguli et al., 2000). A study exploring the effect of race and age on the prevalence of PD reported that there are approximately 40% less melanized nigral neurons in Indian brains when compared to Caucasian brains (Muthane et al., 1998). Furthermore, there was no change in the number of nigral neurons with advancing age in the Asian Indian population (Alladi et al., 2009). Hence the lower prevalence of PD in India despite lower numbers of nigral neurons suggests protective mechanism probably related to the dietary habits. Chronic consumption of spices such as turmeric provide antioxidant defense against various conditions such as diabetes and cancer (Sinha et al., 2003, Srinivasan 2005). We have recently demonstrated that chronic dietary consumption of turmeric ameliorated the MPTP induced neurotoxicity in mice (Mythri et al. 2011). Although curcumin is the most active ingredient of turmeric, it could be more effective in its natural milieu. Also the biological properties of natural extracts would probably be mediated by their various constituents rather than a single compound. Therefore it would be interesting to study the pharmacokinetics and pharmacodynamics of curcumin or the other biologically active components of turmeric in order to completely understand its mechanism of action.

4. Resveratrol

Resveratrol (3,5,4′-trihydroxy-trans-stilbene), a phytoalexin and a stilbenoid derived is a naturally occurring polyphenol with strong antioxidant properties (Baur & Sinclair 2006, Fremont 2000). Resveratrol is a dietary polyphenol found in the skin of grapes, raspberries, mulberries, pistachios and peanuts under normal conditions and synthesized by other medicinal and edible plants under stress conditions (Rocha-Gonzalez et al., 2008, Saiko et al., 2008). It exists as cis- and trans- isomers which are either free or bound to glucose (Mattiivi et al., 1995). The trans-form can undergo isomerisation to the cis-form when exposed to ultraviolet irradiation (Lamuela-Raventos et al., 1995). Experimental evidences in


**4.1 Absorption of resveratrol**

The oral absorption of resveratrol in humans is thought to occur mainly by transepithelial diffusion. However, a major portion of resveratrol administered orally is excreted via feces and urine (Wenzel & Somoza 2005). Following oral ingestion, trans-resveratrol is extensively metabolized in the enterocyte before it enters the blood and target organs. Since trans-resveratrol is photosensitive it is easily oxidized thus presenting unfavorable pharmacokinetics (Frozza et al., 2010). Extensive metabolism in the intestine and liver results in the bioavailability of resveratrol to be <1% and dose escalation and repeated dose administration does not significantly alter the bioavailability. Metabolic studies, both in plasma and in urine, have revealed that resveratrol is modified as glucuronides and sulfates. The sulfated and glucuronidated forms appeared within approximately 15 min of entering the bloodstream, and moderate consumption of red wine results in serum levels of resveratrol that barely reach the micromolar concentrations. In contrast, its metabolites, which may be the active principle, circulate in serum for up to 9 h (Saiko et al. 2008). However, reduced dihydroresveratrol conjugates, in addition to highly polar but unknown products, may account for ~ 50% of resveratrol following oral administration. Apart from the metabolism in the intestine and liver, colonic bacterial metabolism also contributes to resveratrol bioavailability (Walle 2011). Among these, extremely rapid sulfate conjugation by the intestine/liver appears to be the rate-limiting step in resveratrol's bioavailability. Although the systemic bioavailability of resveratrol is very low, accumulation of resveratrol in epithelial cells along the aerodigestive tract and potentially active resveratrol metabolites may still produce *in vivo* effects (North & Verdin 2004). Since low bioavailability of resveratrol is a major obstacle to biomedical applications, efforts are in progress to improve the absorption and slow down its metabolism (Biasutto et al., 2010). Oral and intravenous (i.v.) administration of resveratrol in human volunteers revealed that the absorption following oral administration of 25 mg oral dose was at least 70%, with peak plasma levels of resveratrol and metabolites at 491 ± 90 ng/ml (about 2 μM) and a plasma half-life of 9.2 ± 0.6 h. However, only trace amounts of unchanged resveratrol (<5 ng/ml) could be detected in plasma and most of the oral dose was recovered in urine.

**4.2 Effects of resveratrol in models of PD**

Similar to curcumin, the effectiveness of resveratrol is due to its ability to simultaneously target multiple pathways and proteins and thus may be effective in restoring homoestasis *in vivo*. The most important property relevant in abrogation of neuronal damage is its antioxidant potential. This has been extensively studied in different models and most of them refer to the systemic effects (Kovacic & Somanathan 2010). Experiments from our laboratory have showed that resveratrol prevented peroxynitrite mediated nitration of mitochondrial proteins and this was significantly higher compared to curcumin (Mythri et al. 2007). Apart from its antioxidant properties, resveratrol displays anti-cancer and anti-inflammatory properties. The anti-cancer properties arise due its ability to inhibit multiple target enzymes including kinases, cyclooxygenases, ribonucleotide reductase and DNA polymerases. Similarly, resveratrol protects the cardiovascular system against ischemic-
Therapeutic Potential of Polyphenols in Parkinson’s Disease

reperfusion injury, promotes vasorelaxation, protects the endothelium and displays anti-atherosclerotic properties and other functions thereby strongly supporting its role in cardioprotection. Resveratrol displays different properties depending on its concentration and also on the target cell and its physiology (Saiko et al. 2008). Resveratrol and its derivatives have shown promise in AD therapy and other neurodegenerative disorders (Albani et al., 2010) and more specifically on the inhibition of β-amyloid peptide aggregation (Richard et al., 2011).

Interestingly, resveratrol-mediated intracellular effects and protective mechanisms might be via its effects as an agonist of the sirtuins (also called SIRT or silent information regulator two-proteins), which belong to the histone deacetylase family (Pallas et al., 2008, Wang, F. et al., 2007, Zhuang et al., 2003). Sirtuins possess NAD⁺ dependent deacetylase activity and mono-ribosyltransferase activity and are found in most organisms (North & Verdin 2004, Yamamoto et al., 2007). SIRT1 is a major modulator of metabolism and displays therapeutic properties (Finkel et al., 2009, Harikumar & Aggarwal 2008, Karuppagounder et al., 2009). It has been hypothesized that by activating SIRT1, resveratrol modulates the activity of numerous proteins, including peroxisome proliferator-activated receptor coactivator-1alpha (PGC-1 alpha), the FOXO family, Akt (protein kinase B) and NFκB (Pallas et al., 2009). The beneficial effects of resveratrol as an anti-aging compound are believed to be mediated via the activation of SIRT1 (Hung et al., 2010).

Resveratrol administration in adult mice protected the SN dopaminergic neurons and striatal dopamine against the neurotoxic effects of MPTP (Anderson et al., 2006, Blanchet et al., 2008). Resveratrol administration also significantly protected mice from MPTP-induced motor coordination impairment, hydroxyl radical overloading, and neuronal loss with implications for PD therapy (Lu et al., 2008). In cell culture, treatment with resveratrol protected against MPP⁺ toxicity and oxidative damage by modulating the apoptotic markers (Bournival et al., 2009, Gelinas & Martinoli 2002). It has also been demonstrated that resveratrol effectively blocked the proteolytic cleavage of PKCδ and kinase activity induced by MPP⁺. In BV-2 microglial cells activated with lipopolysaccharide (LPS), pretreatment with resveratrol suppressed pro-inflammatory cytokine production and nitric oxide release.

It could be surmised that resveratrol protects against the dopaminergic neurodegeneration by modulating the PKCδ dependent proapoptotic signaling pathway and the inflammatory response in activated microglia (Gordon, 2010). Resveratrol significantly reversed the toxic effects of MPTP by increasing the levels of dopamine and its metabolites, GSH and activities of GPx and reducing lipid peroxidation along with enhanced behaviour performance indicating that resveratrol could target multiple pathways with therapeutic potential in PD (Anandhan et al., 2010). Interestingly, transfection with SIRT1 siRNA blocked MPP⁺-induced apoptosis in SH-SY5Y cells indicating that the toxicity of MPP⁺ was mediated via SIRT1 and possibly through the regulation of another pro-apoptotic protein BCL2/Adinovirus E1B 19kDa Interacting Protein 2 (BNIP2). In a similar study, Alvira and colleagues showed that although resveratrol prevented MPP⁺ induced death of cortical neurons in culture, the effect was not mediated via SIRT1 activation, since sirtinol - a SIRT1 inhibitor could not attenuate MPP⁺ toxicity (Alvira et al., 2007). Furthermore MPP⁺ decreased the expression of SIRT1 indicating that the antiapoptotic effects of resveratrol against MPP⁺ are independent of the stimulation of SIRT1 and depend on its antioxidant properties.
Other in vitro studies have also demonstrated the effects of Resveratrol against different neurotoxins (Chao et al., 2008, Okawara et al., 2007, Outeiro et al., 2007). The therapeutic role of resveratrol has been demonstrated in 6-OHDA induced in vivo model of PD. Ultrastructural analysis showed that resveratrol alleviated 6-OHDA-induced chromatin condensation, mitochondrial dysfunction and vacuolization of SN dopaminergic neurons. Resveratrol also reduced the levels of COX-2 and TNF-α mRNA in the SN suggesting that resveratrol exerts its effects via reduced neuroinflammatory reaction (Jin, F. et al., 2008). Oral treatment with resveratrol and its liposomal form prevented oxidative damage, prevented neuronal damage and restored motor behaviour in a 6-OHDA rat model of PD (Khan et al., 2010, Wang, Y. et al., 2011). The study also showed that resveratrol liposome offered better therapeutic effect than free resveratrol due to increased bioavailability. Oxyresveratrol, a polyhydroxylated stilbene from mulberry showed improved protection against 6-OHDA compared to resveratrol owing to its antioxidant activity, blood-brain barrier permeability and water solubility (Chao et al. 2008). Pinostilbene, a methylated derivative of resveratrol showed improved cellular bioavailability and better defense against 6-OHDA neurotoxicity in neuronal cell culture (Chao et al., 2010b). Lee et al. demonstrated that resveratrol protected SH-SY5Y neuroblastoma cells from apoptosis induced by dopamine (Lee, M. K. et al., 2007).

Experiments in rat primary midbrain neuron-glia cultures demonstrated that resveratrol protected dopaminergic neurons against LPS-induced neurotoxicity in a dose and time-dependent manner via inhibition of microglial activation and suppression of release of proinflammatory factors. The anti-inflammatory effects and therapeutic effects against LPS toxicity was attributed to the inhibition of NADPH oxidase-dependent generation of reactive oxygen species and attenuation of translocation of the cytosolic subunit (p47) of NADPH oxidase to the cell membrane. This mechanism was substantiated when resveratrol was not effective in cultures from NADPH deficient mice. This is also related to an attenuation of MAPK and NFκB signalling pathways in microglia (Zhang, F. et al., 2010). Bureau and colleagues demonstrated that resveratrol reduced apoptotic neuronal cell death induced by neuroinflammation in an LPS cell model and strongly decreased the mRNA expression of two proinflammatory genes, IL-1α and TNF-α (Bureau et al., 2008). Administration of a botanical extract (Regrapex-R) prepared from whole grape Vitis vinifera and Polygonum cuspidatum, enriched with polyphenols including resveratrol, to a drosophila transgenic model of PD overexpressing alpha-synuclein caused dose-dependent scavenging effects on ROS, protected mitochondria from oxidative damage and improved locomotor dysfunction and extended lifespan (Long et al., 2009). Resveratrol was able to protect neuronal cells in culture against toxicity arising from two aggregation-prone proteins, the AD-involved amyloid-beta (1-42) peptide (Abeta42) and the familiar PD linked alpha-synuclein (A30P) [alpha-syn(A30P)] (Albani et al., 2009).

5. Tea polyphenols

Tea is one of the most frequently consumed beverage of the world. Out of the 2.5 million metric tons of dried tea manufactured annually, ~20 % is green tea, which is mainly consumed in China and Japan and ~78 % is black tea, which is consumed in Western countries (Chen, L. et al., 1997). Although consumption of tea as an ancient beverage and a lifestyle habit has been recognized, its potential pharmacological actions beneficial to human health have been recently reported (Weinreb et al., 2004). Human epidemiological and
animal experiments indicate that consumption of green and black tea might protect the brain against aging and reduce the incidence of dementia, AD and PD, which might be one of the mechanistic factors underlying the significantly lower rates of age-related neurological disorders among Asians compared to European or American populations (Mandel, S. A. et al., 2008). Green tea is non-fermented and contains mainly catechins that accounts for 30-40% of its dry weight. Catechins are polyphenols that have been suggested to contribute to the therapeutic properties of green tea. On the contrary, black and red teas are extensively fermented and oolong tea is semi-fermented which catalyzes the oxidation and polymerization of catechins thereby reducing their content to 3-10% of dry weight. The catechins from green tea are polyphenols representing flavonoids. The eight major catechins of green tea are (+)-catechin (C), (+)-catechin gallate, (+)-gallocatechin, (+)-gallocatechin gallate, (-)-epicatechin (EC), (-)-epicatechin gallate (ECG), (-)-epigallocatechin (EGC), and (-)-epigallocatechin gallate (EGCG) (+ and – represent cis and trans isomers respectively). EGCG is the most abundant catechin (Higdon & Frei 2003). Green tea in addition to catechins contain 2-3% of caffeic acid and 1-6% of amino acids (theanine constitutes about 50% amino acid content) (Higdon & Frei 2003).

5.1 Absorption
The in vivo metabolism of green tea catechins has been very well documented in humans and laboratory animals (Li, C. et al., 2000, Pietta et al., 1998). In humans, orally administered catechins are absorbed, metabolized and largely excreted within 24 h. Catechins were detected in rat plasma and bile as free or sulphated and glucuronidated conjugates (Harada et al., 1999). The bioavailability of green tea polyphenols in vivo depends on the route of administration. After intragastric administration of decaffeinated green tea (DGT) (200 mg/kg) in rats, 13.7% of EGC and 31.2% of EC were shown in the plasma, but only 0.1% of EGCG was bioavailable (Chen, L. et al. 1997). Intravenous injection of DGT (25 mg/kg) showed that, the beta-elimination half-lives of EGCG, EGC, and EC were 212, 45, and 41 min respectively (Chen, L. et al. 1997). Recent studies have suggested that the EC metabolites such as epicatechin glucuronide and 3’-O-methylated epicatechin glucuronide formed after oral ingestion in rats were detectable in the brain (Abd El Mohsen et al., 2002). The results from the distribution study suggest that EGCG is mainly excreted through bile, and that EGC and EC are excreted through both the bile and urine (Spencer 2003).

5.2 Antioxidant functions of tea polyphenols
There is growing evidence that catechins exert protective role in neurodegeneration. Several studies have suggested that the potential curative and therapeutic effects of tea polyphenols could be attributed to their free radical scavenging capacity. Catechins are powerful hydrogen donating antioxidants and scavengers of reactive oxygen and nitrogen species in vitro (Lee, S. et al., 2000, Matsuoka et al., 1995, Oliver et al., 1990). Tea polyphenols may also regulate antioxidant enzymes like SOD and catalase in mouse striatum (Levites et al., 2001). Progressive iron accumulation in SN pars compacta of PD patients is well established and is considered to be a major contributor to oxidative stress (Gerlach et al., 1994, Riederer et al., 1989). Green tea polyphenols can stoichiometrically bind ferric iron to form an inactive iron polyphenol complex (Grinberg et al., 1997). In rat brain tissue, green tea and black tea extracts were shown to inhibit lipid peroxidation promoted by iron ascorbate in homogenates of brain mitochondrial membranes (Levites et al., 2002b). EGCG inhibited
paraquat-induced MDA production in rat liver microsomes by scavenging superoxide radicals and by iron chelating activity (Liou et al., 2001).

### 5.3 Functions of tea and its constituent polyphenols in PD

Several prospective studies have assessed the association between coffee/tea consumption and PD risk, but the results are inconsistent. Kandinov and colleagues carried out a retrospective study on 278 consecutive PD patients and assessed the data on smoking and coffee or tea consumption and found that disease progression was not affected by cigarette smoking, tea or coffee consumption (Kandinov et al., 2007). In contrast, the same group demonstrated that cigarette smoking, coffee and tea drinking may protect against PD and delay the onset of disease (Kandinov et al., 2009). According to their study in human patients, while consumption of more than 3 cups of tea per day delayed the onset of motor symptoms by 7.7 years, coffee consumption advanced the age of PD onset by 4.8 years. In a related study, Barranco Quintana and colleagues compiled data from human studies carried out by different groups and observed a clear protective effect of tea consumption and this protective effect was more evident in the Chinese populations (Barranco Quintana et al., 2009). They also suggest that the low rate of tea consumption among persons with PD could provide valuable insight into the pathology. A population based study in ethnic Chinese to understand the role of environmental factors and dietary intake demonstrated that while there was a dose-dependent protective effect of PD in coffee and tea drinkers and smokers, a history of exposure to heavy metals increased the risk of PD (Tan, E. K. et al., 2003). Tan et al. observed that while black tea, a caffeine-containing beverage, showed an inverse association with PD risk, green tea drinking was unrelated to PD (Tan, Louis C. et al., 2008). A study among Finnish subjects indicated that coffee drinking is associated with a lower risk of PD and increased tea consumption is associated with a lower risk of PD (Hu et al., 2007). Since most of these studies are based on survey among PD patients, it would be difficult to draw significant conclusions.

However, experiments in cell and animal models have clearly indicated the therapeutic effects of tea extracts and its constituent catechin, EGCG against PD. These curative properties might be due to their ability to act as antioxidants, modulators of intracellular neuronal signalling, metabolism, cell survival/death genes, and mitochondrial function (Mandel, S. A. et al. 2008, Mandel, S. A. et al., 2011). With reference to PD therapy, EGCG has a dual therapeutic effect in vivo (Kang et al., 2010). Firstly, EGCG is a naturally occurring catechol-O-methyl transferase (COMT) inhibitor which strongly inhibited human liver COMT-mediated O-methylation of L-DOPA in a dose-dependent manner and decreased the accumulation of 3-O-methylatedopas in the plasma and striatum in vivo following oral administration of L-DOPA + carbidopa. In addition, EGCG also protected against glutamate-induced oxidative cytotoxicity in hippocampal cultures through inactivation of the NFκB-signaling pathway in vitro and against kainic acid-induced oxidative damage and neurodegeneration in the hippocampus in vivo.

Many studies related to therapeutic effects of tea polyphenols in PD have been carried out in the Parkinsonian model involving the neurotoxin 6-OHDA and have obtained varied results. Rats treated with black tea either prior to or after 6-OHDA provided significant protection in terms of behavioural and motor functions, antioxidant levels, apoptotic markers, dopaminergic neuronal numbers and expression of tyrosine hydroxylase (TH). However, the degree of improvement in motor and neurochemical deficits was more
prominent in 6-OHDA rats pre-treated with the extract (Chaturvedi et al., 2006). Green tea polyphenols protected SH-SY5Y neuroblastoma cells against apoptosis induced by 6-OHDA as evidenced by lowered apoptotic markers, restoration of mitochondrial integrity, decreased oxidative damage and intracellular free Ca\(^{2+}\) and inhibition of auto-oxidation of 6-OHDA. Further, the cells were protected against 6-OHDA-induced increase in levels of NO and protein nitration, and over-expression of neuronal NOS (nNOS) and iNOS. These results indicate that green tea polyphenols ameliorate 6-OHDA toxicity via attenuation of the ROS-NO pathway (Guo et al., 2005). The same group confirmed these effects against 6-OHDA in vivo in mid brain and striatum again emphasizing the involvement of the ROS-NO pathway (Guo et al., 2007). In order to identify the active ingredient against 6-OHDA, five catechins [EGCG, ECG, EGC, EC and (+)-C] representing the most prominent tea polyphenols were compared with regard to their effects on 6-OHDA-induced apoptosis in PC12 cells and the protective effects of the five catechins were in the order ECG > EGCG > EC > (+)-C > EGC and the antiapoptotic activities appear to be structurally related to the 3-gallate group (Jin, C. F. et al., 2001, Nie et al., 2002). In a related study, it was showed that EGCG protected against 6-OHDA via stimulation of PKC and modulation of cell survival/cell cycle genes such as Bax, Bad, Mdm2, Bcl-2, Bcl-w, and Bcl-x(L) and also by potent antioxidant and iron chelating actions thereby preventing nuclear translocation and activation of cell death promoting NF\(\kappa\)B (Levites et al., 2002a). Since the stability and bioavailability of EGCG are restricted, Chao et al. generated and tested a pro-drug representing a fully acetylated EGCG (pEGCG) in a PD cell model (Chao et al., 2010a). They found that pEGCG displayed improved protection compared to EGCG against 6-OHDA toxicity in SH-SY5Y neuroblastoma cells probably via activation of Akt pathway and reduced caspase-3 activity. In contrast, when Leaver et al. administered rats with EGCG for 14 days followed by exposure to 6-OHDA, they observed that EGCG produced subtle symptomatic relief in lesioned animals and could not prevent neuronal damage to a significant extent (Leaver et al., 2009).

Tea extracts and EGCG have displayed therapeutic effect in other toxic models of PD. Tea extract and EGCG protected against MPTP mediated dopamine loss and neurotoxicity in vivo and this effect could be mediated by inhibition of NOS (Choi et al., 2002, Kim, J. S. et al., 2010, Li, R. et al., 2006). In a related study, it was demonstrated that green tea extract and EGCG protected against MPTP neurotoxicity via antioxidant effects and inhibition of monoamine oxidase-B (MOAB) (Levites et al. 2001). Mechanistically, green tea polyphenols can ameliorate neuronal damage via inhibitory effects on dopamine transporters (DAT), through which they block MPP\(^+\) uptake and protect dopaminergic neurons against MPP\(^+\)-induced injury (Pan, T. et al., 2003). Similarly, Hou et al. (2008) demonstrated that EGCG attenuated cell death in PC12 cells induced by the herbicide and PD toxin paraquat (PQ) by maintaining mitochondrial membrane potential, inhibiting caspase-3 activity and down regulating the expression of pro-apoptotic protein second mitochondrial-derived activator of caspase (SMAC) in cytosol. However, in rotenone-treated mesencephalic cultures and organotypic striatal cultures, EGCG protected striatal slices to a limited extent by counteracting NO production by rotenone but was not significantly effective against rotenone toxicity in mesencephalic cultures (Moldzio et al., 2010).

Green tea could also protect against environmental toxin-induced cell injury and neuroinflammation with implications for PD. Tai and Truong (2010) reported that EGCG reduced dichlorodiphenyl-trichloroethane (DDT)-induced cell death in dopaminergic SH-
SY5Y cells suggesting that EGCG might activate a protective mechanism that might alleviate organochlorine pesticide-induced cell injury. Likewise, EGCG also inhibited LPS-activated microglial secretion of NO and TNF-α through the down-regulation of iNOS and TNF-α expression thereby protecting against neuroinflammation and injury in dopaminergic neurons (Li, R. et al. 2004). The antioxidant property of EGCG is also beneficial against protein aggregation in PD. Iron induces toxic aggregation of monomeric alpha synuclein leading to neurodegeneration in dopaminergic neurons. In the MPTP model, iron accumulation is also linked to NO-dependent mechanism. Mandel et al. showed that EGCG prevented the accumulation of iron and alpha-synuclein in SN thereby preventing neuronal damage (Mandel, S. et al., 2004). However, green tea and its constituent polyphenols could not protect PC12 cells against L-glutamate and MPP⁺ (Mazzio et al., 2001).

6. Conclusions

Most of the drugs currently used in PD therapy replenish the brain dopamine and provide symptomatic relief during early PD. However, many patients develop motor complications with chronic treatment. Further, these drugs do not slowdown or prevent the progression of the disease since their ability to prevent neuronal damage has not been comprehensively validated in humans. Therefore, novel therapies with possible curative properties are being exploited for adjunctive therapy. It is well documented that neurodegeneration in PD is contributed by several interrelated disease pathways but the molecular mechanisms of their relationships and their chronology have not been completely understood. Therefore, any molecule(s) with therapeutic potential in PD should be simultaneously effective against multiple pathways. In this regard, several in vitro and in vivo studies using PD models have tested natural antioxidants and molecules for their ability to protect against neurodegeneration with implications for therapy. The most prominent are the polyphenolic antioxidants obtained from plant sources. Most of these compounds display antioxidant, anti-inflammatory and anti-cancer properties, cross the blood-brain barrier and prevent neuronal damage in neurological disorders. Recent studies strongly support the clinical application of these compounds in PD. The current chapter explores the therapeutic potential of polyphenols such as curcumin, resveratrol and tea polyphenols in PD. These three polyphenols share common biological properties including antioxidant function, low toxicity and therapeutic benefits in the brain of different PD models. The available data indeed indicate that these polyphenols could have therapeutic potential in PD. Although several studies have been carried out in the recent years, the biological properties and health benefits of these polyphenols have not been completely understood. However, the most important limiting factor for the therapeutic application of these compounds in PD is their limited bioavailability in vivo. Further, the delivery of compounds specifically to mid-brain dopaminergic neurons for maximum therapeutic effect has not been completely standardized. Apart from this, studies are also needed to test the safety and efficacy in humans to translate the experimental results for therapeutic application in PD patients.

7. Acknowledgements

This study was financially supported by the Department of Science and Technology (No. SR/FT/L-152/2005 to MMSB). RBM is supported by an extended senior research fellowship from the Council for Scientific and Industrial Research (CSIR), India. GH is supported by a
senior research fellowship from Indian Council for Medical Research (ICMR), India. NR is supported by a junior research fellowship from CSIR, India.

8. References


Chaturvedi RK, Shukla S, Seth K, Chauhan S, Sinha C, Shukla Y & Agrawal AK (2006) Neuroprotective and neurorescue effect of black tea extract in 6-hydroxydopamine-


www.intechopen.com
Therapeutic Potential of Polyphenols in Parkinson’s Disease


Therapeutic Potential of Polyphenols in Parkinson’s Disease


www.intechopen.com


Sugihara N, Arakawa T, Ohnishi M & Furuno K (1999) Anti- and pro-oxidative effects of flavonoids on metal-induced lipid hydroperoxide-dependent lipid peroxidation in


Parkinson's disease (PD) is characterised clinically by various non-motor and progressive motor symptoms, pathologically by loss of dopamine producing cells and intraneuronal cytoplasmic inclusions composed primarily of ?-synuclein. By the time a patient first presents with symptoms of Parkinson's disease at the clinic, a significant proportion of the cells in the substantia nigra have already been destroyed. This degeneration progresses despite the current therapies until the cell loss is so great that the quality of normal life is compromised. The dopamine precursor levodopa is the most valuable drug currently available for the treatment of PD. However for most PD patients, the optimal clinical benefit from levodopa decreases around five to six years of treatment. The aim of the chapters of this book is to work towards an understanding in the mechanisms of degeneration and to develop disease modifying therapies.

How to reference
In order to correctly reference this scholarly work, feel free to copy and paste the following:
