Review Articles

Efficacy of Mint and Tea Extracts in Regulating Pregnancy Associated Disorders- A Review

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ABSTRACT

Stress during pregnancy is a causative factor for inducing the materno-fetal morbidity and mortality. Efforts to prevent or treat pregnancy stress are not progressive. Delivery remains the ultimate treatment for pregnancy stress associated complications. The current review deals with oxidant/antioxidant status, stress-inducible factor and effect of various antioxidant therapies during pregnancy in particular context to gestational hypertension, gestational diabetes mellitus, preeclampsia, and *Ureaplasmaurealyticum* infection. Different alternative drugs are employed to increase the endogenous antioxidant level. The possible positive effect on administration of mint-black tea, black tea and green tea in managing the oxidative/nitrative stress in pregnancy-associated complications are discussed. The review suggests the protective effect of mint-tea and tea extracts against placental hypoxia-induced oxidative/nitrative stress. Since treatment of the disease with modern medicine is often associated with severe side effects that may affect both mother and fetus,tea and mint-tea extracts with natural medicinal properties can be recommended as an alternative herbal remedy for pregnancy-associated complications to reduce oxidative/nitrative stress and enhance live fetal delivery.

Key Words: Mint, Tea, Pregnancy Associated Disorders, Oxidative Stress

1. OXIDATIVE STRESS AND WOMEN HEALTH

Oxidative stress (OS) results from an imbalance between prooxidants (free radical species) and antioxidants (free radical scavenger) (1). Reactive oxygen species (ROS) are a double-edged sword, which serves as essential signal molecules in both physiological and pathological processes in the female reproductive tract. In a healthy body, ROS and antioxidants remain in balance. When the balance is disrupted towards overproduction of ROS, it may portray the OS. Oxidative stress appears to cause/ participate in the pathogenesis of several human diseases including neurodegenerative disease, cancer, aging, cardiovascular disease, and inflammatory disorders (2). OS in women's health has been of interest only for the last two decades. The gap between the necessary research and the clinical practice has been the main reason for the lack of progress. OS influences the entire reproductive lifespan of a woman and even in menopause period. ROS affect multiple physiological processes from oocyte maturation to fertilization, embryo development, diabetes-related congenital malformations, gestational hypertension, preeclampsia and eclampsia (3).

2. PREGNANCY STRESS

Pregnancy is a state of oxidative stress (OS) originating from an increased elevation of placental mitochondrial activity and overproduction of reactive oxygen species (ROS). In addition, to placenta also produces other stress-inducible factors, which includes ROS interacting with nitric oxide, carbon monoxide, and peroxynitrite. Therefore, embryonic and placental cells are susceptible to oxidative stress (4). Hence, OS may have pronounced effects on placental function such as trophoblast differentiation,

proliferation, and vascular reactivity. The abundance of ROS may exhibit the causes of many pregnancy-related complications. In the 1st trimester of human pregnancy, blood perfusion into the intervillous space may facilitate the burst of OS. Also, OS induces the nitrative stress (NS) in the placenta, resulting alteration in placental vital function. Research suggest that some of the pregnancy complications are associated with placental hypoxia-induced oxidative stress, which is mediated by the raises of ROS(5). Hence, the early pregnancy loss is initiated by an inability to mount an efficient antioxidant defense system and merely induce the exaggeration of OS. However, the late pregnancy may influence enhanced OS in pregnancy-associated complications such as gestational hypertension, diabetes mellitus, preeclampsia and *Ureaplasmaurealyticum* in association with improper trophoblast invasion (apoptosis and deportation) and altered placental vascular reactivity by poor replacement of endothelial cells in the lining of a spiral artery. This results in intrauterine growth retardation of the developing fetus.

3. PREGNANCY STRESS ASSOCIATED COMPLICATIONS

3.1 GESTATIONAL HYPERTENSION

Gestational hypertension (GH) or pregnancy-induced hypertension (PIH) is usually defined as having a blood pressure higher than 140/90 mm Hg measured on two separate occasions, more than 6 hours apart, without the bearing of protein in the urine and diagnosed after the 20th week of gestation. It is more common in women with gestational diabetes, but no consensus has been reached. There is little direct clinical evidence on the reverse issue, but data are presented to suggest that pregnancy-induced hypertension may only predispose to gestational diabetes when its etiology is gestational hypertension and not preeclampsia. The relation between gestational diabetes and pregnancy-induced hypertension is not well understood. Several lines of clinical evidence hypothesized that reduction in uteroplacental perfusion leads to high blood pressure. However, Joey and his colleagues (2001) reported placenta as a central culprit in all pregnancy-related complications which include hypertension (6).

3.2 GESTATIONAL DIABETES MELLITUS

Gestational diabetes mellitus (GDM) is a complex, chronic illness requiring continuous medical care with multifactorial risk reduction strategies beyond glycemic control. It is a glucose intolerance of varying severity with onset during pregnancy and occurs in 1-14% of patients. Both patients with GDM and their offspring have a higher risk of developing type 2 diabetes later in life thereby increasing their fetal morbidity and mortality. Women with type 2 diabetes are at increased risk of hypertension during pregnancy. Several lines of evidence agreed that pathogenesis of GDM is initiated by the significant decrease in insulin sensitivity and insulin secretion and its mediated glucose intolerance (7). The increased insulin resistance is widely caused by the post-insulin receptor events and is probably brought about by the cellular defects exhibited due to the elevated levels of estrogen, progesterone, cortisol, and placental lactogen (8). Therefore, insulin resistance mediated growth promoting hormones are requisite for maturation and development of the placenta. This process usually starts between 20 and 24 weeks of gestation. As the placenta grows, more of these hormones are produced, and the risk of insulin resistance becomes greater. Normally, the pancreas is able to make additional insulin to overcome insulin resistance, but when the production of insulin is not enough to overcome the effect of the placental hormones, results in gestational diabetes (9).

3.3 PREECLAMPSIA

Preeclampsia (PE) is a pregnancy-specific multi-organ disease that adversely affects the mother by vascular dysfunction and the fetus by intrauterine growth restriction. It affects around 6 to 12% of all pregnancies of which most are primigravida (10). Preeclampsia can lead to problems in the kidneys, brain, liver and the clotting system. Risks for the baby include poor growth and prematurity. PE is characterized by high blood pressure, large amounts of protein in the urine and swelling or edema especially in the and face and hands (11). If undiagnosed, preeclampsia may lead to eclampsia, a serious condition that results in both mother and fetus at risk and in rare case causes death. The exact cause of these maternal changes concerning preeclampsia is not clear. Experts believe that it begins in the placenta, the organ that nourishes the fetus throughout pregnancy (12). During early pregnancy, new blood vessels are developed and evolve to transfer blood to the placenta efficiently. In women with preeclampsia, these blood vessels are not properly developed and narrower than normal blood vessels which react differently to hormonal signaling

and limit the blood flow through these vessels (13). Despite extensive research, a proper understanding of the pathogenesis of this disease is enigmatic.

3.3.1 Pathophysiology of preeclampsia

The pathophysiology of PE involves the placental infarcts and sclerotic narrowing of spiral arteries, with a characteristic reduction in endovascular invasion by cytotrophoblasts and inadequate remodeling of the uterine spiral arteries (14). During normal gestation, invasive cytotrophoblasts downregulate the expression of adhesion molecules that are the characteristic feature of epithelial cell and adopt a cell surface adhesion molecule that is the typical nature of endothelial cells, a process that is referred as pseudovasculogenesis (15). However, during preeclampsia ineffective replacement of endothelial cell by cytotrophoblast in the spiral artery occurs leading to adverse effects in both maternal and placental region that may eventually lead to placental hypoxia (16). Therefore, the placenta experience low oxygen due to the restricted blood supply to the fetus and it finally results in immature birth/low weight baby (**Figure 1**).

Figure 1: Normal and stressed pregnancy placentation

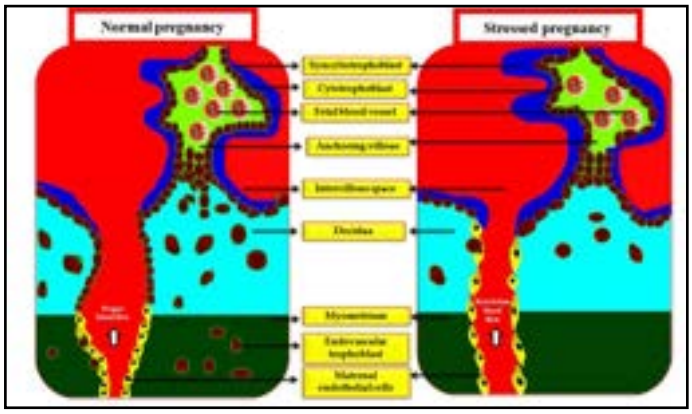
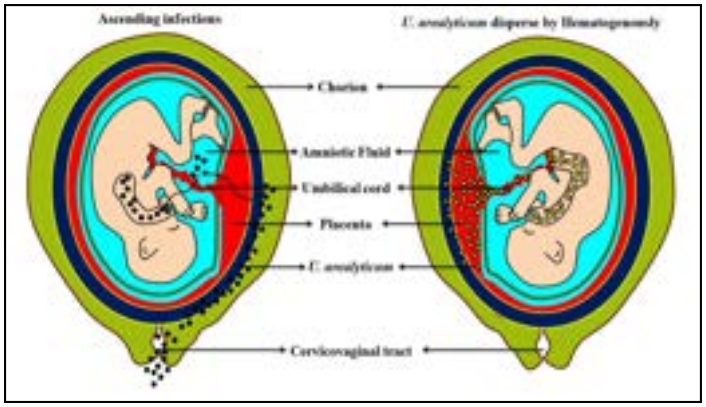


Figure 1represents the trophoblast cells invading the lumen of endothelial cells in the spiral artery and proper materno-fetal circulation during normal pregnancy. However, during stressed pregnancy, a trophoblast cell fails to replace the endothelial cells in a maternal uterine spiral artery, which consequently render the improper invasion of trophoblast leading to improper placentation. It may affect the transportation of nutrients to the fetus leading to diminished fetal organogenesis, fetal growth restriction, and preterm delivery.

3.4 PREGNANCY AND INFECTION

Infection in the placenta can alter its completeness and size (17). There are clinical reports of a wide variety of organisms found to cause intrauterine infections which include Group B *Streptococci, Ureaplasmaurealyticum, Morganella morganii, Listeria monocytogenes, Chlamydia species, Capnocytophaga, Herpes simplex virus, parvovirus, human immunodeficiency virus* and *adeno-associated virus*. Infection can be either restricted to the chorioamnion or cross it to reach the amniotic fluid. The primary process in the etiology of intrauterine inflammation is believed to be ascending bacterial invasion from the cervicovaginal tract, and hematogenously infect the fetus **(Figure 2)** (18). Bacteria may then diffuse across the chorioamniotic membranes and cause chorioamnionitis (19). Chorioamnionitis has severe complications for the fetus through the initiation of preterm delivery. It increases the risk for cerebral palsy to the children (20).

Figure 2: *Ureaplasmaurealyticum*spread by two mode like ascending and hematogenously



3.4.1 Pregnancy and *Ureaplasmaurealyticum* infection

Ureaplasmaurealyticum is a typical commensal of the urogenital tract of humans, is gaining identification as an important opportunistic pathogen on pregnancy (21). It is the most typical organism isolated from the chorioamnion, even in the presence of intact fetal membranes. The colonization of U. urealyticum, a vaginal commensal was also found to be associated with histological chorioamnionitis (22).U.urealyticum is associated with inflammation, premature spontaneous delivery, infertility, spontaneous abortion, stillbirth, and early and perinatal morbidity and mortality, septicemia, meningitis and pneumonia in newborn infants (23). Ureaplasma can be detached from the lower genital tract of 60-80% of parturiency.

They are transmitted from mothers to neonates, either in utero or during passage via the infected birth canal. Kundsin and his colleagues (1996)have shown that a strong association between Ureaplasma infections of the placenta along with fetus may portray the low birth weight of the neonate (24).

3.4.2 Preeclampsia and *Ureaplasmaurealyticum* infection

U. urealyticum invasion in the amniotic cavity in around 50% of the observed preeclamptic subjects as detected through both the culture and PCR technique (25). Similarly, a study in India revealed that the degree of *U. urealyticum* infection in the normotensive subjects, the recovery rate is 56% and a 64% recovery rate in preeclamptic subjects. *U. urealyticum* infection will complicate the normotensive pregnancy with stress, making the condition more similar to preeclampsia and will worsen the existing complication when super-imposed with preeclampsia.

Microbial invasion though results in the inflammatory process, is commonly accompanied by OS (26). Due to the immunosuppressive properties of choriodecidua and the low pathogenicity of *U. urealyticum*, it can colonise the choriodecidua and the amniotic fluid without causing inflammation and consequently inducing preterm birth (27). A highly significant increase in the accumulation of ROS intermediates in the preeclamptic and normotensive tissue with infection has been found to trigger OS (30).Reports suggest that reactive oxygen and nitrogen intermediate produced in response to infection play a major role in pathogenesis of *U. urealyticum*, thereby leading to cell apoptosis(29).*U. urealyticum* has been reported to alter the host catalase activity (29). Preeclamptic and normotensive subjects with infection resulted in a decrease in the antioxidant status when compared with uninfected counterparts (30).

3.4.3 *Ureaplasmaurealyticum* interact with many stress inducible factors

To maintain homeostasis in response to stress, cell apart from inducing antioxidants, increases the level of HSPs (Heat shock proteins) which protects them from damage. HSP70 is one of the major cytosolic chaperones with multiple functions involved in regulating cell homeostasis. We have reported that HSP70 was increased in *U. urealyticum* infected pregnant women (30). Therefore increased expression of HSP70 confers a degree of protection to the cells from apoptosis as confirmed by the ultramicroscopic observations.

Nuclear factor kappa-light-chain-enhancer of activated B cells (NFκB), a primary inflammatory mediator is increased during preeclampsia and normotensive explants with *U. urealyticum* infection suggesting the presence of inflammation under such conditions. Association of preeclampsia with inflammation demonstrates the involvement of infection in the inflammatory complications of preeclampsia. The results indicate that *U. urealyticum* is one of the key factors for the onset of preeclampsia. The increase in TNFα (Tumor necrosis factor-α), an inflammatory and apoptotic protein was noted during both preeclampsia and *U. urealyticum* infection suggests the chances of inflammation and apoptosis under these conditions.

Apoptosis signal regulating kinase 1 (ASK1), the apoptotic mediator was increased significantly in preeclamptic explants with*U. urealyticum* infection suggesting the increasing susceptibility of placental cells to apoptosis during *U. urealyticum* infection. Thec-Jun N-terminal kinase 1 (JNK1) expression was upregulated in all the conditions and alternatively the increase in JNK2 expression was insignificant in all the explants. This emphasizes the antiapoptotic function of JNK1 and proapoptotic function of JNK2 (28). Bcl2 an important antiapoptotic protein is increased significantly in the explants during preeclampsia and *U. urealyticum* infection indicating the presence of a protective mechanism for reducing the mitochondrial cytochrome c release by stabilizing the mitochondrial membrane integrity thereby regulating apoptosis. The concomitant increase in HSP70 noted during preeclampsia and*U. urealyticum* infection might be responsible for the progression of pregnancy towards term through establishment of cell homeostasis and live fetal delivery.

Straszewski and his colleagues (2005) reported that metabolically active placental tissue is vital for the maintenance of pregnancy and alteration in their function is deleterious to the entire gestations leading to preeclampsia (32). It may encounter by the exaggeration of overstress and is more susceptible to ROS mediated apoptosis. We have previously reported that enhancement of HSP70 is favorable for combating pregnancy stress via its antiapoptotic function (31). Hence, the expression of HSP70, JNK, NF-κB and Bcl2 are coherently increased under preeclamptic condition to restore and stabilize the

cellular homeostasis thereby helping the developing fetus from placental hypoxia inbuilt oxidative insult. Thus, the live fetal delivery in spite of the existing complication implicates the defensive role of HSP70 and NF-κB in regulating apoptotic function of JNK and Bcl2, is well documented previously in preeclamptic conditions and normal tissue treated with *U. urealyticum* infection (31).

3.5 PREGNANCY AND ANTIOXIDANT THERAPY

Pregnancy is characterized by the increased generation of pro-oxidants in the placenta. Poor antioxidant reserves can also lift the balance in favor of prooxidant. In normal pregnancy, increasing gestational age promotes low levels of placental lipid peroxidation and increased antioxidant status. In general anti-oxidant defense appears to be depleted during pregnancy related disorders resulting in excessive oxidative stress. Increased vasoconstriction and hypoxia may cause significant high lipid peroxidation products (33) and reduced levels of antioxidant. Hence, management of pregnancy associated complications with natural remedy having anti-stress and vasodilating property is essential to improve placental blood flow and preclude their adverse effects. Various medications including antihypertensive, anticonvulsants and corticosteroids are available for managing pregnancy related complications. However, they are found to cause considerable side effects to both mother and fetus. Hence natural remedy is recommended to manage pregnancy related complications. Indeed,mint and tea (black and green tea)have a wide variety of polyphenols and flavonoids, which are all beneficiary effects under complicated pregnancies.

3.5.1 Natural antioxidant therapy of Mint

Mint is commonly known as pudina in Indian languages, belonging to the genus Mentha in the family Labiatae (Lamiaceace). There are 25-30 species within the genus Mentha, including spearmint, peppermint, wild mint, curled mint etc, of which spearmint is most common of all (34). Mint (Mentha spicata) is used and valued as an aromatic herb for thousands of years (35) and it is considered as stimulant, carminative, antispasmodic, stomachic and diuretic (36). M. spicata commonly known as spearmint is a common constituent of the Indian diet. It is used with spices to give the food a special flavor and fragrance. Mint extract has been found to have antioxidant and anti-peroxidant properties due to the presence of eugenol, caffeic acid, rosmarinic acid, alpha tocopherol and it would enhance error free repair for DNA damage and hence could be anti-mutagenic (37). In addition, Spearmint has been used for its antifungal, antiviral, antimicrobial, insecticide, antioxidant, anti-amoebic, anti-hemolytic, allergenic, CNS depressant, antihelmintic, and anti-encyclostomiasisactivitiy(38). Mint leaves contain rich source of micro-minerals Fe, Mg, Mn, Zn, Co, Se and exhibit beneficial properties by the presence of antioxidants (39).

3.5.2Beneficiary effect of tea

Tea (*Camellia sinensis*) is an evergreen shrub or tree and is a widely consumed beverage in the world. It is extensively used as herbal medicines all over the world for more than 10 decades. It is the second most consumed pleasant beverage in the world and it has been postulated as complementary and alternative medicines because of its anti-stress activities (49). Tea is brimming with antioxidant that scavenges cell-damaging free radicals and also has remarkable healing properties and it is a sort of "wonder drink" that may be even healthier than drinking water. Tea is generally divided into 4 major categories based on how it is processed such as white tea, green tea, oolong tea, and black tea (47).

3.5.2.1 Efficacy of green tea

Green tea (GT), a complex mixture of precious compounds, which includes polyphenols, flavonoids, flavonols and other constituents such as polysaccharides, vitamins, lipids, organic acids, and amino acids. GT also contains gallic acid, quercetin, kaempferol, myricetin and chlorogenic acid and caffeine. The major components of GT polyphenols are catechins (48). Catechins are flavan derivatives, a group of flavonoids differ in the level of oxidation, presence of heterocyclic ring and by good soluble capacity in water. The catechins of green tea are catechin (CA), epicatechin (EC), epigallocatechin (EGC), epigallocatechin-3-gallate (EGCG).GT is commercially available in bottles and sweetened with sugar or as an artificial sweetener, in single tea bags, loose-leaves, and instant-powder. GT supplements are sold in the form of capsules or liquid extracts.

3.5.2.2 Prophylactic effect of black tea

Black tea (BT) is the most broadly consumed beverage in China, India, Sri Lanka, Kenya, and Indonesia. In

black tea, during the fermentation process, most of the catechins the main flavanols of GT are oxidized and polymerized to theaflavins (TF) and thearubigins (TR) (50) which are responsible for the color, flavor, and brightness of tea. The content of individual TF showed a significant correlation and with tea taster scores value (51). Flavonols, present as glycosides, are the other important groups of polyphenols in GT. Whereas, BT contains quercetin, kaempferol, myricetin and glycosides, which all are exhibit antioxidant, anti-inflammatory and vasodilating property(45). Caffeine, theophylline, and theobromine are the main tea alkaloids, important for determining the quality of black tea. Caffeine is the most abundant alkaloid, responsible for the briskness provided by tea (52).

3.6 PREGANCY ASSOCIATED COMPLICATION IS REGULATED BY NATURAL REMEDY

3.6.1 GH and GDM with mint-tea extracts

Mint-tea extracts exhibit the phytochemical constituents like flavonoids and polyphenols. The mint-tea phenolic compounds exhibit the enhancement of succinate dehydrogenase, an enzyme used to identify the mitochondrial changes during the GDM and GH, which is dysregulated under these adverse conditions. The reduction of membrane bound in the releaseenzymes such as LDH, SGOT, SGPT and ALP during GDM and GH of placental tissue and mitochondria suggest that bioactive components present in mint-tea polyphenols is favorable for regulating the membrane integrating enzymes during pregnancy associated complications. In addition, the function of placental cell is dependent on energy supplied by the mitochondria. Therefore, induction of placental oxidative insult may interfere with functional velocity of respiratory complexes and such complex I to IV is well documented (53). Hence, regulation of mitochondria is very crucial for providing the energy rich molecules. The efficacy of mint-tea extractsis able to quench the free radical induced oxidative insult and thereby promoting the enhancement of respiratory enzymes (complexes I to IV). Mint-tea extracts are used to scavenge the lipid peroxidation products like 4-hydroxy nonenol and to reduce the level of nitrative stress markers such as NO_2^- and ADMA (asymmetric dimethyl arginine) in GDM of placental tissue is well documented (53). Hence exaggeration of oxidative/nitrative stress is deleterious to the placentation and induces the abnormal vascular development. However, induction of ADMA is silently inhibiting the potent vasodilator of nitric oxide via their regulating enzymes like endothelial nitric oxide synthase.

However, the increased consumption of mint-tea extracts contributes the improvement in quality of healthy life by increasing the antioxidants and delaying the onset of various degenerative diseases caused by oxidative stress.Kim and David (2008) inferred that both*in vitro* and *in vivo* study suggest that tea and its polyphenols may influence the glucose metabolism and prevent the diabetes via enhancing insulin sensitivity (40). Some human clinical studies alsosuggested that tea polyphenols regulateglucose intolerance mediated changes in diabetic patients. Jayanthy and Subramanian, (2014)reported that mint, a rich source of RosmarinicAcid (RA), a phenolic compound, which helps in treating diabetes, particularly chronic diabetes (41). Hence, their results show that RA may control plasma glucose and ameliorate hyperglycemia. However, RA is an active bioremedy for regulating the diabetes-induced disorders and complications. In addition, Dorman and his colleagues (2003) inferred that RA in the *M. spicata* may be correlated with protection of DNA from inbuilt oxidative insult (46).

ROS activates intracellular signal transduction pathways implicated in the pathogenesis of pregnancy related complications like GDM and GH. Cellular homeostasis requires a balance between the intracellular concentration of ROS with the co-ordinated function of antioxidant and protective activities of HSPs. The induction of genes coding for these proteins is a crucial step in the cellular response to oxidative stress. Expression of HSP70 was significantly increased in GDM and GH of placental tissue and mitochondria are well documented. It is depicting that HSP70 is crucial for rendering cytoprotectionand it may prevent the cell apoptosis via downregulates the activity of apoptotic proteins. In addition of mint-tea extracts decreased the expression of HSP70 which may be due to the restoration of cellular homeostasis via maintaining the antioxidant defence system. This property is attributed by the flavonoids, polyphenols and minerals present in mint-tea extracts. The mechanism of antioxidant activity of flavonoids can be characterized by direct scavenging/ quenching of oxygen free radicals for the inhibition

of oxidative enzymes that generate from reactive oxygen species.

3.6.2 Preeclamptic endothelial cells with mint-tea extracts

The endothelial cell is an exquisite "sensor" which responds to diverse signals present in the blood circulation, which includes regulation of vascular tone, coagulation, fibronolysis, and inflammatory responses to internal and external stimuli (54). Endothelial function is markedly abnormal during preeclamptic conditions. We have previously reported that adverse changes in function and morphology of maternal vascular endothelium account for the altered multisystem damage that occurs in preeclampsia (55).

Hence, management of PE is very essential for protecting the cells from placental hypoxia induced oxidative insult. Efficacy of mint-tea combination effectively modulates the oxidant and antioxidant imbalance in preeclamptic endothelial cell is well established. Therefore, mint-tea polyphenolic compounds enhance the total antioxidant capacity and GRR in preeclamptic endothelial cell. Hence, comparatively lower values for GRR were noted in both mitochondria and an endothelial cell during preeclampsia is well documented. The reduction of tripeptide glutathione is an important non-enzymatic antioxidant protecting cells from toxins such as free radicals (56).The reduction of HSP70 on addition of mint-tea extracts may be due to the restoration of cellular homeostasis through the maintenance of GRR level and antioxidant status under conditions like preeclampsia is well documented (43). The increased expression of HSP70 under such stress conditions favors maintenance of pregnancy by their antiapoptotic function. The increase in HSP70 level leads to the enhancement of antioxidant defense of cells by decreasing ROS content and neutralizing the toxic effects of oxidized proteins. The lower abundance of apoptotic proteins like, ASK1 in the preeclamptic endothelial cells combined with the increased level of Trx and HSP70 (44), which may be responsible for the lower level of phosphorylated JNK2 when compared to JNK1 and this also suggests that forced overexpression of HSP70 is not tolerated without other adaptive changes. The phenolic compounds present in mint-tea is reduction in the apoptotic protein such ASK1, JNK2. This may be due to the polyphenolic compounds present in mint-tea extracts are favorable for restore the cellular homeostasis through their anti-apoptotic and anti-stressor activity under these adverse conditions.

3.6.3Preeclamptic placental explant by black tea infusion

Research on natural products has revealed many unique, active compounds. A class of these known as flavonoids and phytoestrogens have broad implications in preeclampsia. Moreover, being a commonly consumed drink, black tea is considered safe, non-toxic and has potential as a 'natural' therapeutic product to be used as an adjunct to the existing clinical procedures, making it a broad-acting and readily distributed cost-effective agent to promote health benefits especially during pregnancy. The cytoprotective effect of black tea modulates the FRIOS, ORAC, cytokines, steroidogenesis and stress associated signaling molecules in preeclamptic placental explants.

Black tea polyphenols such as theaflavin and thearubigin are involved in the chelation of transition metal ions like iron and copper to prevent their participation in Fenton and Haber-Weiss reactions which in turn preclude the generation of hydroxyl radicals and degradation of lipid hydroperoxides causing reactive aldehyde formation (57). The reduced level of iron ions is unfavorable for free radical generation as well as for decomposition of lipid hydroperoxides. Hence, black tea polyphenols reduce the levels of lipid peroxidation and its byproducts such as conjugated dienes, lipid hydroperoxide and 4-hydroxy nonenol under preeclamptic conditions is well documented. In addition, black tea exhibits gallic acid, serve as a potent hydrogen donor to the free radical resulting in the suppression of protein carbonyl (PC) formation, which is another stress inducer highly evoked during preeclampsia. Recently, we have reported that black tea protects proteins and lipids against FRIOS induced oxidative damage and ameliorate the cell homeostasis (55).

Inflammatory cytokine may participate in the fine tune control of energy and hormonal balance during preeclampsia by extending its action as a metabolic signal to the main organ at the feto-maternal interface. The understanding of these changes may provide valuable knowledge for the root cause of preterm delivery during preeclampsia. Tumor necrosis factor-α (TNF-α) is a pleiotropic cytokine that in addition to its role in infection and immunity, induces

apoptosis in many cell types (58) and are produced by trophoblasts, monocytes and lymphocytes (59). TNF-α regulates trophoblast proliferation-differentiation, cell adhesion tissue remodeling, apoptosis of villous trophoblast and trophoblast hormone production (60). The enhancement of TNF-α in preeclamptic placental explant reveals that exaggeration of cell apoptosis is indirectly regulated by the induction of iHSP70, thereby activating an anti-inflammatory role. Besides,theaflavin is a rich source of black tea polyphenols, which act with anti-inflammatory property. Hence, black tea phenolic compounds are favorable for reduction of inflammatory cytokines. Therefore, promoting the black tea polyphenols portrays the restoration of cellular homeostasis.

The functional status of cellular antioxidant systems and the redox-sensitive survivalsignaling pathways like HSPs can significantly influence the cell-fate decision.

HIF-1α is a master transcription factor that is crucial for the regulation of a variety of cellular functions. HIF-1α is quickly demeaned during normoxic conditions by ubiquitin-mediated proteasome pathway controlled by the tumor suppressor von Hippel Lindau (VHL) (61). The upregulation of HIF-1α is well documented in preeclamptic conditions and is a potent modulator of stress inducible proteins such as HSP70 and ASK1. This condition leads to an increase in the expression of cytoprotective stress proteins like HSP70, which decrease or neutralize the deleterious effects of acute or chronic stresses (62). We have reported that enhancement of HSP70 and insignificant increases of ASK1 in preeclampsia proved that HSP70 acts as a survival mechanism via the downregulation of apoptotic inducers. The antioxidant defense mechanism of Black Tea Polyphenol via quenching the placental oxidative/nitrative insult reduces the levels of HSP70 and ASK1 (63). Therefore, black tea bioactive compounds such as polyphenols and phytoestrogens may aid in the protecting mechanism of apoptotic insult and to combat the oxidative stress developed during preeclampsia. Thus, increased consumption of black tea contributes to the improvement in quality of healthy life by increasing the defense system and delaying the onset of various degenerative diseases during preeclampsia caused by oxidative stress.

3.6.4 Efficacy of black tea and green tea on preeclamptic placental trophoblast

Trophoblasts are specialized, differentiated cells of the placenta with the capacity to migrate, invade the uterine parenchyma and associate with the vasculature. They are found to have a firm relationship with the spiral arteries and their invasion is crucial for the successful placentation (42). Abnormal placentation due to the improper uteroplacental perfusion represents the origin of preeclampsia, leading to the improper fetus organogenesis. Many studies reported that elevated concentrations of ADMA contribute to vascular dysfunction and play a role in the pathophysiology of preeclampsia due to vascular oxidative stress.It is suggesting that ADMA downregulates eNOS activity causes vascular damage and restricts the trophoblast invasion. That the intervention of black tea and green tea is favorable for reduction in the level of ADMA and increase in the level of eNOS in preeclamptic placental trophoblast is well documented (64). Similarly, Tang et al. (2006) described that tea polyphenols protect endothelial cells by increasing DDAH activity, which in turn results in a reduces in ADMA level, which is well observed previously (65). Hence our study suggests that the tea polyphenols like catechins and theaflavin may contribute in reviving of vasculogenic property in trophoblast isolated from preeclamptic placenta(64). ADMA is an anti-angiogenic factor that reduces VEGF expression in endothelial cells and prevents the formation of NO from NOS. ADMA affects the growth factors at their production and activity level (66).

Vascular endothelial growth factor-C plays an essential role in the proliferation, migration and metabolic activity of trophoblasts and also regulates angiogenesis. Borowicz et al. (2007) also reported that both VEGF and eNOS play a crucial role in the development of the placenta and any alteration in their expression may result in abnormal pregnancies (67). The reduction in the level of VEGF-C contributes to the decreased number of small blood capillaries, which in turn minimizes NO bioavailability conduit in hypertension. Placental growth factor (PLGF) is another angiogenic regulating factor which seems to play a synergistic role with VEGF for the formation of the vascular network with the development of the villous tree (68). It exhibits pro-angiogenic effects on the feto-placental circulation and supports trophoblast growth. Hypoxia suppresses transcriptional activity of PLGF in trophoblast and is increased by a normoxic environment pointing to a specific regulatory mechanism and function in these cells (69). Recently,

Nikolaos and his colleagues (2013) reported that low level of PLGF is markedly linked with impaired angiogenesis and placental development, leading to preeclampsia (70).Reduced levels of VEGF-C and PLGF in preeclamptic placental trophoblast depicts the poor placentation, vascularizationand the improper organization of the developing fetus, which results in preterm birth/low birth weight babies. Both black tea and green tea equally alters the expression of VEGF and PLGF in preeclamptic placental trophoblast is already documented. Hence absorption of tea polyphenols is efficiently rescued from placental hypoxia induced oxidative insult. This may be due to the presence of phenolic contents which are able to quench the reactive species. This prophylactic effect of tea extracts is potential in reducing the placental stress-induced cellular damage with profound cellular homeostasis allowing the induction of VEGF-C and PLGF in placental trophoblastand this is well established (64).

Transforming growth factor-β superfamily is evolutionarily conserved and plays fundamental roles in cell growth and differentiation. Many studies reported that failure in the downregulation of TGF-β3 after the 9thweek of gestation depicts shallow trophoblastic invasion, which may predispose the pregnancy to preeclampsia (71). The enhancement of TGF-β3 was observed in preeclamptic placental trophoblast suggesting that this overexpression may be responsible for poor placental perfusion and the associated placental hypoxic insult which together evoke the improper trophoblast invasion leading to fetal growth restriction. Therefore, black tea and green tea exhibit the reduction in the level of TGF-β3 in preeclamptic placental trophoblast suggesting that bioactive compounds attributes to the radical quenching property and inbuilt antioxidants, which ultimately exhibit a beneficial function in preeclamptic trophoblast via suppressing TGF-β3 expression. Anami et al. (1998) indicated that expression of TGF-β gene plays a vital role in the regulation of polyamine synthesis via regulating the expression of ornithine decarboxylase (ODC), an enzyme that plays an important role in cell growth and differentiation (72). Black tea imparts significant effect in altering the expression of TGF-β3 when compared to green tea highlighting that this may be attributed to the presence of catechins in green tea, as catechins are known for their role in inhibiting ODC and the mediated polyamines synthesis. This was also supported from the preeclamptic placental trophoblast incubated with catechins and the mediated expression of TGF-β3. When green tea is compared with black tea, our data show that black tea is daily recommended as the dietary supplement for the management of cellular stress because it is equally potent when compared with green tea.

CONCLUSION

Oxidative stress influences the reproductive phenomenon of a woman. Pregnancy is an important phase in every woman's life. In general antioxidant defences appear to be poor during pregnancy. A condition like preeclampsia and *U. urealyticum* infections increases the oxidative stress and severely alters the antioxidant systems. Increase in the expression of HSPs and its related molecules act as the only means for suppressing the oxidative stress mediated apoptosis. Many antioxidant therapies and alternative medicines are administered to increase the antioxidant deficiencies. As administration of drugs may be delirious to mother and fetus, natural antioxidants rich herbs like Mint and Tea (black and green) can be used to treat pregnancy stress associated complications or taken as general antioxidant therapy. Mint-tea, black and green tea act as potent antioxidant by their inbuilt antioxidant capacity which is attributed by the high phenolic, flavonoids, polyphenols and minerals present in mint and tea extracts which possess radical scavenging activity, reducing power. Hence, our study concludes that pregnancy associated disorders can be regulated by imparting mint and tea extracts as supplements.

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