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Genistein as a Potential Anticancer Agent against Ovarian Cancer

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Abstract

Genistein is known as the major component of isoflavone, which is present in high-soy diets. Genistein has received much attention because of its chemopreventive and therapeutic effects on various types of cancers. Numerous studies have shown that genistein has antineoplastic effects against ovarian cancer. Several epidemiological studies have shown that women who have high consumption of isoflavones have a relatively low incidence of ovarian cancer. Genistein inhibits ovarian carcinogenesis by pleiotropic mechanisms. A higher affinity to estrogen receptor β is one probable explanation for its ability to reduce the risk of ovarian cancer. Genistein also targets multiple cellular signal transduction pathways associated with cell cycle regulation and apoptosis. In addition, genistein has been suggested to have antiangiogenic and antioxidant activities. Herein, we summarize recent results from epidemiological and experimental studies to identify the role of genistein in ovarian cancer. Further studies are needed to achieve conclusive results and determine the clinical applications of genistein.

Key words: Genistein, Isoflavone, Ovarian cancer

Introduction

Previous epidemiological studies have reported that soy products play an important role in reducing the incidence and mortality rates of breast and prostate cancers (Kasiske et al., 1991; Lee et al., 1991; Lee et al., 2003; Magee and Rowland, 2004; Peeters et al., 2003). Soybeans and most soy products contain large amounts of isoflavones called soy phytoestrogens, which mainly consist of genistein (60%) and daidzein (30%) with smaller quantities of glycitein (10%). Phytoestrogens function as natural selective estrogen receptor (ER) modulators, depending on the tissue and the presence of coregulator proteins (Pike et al., 1999). Because of their potential estrogen-antagonistic effects, they have been reported to reduce the risk of hormone-dependent tumors (Myung et al., 2009).

Among the components of isoflavones, genistein has been extensively investigated to determine its chemopreventive and therapeutic activities. Several *in vitro* and *in vivo* studies have shown that genistein inhibits the carcinogenesis of non-hormone-dependent tumors such as colon, gastric, lung, and pancreatic tumors (Andres et al., 2011). Genistein exerts pleiotropic effects through the modulation of genes related to the cell cycle and apoptosis (Banerjee et al., 2008). In addition, genistein has been suggested to inhibit angiogenesis and antioxidant events through its molecular targets.

Among gynecologic malignancies, ovarian and endometrial cancers are related to hormonal and

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reproductive events. Like breast and prostate cancers, ovarian cancer occurs less frequently in Asian countries, where a high-soy diet is consumed, than in Western countries (Adlercreutz et al., 1993; Parkin et al., 1999). Furthermore, numerous studies have shown that genistein has protective effects against ovarian carcinogenesis (Andres et al., 2011; Kim et al., 2011). Herein, we summarize the available evidence on the chemopreventive and therapeutic potentials of genistein in ovarian cancer as follows: first, we discuss the anticancer mechanisms of genistein in ovarian cancer; second, we review epidemiological studies that aimed to determine the relation between soy intake and ovarian cancer risk; third, we review *in vitro* and *in vivo* studies that demonstrate the anticancer effects of genistein.

Anticancer Mechanisms of Genistein in Ovarian Cancer

Ovarian Carcinogenesis

Although the etiology of ovarian cancer is not completely understood, 2 dominant hypotheses regarding the underlying mechanisms of ovarian carcinogenesis have long been suggested. One is the ovulation hypothesis, which states that ovulation causes trauma to the ovarian epithelium, leading to rapid cell proliferation to repair the wound (Fathalla, 1971). Epidemiological studies have shown that oral contraceptive use or pregnancies can reduce ovarian cancer risk, thereby supporting this hypothesis. Another hypothesis concerns gonadotropin stimulation of the ovarian epithelium (Stadel, 1975), which suggests that excess stimulation by hormonal factors could result in abnormal proliferation or malignant transformation of cells. However, neither incessant ovulation nor gonadotropin stimulation of ovarian estrogen gives a completely satisfactory explanation for the pathophysiology of ovarian carcinogenesis.

Recent epidemiological evidence has given rise to a growing interest in the role of inflammation in ovarian cancer (Ness and Cottreau, 1999). Inflammation with rapid DNA damage and repair, oxidative stress, and increased levels of biological substances has shown to induce carcinogenesis (Ames et al., 1995; Dreher and Junod, 1996). Moreover, the ovulation process itself has been suggested to be associated with inflammation at the level of the epithelium as well as the follicle (Kim et al., 2011). Additional risk factors for ovarian cancer, including asbestos and talc exposure, endometriosis, and pelvic inflammatory disease, can cause local inflammation but do not directly affect ovulation and hormone levels. Epidemiological studies have shown that these factors, which can cause local inflammation, can increase ovarian cancer risk (Ness and Cottreau, 1999).

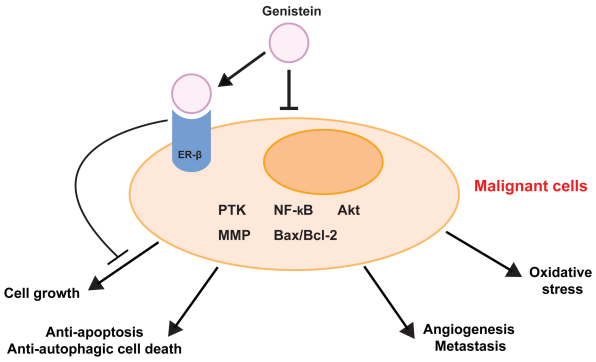

Figure 1. Pleiotropic effects of genistein on the inhibition of ovarian carcinogenesis. The high affinity of genistein to estrogen receptor β has an anticancer effect on ovarian cancer cells. In addition, genistein modulates multiple cellular signal transduction pathways associated with cell proliferation, apoptosis, angiogenesis, metastasis, oxidative stress, and autophagic cell death.

Pleiotropic Actions of Genistein in Ovarian Cancer

Genistein has been shown to inhibit ovarian carcinogenesis and cancer cell growth through its pleiotropic mechanisms against ER, cell proliferation, apoptosis, angiogenesis, metastasis, and oxidation (Figure 1). The hormonal actions of genistein are suggested to exert anticancer effects (Andres et al., 2011; Myung et al., 2009). Genistein has a high affinity for binding to ER, particularly ER-β, which is involved in the suppression of ER-α-stimulated estrogenic signal mechanisms. In addition to having hormonal activity, genistein exerts antineoplastic effects by modulating multiple signaling pathways such as protein-tyrosine kinase (PTK), *Akt*, NF-κB, matrix metalloproteinases (MMPs), and Bax/Bcl-2 (Banerjee et al., 2008; Sarkar and Li, 2002). Genistein has an inhibitory effect toward PTK, which drives signal transduction pathways leading to tumor growth and progression to malignancy (Akiyama et al., 1987). Genistein has also been demonstrated to inhibit the NF-κB and *Akt* signaling pathways, which play important roles in maintaining the homeostatic balance between cell survival and apoptosis (Li and Sarkar, 2002). It has also been found that genistein exerts antioxidant effects on reactive oxygen species by scavenging free radicals (Ruiz-Larrea et al.,

1997). Moreover, genistein can inhibit angiogenesis and metastasis (Li et al., 1999).

Anticancer Effects of Genistein on Ovarian Cancer

Epidemiological Evidence

Despite the extensive literature evaluating the effect of phytoestrogens on breast cancer risk, little attention has been given to the role of phytoestrogens in ovarian cancer. Because large variations exist in isoflavone consumption between Asian population and Western population (who have a low overall consumption), the Asian population is considered an ideal setting for evaluating the role of isoflavones for preventing the risk of cancer (Taylor et al., 2009). In a case-control study in China, consumption of a high-soy diet was shown to have a significant inverse association with ovarian cancer risk. A decreased risk for ovarian cancer was found in women with the highest quartile intake of genistein than in women with the lowest quartile intake (odds ratio [OR], 0.50; 95% confidence interval [CI], 0.30–0.84) (Zhang et al., 2004). In Japan, a prospective cohort study was performed to determine the role of soybean curd containing high levels of phytoestrogen for preventing cancer risk. In this study, dietary habit had no effect on ovarian cancer risk after adjusting for other clinicopathological variables (Sakauchi et al., 2007).

A recent meta-analysis showed that the highest soy intake was associated with a lower ovarian cancer risk than the lowest soy intake (OR, 0.61; 95% CI, 0.42–0.66) (Myung et al., 2009). Due to the growing interest in the role of high-soy diets, several studies in Western countries have been performed to determine the effects of phytoestrogens on ovarian cancer risk. Considering the association between a high-soy diet and breast cancer risk, it is interesting that a slightly stronger effect was observed among Western women than among Asian women (Trock et al., 2006). This may suggest that intervention with a phytoestrogen over a short period can be effective, and prior consumption over a number of years is not required (Taylor et al., 2009). In Italy, the highest quintile intake of isoflavones significantly reduced the development of ovarian cancer when compared with the lowest quintile intake (OR, 0.51; 95% CI, 0.37–0.69) (Rossi et al., 2008). A statistically significant inverse association was found between genistein and ovarian cancer risk in the California Teachers Study cohort (P for trend = 0.07). A 46% reduction in ovarian cancer risk was observed in women who consumed >3 mg/day of isoflavones as compared to women who consumed <1 mg/day of isoflavones (OR, 0.56; 95% CI, 0.33–0.96) (Chang et al., 2007). Moreover, an inverse association was suggested between total phytoestrogen consumption and ovarian cancer risk. The highest tertile intake of total phytoestrogen was significantly associated with a lower risk of ovarian cancer than the lowest tertile intake, after adjusting for other variables (OR, 0.62; 95% CI, 0.38–1.00) (Bandera et al., 2011). In contrast, 3 studies in Western countries showed no significant association between phytoestrogen intake and ovarian cancer risk in Swedish women. These conflicting results might be due to the differences in dietary habits. For example, in the Italian study, the largest dietary contributors to isoflavone intake were soy and soymilk; however, in the Swedish study, the largest dietary contributors to isoflavone intake were beans or vegetables (Hedelin et al., 2011). Out of the 6 studies reviewed by us, we found that 4 studies showed a trend for inverse association (Table 1). There were considerable variations in the amount of soy consumed and confounding variables between each study. Additional cohort studies and randomized controlled trials are needed to confirm the role of genistein in ovarian cancer development.

In vitro Studies

Results from previous epidemiological studies have raised the interest regarding the role of genistein as a chemopreventive agent, and therefore, researchers have attempted to determine the molecular mechanisms of this compound. Numerous studies have shown that genistein has inhibitory effects on ovarian cancer cells and tumor growth *in vitro*. Table 2 shows the anticancer effects of genistein on ovarian cancer cells from recently published *in vitro* studies.

Hormonal Effects

ER has 2 different forms—α and β—that vary with respect to tissue-specific expression and transcriptional activities (Mosselman et al., 1996). The structure of genistein is closely similar that of 17β-estradiol, because of which genistein binds and activates both ER-α and ER-β. ER-α has been mainly found in the endometrium, breast, ovarian stroma, and hypothalamic tissue, whereas ER-β has been mainly documented in the kidney, brain, bone, heart, lungs, intestinal mucosa, prostate, and endothelial cells (Couse et al., 1997). ER-α activation is related to cell proliferation and carcinogenesis in estrogen-dependent tumors (Oesterreich et al., 2001).

Table 1. Effects of genistein on ovarian cancer development from different epidemiological studies

Reference	Study design	Study period	Participants	Race/Country	Measure of soy intake	Adjusted OR (95% CI)
Zhang et al.,2004	CCS	1999–2000	254 EOC cases and 652 age-matched controls	Chinese/China	Soy foods (g/day): ≥136.4 vs. ≤47.0 Isoflavones (mg/day): ≥32.8 vs. ≤11.6 Genistein (mg/day): ≥20.9 vs. ≤6.6	0.50 (0.31–0.82) 0.51 (0.31–0.85) 0.50 (0.30–0.84)
Sakauchi et al., 2007	PCS	1988–2003(15)	77 EOC cases among 63,541 women	Japanese/Japan	Soybean curd (times/week) Almost every day vs. ≤1–2	0.61 (0.26–1.45)
Rossi et al., 2008	CCS	1992–1997	1031 EOC cases and 2411 controls	Italian/Italy	Isoflavone (μg/day) ≥32.5 vs. ≤12.8	0.51 (0.37–0.69)
Chang et al., 2007	PCS	1995–2003(8)	280 EOC cases among 97,275 women	Mainly white (84%), Hispanic (4%),Asian (3%), black (2%), Native American (2%)/U.S.	Isoflavone (mg/day) ≥3 vs. ≤1 Genistein (mg/day) ≥1.1 vs. ≤0.3	0.56 (0.33–0.96) 0.65 (0.42–1.02)
Bandera et al., 2011	CCS	2004–2008	205 EOC cases and 390 controls	White (87%), black(4%), Hispanic (4%)/U.S.	Total phytoestrogen (mg/1000 kcal) ≥1395 vs. ≤534	0.62 (0.38–1.00)
Hedelin et al., 2011	PCS	1991–2007(16)	163 EOC cases (117 invasive cancer, 6 BT) among 47,140 women	Swedish/Sweden	Isoflavone (μg/day) ≥16 vs. ≤0.74	1.26 (0.79–2.01)

CCS: case-control study, PCS: prospective cohort study, EOC: epithelial ovarian cancer, BT: borderline tumor

Table 2. Effects of genistein on ovarian cancer cell lines from *in vitro* studies

Reference	Materials	Cell type	Description	Measure of interest	Results
Choi et al., 2007	Genistein	SK-OV-3	Human ovarian cancer cell	Cell proliferation (MTT assay) Cell cycle distribution (FACS) Cytotoxicity (LDH) Apoptosis (caspase-3 activity)	Inhibition of cell proliferation in a dose- and time-dependent manner Arrest at G2/M phase Increased LDH release Increased caspase-3 activity
Ahmed et al., 2011	ITB-301, genistein derivative	SK-OV-3, ES2, HeyA8, HeyA8-MDR	Human ovarian cancer cell	Cell proliferation (crystal violet staining) Cell cycle distribution (FACS) Apoptosis (caspase-3/7 activity) Microtubule depolymerization (tubulin polymerization assay kit)	Induced microtubule depolymerization in a dose- and time-dependent manner No change in efflux-mediated drug resistance Cytotoxic effect of ITB-301 on a multidrug-resistant cell line
Solomon et al., 2008	Genistein	A2780, C200	Human ovarian cancer cell	Cell survival and apoptosis (MTT assay, histone-DNA ELISA) Indirect measure of apoptosis: Bcl-2, Bcl-xL, c-IAP1, survivin (Western blot analysis) NF-κB (EMSA)	Downregulation of antiapoptotic genes Decreased NF-κB Genistein pretreatment with chemotherapy was effective in both PS and PR ovarian cancer cell lines
Gercel-Taylor et al., 2004	Genistein	5 ovarian cancer cell lines from stage IIIc	Human ovarian cancer cell	Cell growth (sulforhodamine B and colony formation assays) Apoptosis (caspase-3 activity)	Induced caspase-3 activity Additive effect of cell proliferation inhibition with cisplatin, topotecan, and paclitaxel
Luo et al., 2008	Genistein	OVCAR-3	Human ovarian cancer cell	Cytotoxicity (CytoTox 96) Proliferation (CellTiter 96) VEGF (RT-PCR, ELISA)	Induced cell growth Inhibited VEGF expression
Rucinska et al., 2007	G8CG	CHO	Chinese hamster ovary cell	Cytotoxicity (MTT assay) DNA damage (Comet assay) Apoptosis (Hoechst 33258/propidium iodide staining technique) ROS (fluorescence probe)	Cytotoxic at high concentrations Antioxidant properties at lower concentrations
Chen et al., 2001	Genistein	Caov-3, NIH: OVCAR-3	Human ovarian cancer cell	Cell proliferation (MTT assay) ER, GAPDH (RT-PCR) IL-6 (ELISA) TGF-b (immunoassay)	Inhibited cell proliferation Inhibited IL-6 synthesis Increased TGF-b production
Gossner et al., 2007	Genistein	A2780, CaOV3, ES2	Human ovarian cancer cell	Analysis of apoptosis (FACS) Autophagy (microtubule-associated LC3) *Akt* (western blot analysis), glucose uptake	Induced apoptosis Inhibited glucose uptake, reduced phosphorylated *Akt* Induced autophagic cell death

MTT: methyl thiazolyl tetrazolium, FACS: fluorescence-activated cell sorting, LDH: lactate dehydrogenase, ELISA: enzyme-linked immunosorbent assay, EMSA: electrophoretic mobility shift assay, PS: platinum sensitive, PR: platinum resistant, VEGF: vascular endothelial growth factor, RT-PCR: reverse transcription polymerase chain reaction, G8CG: genistein 8-C-glucoside, ROS: reactive oxygen species, ER: estrogen receptor, GADPH: glyceraldehyde-3-phosphate dehydrogenase, LC3: light chain 3

ER-β has been suggested to play a role in suppressing ER-α-mediated cell proliferation (Strom et al., 2004). Furthermore, overexpression of ER-β in cancer cells has been shown to have antitumoral effects (Treeck et al., 2007). Normal ovary contains equal amounts of ER-α and ER-β mRNAs (Brandenberger et al., 1998). However, 90% of ovarian cancers arise from the epithelial layer, which predominantly expresses ER-α, whereas ER-β is expressed predominantly in the granulosa cells (Chu et al., 2000). Previous studies have suggested that ER-α might be associated with the development of reproductive neoplasms. Thus, activation of ER-β could be a promising target to reduce ER-α-regulated gene transcription.

The binding affinity of genistein for ER-α is reported to be 4% and that for ER-β is 87%, in comparison with estradiol (Kuiper et al., 1998). Therefore, it is suggested that genistein blocks the binding of more potent estrogens to receptors, thereby playing a potential role in preventing endocrine-related cancers. In addition to acting as a competitive ligand to estradiol, genistein has a higher affinity for ER-β than ER-α (Harris et al., 2005). This higher affinity to ER-β is considered a possible mechanism for decreasing ovarian cancer risk (Myung et al., 2009).

Effects Genistein on Cell Cycle Regulation

Genistein has been identified as a growth inhibitor of several cancer cells. The inhibition of cancer cell growth could be due to cell cycle arrest, which results in cessation of cell proliferation. By using ovarian cancer cells (SK-OV-3 cells), it was shown that genistein causes cell cycle arrest in the G2/M phase in a dose- and time-dependent manner. The proportion of cells in the G2/M phase after 48 hr exposure increased according to genistein concentration (Choi et al., 2007). In HO-8910 cells, genistein altered the levels of proteins associated with the checkpoint pathway, with a trend of inhibition of cancer cells proliferation (Ouyang et al., 2009). Moreover, the genistein derivative, ITB-301, also inhibits cell proliferation and induces cell cycle arrest in ovarian cancer cells by microtubule depolymerization. Subsequent mitotic arrest by the cytotoxic effect of ITB-301 was also observed in drug-resistant cancer cell lines, suggesting its therapeutic potential against multidrug-resistant cancers (Ahmed et al., 2011).

Effects on Apoptosis

Another antineoplastic action of genistein is programmed cell death (apoptosis). Several molecular mechanisms on how genistein induces apoptosis have been suggested. Some researchers have investigated the relation between genistein and the Bcl-2 family (Banerjee et al., 2008; Kyle et al., 1997; Spinozzi et al., 1994). The Bcl-2 family is the most well-known protein family that mediates apoptosis; Bcl-2 family proteins are classified into 2 main groups according to their functional characteristics: antiapoptotic proteins, such as Bcl-xL and Bcl-2, and pro-apoptotic proteins, such as Bax, Bak, and Bad. Genistein also induces growth inhibition and apoptosis by inhibiting the NF-κB and *Akt* signaling pathways (Brunet et al., 1999; Cardone et al., 1998; Van Antwerp et al., 1996; Wu et al., 1996). NF-κB as a transcription factor is an important regulator of genes involved in cell survival and proliferation. Overexpression of NF-κB is reported to protect cells from apoptosis, whereas inhibition of NF-κB suppresses cell growth and induces apoptosis (Sarkar and Li, 2002). The *Akt* signaling pathway is another transduction pathway that plays an important role in controlling cell survival and apoptosis. Recent studies have shown that *Akt* also regulates the NF-κB pathway through the phosphorylation and activation of molecules involved in the NF-κB pathway (Ozes et al., 1999; Romashkova and Makarov, 1999). Therefore, the *Akt* signaling pathway has been considered an ideal target for apoptosis induction.

Flow cytometry and measurement of caspase-3 activity are usually performed to detect apoptosis. In a previous study, flow cytometry showed that genistein induced caspase-3 activity and increased the apoptotic population in ovarian cancer cells (SK-OV-3 cells) (Choi et al., 2007). Analyses of apoptotic response after genistein administration to ovarian cancer cell lines showed that 20–25% of cells became apoptotic after 24–48 hr of treatment (Gossner et al., 2007). Genistein has an additive effect of inhibiting cell proliferation by inducing caspase-3 activity when used with a chemotherapeutic agent (Gercel-Taylor et al., 2004).

The sensitizing effect of genistein has raised the possibility of overcoming the chemoresistance of ovarian cancer. In both platinum-sensitive (A2780) and platinum-resistant (C200) ovarian cancer cell lines, pretreatment with genistein followed by administration of a chemotherapeutic agent resulted in the inhibition of cell growth by inactivation of NF-κB. Antiapoptotic genes such as Bcl-2, Bcl-xL, survivin, and c-IAP, which are also the downstream genes of NF-κB, were found to be downregulated (Solomon et al., 2008).

Effects on Angiogenesis and Metastasis

Genistein has been shown to inhibit the angiogenesis and metastasis of cancer cells (Fotsis et al., 1995). In breast cancer cells, administration of genistein inhibited MMPs, the proteolytic enzymes involved in cancer invasion (Li and Sarkar, 1999). Angiogenesis of solid tumors is involved in promoting the proliferation, invasion, and metastasis of cancer cells. Therefore, recent studies have focused on antiangiogenesis for cancer therapy and prevention (Carmeliet and Jain, 2000). Vascular endothelial growth factor (VEGF) is one of the most promising targets for ovarian cancer therapy. VEGF and VEGF receptors are expressed in ovarian cancer cells and are associated with angiogenesis and metastasis. Several VEGF-targeting agents have shown to be beneficial in patients with advanced-stage malignancies as well as in adjuvant settings. The effects of flavonoids, including genistein, on ovarian cancer cells (OVCAR-3) have been investigated (Luo et al., 2008). Five of the 12 studied flavonoids not only inhibited cell proliferation but also inhibited VEGF expression. Among the various isoflavones, genistein has the highest potency of inhibiting VEGF protein secretion; interestingly, it is even more potent than cisplatin.

Antioxidant Effects

Several flavonoids are known to function as antioxidants and scavengers of free oxygen radicals (Ruiz-Larrea et al., 1997). The protective effect of genistein against oxidative stress-induced injury depends on the modulation of transcription factors. The activation of the transcription factors Nrf1 and Nrf2, which regulate genes involved in oxidative stress, was evaluated (Hernandez-Montes et al., 2006). In addition, genistein has been reported to inhibit the generation of reactive oxygen species, which are known to increase under conditions of oxidative stress, by modulating NF-κB (Davis et al., 2001). In ovarian cancer, genistein can affect cancer cells differently according to its concentration. Although genistein-8-C-glucoside at high concentrations (10–130 μM) significantly reduced cell viability by inducing apoptosis and DNA damage, genistein at low concentrations (5–7.5 μM) showed antioxidant properties without cytotoxic or genotoxic effect (Rucinska et al., 2007).

Other Effects

The proliferation of ovarian cancer cells is inhibited by regulating cytokine synthesis. Genistein has been shown to inhibit the proliferation of ovarian cancer cells by modulating the cytokine IL-6, which affects immune homeostasis and inflammatory reactions (Chen and Anderson, 2001).

Autophagy, a lysosomal degradation pathway, is a cellular response to environmental stresses such as nutrient starvation. A study investigating the mechanism of genistein-induced cell death in ovarian cancer showed that genistein treatment results in caspase-independent and autophagic cell death (Gossner et al., 2007). Although normal cells utilize oxidative phosphorylation in the presence of oxygen to generate energy, cancer cells depend on glycolysis to supply their metabolic requirements. This accelerated rate of glycolysis in the presence of oxygen is known as the "Warburg effect" (Semenza et al., 2001; Weber, 1977). The oncogene *Akt* has been reported to induce accelerated glycolysis in cancer cells (Elstrom et al., 2004). Genistein may inhibit glucose uptake by inhibiting *Akt* activation, and thereby induce autophagic cell death.

In vivo Studies

Although epidemiological and *in vitro* studies have shown the protective effects of genistein against ovarian cancer, concerns regarding the safety of isoflavones have been raised because of undetermined results from *in vivo* studies. It has been shown that the hormonal activities of genistein may promote increased risks of endocrine-dependent tumors such as breast, endometrial, and ovarian cancers (Allred et al., 2001; Seo et al., 2006). Undesirable results in uterine cancer have also been reported. In ovariectomized rats, uterine weight increased after a high oral genistein dose (54 mg·kg b.w.$^{-1}$·day^{-1} for 3 months) (Rimoldi et al., 2007). Meanwhile, genistein (10 mg·kg b.w.$^{-1}$·day s.c.$^{-1}$ for 3 days) showed an antagonistic effect on estradiol-induced increase in uterine epithelial height (Diel et al., 2006). Moreover, genistein (1 mg·30 g b.w.$^{-1}$·every 4 weeks s.c.$^{-1}$ for 30 weeks) showed a protective effect against the development of endometrial cancer and atypical endometrial hyperplasia in mice by modulating the expression of estrogen-related genes and cytokines (Lian et al., 2001).

For the ovaries, similar results as those for the uterus were observed in female piglets that received isoflavones through a high-soy diet (2.8 mg·kg b.w.$^{-1}$·day^{-1} for 6 weeks). These piglets developed heavier ovaries with more follicles (De Wilde et al., 2007). Genetically susceptible female mice reared on an isoflavone-supplemented diet (125 μg/g each of genistein and daidzein for 8 weeks) developed a higher

granulosa cell tumor frequency (22%) than mice reared on an isoflavone-free diet (11%) (Dorward et al., 2007). However, dietary administration of genistein resulted in a significant reduction in dimethylbenz[a]anthracene (DMBA)-induced ovarian cancer in female Sprague-Dawley rats. DMBA-treated female rats received a supplementary diet containing 250 ppm of genistein for 50 weeks and showed 100% reduction in cancer development (Tanaka et al., 2002).

Conclusion

Collectively, previous epidemiological and experimental studies have suggested that genistein may play promising roles as a chemopreventive or therapeutic agent against ovarian cancer. In addition to its hormonal action, the anticancer effects of genistein are related to multiple cellular signaling pathways such as PTK, *Akt*, NF-κB, MMP, and Bax/Bcl-2. Because the successful treatment of ovarian cancer is limited mainly by the development of chemotherapy resistance, genistein is expected to play a role in sensitizing multiresistant ovarian cancer cells and have additive effects with conventional chemotherapeutic agents. Although some clinical trials are now being performed to identify the role of genistein as an anticancer agent against various types of cancers, there is currently no registered clinical trial on ovarian cancer. Further research should be performed to identify the role of genistein in ovarian carcinogenesis.

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