COMMENTARY

Can selenium be a modifier of cancer risk in CHEK2 mutation carriers?

Satish Gupta1,2, Katarzyna Jaworska-Bienieck1,2, Jan Lubifski1 and Anna Jakubowska1

1International Hereditary Cancer Centre, Department of Genetics and Pathology, Pomeranian Medical University, Polabska 4, 70-115, Szczecin, Poland and 2Postgraduate School of Molecular Medicine, Warsaw Medical University, Zwirki i Wigury 61, 02-091, Warsaw, Poland

*To whom correspondence should be addressed. Department of Genetics and Pathology, Pomeranian Medical University, Polabska 4, 70-115 Szczecin, Poland. Tel: +48 91 466 1821; Fax: +48 91 466 1533; Email: satish1482@gmail.com

Received on May 17, 2013; revised on August 5, 2013; accepted on August 20, 2013

Selenium is an essential trace element for humans, playing an important role in various major metabolic pathways. Selenium helps to protect the body from the poisonous effects of heavy metals and other harmful substances. Medical studies have provided evidence of selenium supplementation in preventing certain cancers. Low and too high selenium (Se) status correlates with increased risk of e.g. lung, larynx, colorectal and prostate cancers. A higher level of selenium and supplementation with selenium has been shown to be associated with substantially reduced cancer mortality. Selenium exerts its biological roles through selenoproteins, which are involved in oxidoreductions, redox signalling, antioxidant defence, thyroid hormone metabolism and immune responses. Checkpoint kinase 2 (CHEK2) is an important signal transducer of cellular responses to DNA damage and acts as a tumour suppressor gene. Mutations in the CHEK2 gene have been shown to be associated with increased risks of several cancers. Four common mutations in CHEK2 gene (1100delC, IVS2+1G>A, del5395 and I157T) have been identified in the Polish population. Studies have provided evidence that CHEK2-truncating and/or missense mutations are associated with increased risk of breast, prostate, thyroid, colon and kidney cancers. The variability in penetrance and cancer expression in CHEK2 mutation carriers can probably be explained by the influence of other genetic or environmental factors. One of the possible candidates is Se, which together with genetic variations in selenoprotein genes may influence susceptibility to cancer risk.

Selenium

Humans and animals require selenium for the function of a number of selenium-dependent enzymes, also known as selenoproteins. Selenium behaves as an antioxidant and anti-inflammatory agent. The antioxidant properties of selenoproteins help prevent cellular damage from free radicals by reducing the production of inflammatory prostaglandins and leukotrienes through the nuclear factor kappa-B (NF-kB) signalling pathway (1). Selenium plays an important role in the regulation of thyroid activity (1), immune system functioning and normal development of spermatozoa and is required for sperm motility (1). Selenium has been linked to heart disease, adverse mood states, Alzheimer’s disease, schizophrenia, juvenile cardiomyopathy, Keshan disease, Kashin–Beck disease (which results in osteoarthropathy) and myxedematous endemic cretinism (which results in mental retardation) and it has been extensively analysed in relation to cancer risk (1,2). Baseline selenium level in the body is mainly dependent on its content in diet, which is related to geographical location.

Selenium and cancer

It has been reported that the optimal selenium level in the body is beneficial in suppression of tumour formation. Both high and low levels can promote tumour formation (2). Selenium has been shown to protect against oxidative DNA damage and to enhance DNA repair, which clearly indicates their important role in carcinogenesis (3). There is evidence of the beneficial effect of selenium in terms of cancer-specific mortality and overall mortality (4). Several prospective and epidemiological studies have suggested the significant association of selenium intake with decrease in risk of breast (4), lung (5), bladder (6), colorectal (7), liver (8), oesophageal (9), gastric cardia (9), thyroid (10) and prostate (11) cancers. It is also important to note that selenium supplementation can be potentially beneficial only for those who have relatively low baseline Se level. For individuals with higher baseline Se concentration, additional intake has no effect or can even be harmful. This was particularly visible in two large case–control clinical trials, Nutritional Prevention of Cancer (NPC) (12) and Selenium and Vitamin E Cancer Prevention Trial (SELECT) (13). The NPC study was a randomized clinical trial designed to evaluate the efficacy of selenium supplementation in preventing the recurrence of non-melanoma skin cancer among 1312 volunteers with a previous history of non-melanoma skin cancer from Southeastern USA. They found that participants with baseline plasma selenium concentrations in the lowest tertile (<105.2 ng/ml) had almost two times lower total cancer incidence in comparison to those in the highest tertile (>121.6 ng/ml). There was also reduction in total cancer mortality (12). The SELECT was designed to investigate the effect of selenium and vitamin E supplementation on prostate cancer risk in 35 533 American men. In this study, reduction in cancer incidence or mortality was not observed, however the baseline Se level in participants was >122 µg/l (13).

Selenoproteins

Selenium is incorporated as selenocysteine (Se-Cys) at the active site of a number of proteins (selenoproteins), which are involved in different biological processes, ranging from DNA synthesis to protection against oxidative stress. In the human genome, 25 selenoproteins have been identified, including glutathione peroxidases (GPX1, GPX2, GPX3, GPX4 and GPX6), thioredoxin reductases (TXNRD1, TXNRD2 and...
TXNRD3), iodothyronine deiodinases (DIO1, DIO2 and DIO3), endoplasmic reticulum selenoproteins (SEP15, SELK, SELM, SEPN1, SELS and SELT) and other selenoproteins (SEPP1, SEPW1, SELR, SPS2, SELH, SELI, SELO and SELV). They are located in different parts of the cell and are involved in reduction of peroxides, contributing to extracellular antioxidant status (GPXs, SEPP1, SEPW1, SELR, SPS2, SELH, SELI, SELO and SELV). They regulate thyroid hormone and embryogenesis (DIOs), sperm maturation and male fertility (GPX4), inhibition of apoptosis and cell growth factors (TXNRDs), protein folding (SEP15, SELM and SELS), muscle development (SEPN1), transport of selenium to brain and testes (SEPP1) and inflammation and immune response (SELS) (14). The function of some selenoproteins (e.g. SELH, SELI, SELT and SELO) is still under investigation (14).

There are several studies, which reported the expression pattern and occurrence of polymorphisms in selenoprotein genes relevant to cancer biology. There is reduced expression of these enzymes in various types of cancer (15), especially when associated with low intake of selenium. Alternatively, the expression of selenoprotein genes are highly induced by supplemental selenium in cancer cells (16). The differential expression profile of SEP15 (15), GPXs (17) and TXNRDs (18) are associated with different cancers and can be a potential target in cancer therapeutics. Single nucleotide polymorphisms are a potential tool to improve cancer diagnosis and treatment planning. Genetic variants in selenoprotein genes are significantly associated with risk of cancer of the breast (19,20), prostate (21,22), colorectal (23,24) and lung (25,26).

Checkpoint kinases

Checkpoint kinases (CHEKs) are serine/threonine kinases that are involved in the regulation of the cell cycle. Two subtypes have been identified thus far, CHEK1 and CHEK2. They are essential components to delay cell cycle progression in normal and damaged cells and can act at all three cell cycle checkpoints, i.e. G1, S and G2. CHEKs have a role in the physiological stress of hypoxia/reoxygenation (27).

CHEK2 protein includes three characteristic domains: an N-terminal SQ/TQ cluster domain (SCD; amino acid residues 20–75), a forkhead-associated (FHA) domain (residues 112–175) and a serine/threonine kinase domain (residues 225–490) (28) (Figure 1b). The SCD is a regulatory domain containing serine or threonine residues followed by glutamine. The SCD consists of seven SQ/TQ (Ser-Gln/Thr-Gln) motifs that are characteristic of ataxia telangiectasia mutated (ATM) phosphorylation sites, with Thr68 being the primary site that gets phosphorylated in response to DNA damage (28). The FHA domain is a protein–protein interaction domain that binds to phosphothreonine-containing peptides, including the phosphorylated Thr68 segment of SCD (28). The kinase domain also has two important autophosphorylation sites (Thr383 and Thr387), which are important for CHEK2 activation (28) (Figure 1b).

CHEK2 gene contains 14 exons (Figure 1a). The CHEK2 gene sequence (Genbank: NG_008150) contains Se-Cys insertion sequence (SECS) element (Figure 1a) and also possesses a UGA codon, which codes for Se-Cys (29). However, there is no evidence of insertion of Se-Cys residue in CHEK2 protein.

CHEK2 and cancer

Mutations in CHEK2 gene were first discovered in patients with Li–Fraumeni syndrome, a highly penetrant familial cancer phenotype (30). Several mutations have been identified in CHEK2 gene, but few of them have become important and have been analysed in numerous research studies. Three protein-truncating mutations (1100delC, IVS2+1G>A and del5395) and one missense mutation (I157T) in CHEK2 gene have been the subjects of extensive research. Another important CHEK2 variant is S428F, residing in the kinase domain, which abrogates CHEK2 function and is associated with a 2-fold increase in breast cancer risk among Ashkenazi Jews (31). CHEK2 is a multiorgan cancer susceptibility gene, and mutations in CHEK2 have been reported to be associated with higher risk of several cancers (32). The protein-truncating mutations are significantly associated with increased risk of cancer of the breast (32–36), prostate (32), thyroid (32), gastric (37) and colorectal (38). The missense variant I157T is significantly associated with increased risk of cancer of the breast (32,39), prostate (32), colon (32), kidney (32), thyroid (32) and colorectal (40). However, the risk of cancer is much higher with protein-truncating mutations in comparison with missense mutation (32). The observed cancer risk is enhanced with positive family history of cancer (41). CHEK2 I157T mutation was also reported to be associated with reduction of risk of lung or upper aerodigestive cancers in Central Europe (42) and in Poland (43).

Modifiers of CHEK2

A few studies have indicated that cancer risk in CHEK2 carriers can be modified by several factors, e.g. family history (44), hormonal imbalance (45), genetic factors (46), environmental and nutritional uptake (47). Various dietary antioxidants have shown considerable promise as effective agents for cancer prevention by reducing oxidative stress, which has been implicated in the development of many diseases, including cancer. Research evidence suggests that many dietary factors may be used alone or in combination with traditional chemotherapeutic agents to prevent the occurrence of cancer and their metastatic spread or even to treat cancer. The reduced cancer risk and lack of cytotoxicity associated with high intake of fruits and vegetables suggest that specific concentrations of antioxidant agents from these dietary sources may produce cancer chemopreventive effects (47). One such active agent is gallic acid (3,4,5-trihydroxybenzoic acid). It is a naturally occurring phenolic acid found in gallnuts, sumac, witch hazel, tea leaves and oak bark and is also obtained by the hydrolysis of tannins. Gallic acid is known to show some pharmacological activities and cytotoxicity against cancer cells (48). Ou et al. (49) have shown that gallic acid can induce cell cycle arrest at G2/M phase via CHEK2-mediated phosphorylation of CDC25C in a bladder transitional carcinoma cell line. These authors also demonstrated that extracts of Paeonia lactiflora Pall [radix paeoniae alba (RPA)], a traditional Chinese herbal medicine (50), inhibit growth of bladder cancer via induction of apoptosis and cell cycle arrest. RPA-mediated growth inhibition of bladder cancer was correlated with activation of CHEK2 (51).

Evidence of selenium and CHEK2 interactions

There is a weak evidence of interaction between selenium and CHEK2 in the literature. However, the available data suggest the influence of selenium and selenium compounds with CHEK2 in cancer risk (52–56).
Selenium: a modifier of cancer risk in CHEK2 mutation carriers

Selenium compounds
Methylselenocysteine (MSC) is an organic selenium compound synthesized by plants such as garlic, onions and broccoli. Yin et al. (52) have demonstrated that MSC with combination of 7-ethyl-10-hydroxycamptothecin (SN-38) results in increased CHEK2 phosphorylation at Thr68 and down-regulation of DNA replication-associated proteins CDC6, MCM2 and CDC25A. This is accompanied by the induction of pre-apoptotic DNA fragmentation in human head and neck cancer cell lines (56).

Methylseleninic acid (MSA) inhibits the growth of prostate cancer cell lines. The expression change of CHEK2 gene was considered to be the target of MSA in impeding cell cycle progression. Results also provided valuable insights into novel biological effects of selenium, such as inhibition of cell invasion, DNA repair and stimulation of transforming growth factor beta signalling (53).

Recently, an interesting study performed on human breast carcinoma cell lines (MCF-7 and its derivative MCF-7GPx-1) indicates that selenium may increase CHEK2 phosphorylation, as well as cell viability following exposure to DNA damage. The enhancement of CHEK2 phosphorylation by selenium supplementation was considerably more dramatic in GPX1-expressing cells (MCF-7GPx-1) (54). Selenium’s ability to elevate GPX1 protein level indicates a possible mechanism towards prevention of cancer by protecting against toxic and carcinogenic effects of diverse DNA-damaging agents.

ATM-dependent pathway
CHEK2 is an important constituent of ATM-dependent pathway and activated by ATM through phosphorylation.

Wu et al. (55) has suggested a novel role of selenium in induction of ATM-dependent senescence response, which depends on reactive oxygen species. In a similar study, Qi et al. (56) demonstrated the new role of selenium in mitigating tumour genesis by targeting the mismatch repair pathway in colorectal cancer cells through ATM-dependent pathway.

Is CHEK2 a selenoprotein?
As was already mentioned, selenium is incorporated to the proteins as Se-Cys. The mechanism for adding a Se-Cys residue to a growing polypeptide chain is complex. It involves interactions between the SECIS located in the 3′-untranslated region and two trans-acting components: a SECIS-binding protein (SBP2) and a Se-Cys-specific elongation factor (eEFSec) (57). Interestingly, CHEK2 gene sequence (Genbank: NG_008150) contains SECIS element (Figure 1a) and also possess a UGA codon, which codes for Se-Cys in the selenoprotein family (29). However, there is no evidence of insertion of Se-Cys residue in CHEK2 protein. As selenium independently and under influence of GPX1 can increase phosphorylation of CHEK2 (54), there may be a possibility of interaction of GPX1 with CHEK2.

Moreover, CHEK2 plays a role in physiological stress of hypoxia/reoxygenation (27), a typical function of GPXs and TXNRDs. The similarity in function of CHEK2 and selenoproteins provides an indication towards important interaction machinery of CHEK2 with the selenoprotein family in cancer biology.

Conclusion
Suboptimal selenium concentrations, as well as variations and expression of selenoproteins, are associated with cancer risk. Studies have shown that selenium has a protective effect in the prevention of cancer. Selenium has been proved to be an inducing agent for ATM-dependent DNA damage response. There is no experimental evidence for an association between blood selenium level and cancer risk in CHEK2 carriers yet. However, CHEK2 is an important constituent of the ATM-dependent DNA damage pathway, and the relation of selenium...
in the context of CHEK2 would be an interesting topic in cancer research.

Acknowledgements

S.G. is a fellow of International PhD program, Postgraduate School of Molecular Medicine, Warsaw Medical University, supported by the Polish Foundation of Science.

Conflict of interest statement: None declared.

References


