

**SELENIUM COMPOUNDS IN THE ENVIRONMENT;
THEIR EFFECT ON HUMAN HEALTH**

BARBARA WACHOWICZ, HALINA M. ŻBIKOWSKA
and PAWEŁ NOWAK

Department of General Biochemistry, University of Łódź, 90-237 Łódź, Poland

Selenium (Se) was discovered by Berzelius in 1818 and for the next 140 years the toxicity of Se was all that concerned biologists. In the 1950s Se was considered to be a highly toxic element with possible carcinogenic properties. The necessity of Se was shown in 1957 [1]. Since then there has been increasing interest in this trace element; the literature covering its biological importance is very comprehensive. One of the reasons for the relatively late discovery of the necessity of Se (for animals and humans) was the lack of a sufficiently sensitive analytical method to detect low concentrations of Se. In 1973 Se was discovered in the enzyme - glutathione peroxidase (GSH-Px; EC 1.11.19) as the biologically active structural component [2]. It is well known that Se is not only bound to GSH-Px [3]. The list of selenoproteins, defined as proteins containing selenium in stoichiometric amounts, has grown. At least 12 animal selenoproteins have been identified and their biochemical functions characterized [3-6]: four isoforms of glutathione peroxidase (1. cellular:cGSH-Px or GSH-Px1; 2. extracellular or plasma pGSH-Px2; 3. phospholipid hydroperoxide:PHGSH-Px or GSH-Px3; 4. gastrointestinal:GSH-Px4), three isoforms of iodothyronine deiodinase (types I, II, and III), thioredoxin reductase, selenoprotein P, and selenoprotein W. It can be predicted that more proteins will be discovered. Progress in the discovery of bacterial selenoproteins has been even greater. Presently, Se is regarded as an essential trace element for humans and a variety of other animal species. However, it should be supplied to the organism in a relatively narrow range of concentrations. The requirement for Se in human nutrition is now well-established. The biological activity of selenium is dependent on its chemical form and not the element per se. Se is found in four oxidation states; selenate (SeO_4^{2-}) and selenite (SeO_3^{2-}) are highly soluble in water and are known to be toxic to biological systems at relatively low concentrations. By contrast, elemental Se (Se^0) is essentially non-toxic and highly insoluble in water. Selenide (Se^{2-}) is both highly toxic and reactive but is readily oxidized to Se^0 . Thus, Se behaves as a double-edged sword in biological systems, in which a fine line must be negotiated between concentrations that are physiologically essential and those that are toxic.

Metabolism of selenium

Se is supplemented in food and water, either in inorganic compounds (selenite, selenate) or organic compounds (selenomethionine, selenocysteine) - selenoaminoacids in which S is replaced by Se. The level of Se in plants, and in turn in animals depends on the amount of biologically available Se in the soil where Se content varies greatly [7]. The mammalian organism has rather limited means of Se storage, Se concentration depends upon a continuous supply of this element. The metabolism of Se depends on the chemical form of Se ingested. Some Se will be used to produce selenoproteins, proteins that require Se for catalytic activity and incorporate Se as selenocysteine (SeCys) into their polypeptide chain using UGA as the encoding codon [8]. Other forms of Se primarily go into Se-containing proteins (proteins that do not require Se for catalytic activity) which apparently incorporate Se randomly by substituting selenomethionine (SeMet) for methionine [3, 4]. Finally, all the forms of Se can be methylated to methylselenol, dimethylselenide and trimethylselenonium and excreted in the urine and through the lungs [9]. Selenite (selenate after reduction) enters cells, and reacts with thiols; glutathione GSH is considered to be the main component of the Se metabolism pathway taking part in the first of a series of reduction reactions which convert selenite to hydrogen selenide (H_2Se). GS-Se-SG (selenodiglutathione) is the major product formed. A consequence of the reaction of selenite with glutathione is the production of H_2O_2 and $O_2^{\bullet-}$ [10, 11]. $4RSH + H_2SeO_3 \rightarrow RS-Se-SR + RSSR + 3H_2O$. The biological activity of selenite is probably governed by its metabolism. Glutathionylselenol (GS-SeH) is an intermediate product in the reaction between GS-Se-SG and GSH. Similar reactions may also take place with other biologically important thiols, such as cysteine. The reduction of selenite to selenide and the subsequent methylation is a well-characterized Se metabolism pathway in animals. Selenide appears to be a common and perhaps important intermediate in the metabolism of both inorganic and amino acid forms of Se [10, 11]. Methylation is a common end-stage of selenium metabolism [9]. Methylated forms of Se have a distinctive garlic-like odor. With a high intake of selenite or SeMet, the levels of methylated metabolites, including methylselenol, dimethylselenide (exhaled) and trimethylselenonium (excreted in the urine), increased. The formation of H_2Se is not essential for the expression of the anticarcinogenic activity of Se. Precursor Se compounds which are able to produce a steady stream of monomethylated metabolite have chemopreventive activity [10-12]. The fully methylated form, trimethylselenonium is totally ineffective. The degree of methylation is an important factor for the anticarcinogenic activity of Se compounds [10-12]. Much plant material contains Se in the form of SeMet, which is incorporated into Se-containing proteins to a greater extent than Se salts are, although it also can be degraded to selenide and then be incorporated into selenoproteins. SeMet

is incorporated non-specifically into body proteins in place of methionine, because met-tRNA cannot distinguish between methionine and selenomethionine [3, 4]. Some plants contain Se in a form other than SeMet, and the metabolism of Se in those plants is unique. Selenium Methylselenocysteine (SeleniumMC) is a naturally occurring selenoaminoacid synthesized by plants such as garlic, broccoli and onions [12]. SeleniumMC metabolism has been studied in animals and the chemoprevention effect of SeleniumMC is believed to occur due to the generation of monomethylated selenium species by endogenous enzymes [13, 14]. Monomethylated Se species have been shown to generate superoxide and induce apoptosis in cancer cells in culture [15]. Unlike L-selenomethionine (the form found in yeast and meat) which is incorporated into proteins in place of methionine in vivo, SeleniumMC is not incorporated into any proteins therefore is fully bioavailable for chemoprevention Se. It is also less toxic than L-selenomethionine. It can be used as a human and animal food supplement.

SeleniumMC from extracts of Se-enriched garlic is the active ingredient in the chemoprevention of mammary cancer [14, 15]. Plants are known to convert inorganic Se in the soil to organic Se compounds via the sulphur assimilatory pathway. Selenized yeast contains a cocktail of Se in a variety of chemical forms, with SeMet as the major constituent. Se is present in various amounts in all tissues, predominantly in the kidneys, liver, testes and muscles. The rate of accumulation in tissues is the highest in the kidneys, liver and testes, and much lower in the muscles. In the cell Se is predominantly located in the nucleus, less commonly in the cytosol, bound to various proteins. The majority of Se in human plasma is incorporated in three proteins: GSH-Px, selenoprotein P and albumin.

Anticancer properties of selenium

Geographical studies suggest a relationship between Se status and cancer incidence. Observations have indicated that cancer death rises when the dietary intake of Se is low. Se may help prevent certain types of cancer: lung, colon, prostate and rectum, but not skin cancer. Evidence for the possible anticancer benefits of Se comes from large-scale Chinese studies showing that giving Se supplements to people who live in Se-deficient areas reduces the incidence of cancer [16-18]. Se compounds have been shown to have antitumorigenic activities in animal models when the drug is administered at levels greater than those associated with nutritional needs. Several hypotheses have been proposed to explain the inhibition of tumorigenesis by supplemental Se, including: protection against oxidative damage involving the Se as an essential component of antioxidant enzymes, alterations in carcinogen metabolism, effects on the endocrine and immune systems, production of cytotoxic selenium metabolites, generation of free radicals, inhibition of protein synthesis, inhibition of specific enzymes and stimulation of apoptosis [15, 18-20]. The progress in basic

research on Se and cancer prevention during the past decade is observed. Special emphasis has been placed on chemical forms of Se and their anticarcinogenic activity, selenium-enriched food, in vitro effects of selenite and monomethylated selenium and the role of aromatic selenium compounds (very effective chemopreventive agents in experimental mammary cancer models), particularly ebselen. More than 90% of Se cancer chemoprevention experiments have used either sodium selenite or selenomethionine (commercially available). Both of these compounds are known to suppress carcinogenesis in many animal models. SeMet was less active than selenite in cancer inhibition.

It is almost impossible to increase Se intake by eating certain types of food because most common foods have a very low selenium content. Ip & Lisk [14] tried to enrich garlic with Se by fertilizing the crop with water-soluble selenite salt. Because garlic contains an abundance of sulphur derivatives, it might be able to accumulate high levels of Se. Se-garlic with SeMC as the major Se-constituent was superior to SeMet in terms of its anticarcinogenic efficacy [13, 14]. Unlike SeMet, which produced large increases in tissue accumulation, Se-garlic caused only modest elevation. Se-garlic has many desirable characteristics. Because garlic is used primarily in flavoring food, there is less danger of overconsumption. At nutritional levels of Se intake, Se-garlic provides bioavailable Se for the maintenance of selenoenzymes. At higher levels, it has potent anticancer activity, and does not cause excessive Se accumulation because its predominant Se-methylselenocysteine is rapidly metabolized to di- and trimethylated excretory products. Because Se-methylselenocysteine cannot be incorporated non-specifically into proteins, the amount of total Se decays quickly in various tissues upon discontinuation of Se-garlic feeding [13-15].

Biological markers of selenium

The growing complexity of Se biochemistry makes it rather difficult to identify the most reliable indicators of Se status and define Se requirements. To assess Se status different approaches are used [21, 22].

Se concentration in biological fluids: Se concentrations vary substantially in healthy subjects, mainly according to geographic location diet and age. The mean serum Se concentrations for healthy adult subjects in different parts of the world vary from 40 to 200 µg/l. The determination of the whole blood or urine Se content poses analytical problems. Convenient indicators of long-term Se exposure can be Se in hair or toenail clippings in populations where blood or urine samples are difficult to collect. A significant positive correlation between GSH-Px activity and Se is observed only for relatively low Se concentrations. It disappears in the higher range of Se concentrations. The response of GSH-Px activity to Se supplementation is measured in blood platelets, which appear to be a very sensitive index of Se exposure, mainly as a consequence of the rapid

turnover of these cells in the circulation. Platelet GSH-Px activity increased in populations which consumed less than 50 µg Se/day and reached a stable plateau after several weeks of supplementation. No increase in platelet and erythrocyte GSH-Px activities was observed in a group consuming 100-110 µg Se/day. Several authorities established dietary standards for Se based on the saturation of GSH-Px activity. Recommended Dietary Allowances (RDA): in the USA - 55-70 µg Se/day, in the UK - 60-75 µg Se/day, in Australia 70-85 µg Se/day. The isoforms of GSH-Px have a different Se requirement [21]. The recent discovery of isoforms of GSH-Px, different from the classical enzyme, and new selenoenzymes offers new markers of Se status.

The effects of the selenium on blood platelets

Blood platelets are very rich in Se. The unusually high concentration of Se in these cells suggests a particular role for this element in platelet function and in platelet-related diseases.

Blood platelets are the first target of Se action in blood. Selenite exerts an inhibitory effect on blood platelet activation. In toxic doses it inhibits thrombin- and ADP-induced platelet aggregation, decreases the release of adenine nucleotides stored in platelet granules and reduces arachidonate metabolism in platelets stimulated by thrombin [23, 24].

Selenite reacts with GSH and protects against the inhibitory action of cisplatin on platelet activation and protects against complex GS-Pt formation.

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