The Neurobiology of Selenium: Lessons from Transgenic Mice1,2

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ABSTRACT The brain represents a privileged organ with respect to selenium (Se) supply and retention. It contains high amounts of this essential trace element, which is efficiently retained even in conditions of Se deficiency. Accordingly, no severe neurological phenotype has been reported for animals exposed to Se-depleted diets. They are, however, more susceptible to neuropathological challenges. Recently, gene disruption experiments supported a pivotal role for different selenoproteins in brain function. Using these and other transgenic models, longstanding questions concerning the preferential supply of Se to the brain and the hierarchy among the different selenoproteins are readdressed. Given that genes for at least 25 selenoproteins have been identified in the human genome, and most of these are expressed in the brain, their specific roles for normal brain function and neurological diseases remain to be elucidated. J. Nutr. 134: 707–710, 2004.

KEY WORDS: ● selenoprotein P ● glutathione peroxidase ● tRNASec ● MarB ● thioredoxin reductase

Observations on the Importance of Se for Brain Function. Selenium (Se) has been implicated in a number of health issues [for review see (1,2)]. Here, we focus on the effects of Se on the brain. The first clinical reports directly linking Se status and neurological conditions showed that a form of intractable seizures in infants was associated with low blood Se levels and could be treated by Se supplementation (3,4). Before, mainly circumstantial evidence was available pointing to the lack of Se as a risk factor for patients receiving total parenteral nutrition to develop a Se deficiency syndrome including seizures (5,6). As yet, there is no consensus whether Se is involved in neurodegenerative disorders such as Alzheimer’s or Parkinson’s disease. One key problem is that we do not know how blood Se variables correlate with brain Se status. A compilation of available data was published recently (7). In animal models of neurological disease, Willmore showed that Se administration had a beneficial effect after iron-induced epilepsy in rats (8,9). Later, a number of studies reported an increased sensitivity of rodents fed a low-Se diet to drug-induced nigrostriatal degeneration (10,11). Se supplementation, in turn, prevented dopamine loss, degeneration of neurons in the substantia nigra, and reduced lipid peroxidation (12–14). In addition, kainic acid injection into rats fed a low-Se diet led to increased hippocampal cell death and more pronounced seizures (15).

Hierarchical Biosynthesis of Selenoproteins in the Brain. Biochemical analysis provided evidence that Se in tissues exists mainly in a protein bound form (16). Moreover, Se exerts its biological effects predominantly after incorporation into selenoproteins as the rare amino acid selenocysteine (Sec)4 in which sulfur is replaced by Se (17). In eukaryotes, Sec is incorporated into proteins at UGA (STOP) codons in conjunction with a selenocysteine-insertion-sequence (SECIS), i.e., an RNA stem loop structure in the 3’-untranslated region of the mRNA. A recent review covers this issue in detail (18). In brief, several specific factors were identified that are essential for selenoprotein biosynthesis, i.e., a SECIS-binding protein, a Sec-specific tRNA Sec, and a tRNA Sec-specific elongation factor, EFSec. Importantly, Sec is not loaded onto tRNA Sec by an aminoacyl-tRNA synthetase, but is synthesized instead from seryl-tRNA Sec and selenophosphate. Two different proteins were identified in mammals that synthesize selenophosphate from ATP and selenide, one of which is a selenoprotein itself (SPS2). The tRNA Sec can be methylated at the wobble position of the anticodon by a specific 2’-O-methyltransferase. During Se deficiency, the ratio of methylated to nonmethylated forms decreases, but not in brain, indicating brain-specific regulation of selenoprotein biosynthesis (19). Another indication for an exceptional Se metabolism in the brain was first characterized by Behne and colleagues (20). They realized that the different tissues retain their Se to a different extent under conditions of Se depletion. The brain and endocrine organs rank at the top of this hierarchy such that dietary Se depletion results in only mildly reduced Se levels in these tissues, whereas organs such as the liver and skeletal muscle lose most of their Se. This finding also suggests that Se plays a more vital role in the brain and in endocrine glands than in other organs.

A second hierarchy exists in Se metabolism. It refers to the fact that the expression of individual selenoproteins is differentially affected by cellular Se content. For example, cellular glutathione peroxidase (GPx1), the first mammalian selenoenzyme identified (21,22), is generally more sensitive to Se restriction than is type I thyroid hormone deiodinase, the second selenoenzyme characterized (23–25). Attempts to explain this hierarchy with the “strength” of respective SECIS elements were partially successful, but cannot yet explain all observations (26–28). As a working hypothesis, it is assumed

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that selenoproteins ranking high in this hierarchy are more critical for basic cellular functions.

It is becoming more and more obvious that selenoprotein expression is not controlled solely by mRNA transcription, but rather by a combination of transcript availability and translational regulation (18). Because the UGA codon can in principle be interpreted both as a STOP and as a Sec codon, it is evident that some form of competition must exist between the termination machinery and the EFSec–tRNA\(^{\text{Sec}}\) complex. Because the formation of the EFSec–SPB2–SECIS complex also requires tRNA\(^{\text{Sec}}\) (29), an increased Se supply, via an increased pool of Sec–tRNA\(^{\text{Sec}}\), may simply enhance translational readthrough, whereas a limited supply of Sec–tRNA\(^{\text{Sec}}\) may directly increase the rate of termination. A quantitative analysis showed that only 5% of attempts to translate 5'-deiodinase type I mRNA are successfully completed beyond the UGA codon (30), a fraction well in line with results from a reporter gene study (28).

What Can Be Learned from the Transgenic Mouse Models? Transgenic overexpression of GPx1 in mouse brain confers increased resistance to neurotoxins (31) and experimental stroke (32), and protects electrophysiologic variables in an in vitro model of brain hypoxia (33). Accordingly, genetic inactivation of GPx1 in mice resulted in increased hydroxyl radical generation, cell death, and dopamine loss in models of chemically induced striatal degeneration (34). Similar observations were made in a model of transient brain ischemia in which GPx1 knockout (KO) mice displayed a dramatically increased infarct volume associated with increased apoptosis and increased lipid peroxidation (35). Thus, it seems conceivable that part of the protection exerted by Se is mediated through increased expression of the antioxidant enzyme GPx1. However, GPx1-deficient mice do not exhibit signs of neurological deficits if not experimentally challenged (36).

Selenoprotein P (SePP) is a glycosylated protein that carries up to 10 Sec residues per molecule and may represent the main carrier of Se in plasma (37). Interestingly, it was purified as a neurotrophic factor from bovine serum, and evidence exists that its Sec-rich C-terminus may be a preferred source of Se for cultured cells (38,39). Mice with a targeted inactivation of its gene do exhibit signs of a neurological disorder, including seizures and a movement disorder (40). SePP was suggested to transfer dietary-derived Se from liver to extrahepatic organs (41). Burk and colleagues (42) showed that rats injected with \(^{75}\)Se-labeled SePP accumulated the radiotracer rapidly in brain, indicative of a specific uptake mechanism for SePP by the central nervous system. This hypothesis is now supported by findings in SePP-deficient mice in which Se transport to extrahepatic organs, including the brain, is severely impaired (40,43). Apart from a drastic reduction of brain Se, the activities of the selenoenzymes GPx and thioredoxin reductase (TrxR) are diminished in SePP-deficient mice (40). Our own data (44) and those from Hill (43) also demonstrate that the SePP-dependent Se transport mechanism can be by-passed by increasing amounts of dietary Se, eventually rescuing the neurological phenotype of SePP-deficient mice. Interestingly, the chemical form of the supplement matters, i.e., selenite supplementation was more efficient than selenomethionine (45), possibly pointing to different uptake mechanisms of Se compounds into the brain. Hence, available data support the hypothesis that the preferential supply of the brain with Se may involve primarily, but not entirely, a SePP-dependent mechanism.

Using transgenic mice overexpressing a mutant tRNA\(^{\text{Sec}}\) that lacks the isopentenyl-modification at position 37, Hatfield and co-workers demonstrated a shift in selenoprotein expression, i.e., GPx1 activity decreased, whereas TrxR3 activity increased (46). Thus, the hierarchy among selenoproteins may be related in part to differential usage of different forms of tRNA\(^{\text{Sec}}\). Until now, no neurological phenotype has been reported in mice overexpressing the mutant tRNA\(^{\text{Sec}}\). Complete lack of tRNA\(^{\text{Sec}}\) in mice is embryonic lethal (47), but mice with a tissue-specific knockout of tRNA\(^{\text{Sec}}\) are viable (48). Moreover, there are no pathological changes in mammary glands from these mice. This finding comes as a surprise; the selenoenzyme TrxR was believed absolutely essential for cell proliferation because thio- reductin provides reducing equivalents to the key enzyme of DNA synthesis, ribonucleotide reductase. Whether selenoproteins in general have a pivotal role in neurons will have to await the generation and analysis of mice with a neuron-specific inactivation of tRNA\(^{\text{Sec}}\).

The most recently enzymatically characterized selenoprotein, SePR, is a methionine sulfoxide reductase [MsrB, SePR; (49)]. This enzyme, as well as its non-Se–containing counterpart, MsrA, reduces oxidized methionine residues in proteins. MsrA-deficient mice have a peculiar neurological phenotype, i.e., “tip-toe walking,” and a reduced life-span (50). In addition, low-Se diets reduced the activity of SePR in brain, eventually leading to an increased cellular methionine sulfoxide content (51). One of the potential physiologic consequences may be a modulated potassium flux because potassium channels of the shaker type can be regulated by reversible methionine oxidation (52).

The three types of thyroid hormone deiodinases are selected by thyroglobulin-dependent Se transport mechanism can be by-passed by increasing amounts of dietary Se, eventually rescuing the neurological phenotype of SePP-deficient mice. Interestingly, the chemical form of the supplement matters, i.e., selenite supplementation was more efficient than selenomethionine (45), possibly pointing to different uptake mechanisms of Se compounds into the brain. Hence, available data support the hypothesis that the preferential supply of the brain with Se may involve primarily, but not entirely, a SePP-dependent mechanism.
Open Questions. Metabolic labeling suggests the presence of the pituitary gland. Moreover, the lack of a neurological defect was in contrast to developmental or behavioral phenotype was reported (54).

Although recent years have seen tremendous progress in the molecular biology and metabolism of Se, we still know little about the cell type-specific and temporal pattern of selenoprotein expression in the brain. Transgenic mouse technology will certainly contribute to the elucidation of the functions of new selenoproteins in the near future. Thus, it seems likely that we are seeing the emergence of a new field at the intercept of nutritional sciences, molecular biology, and neurobiology that may shine new light on the issues of endogenous neuroprotective mechanisms and pathomechanisms in neurological disorders.

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LITERATURE CITED


TABLE 1

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<thead>
<tr>
<th>Gene modified</th>
<th>Major phenotype reported (reference)</th>
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<tr>
<td>Dio2 KO</td>
<td>Impaired thermogenesis in brown adipose tissue, impaired pituitary thyroxine feedback, minor growth retardation (54).</td>
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<tr>
<td>GPx2 KO</td>
<td>Colitis if double knockout with GPx1 (62).</td>
</tr>
<tr>
<td>GPx4 KO</td>
<td>Embryonic lethal (57, 63).</td>
</tr>
<tr>
<td>tRNA&lt;Sec&gt; KO</td>
<td>Embryonic lethal (47, 48).</td>
</tr>
<tr>
<td>i6A&lt;sup&gt;1&lt;/sup&gt;-tRNA&lt;sup&gt;Sec&lt;/sup&gt; KOTransgenic&lt;sup&gt;1&lt;/sup&gt;</td>
<td>Impaired GPx expression, enhanced TrxR expression (46); muscular hypertrophy after synergist ablation; increased protein phosphorylation in the mammalian target of rapamycin pathway (58).</td>
</tr>
<tr>
<td>Mammary-specific tRNA&lt;sup&gt;Sec&lt;/sup&gt; KO</td>
<td>Altered expression of p53 and BRC1A1 (48).</td>
</tr>
<tr>
<td>Liver-specific tRNA&lt;sup&gt;Sec&lt;/sup&gt; KO</td>
<td>No selenoprotein expression in liver; severely reduced SePP in plasma, liver degeneration between 1 and 3 mo associated with death (61).</td>
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<sup>1</sup>i6A<sup>1</sup>: do not contain the 6-isopentenyl-modified adenosine at position 37.
redistribution of the selenocysteine tRNA population in a tissue specific manner. Biochim. Biophys. Acta 1359: 25-34.