The Possible Neuroprotective Role of Selenium in Arsenic-Induced Midbrain Substantia Nigra Neurotoxicity in Guinea Pig Model: Biochemical and Histological Study

Original Article

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ABSTRACT

Introduction: Arsenic (As) is one of the heavy metals and it is widely present in our environment polluting the food and water consumed by the human. Chronic exposure to As leads to arsenic toxicity that produces certain undesirable effects on the human body including substantia nigra (SN) dopaminergic cells neurotoxicity. Selenium (Se) has many protective effects on the human body against toxic substances that include antidegenerative effects proved on the substantia nigra of the midbrain.

The Aim of the Work: The current study was carried out for evaluation of the possible neuroprotective role of selenium in arsenic-induced midbrain substantia nigra neurotoxicity in guinea pig model.

Material and Methods: In the current study, sixty adult male guinea pigs were used. The animals were left for 7 days to acclimate in the new environment then, divided randomly into equal 4 groups with fifteen animals per group. Group A (Control group); received distilled water orally daily. Group B (Se-treated group); received sodium selenite at a single daily orally dose of 0.25 mg/kg/day. Group C (As-treated group); received sodium arsenite at a single daily orally dose of 2.5 mg/kg/day. Group D (As & Se-treated group); received sodium arsenite at a single daily orally dose of 2.5 mg/kg/day followed after 2 hours by sodium selenite at a single daily orally dose of 0.25 mg/kg/day. The drugs were given daily for successive 8 weeks.

Results: The Arsenic exposure resulted in deformed dopaminergic neurons, many neurons with vacuolated cytoplasm and congested blood vessels. However, the co-administration of selenium showed normal multiple pyramidal dopaminergic neurons, no obvious cytoplasmic vacuoles or dilated vessels.

Conclusion: Selenium proved a protective effect on neurodegenerative toxic morphological changes induced by arsenic in dopaminergic neurons of SN of midbrain in guinea pig model.

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Key Words: Arsenic, dopaminergic cells, midbrain, selenium, substantia nigra.

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INTRODUCTION

The midbrain is the most cranial part of the brain stem and consists of a large anterior part; the cerebral peduncles and a smaller posterior part; the tectum. The cerebral peduncles consist of three parts from anterior backwards; they are the crura cerebri, substantia nigra (SN) and the tegmentum[1]. Substantia nigra consists of two different zones; pars reticulata anteriorly and pars compacta posteriorly[2].

The cellular component of pars compacta of SN consists mainly of dopaminergic cells that are darkly pigmented due to the presence of melanin granules that accumulate with age as a by-product of catecholamine synthesis. These dopaminergic cells secrete dopamine as a neurotransmitter that projects to the corpus striatum[2].

The normal dopaminergic neurons' function is to secrete dopamine in the brain that is required for the control and maintenance of the normal motor activity. Neurodegeneration of the dopaminergic cells leads to decreased level of dopamine in the brain leading to motor manifestations such as tremors, rigidity, bradykinesia and psychological disturbances[3].

Arsenic (As) is one of the naturally occurring metalloids with toxic properties on the human health. As is present in the environment either in a pure form or combined with other metals or sulfur. As is found in black, yellow and gray forms with the gray As is the commonest, the most stable and the only form with an industrial importance and a metallic picture[4]. The gray As loses its stability when exposed to humidity and forms a black layer of arsenic trioxide on the surface[5].

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The industrial uses of the gray As include mainly the production of car batteries and agricultural insecticides and pesticides[6]. However, the use of arsenic is generally diminishing because of its toxic and carcinogenic effects[7].

Several millions of individuals all over the world suffer from As toxicity[8] mainly through As-contaminated drinking water with As concentrations above 10 µg/L that represents a worldwide health problem[9,10]. Chronic As toxicity leads to numerous diseases affecting different body systems such as cancers, renal problems, hepatic affection[8,11] and neurodegenerative diseases[12].

Chronic exposure to As. causes oxidative stress that produces cell damage through enhancement of lipid peroxidation with the production of malondialdehyde (MDA), carbonylated proteins and reactive oxygen species (ROS)[13]. Moreover, As causes reduction of the level of glutathione (GSH) that is a strong antioxidant having the ability to protect against different oxidizing compounds including As[14]. As has numerous neurotoxic effects on the central nervous system including dopaminergic neuronal destruction affecting the SN of the midbrain through oxidative stress-mediated destruction of the mitochondria and neuronal degeneration as a result of decreased energy production, destruction of the intracellular microtubules and deposition of the protein aggregate alpha-synuclein[12].

The inevitable As exposure should be accompanied by administration of essential vitamins, probiotics, polyphenols or essential antioxidant micronutrient elements such as selenium (Se) that are needed to reduce the hazardous effects of As and prevent As toxicity[15].

Selenium is a vital micronutrient metalloid considered as an important antioxidant agent[16] and present in two forms; organic and inorganic. The organic Se is found in meat, seafood and cereals[17] while the inorganic Se is found mainly in the groundwater[18].

Selenium is required for normal body systems and functions such as brain[19], skeletal muscles[20], liver, kidney[21], immune system[22] and normal body growth and metabolism[23]. For normal body functions and avoidance of Se deficiency, the healthy adult females need a daily Se intake not less than 26 µg while the adult males need a daily Se intake not less than 34 µg[15]. Selenium has protective properties against the oxidative stress-induced cell destruction[24].

AIM OF THE WORK

The current study was carried out for evaluation of the possible neuroprotective role of selenium in arsenic-induced midbrain substantia nigra neurotoxicity in guinea pig model.

MATERIAL AND METHODS

Chemicals

Sodium arsenite: Arsenic extra pure powder 500 gm with a concentration 98%. LOBA CHEMIE PVT.LTD. This was purchased from Sigma Aldrich, Cairo, Egypt.

Sodium selenite: Selenium-ACE. Film coated tablets. Hocher Pharmaceutical Industries, Badr city, Egypt.

Animals

Sixty healthy adult male guinea pigs (Caviaporcellus) were used for the experiment. The animals were bred in the animal house (Faculty of Medicine, Ain shams University). At the beginning of the study, the animals were 850 – 950 grams and of two years of age with no diseases; no motor disabilities, skin diseases or nutritional problems[14]. The animals were bred according to the ethics of experimental research center in the Ain Shams University with a code number in experimental animal research unit is (RE (191)23).

Food and environment

The guinea pigs were housed in a suitable room with good ventilation and a temperature maintained at 26 ± 2°C and received a standard mixture diet comprised of 0.05 % ascorbic acid, 1 % common salt, 2 % vitamin-mineral mixture, 5 % soybean, 10 % fish meal, 20 % ground gram grain, 27 % ground maize grain and wheat bran (35 %)[14,23]. The animals were left for one week for acclimatization, and then divided into 4 equal groups randomly; each of them consisted of fifteen guinea pigs.

Experimental design

Group A (Control group): Received distilled water by gastric gavage daily orally for 8 weeks.

Group B (Se-treated group): Received sodium selenite at a single daily orally dose of 0.25 mg/kg/day for 8 weeks[10].

Group C (As-treated group): Received sodium arsenite at a single daily orally dose of 2.5 mg/kg/day for 8 weeks[25].

Group D (As and Se-treated group): Received sodium arsenite at a single daily orally dose of 2.5 mg/kg/day[25] followed after 2 hours by sodium selenite at a single daily orally dose of 0.25 mg/kg/day[10]. Both sodium selenite and sodium arsenite were given by gastric gavage after dissolution in distilled water. They were given daily for successive 8 weeks[10,25]. At the end of the experiment that lasted for eight weeks, the animals were sacrificed by intraperitoneal overdose of phenobarbital then; the head and brain were dissected.

Biochemical analysis

The biochemical analysis procedures were done at the biochemistry department, Faculty of Medicine, Al-Azhar University, Cairo, Egypt .The skulls and brains of five animals of each group were dissected and the midbrains were identified by the presence of the characteristic corpora quadrigemina on the posterior surface[1]. The midbrains were isolated by separating them from the pons, diencephalon and cerebellum, then washed in cold saline and used for biochemical analysis. These specimens were

homogenized by centrifugation at 5000 rpm for 10 min in a homogenizer containing sodium phosphate buffer then, used to evaluate the levels of GSH, proteins carbonyls and MDA that are indicators for oxidative stress-mediated cell destruction[25]. The rest of the specimens were processed for histological evaluation.

Histological evaluation of midbrain

The midbrains of ten guinea pigs of each group were processed for histological evaluation; half of the samples were prepared as paraffin sections for light microscopic examination while the samples of the other half were processed for ultrastructural examination by electron microscopy.

Light microscopy

The midbrains were put immediately in 10% formalin that hardened the specimens. Then, the specimens were dehydrated by ascending concentrations of alcohol then, embedded in paraffin. Finally, the specimens were cut by the microtome to give 5 µm thick sections that were mounted on glass slides and subjected to the following staining techniques:

Haematoxylin and eosin: - For assessment and detection of the morphological changes of SN[26].

Immunohistochemical staining using tyrosine hydroxylase (TH) antibody: for detection of the dopaminergic cells of SN[27]. Tyrosine hydroxylase is a polyclonal antibody (Thermofisher Scientific Corporation, USA, and Catalog No.: PA1-18315). Positive control: was the brain (striatum, substantia nigra) dopaminergic neurons using tyrosine hydroxylase antibody. Negative control: Additional specimens of midbrain were processed in the same way like the positive control but omitting the step of primary antibody.

Transmission electron microscopy (TEM)

The electron microscopy specimens were processed, examined and photographed at the electron microscope unit, Al-Azhar University, Cairo, Egypt. The midbrains were cut into small parts of about 1 mm^3 size. These parts were put in 2% gluteraldehyde for fixation then immersed for 2 hours in 0.1 mol/l phosphate at pH 7.4. The specimens then dehydrated by ascending grades of alcohol. Then, the specimens were incubated in resin mixed with propylene oxide. Finally, the specimens were put in BEEM capsules and cut by the ultramicrotome to give ultrathin sections then, stained by uranyl acetate and lead citrate. Electron microscopic examination of the sections was done for ultrastructural evaluation of the SN dopaminergic neurons[28].

Morphometric study and Statistical analysis

Morphometric study was done at the Histology department, Faculty of Medicine, Al-Azhar University, Cairo, Egypt by Leica "Qwin 500C" image analyzer. The TH- positive dopaminergic neurons mean count was measured using interactive measurements menu/µ2 in the SN pars compacta in the four groups.

Statistical Package of Social Sciences (SPSS) with the assistant software version 7.7 was used for statistical analysis of the measurements found in the present study. The means of the measurements obtained were compared along the four groups through the one way analysis of variance (ANOVA). *P-value* <0.05 was statistically significant[29].

RESULTS

Biochemical results

Treatment with Se alone caused no significant changes in MDA, proteins carbonyls and GSH levels when compared to the control group. As administration resulted in a significant elevation of MDA and protein carbonyls level with a significant reduction of GSH level in the As group when compared to the control. However, co-treatment with Se resulted in restoration of MDA, proteins carbonyls and GSH levels towards the normal levels in the combined As and Se-treated group. In addition the combined As and Se-treated group showed a significant decreased values of MDA, proteins carbonyls and a significant elevation of GSH level as compared to the As group (Table1, Figure 1).

Histological results

i- Hematoxylin and eosin sections

Pars compacta of the SN of the control group showed the normal appearance of SN with densely packed dopaminergic neurons; multiple of them were pyramidal in shape and few were nonpyramidal. The Se treated group showed densely packed dopaminergic neurons; multiple of them were pyramidal in shape and few were nonpyramidal. The As treated group showed deformed fewer dopaminergic neurons, vacuolated neuropil and cytoplasm of neurons, in addition to congested blood vessels. The combined As and Se group showed apparently normal pyramidal dopaminergic neurons, no obvious vacuolations and no obvious dilated vessels (Figures 2,3).

ii-Tyrosine hydroxylase immunohistochemistry sections

The control group revealed densely packed dark brown TH immune-positive neurons. Selenium-treated group showed densely packed dark brown immune- positive neurons. The As-treated group showed few positive dopaminergic neurons. The combined As and Se group showed multiplicity of the TH positive dopaminergic neurons (Table 2, Figures 4,5).

iii- Transmission electron microscopy sections

The control and Se group showed normal dopaminergic neurons having regular nuclei with fine chromatin and regular non-distended mitochondria. The axons were surrounded by normal; regular thick continuous myelin sheaths. However, the As-treated group showed the dopaminergic neuron constituting of irregular

heterochromatic nucleus and aggregated condensed nuclear chromatin. There were cytoplasmic vacuoles, mitochondria with destroyed cristae and the axons were surrounded by thin myelin sheaths. In the As and Se group, the dopaminergic neuron demonstrated normal regular nucleus , fine chromatin, regular mitochondria and few thin myelin sheaths (Figure 6).

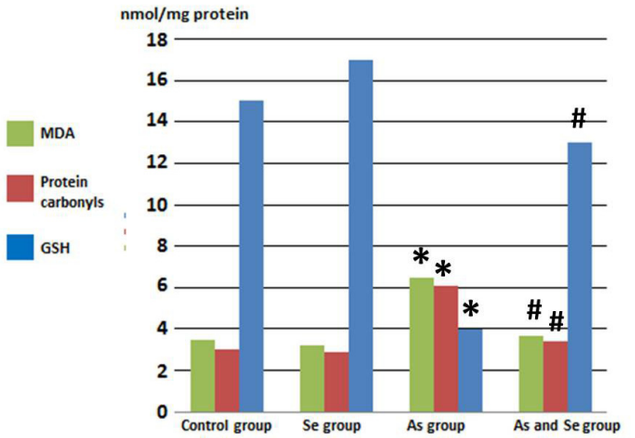

Fig. 1: A histogram showing the MDA, proteins carbonyls and GSH mean levels in nmol/mg protein.
* Significant to the control.
# Significant to As group.

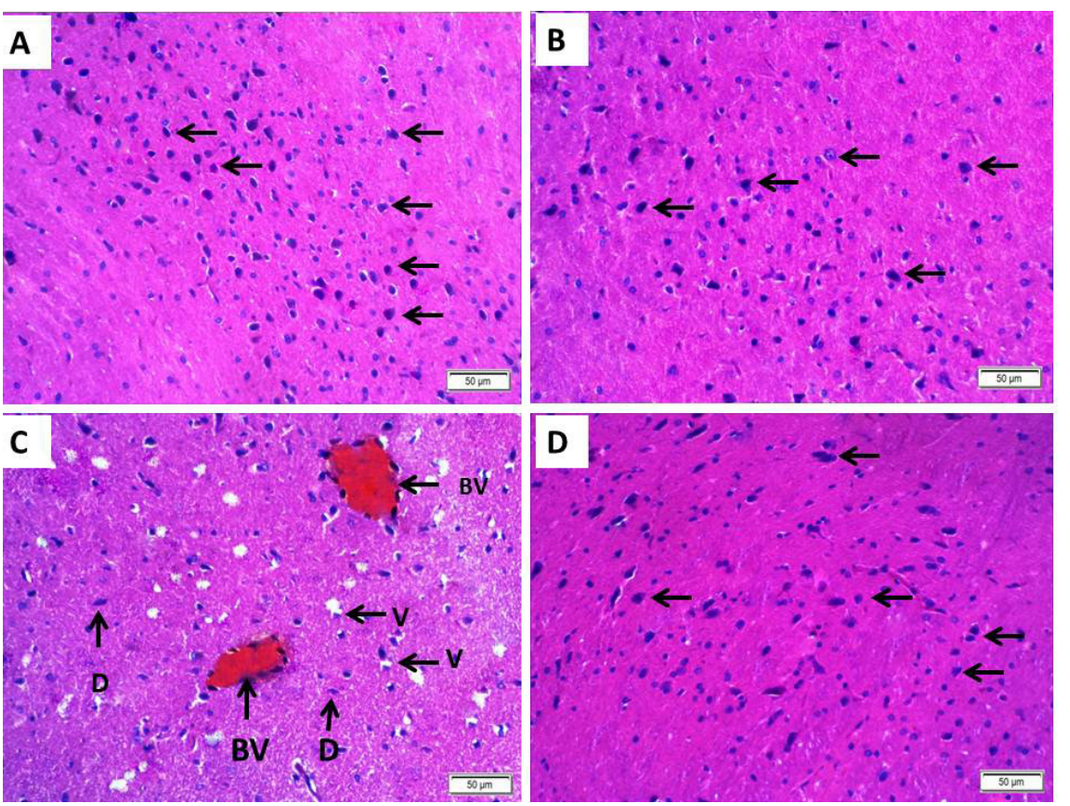

Fig. 2: The photomicrograph A (control group) shows the normal appearance of SN with densely packed dopaminergic neurons (arrows). Photomicrograph B (Se group) shows densely packed dopaminergic neurons (arrows). Photomicrograph C (As group) shows deformed fewer dopaminergic neurons (D), vacuolated neuropil (V) and congested blood vessels (BV). Photomicrograph D (As and Se group) shows multiple pyramidal dopaminergic neurons (arrows), no obvious vacuolations and no obvious dilated blood vessels. (H & E; X 200).

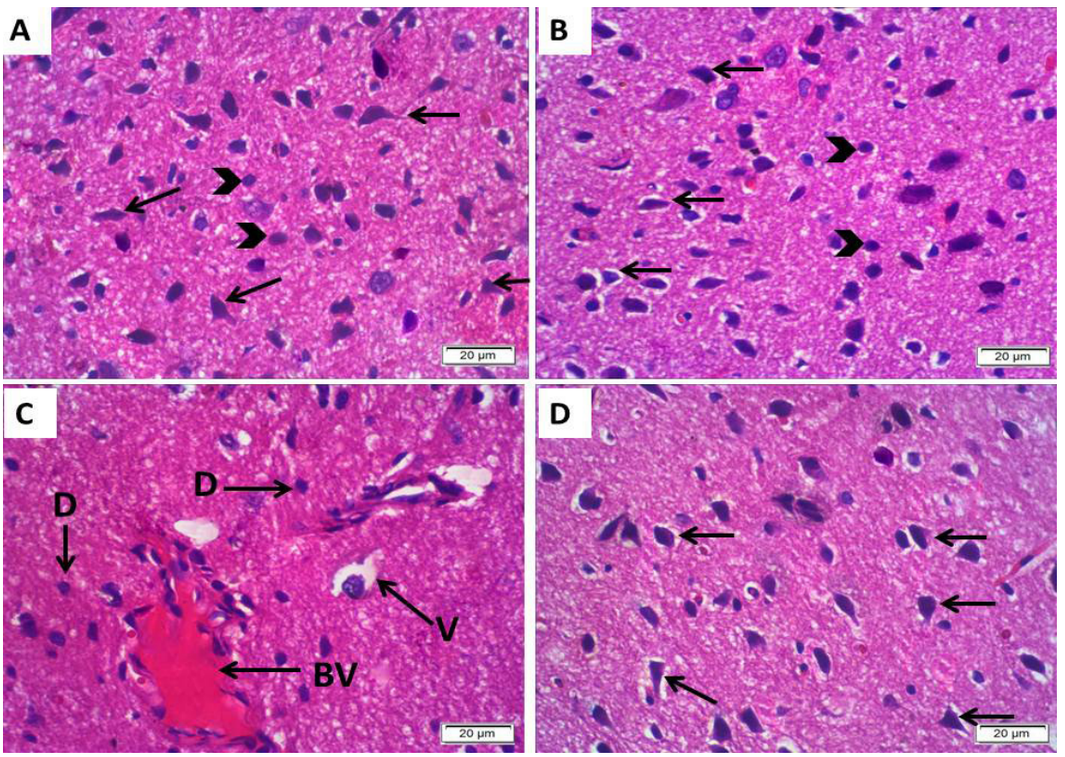

Fig. 3: The photomicrograph A (control group) shows the normal appearance of SN with dopaminergic neurons; multiple of them are pyramidal in shape (arrows) and few are nonpyramidal (arrowheads). Photomicrograph B (Se group) shows multiple pyramidal (arrows) and few nonpyramidal (arrowheads) dopaminergic neurons. Photomicrograph C (As group) shows deformed dopaminergic neurons (D), vacuolated cytoplasm of a neuron (V) and congested blood vessels (BV). Photomicrograph D (As and Se group) shows multiple apparently normal pyramidal dopaminergic neurons (arrows), no obvious vacuolations and no obvious dilated vessels. (H & E; X 400).

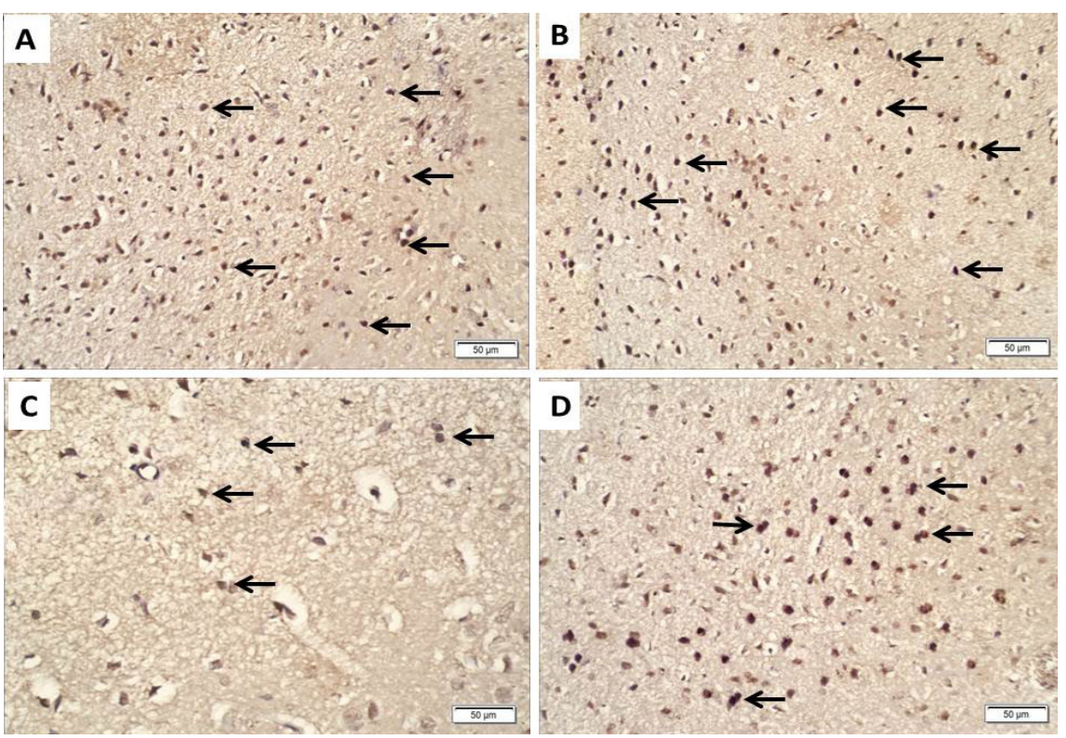

Fig. 4: The photomicrograph A (control group) shows densely packed dark brown TH immune-positive neurons (arrows). Photomicrograph B (Se group) shows densely packed dark brown immune- positive neurons (arrows). Photomicrograph C (arsenic group) shows few positive dopaminergic neurons (arrows). Photomicrograph D (As and Se group) shows multiplicity of positive dopaminergic neurons (arrows). (TH immunostaining; X 200).

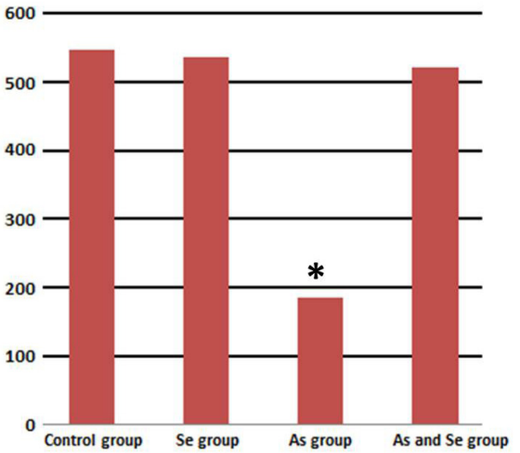

Fig. 5: A histogram showing mean count of the TH+ dopaminergic cells/ µ2 in the SN pars compacta of the four groups

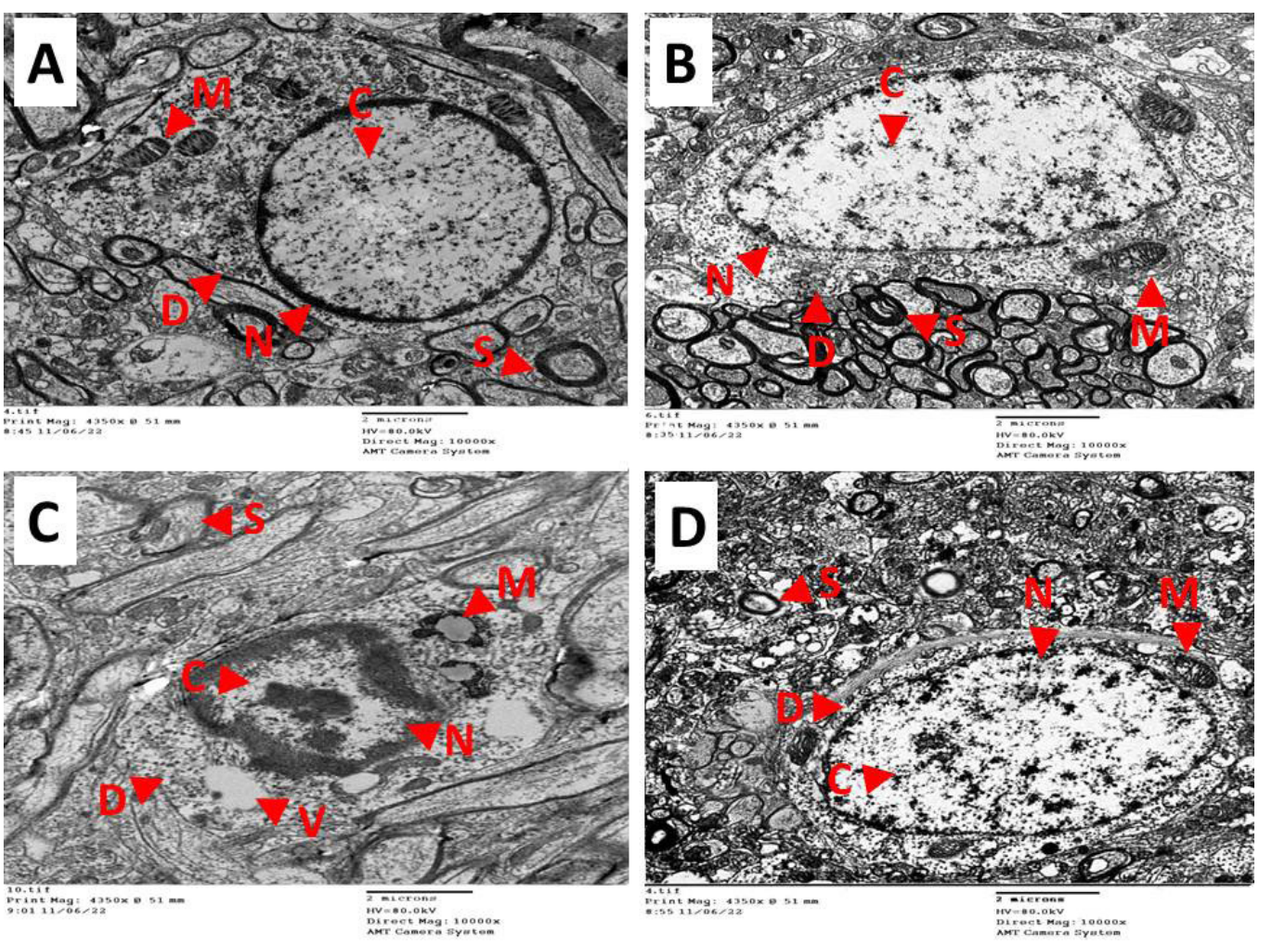

Fig. 6: The photomicrographs A and B that represent the control and Se group respectively show normal dopaminergic neurons (D) having regular nuclei (N) with fine chromatin (C) and regular non-distended mitochondria (M). The axons are surrounded by normal; regular thick continuous myelin sheaths (S). The photomicrograph C (As group) shows the dopaminergic neuron (D) constituting of irregular heterochromatic nucleus (N) and aggregated condensed nuclear chromatin (C). There are cytoplasmic vacuoles (V), mitochondria with destroyed cristae (M) and the axons are surrounded by thin myelin sheaths (S). In the photomicrograph D (As and Se group), the dopaminergic neuron (D) demonstrates normal regular nucleus (N), fine chromatin (C), regular mitochondria (M) and few thin myelin sheaths (S). (Scale bar 2 mm; X 10000).

Table 1: MDA, proteins carbonyls and GSH mean levels in nmol/mg protein.

	Control Group	Se Group	As Group	As + Se Group	*P value*	Significance
MDA	3.5	3.4	6.3*	3.8 #	<0.002	Highly significant
Protein carbonyls	3.1	3.1	6.1*	3.5 #	<0.002	Highly significant
GSH	15	16.8	4*	13 #	<0.002	Highly significant

* Significant to the control.

Significant to As group.

Table 2: Dopaminergic (TH +ve) neurons count/ μ^2 in the SN pars compacta in the four groups showing highly significant decrease in mean count in As group compared to the other groups.

TH + cells	Control Group	Se Group	As Group	As + Se Group	*P value*	Significance
The mean number	537	522	183*	519	<0.004	Highly significant

DISCUSSION

In the present study, the neuroprotective role of selenium (Se) in arsenic (As) -induced neurotoxicity of substantia nigra dopaminergic neurons in guinea pigs was assessed because the neurotoxic effects of As represent a global problem affecting children and adults[30,31]. In chronic As exposure, it accumulates in the brain as it crosses the blood brain barrier. The brain represents one of the important target organs in cases of chronic As toxicity producing neurotoxicity and neurological deficits including behavioral, cognitive, intellectual, learning and memory impairment[25]. Arsenic toxicity affects the brain easily because the brain tissue is deficient in glutathione (GSH) that is the main antioxidant agent against the ROS hence; the brain tissue is highly sensitive to the As-induced oxidative stress[32].

The obtained data in the present study revealed that As administration led to SN dopaminergic neurodegenerative changes mediated by As-induced oxidative stress. This was marked by increase MDA, increase protein carbonyl and decreased GSH levels in the brain. This was in agreement with previous studies detected that As toxicity produces oxidative stress due to increased lipid peroxidation[33]. In addition, the As-induced reduction of GSH concentration in the present work might be attributable to As capacity to interfere with GSH metabolism[34] through alteration of the activity of GSH reductase and GSH peroxidase enzymes that are involved in GSH metabolism[35]. This was in correlation with numerous past studies reported As-mediated depletion in GSH level in guinea pigs[14], rats[36] and human[37].

In the current study, the co-administration of Se reversed the alterations induced by As. The neuroprotective effects of Se were indicated biochemically by restoration of the levels of the oxidative stress markers towards the normal levels. MDA level, carbonylated protein level decreased and GSH level increased significantly near the normal levels. This indicates the ability of Se to attenuate the As-mediated oxidative neurodegenerative changes. It was proved that, Se prevents the As-mediated oxidative stress in the brain tissue by inhibition of lipid peroxidation, oxidative destructive carbonylation of proteins and GSH depletion mediated by As[10,38]. These neuroprotective effects of Se were further explained by its ability to regulate the balance between the oxidants and antioxidants activities, also Se prevents interleukin-1β that is a pro-inflammatory cytokine and increases the level of interleukin-4 that is an anti-inflammatory cytokine[39].

In the present work, the neurodegenerative effects of As on SN were obvious and marked by significantly reduced dopaminergic neurons multiplicity with the appearance of deformed dopaminergic neurons and many neurons had vacuolated cytoplasm. This was confirmed by mitochondria exhibiting destroyed cristae indicating degeneration. In addition, the nuclei were irregular with aggregated condensed chromatin and the axons were surrounded by thin myelin sheaths.

These results were in line with numerous previous studies proved the destructive effects of As on the brain through inducing oxidative stress and destroying the mitochondria[40]. Also, it was previously confirmed that, As-induced neurotoxicity leads to degenerative changes affecting the myelin sheaths around the axons, synaptic disruption and formation of degenerative vacuoles[41]. This was mediated by disruption of the dopaminergic and cholinergic signaling pathways and enhancement of chromatin damage and apoptosis[42]. These neurotoxic effects of As were attributable to the As-induced overproduction of ROS that accumulate inside the mitochondria. Hence, the chronic exposure to As destroys the brain cells' mitochondria leading to consecutive brain cell damage because the normal brain cells' function depend on intact mitochondria due to their high energy requirements[43].

The neuroprotective properties of Se were indicated histologically as in the combined As and Se treated group, multiple pyramidal dopaminergic neurons were detected. In addition, no obvious cytoplasmic vacuoles or dilated blood vessels were observed. These observations were confirmed by immunohistochemical staining where

multiplicity of the TH positive dopaminergic neurons was obvious. In the TEM sections, the dopaminergic neurons showed normal regular nucleus, fine chromatin, regular mitochondria and few thin myelin sheaths. The neuroprotective effects of Se detected in the present study were in correlation with previous findings detected that; Se has neuroprotective properties through its antiapoptotic and antioxidant capacities[44].

In addition, it was reported that Se has neuroprotective effects against As-mediated motor and locomotor insufficiencies through inhibition of the arsenic-induced oxidative stress mediated damage of the dopaminergic cells[10]. These results were explained by the previously proved importance of Se for the normal antioxidant status in the brain tissue because Se is essential for the formation of selenoproteins that are required for the antioxidant activity[45]. Moreover, it was previously proved that, Se exhibits anti-inflammatory and antioxidant properties against As-mediated neurotoxicity through enhancement of As methylation to decrease the level of As in the blood and brain tissue[46].

CONCLUSION

Selenium proved a protective effect on neurodegenerative toxic morphological changes induced by arsenic in dopaminergic neurons of SN of midbrain in guinea pig model.

CONFLICT OF INTERESTS

There are no conflicts of interest.

REFERENCES

1. Junior L.S., Lemos N.B., Gonçalves L.F., Dias A.J., Neto O.C., Sousa C.C., Diniz A.M., Rabelo N.N., Barroso L.K., Valenca M.M. and Filho H.R.: The anatomy of the brain – learned over the centuries. Surg. Neurol. Int. (2021) 12: 319.
2. Parraga R.G., Possatti L.L., Alves R.V., Ribas G.C., Ture U., de Oliveira E.: Microsurgical anatomy and internal architecture of the brainstem in 3D images: surgical considerations. J. Neurosurg. (2016) 124(5):1377-1395.
3. Darabi S., Noori-Zadeh A., Abbaszadeh H., Rajaei F. and Bakhtiyarie S. : Trehalose Neuroprotective Effects on the SubstantiaNigra Dopaminergic Cells by Activating Autophagy and Non-canonical Nrf2 Pathways. Iran J Pharm Res. (2019) 18(3):1419–1428.
4. Schoo C., Bestgen S., Egeberg A., Seibert J., Konchenko S., Feldmann C. and Roesky P.: Samarium Polyarsenides Derived from Nanoscale Arsenic. Angewandte. (2019) 58:4386-4389.
5. Zhao L., Xu Q., Wang X., He J., Li J., Yang H., Long Y., Chen D., Liang H., Li C., Xue M., Ren Z., Lu L., Weng H., Fang Z., Dai X. and Chen G.: Magneto transport properties in a compensated semimetal gray arsenic. Physical Review B. (2017) 95:11-15.
6. Zhu Z., Guan J. and Tománek D.: Strain-induced metal-semiconductor transition in monolayers and bilayers of gray arsenic: A computational study. Physical Review B. (2015) 91:15-16.
7. Asere T.G., Stevens C.V. and Laing G.D.: Use of (modified) natural adsorbents for arsenic remediation: A review. Science of the total environment. (2019) 676:706-720.
8. Chen Q.Y.And Costa M.: Arsenic: A global environmental challenge. Annual review of Pharmacology and Toxicology. (2021) 61:47-63.
9. Hashmi MZ, Kanwal A, Murtaza R, Siddique S, Su X, Tang X and Afzaal M.: Arsenic in Untreated and Treated Manure: Sources, Biotransformation, and Environmental Risk in Application on Soils. Soil Biology. (2018) 53:179-195.
10. Adedara I.A., Fabunmi A.T., Ayenitaju F.C., Atanda O.E., Adebowale A.A., Ajayi B.O., Owoeye O., Rocha J.B.T. and Farombi E.O.: Neuroprotective mechanisms of selenium against arsenic-induced behavioral impairments in rats. Neurotoxicology. (2020) 76:99-110.
11. Singh G., Thaker R., Sharma A. and Parmar D.: Therapeutic effects of biochanin A, phloretin, and epigallocatechin-3-gallate in reducing oxidative stress in arsenic-intoxicated mice. Environmental Science and Pollution Research. (2021) 28:20517-20536.
12. Cholanians A.B., Phan A.V., Ditzel E.J., Camenisch T.D., Lau S.S. and Monks T.J.: Arsenic Induces Accumulation of α-Synuclein: Implications for Synucleinopathies and Neurodegeneration. Toxicological sciences. (2016) 153:271-281.
13. Ogun M., Ozcan A., Karaman M., Merhan O., Ozen H., Kukurt A. and Karapehlivan M.: Oleuropein ameliorates arsenic induced oxidative stress in mice. Journal of Trace Elements in Medicine and Biology. (2016) 36:1-6.
14. Mohanta R.K., Garg A.K., Dass R.S. and Behera S.K.: Blood Biochemistry, Thyroid Hormones, and Oxidant/ Antioxidant Status of Guinea Pigs Challenged with Sodium Arsenite or Arsenic Trioxide. Biological Trace Element Research. (2014)160:238–244.
15. Zwolak I.: The Role of Selenium in Arsenic and Cadmium Toxicity: an Updated Review of Scientific Literature. Biological Trace Element Research. (2019) 193:44–63.
16. Rahbardar MG.,Farmad HC., Hosseinzadeh H. and Mehri S.: Protective effects of selenium on acrylamide-induced neurotoxicity and hepatotoxicity in rats. Iranian Journal of Basic Medical Sciences. (2021) 24(8):1041-1048.

17. Ullah H., Liu G., Yousaf B., Ali MU., Abbas Q., Munir MAM. and Mian MM.: Developmental selenium exposure and health risk in daily foodstuffs: a systematic review and meta-analysis. Ecotoxicol. Environ. Saf. (2018) 149: 291-306.

18. Hu T., Liang Y., Zhao G., Wu W., Li H. and Guo Y.: Selenium biofortification and antioxidant activity in cordycepsmilitaris supplied with selenate, selenite, or selenomethionine. Biol. Trace Elem. Res. (2019) 187:553-561.

19. Solovyev N.D.: Importance of selenium and selenoprotein for brain function: From antioxidant protection to neuronal signaling. Journal of Inorganic Biochemistry. (2015) 153:1-12.

20. Hillamy K.E., James K.M., Raymond M.M. and Burk F.: Combined selenium and vitamin C deficiency causes cell death in guinea pig skeletal muscle. Nutrition research. (2009) 29(3):213-219.

21. Mehdi Y., Hornick J., Istasse L. and Dufrasne I.: Selenium in the environment, metabolism and involvement in body functions. Molecules.(2013) 18(3):3292-3311.

22. Bunglavan S.J, Garg A.K., Dass R.S., Shrivastava S. and Chaturvedi V.K.: Effect of supplementation of different sources of selenium on humoral immunity in guinea pigs. Journal of Immunology and Immunopathology.(2013) 15(2):169-174.

23. Chaudhary M., Garg A.K., Mittal G.K. and Mudgal V.: Effect of Organic Selenium Supplementation on Growth, Se Uptake, and Nutrient Utilization in Guinea Pigs. Biological Trace Element Research. (2009) 133:217-226.

24. Zhao X., Zhou W., Jian-jun L., Chen C., Ping-chuan Z., Lu D., Jing-hong C., Qun C., Xiao-tian Z. and Zhi-lun W.: Protective effects of selenium on oxidative damage and oxidative stress related gene expression in rat liver under chronic poisoning of arsenic. Food and Chemical Toxicology. (2013) 58:1-7.

25. Soni M., Prakash C., Dabur R and Kumar V.: Protective effect of hydroxytyrosol against oxidative stress mediated by arsenic-induced neurotoxicity in rats.Applied Biochemistry and Biotechnology. (2018)186:27–39.

26. Liudan C., Ke C., Liang A., Linhui C., Bin Z., Xue L., Mengru K. and Jianjun L.: Effect of electro acupuncture on dopaminergic neurons in a rat model of Parkinson's disease based on the α-synuclein pathway. Materials Express. (2020) 10(1):62-69.

27. Bigham M., Mohammadipour A., Hosseini M., Malvandi A.M. and Ebrahimzadeh-Bideskan A.: Neuroprotective effects of garlic extract on dopaminergic neurons of substantia nigra in a rat model of Parkinson's disease: motor and non-motor outcomes. Metabolic Brain Disease. (2021) 36:927-937.

28. Biesemeier A., Eibl O., Eswara S., Audinot J., Wirtz T., Pezzoli G., Zucca F.A., Zecca L. and Schraermeyer U.: Elemental mapping of Neuromelanin organelles of human substantia nigra: correlative ultrastructural and chemical analysis by analytical transmission electron microscopy and nano-secondary ion mass spectrometry. Journal of Neurochemistry. (2016)138(2):339-353.

29. Thilakaratne M., Falkner K. and Atapattu T.x.: A Systematic Review on Literature-based Discovery: General Overview, Methodology &Statistical Analysis. ACM Computing Surveys. (2020) 52(6):1-34.

30. Tolins M., Ruchirawat M. and Landrigan P.: The developmental neurotoxicity of arsenic: cognitive and behavioral consequences of early life exposure. Ann Glob Health. (2014) 80:303-314.

31. Signes-Pastor A.J., Vioque J., Navarrete-Munoz E.M., Carey M., Garcia-Villarino M., Fernández-Somoano A., Tardon A., Santa-Marina L., Irizar A., Casas M., Guxens M., Llop S., Soler-Blasco R., García-de-la-Hera M., Karagas M.R. and Meharg A.A. : Inorganic arsenic exposure and neuropsychological development of children of 4-5 years of age living in Spain. Environ. Res. (2019) 174:135-142.

32. Cobley J.N., Fiorello M.L. and Bailey D.M.: 13 reasons why the brain is susceptible to oxidative stress.Redox Biology. (2018) 15:490-503.

33. Zabel M., Nackenoff A., Kirsch W.M., Harrison F.E., Perry G. and Schrag M.: Markers of oxidative damage to lipids, nucleic acids and proteins and antioxidant enzymes activities in Alzheimer's disease brain: A meta-analysis in human pathological specimens. Free radical Biology and Medicine.(2018) 115:351-360.

34. Bashir S., Sharma Y., Irshad M., Gupta S.D. and Dogra T.D.: Arsenic-Induced Cell Death in Liver and Brain of Experimental Rats.Basic and Clinical Pharmacology and Toxicology. (2006) 98(1):38-34.

35. Yen CC., Ho TJ., Wu CC., Chang CF., Su CC., Chen YW. And Huang CF.: Inorganic arsenic causes cell apoptosis in mouse cerebrum through an oxidative stress-regulated signaling pathway. Arch. Toxicol. (2011) 85(6):565-575.

36. Samad N.,Rao T.,Rehman M.H., Bhatti S.A. and Imran I.: Inhibitory Effects of Selenium on Arsenic-Induced Anxiety-/Depression-Like Behavior and Memory Impairment. Biological Trace Element Research. (2021) 200:689-698.

37. Bjorklund G.,Oliinyk P.,Lysiuk R.,Rahaman M.S., Antonyak H., Lozynska I., Lenchyk L. and Peana M.: Arsenic intoxication: general aspects and chelating agents. Archives of Toxicology. (2020) 94:1879-1897.

38. Yang J.Y., Du J.J., Zhang Y.F. and Li Y.X.: Vitamin E and selenium partially prevent cytotoxicity, oxidative stress and DNA damage induced by T-2 toxin in bovine Leydig cells. Theriogenology. (2022) 189:255-261.

39. Senol N., Nazıroglu M. and Yuruker V.: N-acetylcysteine and selenium modulate oxidative stress, antioxidant vitamin and cytokine values in traumatic brain injury-induced rats. Neurochemical research. (2014) 39:685-692.

40. Saha S., Sadhukhan P., Mahalanobish S., Dutta S. and Sil P.: Ameliorative role of genistein against age-dependent chronic arsenic toxicity in murine brains via the regulation of oxidative stress and inflammatory signaling cascades. Nutritional Biochemistry.(2018) 55:26-44.

41. Srivastava P., Yadav R.S., Chandravanshi L.P., Shukla R.K., Dhuriya Y.K., Chauhan L.K., Dwivedi H.N., Pant A.B. and Khanna V.K.: Unraveling the mechanism of neuroprotection of curcumin in arsenic induced cholinergic dysfunctions in rats. Toxicol. Appl. Pharmacol. (2014) 279:428-440

42. Lu T.H., Tseng T.J., Su C.C., Tang F.C., Yen C.C., Liu Y.Y., Yang C.Y., Wu C.C., Chen K.L., Hung D.Z. and Chen Y.W.: Arsenic induces reactive oxygen species-caused neuronal cell apoptosis through JNK/ERK-mediated mitochondria-dependent and GRP 78/CHOP-regulated pathways. Toxicol. Lett. (2014) 224:130-140.

43. Prakash C., Soni M. and Kumar V.: Mitochondrial oxidative stress and dysfunction in arsenic neurotoxicity: A review. Journal of Applied Toxicology. (2016) 36(2):179-188.

44. Wei D., Chen T., Yan M., Zhao W., Li F., Cheng W. and Yuan L.: Synthesis, characterization, antioxidant activity and neuroprotective effects of selenium polysaccharide from Radix hedysari. Carbohydrate Polymers.(2015) 125:161-168.

45. Brauer AU. and Savaskan NE.: Molecular actions of selenium in the brain: Neuroprotective mechanisms of an essential trace element. Reviews in the Neurosciences. (2004) 15(1):19-32.

46. Su C.T., Hsieh RL., Chung CJ., Huang PT., Lin YC., Ao PL. and Hsueh YM.: Plasma selenium influences arsenic methylation capacity and developmental delays in preschool children in Taiwan. Environmental research. (2019) 171, 52-59.

الملخص العربى

الدور الوقائي العصبي المحتمل للسلينيوم في التسمم العصبي المستحدث بالزرنيخ في المادة السمراء للمخ المتوسط في نموذج خنزير غينيا : دراسة كيميائية حيوية و هستولوجية

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الخلفية: يعتبر الزرنيخ من المعادن الثقيلة و هو واسع الانتشار في بيئتنا والذي يتسبب في تلوث الطعام ومياه الشرب التي يستهلكها الانسان. التعرض المزمن للزرنيخ يؤدي إلى التسمم بالزرنيخ و الذي ينتج عنه بعض الآثار الغير مرغوب فيها على جسم الإنسان والتي تشمل التسمم العصبي على الخلايا الدوبامينية للمادة السمراء . السيلينيوم له الكثير من الآثار الوقائية على جسم الإنسان ضد المواد السامة و التي تشمل الآثارالتدميرية على المادة السمراء للمخ المتوسط.
الهدف من البحث: أجريت الدراسة الحالية لتقييم الدورالوقائي العصبي المحتمل للسلينيوم في التسمم العصبي المستحدث بالزرنيخ في المادة السمراء للمخ المتوسط في نموذج خنزير غينيا.
المواد والطرق: في الدراسة الحالية تم استخدام ستين ذكرا بالغا من خنازير غينيا. تركت هذه الحيوانات لمدة اسبوع للتأقلم في البيئة الجديدة ثم تم تقسيم الحيوانات عشوائيا إلى أربع مجموعات متساوية تضم كل منها خمسة عشر حيوانا. المجموعة أ (المجموعة الضابطة) تلقت الماء المقطر يوميا عن طريق الفم. المجموعة ب (المجموعة المعالجة بالسلينيوم) تلقت سيلينيوم الصوديوم بجرعة واحدة يوميا ٠,٢٥ مج/كجم عن طريق الفم. المجموعة ج (المجموعة المعالجة بالزرنيخ) تلقت زرنيخ الصوديوم بجرعة واحدة يوميا ٢,٥ مج/كجم عن طريق الفم. المجموعة د (المجموعة المعالجة بالزرنيخ و السلينيوم) تلقت زرنيخ الصوديوم بجرعة واحدة يوميا ٢,٥ مج/كجم و سيلينيوم الصوديوم بجرعة واحدة يوميا ٠,٢٥ مج/كجم بفارق ساعتين يوميا . تم تلقي العلاج يوميا لمدة ثمانية اسابيع متصلة.
النتائج: التعرض للزرنيخ أدى إلى تشوه الخلايا العصبية الدوبامينية مع ظهور فجوات سيتوبلازمية في الخلايا وظهور الاوعية الدموية المحتقنة . ومع ذلك فان العلاج المزدوج بالسيلينيوم أظهر العديد من الخلايا الدوبامينية الهرمية الشكل وعدم ظهور فجوات سيتوبلازمية أو أوعية دموية متسعة.
الإستنتاج: السيلينيوم أعطى تأثيرا وقائيا ضد التاثيرات التدميرية العصبية للزرنيخ على الخلايا الدوبامينية للمادة السمراء للمخ المتوسط في نموذج خنزير غينيا.