

Nanoparticle systems are comprised of ultra-fine materials ranging from 1 to 100 nm which possess unique physico-chemical properties different from properties of their bulk material. Silver nanoparticles (AgNPs) exhibit strong antibacterial activity. Thus, they are commonly used in many medical products (wound dressings, implants, catheters, surgical instruments, antibacterial fluids), so as in cosmetics and household goods (underwear, cleansers, toothpastes, nursing bottles, food containers). Currently AgNPs are one of the most commonly used nanomaterials and the number of AgNP-containing products still increases. Widespread use of AgNPs exhibiting cytotoxic effect towards bacteria and viruses, rises question about potential adverse effects on living organisms indicating the need for research on the mechanisms of nanosilver toxicity. Moreover, the release of AgNPs from products was shown, pointing out the risk of environmental exposure. Therefore, it is very important to determine the toxic effects of nanosilver in animal experimental models. The aim of the proposed project is to examine neurotoxic effect of low doses of AgNPs in immature rats under conditions of prolonged exposure via oral route with particular emphasis on the role of glutamate receptors (NMDA). Previous studies showed that nanosilver preferentially accumulates in brain, damaging microvasculature. Moreover, in nanosilver-exposed animals increased production of reactive oxygen species and oxidative stress have been observed, leading finally to cell death. Our preliminary studies on cultured cerebellar granule cells revealed the involvement of glutamate receptors (NMDA) into AgNP-induced cell death. As it is known, overactivation of NMDAR is connected with oxidative stress and cells death. Thus, possibly overactivation of these receptors may be involved in mechanisms of AgNP-induced neurotoxicity *in vivo* as well. Glutamate NMDA receptors participate in a variety of pathological states of brain but they are also involved in many physiological functions, including learning and memory. Therefore, determination of possible adverse effects in immature organisms is of particular importance. The research proposed in the current project will emphasize on determination of expression and activity of NMDA receptors in immature rat brains, so as the function and expression of glutamate transporting systems and the possible effect of NMDAR overactivation which is neuronal death. The behavioral tests will be conducted which may expressed changes in NMDAR function. The contribution of NMDAR will be verified by administration of substance blocking the receptor's activity. In order to approach experimental design to the environmental conditions, the long-term effects of exposure will be determined. Studies will be conducted in two temporal variants - early (just after exposure) and late (90 days). General work plan assumes prolonged administration (21 days) of nanosilver in a dose of 0.2 mg/kg b.w. via oral route for immature (14 day-old) rats and estimation of its bioavailability by measuring silver concentration in blood and tissues. Further, early and late after exposure, behavioral tests will be performed. In brains of exposed animals expression of the receptor will be determined. In isolated fractions receptors' activity will be analysed by measuring biochemical markers of receptor's activation. Further expression and function of glutamate transporter systems will be analysed in glutamate uptake assay performed on isolated brain fractions. Disturbances in the proper function of transporters may lead to the excessive extracellular level of glutamate, overactivation of glutamate receptors and subsequent death of neurons. The latter will be determined in immunohistochemical studies. The results of proposed studies will extend the knowledge about neurotoxic mechanisms of nanosilver during development. Such research have not been conducted as far. Moreover, scientific problem presented in the project is valuable from the social and environmental points of view. Current knowledge concerning the influence of nanosilver on human is very limited, so it is very difficult to predict long-term effects of environmental exposure. Obtained results will extend our view on the nanotoxicology of AgNPs and may contribute to draw attention to the increasing risk of both environmental and health hazards by nanoparticles produced in nanotechnological processes and released from nanoproducts.