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Oxidative Stress Induced by Organo Sliver Clay Particles in Mice

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Abstract: Toxicity of organo sliver clay particles (OSCPs) is dependent upon chemical substance make up, atomic arrangement and particle size. With this research, the toxic effects of distinct measurement (OSCPs) 50, 100 as well as 200 nanometer with various concentration had been examined on liver enzyme activities as well as oxidant/antioxidant condition of mice treated every day with OSCPs for twenty eight consecutive days, One hundred mice had been arbitrarily split into 10 groups: G1 (control with no treatment), T1, T2, T3 injected intraperitoneally with exactly the same receiving of 0.25 mg/kg. body weight of OSCPs at 50 nm, 100 nm as well as 200 nm sizing diameter, T4, T5, T6 injected together with the exact same dose of 0.50 mg/kg.b.w at 50 nm, 100 nm and 200 nm size diameter and T7, T9 and T8 injected with the same dose of 1.0 mg/kg.b.w at 50 nm, 100 nm and 200 nm size diameter. Blood samples for sera formulations have been gathered up as well as saved at -20°C till additional evaluation. Outcomes from this particular analysis demonstrated that AST, ALT and ALP enzymes belonging in just about all medicated groups have risen around comparability together with the control and more considerably in excessive serving with lower measurement diameter of OSCPs handled. Negative impacts on oxidant/antioxidant condition had been noticed inside an impressive dose treated group (1.0 mg/kg.b.w) with fewer sizing diameter (50 nm), From outcome gotten, it had been apparent that lipid peroxidation marker; Malondialdehyde (MDA) amounts was more while superoxide dismutase (SOD), catalase (CAT), glutathione (GSH) and also glutathione peroxidase (GPX) quantities decreased significantly (p<0.001) within all of mice injected with OSCPs in comparability with the standard settings, but probably the most modification was related to increased serving with a reduced diameter sizing (1.0mg/kg.b.w from 50 nm). Based on the final results, it's recommended the impact of OSCPs on the tissue cells could cause body organ toxicity of mice.

Keywords: Organo sliver clay particles, liver function, oxidant/ antioxidant, hypotoxicity.

1. Introduction

Clay minerals are believed to be as all-natural content with particle dimensions < 2 µm Smectite, which classified as 2:1 phyllosilicate clays, use a crystal lattice product created by a single alumina octahedral sheet sandwiched between two silica tetrahedral sheets. An adversely recharged floor will provide an increase whenever the ion substitution or maybe the site vacancies in the tetrahedral and also octahedral sheets occur. The exchangeable cation in between the levels compensate the negative charge as well as might be quickly replaced by some other metallic cation, detailing the higher ion exchange capabilities of the minerals (70-120 meq/100)g. As a result of this particular crystalline plan, smectites are competent to increase as well as contract the while keeping the two crystallographic integrity. Water and cations molecules are contained by the interlayer between units. Montmorillonite is several of the members of the smectite family [1].

Clays are utilized in veterinary and human healthy formulations as all-natural materials as active substances or percipients. The interaction in between the medication and the excipients of its might hold off the drug discharge and also consequently, the absorption of its, by decreasing the drug amounts within the blood. This particular function is usually advantageous once the sluggish, controlled desorption on the medication has a good impact on the therapeutic action of its [2]. Along with the supplies, montmorillonites are already essentially the most thoroughly pre-owned minerals within this particular area of program. Montmorillonites display big certain surface area as well as colloid qualities and substantial cation exchange capability, which provide rise to the best possible adsorbents of inorganic and organic materials. Metal ion exchanged

montmorillonite dispersed within aqueous solution has proven to appeal to germs by fluoromicas, montmorillonites, and electrostatic forces [3]. Furthermore intercalated with organic and natural ingredients with antibacterial consequences including cetylpyridinium, cetyltrimethylammonium, norfloxacin tetracyclines happen to be additionally analyzed. The fluoromicas as well as organo montmorillonites exhibited superb antibacterial activities [4, 5].

Clay mineralsa are utilized within a multitude of manufacturing programs [6] due to two essential as well as contrasting properties: inertia and reactivity and stability as well as catalytic undertaking. The prevalent utilization of clay nutrients responds to the extensive distribution of theirs within the atmosphere, the simple and easy extraction as well as financial feasibility they show. Large volumes of the substances are utilized around purification tasks of clarification and bleaching of different devices for animal feed pellets, inside inorganic pesticides formula what about water filtration. They're additionally utilized as fat and also oils adsorbents, so when catalyst or maybe help catalysts to come down with a number of natural syntheses [6, 7]. The modification on the surface attributes is of value to lengthen the clay programs and has now thus received a lot of interest [7]. The hydrophobic change on the montmorillonite is able to boost the thermal, rheological, as well as physical qualities of resulting substances. Though the changes of clay minerals could be done by a number of techniques, the ion exchange of inorganic cations with organic and natural cations, typically with quaternary ammonium elements, enables replacing the area qualities from hydrophilic to hydrophobic. Thus, these gotten organoclays hence received have already been thoroughly examined for hydrophobic contaminants immobilization. This particular analysis

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concentrates on toxological influence of numerous complexes created between quaternary ammonium elements as well as silver with kaolinite, within its modified and natural type. The existing analysis was completed to take a look at the particle size impact of OSCPs about the hepatic enzyme activities as well as modification in oxidant/antioxidant condition of animals injected with those particles to make an effort to discuss as well as fully grasp potential threat and the toxicity of the diagnostic and therapeutic usage of relation with time of exposure.

2. Materials and Methods

Sliver modified clays (AgKaOF) was first prepared by the addition of 10 g powdered clays 500 mL AgNO3 (1000 ppm). The combination was stirred for sixteen hours at room tempreture then filtered to distinct the liquid and solid fractions. The fraction proportion was next dehydrated within an oven from 90°C immediately and this was employed for the preparing of AgKaOF was modified by CTAB (CTAB AgKaOF). About 2 g of dried out samples was put in with 200 mL of 2 mM CTAB strategies. This particular concentration was selected since it's somewhat greater compared to CMC (Critical Micelle Concentration) importance of CTAB (CMC CTAB: 1.0 mM). Following which, the combination was stirred for 16 hours, filtered and lastly the fraction proportion was dehydrated within an oven immediately.

Characterization Techniques

Ag-clays are recognized by X-Ray Diffraction (XRD), along with XRD patterns have been captured. Fourier Transform-Infrared (FT-IR) spectrometer designed with OMNIC program was used-to identify the existence of CTAB on the well prepared samples. The existence of silver in sliver modified clays was recognized by using Energy Dispersive X-Ray (EDX) analyzer outfitted along with Field Emission Scanning Electron Microscopy (FESEM). The sizing division of synthesized OSCPs during fifty nm, 100nm along with 200 nm had been examined by microscope AFM.

Lab animals: Female and male albino mice together with the mean weight of (28.3 ± 1.2) g was utilized, the mice had been prepared by using AL Nahrain University; they had been stored in the house of animals with fixed conditions (twelve hour darkness, twelve hour weightlessness, heat of 25°C as well as appropriate humidity). Mice were given by food that is standard with no limitation.

Animals' treatment: The mice had been split to ten groups (ten mice in each group, five male and also the various other female randomly). Every group was stored in different cage. G1 set as control group only gotten food and water. T1, T2 as well as T3 class have gotten OSCPs with serving of 0.25

mg/kg intraperitoneally within twenty eight successive times at 50, 100 as well as 200 nm. T4, T5 and T6 class gotten OSCPs with serving of 0.5 mg/kg at 50, 100 as well as 200 nm, and lastly T7, T8 and T9 class received OSCPs with dose of 1.0 mg/kg during 50, 100 and 200 nm. During the end of experimental time throughout the mice had been anaesthetized by chloroform as well as blood sampling was carried out from heart of mice. Blood was gathered up in laboratory tubes carefully and kept for fifteen minutes in the laboratory temperature. After coagulating the blood, serum was divided by centrifuge at 1500xg. For analyzing evaluating liver perform within several groups, serum transaminases such as AST, ALT, and also ALP had been calculated colorimetrically utilizing immediate spinreackt systems (Spain). Serum lipid peroxidation marker, malondialdehyde (MDA) was assessed by precipitating lipoproteins with trichloroacetic acid as well as boiling with thiobarbituric acid getting orange colour as per the technique [8]. Superoxide dismutase (SOD) was assessed by making use of RANSOD package as well as Glutathione peroxidase (GPx) was measured by making use of RANSEL package (Randox Laboratories Ltd. Crumlin, United Kingdom) This technique is grounded on Valentine and Paglia [9]. Catalase activity was based on Goth technique [10]. Decreased glutathione by technique for [11].

Statistical analysis: The outcome was conveyed as mean \pm SD. Differences between implies have been examined utilizing one way ANOVA, after which the mean had been in contrast to Duncan. P- values of 0.05 reduced had been considered as remaining statistically considerable. Information have been examined making use of variation sixteen of SPSS application (SPSS Inc., IL, Chicago, USA).

3. Results

Serum AST amounts in male mice exhibited statistically elevated (P<0.01) when compared with control, which expansion based on serving of OSCPs administrated intraperiotally. Via figure (1) excessive degree of serum AST (70 ± 11) IU/L was noticed around cluster of animals injected with 1.0 mg/kg.b.w during fifty nm sizing diameter of molecules in contrast to (62 \pm 6) IU/L of mice treated with 0.5 mg/kg.b.w as well as (51 ± 5) IU/L of mice injected with 0.25 mg/.kg.b.w of OSCPs, while influence mice with no therapy had amounts of AST: (42 ± 4) IU/L. Distinct details have been seen within female mice injected with exactly the same serving of sliver clay particles though it wasn't significant (P>0.06) in contrast to male mice groups. On the othe side serum amounts of ALP and ALT had been enhanced as seen in figure (2, 3) but the levels of ALP elevated extremely both only in female or male groups in contrast to command mice. Most therapy groupss suggest statistically improved tasks to the control of theirs (C1).

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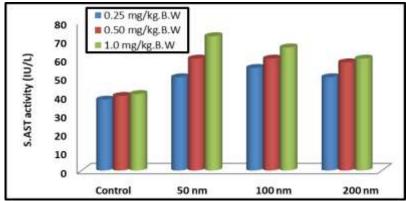


Figure 1: Mean sera levels of aspartate transferase in male mice treated with, three distinct doses (0.25 mg/kg, 0.50 mg/kg as well as 1.0 mg/kg) of OSCPs at 3 distinct measurements (50, 100 along with 200 nm) after 28 days.

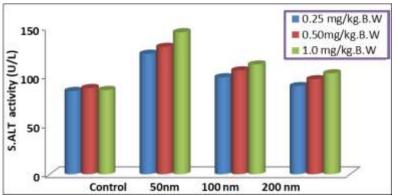


Figure 2: Mean sera levels of alanine transferase in male mice treated with, three distinct doses (0.25 mg/kg, 0.50 mg/kg and 1.0 mg/kg) of OSCPs at 3 distinct measurements (50, 100 along with 200 nm) after 28 days

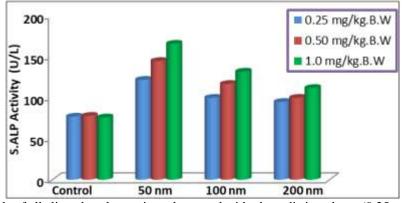


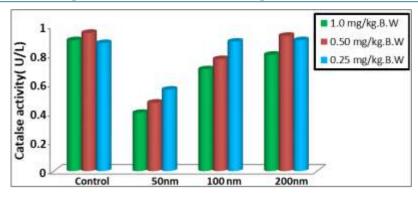
Figure 3: Mean sera levels of alkaline phosphatase in male treated with, three distinct doses (0.25 mg/kg, 0.50 mg/kg and 1.0 mg/kg) of OSCPs at 3 distinct measurements (50, 100 along with 200 nm) after 28 days

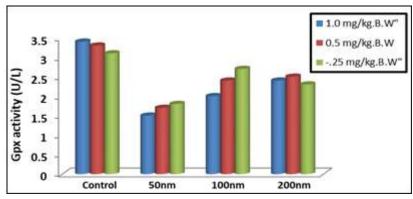
Results of oxidant /antioxidant condition displayed a big modification within most biochemical parameter assessed. MDA was significantly improved when mice injected by using an impressive serving (1.0 mg/kg.b.w) of OSCPs during 50nm dimensions (1.0 > 0.5 > 0.25 mg.kg.b.w). So we realized the influence was based upon size and dose as revealed in figure (4). On another hands decreased glutathione was significantly reduced and this also lessening

seen naturally during 1.0 mg/kg.b.w as well as during fifty nm scale of OSCPs as shown in figure (5). Data from figures (6, 7 and 8) noticed a very substantial decline in most enzymatic antioxidant GPx, SOD as well as Cat once the mice taken care of extremely serving of OSCPs 1.0 mg/kg.b.w with size that is small (50 nm). Therefore the alteration within the tasks of the enzymatic anti-oxidants relies on size and dose.

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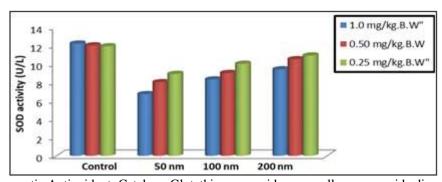


Figure 4: Serum enzymatic Antioxidant: Catalase, Glutathione peroxidase as well as superoxide dismutase in male treated with, three distinct doses (0.25 mg/kg, 0.50 mg/kg and 1.0 mg/kg) of OSCPs at 3 distinct measurements (50, 100 along with 200 nm) after 28 days

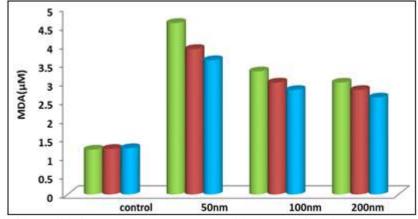


Figure 5: Mean Sera lipid peroxidation marker (MDA) treated with, three distinct doses (0.25 mg/kg, 0.50 mg/kg and 1.0 mg/kg) of OSCPs at 3 distinct measurements (50, 100 along with 200 nm) after 28 days

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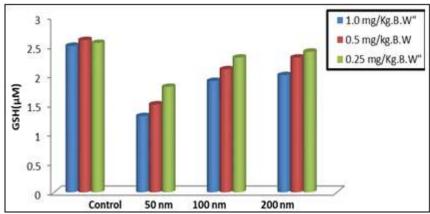


Figure 6: Mean Sera reduced glutathione treated with, three distinct doses (0.25 mg/kg, 0.50 mg/kg and 1.0 mg/kg) of OSCPs at 3 distinct measurements (50, 100 along with 200 nm) after 28 days

4. Discussion

There's absolutely no opinion on the cytotoxicity of Nanosilver, nonetheless, the majority of publications do clearly show decreased cellular viability following exposure. Extra toxological consequences observed in vitro scientific studies are glutathione depletion, mitochondrial deviations or maybe damage and destruction to cellular membranes. Nanosilver goes into the entire body with the epidermis, gastrointestinal tract and respiratory system. The most crucial method to get hold of it [12], particularly within the gastrointestinal tract is colloidal form [12]. With this research much more increased AST amounts in male mice, ended up being noticed within greater serving of 1.0 mg/kg with 50 nm sizing diameter set in comparability to various other doses, 0.25 mg/kg as well as 0.50 mg/kg.b.w Absorbed OSCPs bind to plasma protein which enable it to go into the cells, they're sent out in organs including liver, lung, brain, lymph nodes, heart, kidney, tummy along with testicles based on the path of administration [13]. Assimilated Nanosilver from gastrointestinal area get into liver because of the portal vein as well as may have effect on the liver because the liver can serve as the first checkpoint for all the things assimilated just before getting systemic. Liver can definitely get rid of ingredients coming from the bloodstream and also change them to chemical substance types which may effortlessly be excreted. It's a rational assumption which ingested OSCPs may have impact over the liver.

Within the existing study, biological negative effects of OSCPs were examined during different size and different doses on serum AST, ALT, ALP in female and male mice. Hepatic deterioration caused by intraperitoneal injection of OSCPs in mice, has perhaps brought about serious irritation of oxidant process inside the cells. The more compact the diameter on the OSCPs (50 nm) is, the more often the influence of it's to cells and the molecular effects of it's about the intracellular systems increases. Earlier scientific studies, demonstrated that reactive oxygen species (ROS) caused by nanoparticles are able to bring about damage of red blood cells [14, 15] and also the information exhibited various consequences based on switches within the diameter of nanoparticles, the distribution of theirs in the route and body tissues of consumption as well as time period of exposure. Actually ROS through the of OSCPs have assaulted hepatocytes and also available ALT kept in them putting in into the blood serum; whereas; the immune effect of mice to an outside element continues to be the expansion on the variety of white blood cells for phagocytosis of Nanosilver particles [16]. Similar details in this research had been noticed in female mice for AST, ALT and ALP in most therapy groups elevated in comparison with control.

Thinking about the benefits of role of hepatocytes in detoxification, any kind of modifications made in their number and structure is able to bring about big physiological modifications for man body. On the other side, great usage of various Nanosilver OSCPs within the entire earth calls for much more correct as well as extensive scientific studies on the consequences of the particles on blood cells. The utilization of lab mice as animal model, along with different therapy techniques as well as OSCPs with various mixtures and diameters presents brand new horizons for additional study to take a look at uses of nanotechnology in physiology [17]. Also this research observed a rise inside ALP amounts in male mice G1 and G2 groups in comparability to control which may be justified with inflammatory progression as well as damage of hepatocytes [18]. In accordance with research by change that demonstrated that repeated dental doses of nanosilver for twenty eight days did cause liver toxicity, as found by increases in serum activity of ALP [19]. Malondialdehyde MDA amounts had been much higher within the research group than control figure (4). The improved MDA amount, as assessed by Thiobarbituric acid research substance (TBARS) technique, inside mice injected with OSCPs is a great sign of oxidative harm. Additionally, MDA, something of lipid peroxidation, is produced within extra quantities determined by small size and oral dose. As a consequence of constant day serving for twenty eight days, the animals could be put through per oxidative cells injuries by the OSCPs overload. These findings could help support the concept that OSCPs injected in animal's results in an enhanced model of reactive oxygen species as well as oxidative stress.

Information found that all mice injected with OSCPs are afflicted by lower amounts of superoxide dismutase (SOD), catalase (CAT),glutathione peroxidase (GPX) and also glutathione reductase, when as opposed with this decrease and control animals was significantly clear (P<0.001). Superoxide dismutase will be the protein co factor

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with copper, manganese, zinc, nickel or iron. Inside mice, it is present in 3 various forms such as SOD1 present in cytoplasm, SOD2 contained in cytoplasm, as well as SOD3 is extracellular. Superoxide stands out as the primary reactive oxygen species, that behave with nitric oxide radical and also forms peroxynitrite; therefore, leading to cellular damage and oxidative stress. SOD is definitely the crucial antioxidant which reduces the development of oxidative stress and ROS; as a result, defending the cells at harm. The erythrocyte by becoming harmed while in oxidative stress is protected by Erythrocyte SOD [20]. Grass activity of mice injected with OSCPs is reduced, leading to pronounced inhibition within the enzymatic blood antioxidant.

Catalase (CAT), commonly distributed in whole cells, occurs in very high quantities within erythrocytes. It's an intracellular enzyme comprised of four polypeptide chains with four porphyrin heme organizations. Catalase is liable for cleansing of hydrogen peroxide within the cells [21]. For existing analysis, the enzymatic antioxidants CAT had been drastically reduced in mice injected with extremely serving (1.0 mg/kg.b.w) at little measurement diameter 50 nm. (44.88 \pm 19.87) as in contrast to nourishing untreated mice (120.29 \pm 25.2). Reduction during the activity of CAT might be because of boost within the lipid peroxidation merchandise malonyldialdyhyde that may create crosslink's treatment inactivating a number of membrane bound enzymes [22].

Glutathione peroxidase (GPX) belong to cluster of antioxidant selenoenzymes which protects the cellular injury by catalyzing the decrease of lipid hydro peroxides. The existence of glutathione is required by this action. Glutathione peroxidase amounts within the body happen to be in good relation together with the glutathione, and that is the most crucial antioxidant that is within the cytoplasm on the cells [23]. The existing analysis shown considerable decrease in GPX found in mice injected every day higher serving of OSCPs at reduced measurement 50 nm, as in contrast to untreated mice. A reduced amount of GPX is a result of inactivation by the improved great oxide anion generation resulting in a rise within oxidative stress [24]. Reduced degree of GPX appears to originate from the enzyme inhibition or even decreased actions because of substantial creation of hydrogen peroxide. This particular analysis confirmed drastically reduced amounts of all of the anti-oxidants GSH, SOD and GPX in mice intake OSCPs compared with the matched untreated controls.

A deficiency in decreased GSH quantities was reported by the existing analysis. GSH, that injected being a serious intracellular lowering representative, and that is extremely vulnerable to oxidative stress and possesses many crucial features like as: safeguard from oxidative stress, regulation of gene expression, induction of apoptosis activation as well as proliferation of T lymphocytes. To conclude, the oxidative stress of mice injected with OSCPS is primarily brought on by peroxidative injuries. Creation of free radical groups by OSCPS consumption, modification in serum antioxidant enzymes condition engages in a crucial role in the cytotoxicity of the OSCPs injection. Impairment of antioxidant condition is related with heightened plasma amounts of malondialdehyde. These outcomes propose that

the noticed expansion of impulsive alternation within the liver feature might originate from oxidative tension was induced by OSCPs. Entirely, the outcomes propose that OSCPs in higher doses are able to inducing oxidative tension that is responsible for hepatotoxicity within experimental animals. Additionally, impulsive alternation within the oxidative strain biomarkers signifies toxicity of OSCPs accumulation. This information proposes that OSCPs is able to induce oxidative harm by way of a ROS mediated procedure. Nevertheless, it continues to be to become examined if OSCPs cause free radical groups indirectly or directly through depletion of antioxidant safeguard systems based serving e.g. a result of interactions with antioxidant methods [25, 26]. The latest analysis have found that little serving of OSCPs tend to be more useful antioxidant than big OSCPs. Within the existing analysis OSCPs sizing is diverse with small doses, large doses with lesser measurement tend to be more toxic.

5. Conclusion

The present study obviously exhibited that, intraperitoneal administration of OSCPs to female and male mice causes alteration in liver enzymes and alteration in oxidant/antioxidant status.

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