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Dermal Safety of Virgin and Used Engine Oil Enriched with Copper Nano Particles in Rabbits

[1]Mohaddeseh Abouhosseini, [1]Goudarz Sadeghi Hashjin, [2]Javad Malakootikhah,
[3]Ahmad Roshani, [1]Ebrahim Shahroziyan and [1]Bahar Badri

[1]Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran
[2]Faculty of New Science and Technologies, University of Tehran, Tehran, Iran
[3]Industrial and Environmental Protection Division,
Research Institute of Petroleum Industry (RRIPI), Tehran, Iran

Abstract: Acute dermal toxicity of virgin and used conventional engine oil (vCEO and uCEO) and nano-enriched engine oil (vNEO and uNEO) was evaluated in rabbits according to the OECD 404-2004 guideline. Rabbits were exposed dermally to 0.5 mL of either of the products using dermal patches on the dorsal area for 3 min - 4 h. The area was monitored up to 14 days after termination of the application. The results determined that none of the products led to a harmful effect during 3 min - 4 h of application or 1 h - 14 days after ceasing the application.

Key words: Engine oil · Nano particles · Rabbits

INTRODUCTION

Conventional products are gradually replaced by new industrial products combined with nano particles. Recent studies indicated the toxicity of nano materials on aquatic animals and rodents, however, many of their hidden effects are still unknown. Therefore, more investigations are required to distinguish the influences of nano products on human health and environment [1].

Currently, limited understanding exists about the adverse effects of nano materials in nature and wildlife as well [2]. Huge amount of data is needed for systematic studies of nano materials' toxicity in different species in water and soil environments [1] Much of our information about toxicity of nano materials is on aquatic organisms; thus, there is a need for further studies on toxicity of nano-materials on land animals as well. The skin, the largest organ and the cover of the body is highly exposed to the surrounding environment and has been the subject of enormous bulk of studies over the impact of chemicals on the body. Due to the increased inventions and applications of nano particles, researchers have started to study their effects on the skin. Because of the their potential toxicity as well as little information available in this field, their potential hazardous effects are required to be identified.

Copper nano particle is one of the nano metals, which is commercially manufactured for application in various industries. For instance, due to its high conductivity, low cost and recycling properties, copper nano particle is used in the anode pole materials of the lithium batteries [3]. Moreover, as a bioactive cover, it is capable of inhibiting microorganisms such as *Sacromaysis*, *E. coli*, *S. aureus* and *Listeria* [4]. One of the other characteristics of copper nano-particles is their plasticizing feature when mixed with used oils in the oil industry [5]. In general, it can be said that nano particles have restorative properties in abrasion surfaces, which can cause the friction to be reduced. Nano copper particles are widely used as additives into engine oil to reduce friction and improve lubrication. However, the difference between the effects of virgin and used conventional engine oil (vCEO and uCEO) and the virgin and used nano-enriched engine oil (vNEO and uNEO) on the dermal toxicity is not known.

Corresponding Author: Goudarz Sadeghi-Hashjin, Faculty of Veterinary Medicine,
University of Tehran, P.O. Box: 6453, Tehran 14155, Iran.
Tel: +98-2161117190, Fax: +98-21-66933222, E-mail: gsadeghi@ut.ac.ir.

The aim of the present study was to compare the effects of 4 types and conditions of the engine oil on the albino rabbits. The experiments were performed in accordance with the recommended protocols of OECD [6].

MATERIALS AND METHODS

Test Substances: The engine oil used in this study was a commercial brand manufactured in Iran and enriched with copper nanoparticles by researchers working at Research Institute of Petroleum Industry (RRIPI), Tehran, Iran. The product was tested in 4 different conditions: virgin conventional engine oil (vCEO), used conventional engine oil (uCEO), virgin nano-enriched engine oil (vNEO) and used nano-enriched engine oil (uNEO). The dimensions of the particles were between 6-20 nm, with an average of 12 nm. The type of engine oil was SAE20W-50W with oil basis and a density of 0.898. Used products had a performance of 8000 km.

Animals: Twelve healthy adult male and female albino rabbits were housed individually in stainless steel cages. Temperature was kept at 20±2°C and the humidity of the room at 50-60%. A 12 h light/dark cycle was provided for the animals. Commercial chow was purchased from Pars Dan Company (Tehran, Iran) and the rabbits had free access to the feed and tap water. They were kept in the animal house for 2 weeks for adaptation and acclimation before the experiments.

Experimental Protocol: Acute dermal toxicity (irritation / corrosion) test was accomplished according to the guidelines of OECD 404-2004. Rabbits were divided into 4 groups of 3 each and were exposed dermally to a single dose of 0.5 mL vCEO, uCEO, vNEO, or uNEO for 3 min, 1 h and 4 h on different points of their clipped, dorsal body areas. The dimensions of the exposed points were 2 × 3 cm. Nothing was applied on control points of the body. At the end of exposure, the area was rinsed thoroughly with water (Figure 1).

Fig. 1: Methods of fixing Elizabeth Collar around the head (left) and engine-oil containing path on the dorsal side (right) of the rabbit

Clinical Observations: At the end of exposures (3 min, 1 h and 4 h) as well as 1 h, 24 h, 48 h, 72 h and 14 days after termination of the exposures, the areas were examined macroscopically for the presence or absence of erythema and edema.Dermal reactions were graded according to the OECD 404-2004 instructions.

RESULTS AND DISCUSSIONS

No mortality, abnormal behaviour and obvious differences in body weight gain were found within the 2 weeks in any experimental group. No signs of edema, erythema or escar were observed in any 4 group of rabbits treated with either vCEO, uCEO, vNEO, or uNEO during the presence of the patches on the skin at the time points of 3 min, 1 h and 4 h. After removal of the patches, signs of edema, erythema or escar did not appear as monitored at the time points of 1 h, 24 h, 48 h, 72 h and 14 days (Figure 2).

The present study clearly showed that, like conventional engine oil, the product with copper nano particles did not exert any harmful effect on the skin of the rabbit based on the standard protocol of OECD 404-2004. This fact applied not only to the virgin but also to the used engine oil of both types.

Most of the studies performed on the effects of nano particles on the skin are related to nano particles used in cosmetics [7]. Titanium oxide and liposome are two kinds of nano particle products which have been widely studied because of their widespread usage to increase transparency in cosmetic products and to reduce

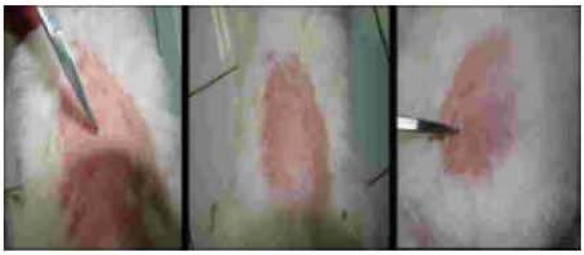

Fig. 2: Three samples of rabbit skin exposed to different types of engine oil. No erythema or edema were observed during and/or following exposure with different protocols explained in the materials and methods section

absorption of ultraviolet light [8]. It was shown that the time of exposure and the size of nano products may influence the toxicity induced by these substances in the rabbit skin. In our study, however, there was no choice over the particle size although different time points were used for the exposure and evaluation purposes [9].

Our study does not claim that the engine oil of any type and condition is completely safe from every point of view. Instead, it was shown long time ago that the used engine oil may lead to a kind of carcinogenic outcome in the skin of the mouse [10].

Carcinogenic and mutagenic effects are possibly caused by formation of DNA adducts in the skin tissue as shown in the mice dermally exposed to used gasoline engine oil [11]. It is interesting that dermal exposure with engine oil-containing cleaners may be even harmful for other organs, including the lung, as they may diffuse through the skin into the general circulation [12]. The harmful effects of engine oil are not limited to the few experimental models reported so far. A recent case report showed that it may be involved in the pathogenesis of extra-mammary Paget's disease (EMPD), a rare intraepithelial adeno-carcinoma, in humans as well [13]. We have only focussed on a standard method for the irritation induced by chemicals, which is acceptable by legal authorities in many countries. In this context, it can be claimed that there is no difference between virgin and used product of either conventional or nano-enriched engine oil.

In conclusion, the engine oil enriched with copper nano particles seems to be as safe as the conventional product. This is a value added aspect to the nano product, which is believed to possess technical advantages over the conventional products.

Conflicts of Interest: The authors declare that there are no conflicts of interest in regard to the data published in this research work.

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