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Changes in the behavior of laboratory rats after exposed exhaust gases

The research of animal behavior when exposed to gasoline fumes using test systems: «Open field» and «The elevated plus maze». Descriptions characteristic for acute inhalation toxicity of gasoline, which is reflected in the subsequent behavioral responses in tests «Open field» and «The elevated plus maze». Features of the test systems and their differences. Use of vitamin D3 as correction upon exposure to polycyclic aromatic hydrocarbons.

Key words: gasoline fumes, the acute inhalation toxicity of petrol vapor, the test system is «Open field», «Elevated plus maze».

Introduction

The primary route of human exposure to benzene is inhalation of ambient air. Benzene is present in the atmosphere both from natural sources, which include forest fires and oil seeps, and from industrial sources, which include automobile exhaust, industrial emissions, and fuel evaporation from gasoline filling stations [1].

Human exposure to benzene has been associated with a range of acute and long-term adverse health effects and diseases, including cancer and aplastic anaemia. Benzene is highly volatile, and exposure occurs mostly through inhalation [2].

Benzene has been widely used as a multipurpose organic solvent. This use is now discouraged due to its high toxicity, including carcinogenicity. Present uses include use as a raw material in the synthesis of styrene, phenol, cyclohexane, aniline, and alkyl benzenes in the manufacture of various plastics, resins, and detergents. Syntheses of many pesticides and pharmaceuticals also involve benzene as a chemical intermediate. Impairment of immune function and/or various anemias may result from the hematotoxicity. The hematologic lesions in the bone marrow can lead to peripheral lymphocytopenia and/or pancytopenia following chronic exposure [3].

Releases of benzene to the environment are largely to air, which is due to its volatile nature.

Major sources of releases to air include gasoline vapor, auto exhaust, and manufacturing industries. Tobacco smoke can be a significant source of exposure (a dose of 1 milligram (mg) for each pack smoked). An estimated 99 percent of total human exposure to benzene is by inhalation [4].

Workers involved in the transport of crude oil and gasoline and in the dispensing of gasoline at service stations, as well as street workers, taxi drivers and others employed at workplaces with exposure to exhaust gases from motor vehicles also experience exposure to benzene [5].

Concentration ranging from 0.02 ppb (0.06 µg/m³) in a rural area, to 112 ppb (356 µg/m³) in an urban area. Exposure to benzene is highest in areas of heavy motor-vehicle traffic and around gasoline filling-stations. Based on an average benzene concentration of 12.5 ppb (40 µg/m³) in the air and an exposure of 1 hour per day, the daily intake of benzene from driving or riding in a motor vehicle is estimated to be 40 µg. Exposure is higher for people who spend significant time in motor vehicles in areas of congested traffic [5].

Materials and methods

Selection of experimental animals and standard procedures. The experiments were conducted on laboratory outbred rats. Adult males weighing 250–320 grams. During experiments, animals were maintained under similar conditions as before the experiment. Water and food were given without limitation. Temperature $T = 21\text{--}22^{\circ}\text{C}$ relative humidity, light regime day and night 09/15, in accordance with the mode of operation of the vivarium staff (SRI Transfusiology Astana, Kazakhstan), while the light started at 8.00 am. Animals were housed 5 animals in each cage in accordance with СанПиН РК [6].

The experiment used 30 laboratory rats. Which were divided into three groups of 10 in each group. The first group of the control group (animals pre-up experiments have not been introduced — or drugs), the second group received a dose of gasoline inhalation (LC50 values (10,000 ppm / 7d) 7 exposure of 4 hours. The third group received gasoline dose inhalation (LC50 (10,000 ppm / 7d) 7 exhibit 4 hours and received vitamin D3 dose according to the dosage for humans 500 IU as a correction. One drop of vitamin D3 diluted in one ml of water and was administered orally by catheter after air seeding gasoline vapors.

The elevated plus-maze test

The elevated plus maze consists of two closed (the analog hole) and two open (potentially dangerous) sleeves. The test measures the number of entries and time spent in the open and closed arms; More alarming mouse prefer closed arms. During the experiment, the number of visits recorded both open and closed arms. The residence time in the arms, the act of grooming.

Open Field test

«Open Field» to estimate the total (locomotor) research activity (Hall, 1934). The apparatus consisted of the arena, divided into equal squares. The animal facility was placed, was recorded and number of squares crossed thus traveled for some time (2 minutes), the distance.

During the experiment, we take into account such factors as: locomotion, grooming, stand with support, stand without support, Immobility, movement in place, defecation, urination. Behavioral tests were conducted at a set time (9 am), and each test was allocated a single day (Table 1).

Results

Table 1

Number of acts

Indicators	Control group	Second group	Third group
Locomotion	8,6±0,61	11,4±0,65	8,6±1,1
Sniffing	4,4±1,15	2,6±0,82	6,6±0,98
Stand with support	5,8±0,60	7,4±0,52	9,4±1,29
Stand without support	1,2±0,51	0,4±0,17	0,8±0,26
Movement in place	1,2±0,26	1,8±0,41	2,6±0,47
Immobility	1,2±0,51	0,6±0,17	0,6±0,17
Grooming	0,4±0,17	0,3±0,14	1,2±0,14
Defecation	0,4±0,17	0,4±0,17	1,2±0,26
Urination	-	-	0,8±0,26

Note. $P < (0,01)^{**}$; $(P < 0,001)^{***}$; * mark in this article correspond to the following parameters.

Compared with the control group in the second group by the number of the done an act of locomotion observed increasing rates of 27.9 %, in the third group on this indicator as compared to the control group experienced higher performance by 6.9 %.

By sniffing act compared to the control group in the second group act sniffing observed reduction rates of 40.9 % in the third group there is the act of increasing by 50 % compared with the control group (Figure 1).

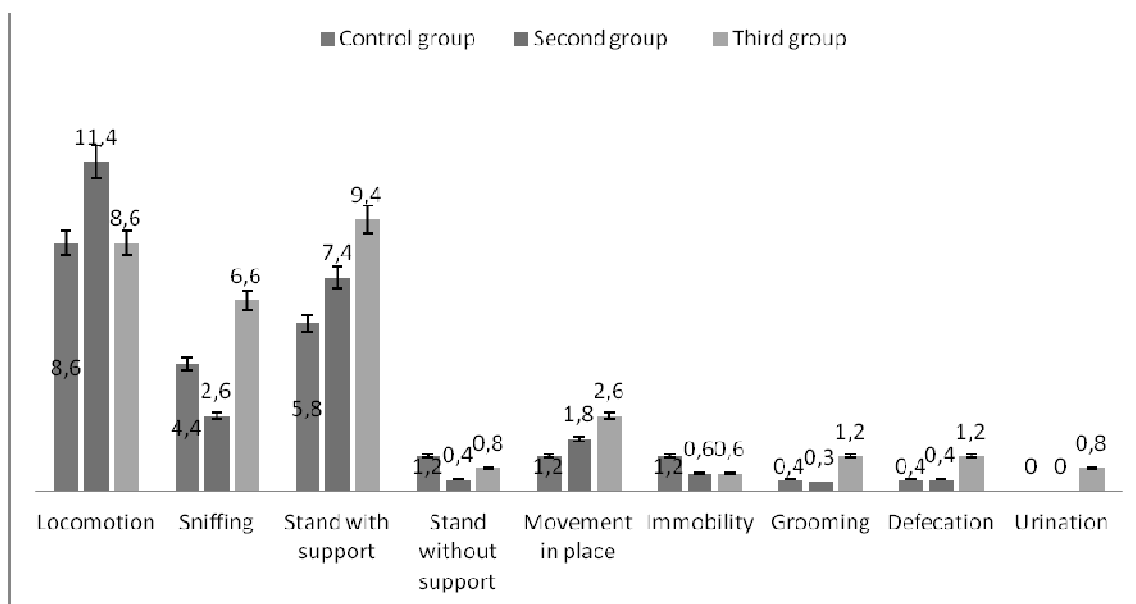


Figure 1. Indicators number of acts carried out in the test «open field» by influence of traffic fume

In quantitative terms the act of «stand with support» in comparison with the control group there was an increase of 27.5 % on the figures, there has been a slight decrease in performance of 6.8 % in the third group of the present Act.

According to the act stand in comparison with the control group in the second group there is a decrease in the rates of 66.7 %, in the third group of the present act contrary experienced higher performance by 16.6 %.

Movement by the act in place compared to the control group in the second group, there is an increase of 50 % on the performance, as in the third group on the indicator, a sharp increase in performance of 116 % compared with the control group.

When comparing figures act immobility in the second and third groups observed decline in the same 50 % compared with the control group.

Quantitative indicator grooming act in the second group compared with the control group decreased by 25 %, a sharp increase of 200 % in the third group, as compared with the control group on the indicator. That is a sharp prevalence of this indicator in the third group.

Indicators defecation, compared to the control group in the second group-or any change was not observed, but in the third group, as compared with the control group there is a sharp increase in the rate by 200 %. According to this act as a sharp prevalence of this indicator compared to the control group.

According to the act of urination in the control group and the second major acts of urination is not observed, the act of urination was observed only in the third group (Table 2).

Table 2

Performance test the open field: duration of intercourse

Indicators	Control group	Second group	Third group
Locomotion	39,6±4,54	46,6±4,60	41,4±4,72
Sniffing	27,6±7,80 *	15,4±2,22	25,8±3,81
Stand with support	26,8±2,45	44,6±2,26	35,8±4,34
Stand without support	6,2±1,90	3,0±2,12	3,8±2,02
Movement in place	8,0±1,59	14,6±3,56	7,6±1,52
Immobility	6,2±1,92 **	1,8±0,64 *	3,6±1,71
Grooming	0,6±0,28	0,2±0,14	1,2±0,41
Defecation	0,8±0,34	1,5±0,82	1,4±0,65
Urination	-	-	0,2±0,14

Note. P<(0,01) **; (P<0,001) *; * mark in this article correspond to the following parameters.

Compared with the control group in the second group for the duration of the done an act of locomotion observed increasing rates of 17.6 %, in the third group on this indicator as compared to the control group experienced higher performance by 4.5 %.

For the duration of the act of sniffing as compared to the control group in the second group observed decrease in the act of sniffing indices by 55.7 % ($P < 0,001$) in the third group also observed a decrease of the act by 6.5 % compared with the control group.

For the duration of the act of «stand with support» in comparison with the control group there was an increase in the indicators 66,4,5 %, in the third group on the duration of the act experienced higher performance by 33.5 % (Figure 2).

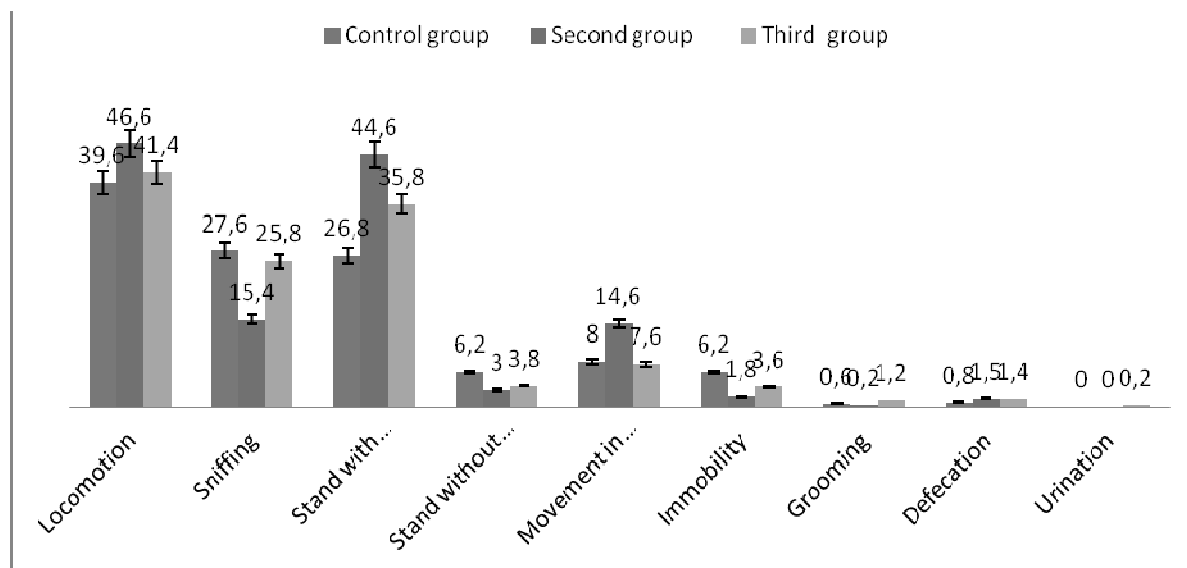


Figure 2. Totally made acts in the test «open field» by influence of traffic fume

According to the act stand in comparison with the control group in the second group there is a decrease in the rates of 51.6 %, in the third group for the duration of the act also observed reduction in rates by 38.7 %. For the duration of motion act on the spot as compared to the control group in the second group observed increase performance by 82.5 % in the third group of the present figure there is a slight decrease in performance by 5 % compared with the control group.

If we compare the performance duration of immobility of the act, in the second and third groups observed the same decline in 70.9 % ($P < 0,01$) and 58.1 % ($P < 0,001$), respectively, compared with the control group.

Act grooming duration in the second group compared with the control group decreased by 33.4 % in the third group, as compared with the control group for the duration of this figure there is a sharp increase of 200 %. That is a sharp prevalence of this indicator in the third group.

Indicators defecation, compared to the control group in the second group and the third group compared with the control group there is a sharp increase in the rate at 87.5 % and 75 % respectively. According to this act as a sharp prevalence of this indicator compared to the control group.

According to the act of urination in the control group and the second major acts of urination is not observed, the act of urination was observed only in the third group (Table 3).

Table 3

Number of indicators elevated plus maze

№	Number of visits		The residence time		Grooming		Fall over from sleeve
	Open sleeve	Closed sleeve	Open sleeve	Closed sleeve	Open sleeve	Closed sleeve	The open sleeve only
1	2	3	4	5	6	7	8
Control	3,4±2,7	3,2±0,48	80,4±4,36	38,8±4,06	0,6±0,16	2,4±0,47	16,2±1,05

1	2	3	4	5	6	7	8
2 Group	3,6±0,17	2,1±0,21*	87,01±6,12	17,4±1,6	-	0,8±0,41 ***	13,4±1,19 **
3 Group	3,2±0,26	2,6±0,26 **	109,4±1,21	10,6±1,27	-	0,6±0,17 **	15,6±0,61

Note. $P < (0,05)$ ***; $P < (0,01)$ **; $P < (0,001)$ *; * mark in this article correspond to the following parameters.

Compared with the control group in the second group in the number of visiting open sleeves elevated plus maze was a slight increase in the rate of 5.8 %, in the third group on this indicator, as opposed to the above data is observed decline of 5.8 %.

By the number of visiting a sleeve closed relative to the control group observed decline in the second group, 34.5 % in the third group, and 12.5 %, respectively.

As the total time spent in the open arm as compared to the control group in the second group there is a slight performance increase by 8.2 % in the third group of the present figure there is a sharp increase in the rate of 36 % compared with the control group.

Residence time in the closed arm of the elevated plus maze compared with the control group in the second group was observed at a low reading of 55.2 % ($P < 0,001$) and in the third group, as compared with the control group there was a sharp decline on the 72.6 % ($P < 0,01$).

Grooming in the open arm was observed only in the control group, the other groups in the open arm act grooming observed laboratory animals did not commit.

Grooming act in the elevated plus maze compared with the control group in a closed sleeve in the second group indices fell by 33 % ($P < 0,05$), in the third group in comparison with the control group also decreased by 25 % ($P < 0,01$). In terms of die and fall with open sleeves elevated plus maze compared with the control group was observed decline by 17.2 % ($P < 0,01$) and in the third group on the act as there is a slight decline of 3.7 %.

Conclusion

In conclusion I would like to note that the tests «open field» and elevated plus maze on the results of the test may at first glance show as discrepancy between the results and the logical explanation of the experiments. According to the data of the experiment in the test «open field» to take an active reduction in acts of locomotion and sniffing, as the duration and by the multiplicity of execution of the act in the second group. In this regard, it can be assumed on the effect on gasoline vapor exploratory activity that is observed in the third group of animals. But in the acts stand with support is increasing research activity in test animals the second and third groups, respectively. Such differentiation results from the opinions of other authors (9), can be attributed to a complex adaptive response associated with the mismatch in emotional terms. What can explain the decline in indicators such as rack, movement in place, grooming, defecation and urination. The elevated plus maze test performance increased significantly and stay open sleeves visits compared with the control group. There was a significant decline in stay in the closed sleeve in test groups, indicating that the experimental stress adaptation. Grooming act in this test was observed in the control group of rats, and was not observed in the subjects of the second and third groups, indicating that the alarm state test, where the animals were subjected to inhalation seed.

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Р.Р. Бейсенова, М.Р. Хантурин, У. Февзи, Р.С. Мустафа

Жағармай жартылай жану өнімімен улану жағдайындағы лабораториялық егеуқұйрықтардың мінез-құлықтарының өзгерістері

Мақалада лабораториялық жануарлардың мінез-құлықтарының бензин буының әсерінен пайда болатын өзгерістерді «Ашық алаң» және «Көтеріңкі крест тәрізді лабиринт» тестілерімен зерттеу нәтижелері берілген. Аталған тестілерде көрініс тапқан шұғыл ингаляциялық улану жағдайындағы сипаттамалар, тестілеу жүйесінің ерекшеліктері мен айырмашылықтары көрсетілген. Полициклді ароматты көмірсутектердің әсерін түзетуге арналған препарат есебінде витамин D3 қолданған.

Р.Р. Бейсенова, М.Р. Хантурин, У. Февзи, Р.С. Мустафа

Изменения в поведенческих реакциях лабораторных крыс под воздействием выхлопных газов

В статье приведено исследование поведения животных под воздействием паров бензина с использованием тест-систем «Открытое поле» и «Приподнятый крестообразный лабиринт». Представлены описания, характерные для острой ингаляционной токсичности выхлопных газов, что нашло отражение в последующих поведенческих реакциях на тест-системах «Открытое поле» и «Приподнятый крестообразный лабиринт». Выделены особенности тест-систем и их различия. Показано использование витамина D3 в качестве коррекции при воздействии полициклических ароматических углеводородов.

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