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Evaluating the Impact of Vitamin E Treatment on Paraquat Induced Toxicity in Liver

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Abstract: Paraquat is a weed destroyer utilized by agriculturists to safeguard their harvests against weed and increase crop yields. Paraquat has high gastrointestinal absorption rate and being very poisonous, it harms the liver, kidney, and the lungs by delivering free radicals, NF-KB activation, and apoptosis in numerous cells. The experimental study used. The average weight of 200 male Wistar rats was 200±20g. The two hundred rats were divided into four groups of fifty. The groups were labelled as A, B, C, and D. A control group received no paraquat, while groups B, C, and D received either 0.02g, 0.04g, or 0.06g of paraquat per kilogramme of rat every two weeks for three months. Each group consisted of two smaller groups. There were subgroups "A0" and "AVE" within the "A" group, "B0" and "BVE" within the "B" group, "C0" and "CVE" within the "C" group, and "D0" and "DVE" within the "D" group. Subgroups "A0," "B0," "C0," and "D0" did not receive vitamin E, while those in "AVE," "BVE," "CVE," and "DVE" did. subgroups weekly for one month after paraquat chronic treatment. Blood was acquired and assayed for liver function test. Aside from A0 grounp which is the control group, B0, C0, and D0 being the test groups (paraquat treated groups) as well as the BVE, CVE, and DVE vit. E treatment subgroups had a measurably significant difference, p-value≤0.05, in SGOT, SGPT, ALP, and GGT, a result which affirmed the toxic potential of paraquat and the ameliorative impact of vit. E. The outcomes showed that vitamin E treatment is strong against paraquat instigated liver toxicity on a one month weekly treatment plan.

Keywords: Paraquat, toxicity, liver, vitamin E.

1. Introduction

Paraquat is a weed destroyer utilized by ranchers to safeguard their harvests against weed and increase crop yields [1]. It is a very harmful compound normally utilized as herbicide by farmers in the US of America where just qualified and authorized people are allowed to utilize it [2]. Paraquat has absorption ability into the body and being very harmful, it affects the liver, kidney, and the lungs by delivering free radicals, NF-KB activation, and apoptosis in numerous cells [2, 3]. Ingestion of high amount of paraquat prompts organ impairment and even death within hours to days, while little amount consumed prompts impairment in two main organs; the lungs and kidney within 2-6days. As reported by Centre of Disease Control and Prevention, exposure to paraquat happens through the ingestion of paraquat contaminated food, air via

inhalation, or skin exposure [4]. Exposure to paraquat presents with side-effects like painful mouth and throat expansion [4]. Gastrointestinal side-effects, like nausea, emesis, stomach uneasiness, and loose bowels, are the following indications of disorder after use (which might turn out to be bloody) [4].

As per the report by Raja et al. (1992), the plasma activity of transaminase enzymes, alkaline phosphatase, and liver transketolase were notably reduced after paraquat exposure [5]. In one more study led by Noriega et al. (2002), it was found that the administration of paraquat, on guineas pigs showed a huge increase in lipid peroxidation and a drop in glutathione (GSH) levels [6]. The activity of antioxidant enzymes in the liver, for example, superoxide dismutase, catalase, and glutathione peroxidase, was diminished 3 hours after paraquat treatment [6].

Vitamin E is a group of eight fat-soluble vitamins with rich and intense cell antioxidative properties [7]. Of the eight groups, people's dietary necessities are best met by alpha-tocopherol (vitamin E) [7]. Vitamin E is notable for its capacity to shield the body against oxidative stress from free redicals otherwise called reactive oxygen species [7]. Rizvi et al., (2014) in their examination noticed that vitamin E is mainly present in cell and membranes of organelles, where it offers protection against oxidative damage[8]. It safeguards cell films from free radicals insults and goes about as the main line of protection against lipid peroxidation [8]. As per Bridges et al. (2021), vitamin E is stored in the body inside the fat tissues and the liver [9]. Vitamin E can is available in a many food sources and oils. Elevated degrees of alpha-tocopherol can be found in nuts, seeds, and vegetable oil, and a good amount can likewise be found in green vegetables and fortified grains [8].

2. Materials And Method

2.1. Experimental Study

Two-hundred male Wistar rats weighing 0.200.02kg were used in the investigation. Fifty rats were randomly assigned to each of four groups (A, B, C, and D) made up of the total of 200 rats. Paraquat was not administered to the "A" group, but 0.02g, 0.04g, and 0.06g were administered to the "B," "C," and "D" groups, respectively, every two weeks for three months. In every category, there were two further classifications ("0" and "VE"). In the "A" group, there were the "A0" and "AVE" subgroups; in the "B" group, the "BVE" and "C0" subgroups; in the "C" group, the "C0" and "CVE" subgroups; and in the "D" group, the "D0" and "DVE" subgroups. Although the "A0," "B0," "C0," and "D0" subgroups received no vitamin E supplementation, the "AVE," "BVE," "CVE," and "DVE" subgroups received 500mg of vitamin E once a week for a month following paraquat induction. It was decided to take a blood sample and analyse it for liver function.

Rat Source

Rivers State University of Science and Technology's Animal House provided 200 rats averaging 20020g in weight. The rats were given two weeks to acclimatise to their new environment before the experiment began. The research was conducted at Rivers State University of Science and Technology's Department of Medical Laboratory Science.

Sample Collection

Blood was collected for liver function test. 2ml of blood was taken through a heart puncture and transferred into plain containers by means of needle and syringe. The serum was separaated by spinning the blood at 4000 rpm for 5 minutes after it had coagulated. The serum was tested for liver enzyme.

3. Treatment Methods

3.1. Method for Paraquat Treatment

Oral gavage was utilised in the administration of paraquat. Group-specific dosing aside, all paraquat-treated groups received therapy every two weeks for a total of three months.

The mice were held by the skin over the head, and then twisted so that their mouths faced upward and their bodies faced the person holding them. After that, we positioned the slanted needle inside the rat's mouth horizontally, making sure to avoid the teeth in the middle. Paraquat was slowly dispensed from the syringe into the rat's mouth [10].

3.2. Method for Vitamin E treatment

For one month, 500 mg doses of vitamin E were approved for daily oral consumption [10].

Laboratory Analysis

Serum glutarate-oxaloacetate-aminotransferase (AST/SGOT) method: by Okolonkwo et al. (2022) [10].

Procedure

For the enzyme activity test, 0.5 mL of buffered-L-aspartate and -oxoglutarate solutions were added to two glass tubes labelled "Reagent Blank" and "Test," followed by 0.1 mL of distilled water and the sample, and then the tubes were shaken for 30 minutes at 370C. After thoroughly mixing, we added 0.5 ml of a 2, 4-dinitrophenylhydrazine (2 mmol/L) solution to each test tube and left them at 20 250C for 20 minutes. At the end of the period, 5.0mL sodium hydroxide (0.4mol/L) was added to enhance colour development at alkaline pH. After waiting for 5 minutes, the absorbance in the 'Test' (Atest) tube was compared to that in the 'Reagent blank' tube. The serum AST activity can be calculated using the previously established data plotted against enzyme activities. If there has been any hemolysis, the results will be inaccurate.

Serum glutarate-pyruvic-aminotransferase (SGPT) by Okolonkwo et al. (2022) [10].

Procedure

To determine the enzyme's activity, 0.5 mL of a buffered-L-alanine and -oxoglutarate solution was added to 0.1 mL of distilled water and 0.1 mL of sample in two separate

glass tubes labelled "Reagent Blank" and "Test," respectively. The mixture was then incubated at 370C for 30 minutes. Each test tube was given a thorough mixing before receiving 0.5 ml of a 2, 4-dinitrophenylhydrazine (2 mmol/L) solution. The tubes were then left at 20 250C for 20 minutes. Finally, 5.0mL sodium hydroxide (0.4mol/L) was added at the conclusion of the time period to promote greater colour development in an alkaline environment. The absorbance in the 'Test' (Atest) tube was compared with that in the 'Reagent blank' tube after 5 minutes had passed.

Calculation

Serum ALT activity can be calculated using the values shown against activities table. Haemolysis complicates the test's results.

Alkaline phosphatase (ALP) method by Okolonkwo et al. (2022) [10].

Procedure

After a new Gain calibration was performed in flow cell mode, fresh double distilled water (ddH2O) was inhaled. The previous sample run's apparatus has been reset to zero. After selecting ALP on the Run Test Screen and performing a water blank test, we dispensed 0.02 mL of sample and 1.0 mL of reagent (Diethanolamine buffered p-nitrophenylphosphate) into a test tube, mixed for 2 minutes, and read the results. Once that was done, the liquid was injected into the Rx Monza. After around 2 minutes, the test sample result was printed out using a connected printer.

In 1 hour, using S. I. unit = IU/L, the system can process and analyse 200 samples.

Manual calculation

Manually determining ALP requires the following formula: IU/L = 2760 x A 405 nm/min.

Gamma-Glutamyltransferase (GGT) method: by Okolonkwo et al. (2022) [10].

Procedure

A cuvette was filled with 0.1 ml sample and 1 ml reagent (Buffered Glycylglycerine and L-gammaglutamyl-3-carboxy-4-nitrolide), and the timer was set to run from 400 to 420 nm while the mixture was agitated. The absorbance was checked again at 1, 2, and 3 minutes.

To convert IU/L, multiply 1158 XΔA by A (405 nm/sec).

Results

Table 1. shows the mean values of liver enzymes after three month dose-dependent paraquat treatment. The results show that there were significant rise (p-value<0.05) in liver enzymes among the groups with increasing paraquat dose.

Table 1. Changes in liver enzymes after three months treatment period with paraquat

Subgroup	SGOT (IU/L)	SGPT (IU/L)	ALP (IU/L)	GGT (IU/L)
A_0	4.20 ± 0.14	1.35 ± 0.06	1.96 ± 0.02	12.75 ± 0.08
B_0	126.50 ± 2.05[a]	99.00 ± 0.01[a]	1028.00 ± 4.42[a]	87.00 ± 1.30[a]
C_0	115.50± 3.15[a,b]	191.00 ± 2.40[a]	1348.50 ± 2.75[a]	128.50 ± 0.85[a]
D_0	342.50 ± 4.35[a]	167.50 ± 1.25[a]	1570.00 ± 6.80[a]	170.00 ± 6.40[a]
p-value	<0.05	<0.05	<0.05	<0.05

Table 2. shows the mean value of liver enzymes activities after vitamin E treatment. The results revealed that were significant drop (p-value<0.05) in the liver enzymes activities in all groups treated with vitamin E after prolong paraquat induction.

Table 2. Changes in liver enzyme activities after Vitamin E treatment

Subgroup	SGOT (IU/L)	SGPT (IU/L)	ALP (IU/L)	GGT (IU/L)
A_0	4.20 ± 0.14	1.35 ± 0.06	1.96 ± 0.02	12.75 ± 0.08
A_{VE}	6.20 ± 0.34	8.30 ± 0.19	3.21 ± 0.01	22.95 ± 0.09
B_0	126.50 ± 2.05	99.00 ± 0.01	1028.00 ± 4.42	87.00 ± 1.30
B_{VE}	114.00 ± 1.80*	64.50 ± 0.05*	391.50 ± 4.54*	54.00 ± 1.40*
C_0	115.50 ± 3.15	191.00 ± 2.40	1348.50 ± 2.75	128.50 ± 0.85
C_{VE}	101.50 ± 0.55*	59.50 ± 0.25*	1294.50 ± 1.54*	65.50 ± 0.95*
D_0	342.50 ± 4.35	167.50 ± 1.25	1570.00 ± 6.80	170.00 ± 6.40
D_{VE}	199.00 ± 0.10*	123.50 ± 1.65*	659.50 ± 2.23*	122.00 ± 2.40*

Statistical significance: $p < 0.05$

*: signifies significant difference within groups

4. Discussion

This study assessed the ameliorative impact of vitamin E on liver enzymes in paraquat induced liver toxicity in male rats.

The outcome showed that there was a dose-dependent rise in the mean values of the liver enzymes in paraquat treated group. This means that the degree of liver injury was dependent of the dose of paraquat administered, indicating the lowest dose group experienced less liver harm than the highest dose group. This was expressed in the values of the liver enzymes activities. The reason for these outcomes could be a result of the toxic effect from oxidative stress initiated by paraquat on the liver organ of the rats. This outcome is in concurrence with works of Raja et al. (1992) and Noriega et al. (2002) which detailed that paraquat made a change in the liver function in guineas pigs utilized in their research works [5, 6].

Significant differences (p 0.05) were seen between subgroups B_0 and B_{VE}, C_0 and C_{VE}, and D_0 and D_{VE} for all four measures (SGOT, SGPT, ALP, and GGT), but subgroup A_0 and A_{VE} showed little change. The difference found in the subgroups B_0 versus B_{VE}, C_0 versus C_{VE}, and D_0 versus D_{VE} might be because of the role of vit. E in

decreasing the peroxidation brought about by paraquat in the B_0, C_0, and D_0 subgroups. As there was no oxidative damage presented in the A_0 versus A_{VE} subgroups, the administration of vitamin E yielded no outcome as there was no injury to mend, affirming again that vitamin E has ameliorative capacity as displayed in the research works of Richter et al. (2022) and Rizvi et al. (2014) [7, 8].

Conclusion

In all, the consequences of this trial showed that vitamin E treatment is strong against paraquat prompted liver damage within one month of weekly treatment.

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