THE FIBRINOLYTIC ALTERATION ASSOCIATED WITH DAILY ADMINISTRATION OF SILDENAFIL

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ABSTRACT

Objective: The objective of this study was to determine the fibrinolytic alteration associated with daily administration of sildenafil.

Methods: A total of 12 adult male rabbits without mortality rate had been fed standard and subdivided into four groups; their average weight was 1.5, 2.5, 1.9, and 2 kg randomly selected during the period of March 2012–July 2013. Depending on weight, the control groups (2.25 mg/1.5 kg day) and sildenafil groups (3 mg/2 kg/day, 2.85 mg/1.9 kg/day, and 1.7 mg/2.5 kg/day) were injected by normal saline and sildenafil concentration, respectively to create four groups, every group was composed of three rabbits; saline rabbit (control group, n=3) and sildenafil rabbits (sildenafil group, n=9). All rabbit's plasma samples have been investigated for prothrombin time, activated partial thromboplastin time, fibrinogen, plasminogen activator inhibitor-1 (PAI-1), prothrombin fragment 1+2, tissues plasminogen activator (tPA), plasmin antiplasmin (PAP), plasminogen, and D-dimer after 24 h of administration.

Results: The PAP level was significantly ($p<0.05$) decreased following sildenafil injection. Sildenafil-injected (3 mg/ml) rabbits had decreased the means of PAI-1 and mean tPA, as early as 1-day post-injection, with a considerable lower PAP first determined 3 days after injection that continued into each rabbit 2 and 3.

Conclusion: Better strategies are to initiate and manipulate this drug ought to reduce the chance of each thrombosis and hemorrhage, at the same time as minimizing the need for laboratory monitoring with the aid of the use of PAI-1, tPA, and PAP checks.

Keywords: Fibrinolytic, Alteration, Administration, Sildenafil.

INTRODUCTION

Clinical studies support the use of sildenafil as a treatment for various forms of pulmonary arterial hypertension in both children and adults [1,2] and for prevention or management of the right heart failure [3,4]. The observation of an antiplatelet effect of sildenafil and its influence in improving the microcirculation during wound healing in animal and human studies has also been encouraging [5-7]. Other animals and clinical studies have tested capability advantages of sildenafil in situations associated with diminished skin blood go with the flow and bad vascularization, which includes Raynaud's phenomenon and virtual ulcers in patients with systemic sclerosis [8-11]. Different studies have shown beneficial effects of low doses of sildenafil on portopulmonary hypertension following liver transplantation [12] or on systemic sclerosis associated with pulmonary hypertension [13]. Data from animal models on the beneficial effects of sildenafil on wound healing are promising, but human data are currently lacking [14]. Several research studies have been focused on different effects of sildenafil on tissue repair, but the alteration in fibrinolysis pathway associate to vascular defect was not done before. In the present study, the impact of sildenafil on wound healing in all clinical experimental is discussed by determining fibrinolytic alteration associated with daily administration of sildenafil.

METHODS

All experiments were authorized using the animal coverage committee of the College of Applied Medical Science, Aljouf University. 12 adult male rabbits without mortality rate had been fed standard and subdivided into four groups; their average weight was 1.5, 2.5, 1.9, and 2 kg, respectively, and randomly selected during the period of March 2012–July 2013. Depending on weight, the control groups (2.25 mg/1.5 kg/day) and sildenafil groups (1.7 mg/2.5 kg/day, 2.85 mg/1.9 kg/day, and 3 mg/2 kg/day) were injected by normal saline and sildenafil concentration, every group was composed of three rabbits; saline rabbit (control group, n=3) and sildenafil rabbits (sildenafil group, n=9). About 1–1.5 ml of blood was collected from each animal by aspirated from the ear or thigh vein according to the clarity of the vein and added to the anticoagulant (3.2%, 0.105 M) buffered sodium citrate, then gently mixed. The samples were centrifuged at 1700 g for 15 min to obtain platelet-poor plasma. We consequently used the higher dosage (3 mg/ml) to examine the consequences of treatment on fibrinolytic feature caused using sildenafil overdose as these results were greater obvious with the more extreme fibrinolytic alteration caused using the better dosage. All rabbit's plasma samples have been investigated for prothrombin time, fibrinogen, plasminogen activator inhibitor-1 (PAI-1), prothrombin fragment 1+2, tissues plasminogen activator (tPA), plasmin antiplasmin (PAP), plasminogen, and D-dimer (DD) after 24 h of administration. The statistical software SPSS 17 for windows was used for data analysis. Statistics is provided as mean ± standard deviation. Differences between groups were analyzed through one-way ANOVA test for more than one comparison, with a probability value $p<0.05$ appeared as significant.

RESULTS AND DISCUSSION

To establish a time path of sildenafil - brought on fibrinolytic alteration, we tested rabbits 1–3 days after injection with sildenafil and compared them to saline-injected control animals that have been examined on the primary day of the check. When compared sildenafil-injected groups to these controls, two rabbits out of 12 had been died after completing this step. The purpose of death may be due to hypovolemia as approximately

1–1.5 ml of blood was aspirated. The laboratory data were analyzed statistically for the level of significant. Table 1 summarizes that there was a significant difference (p<0.05) between the post-operative activated partial thromboplastin time (APTT) and the APTT control.

The PAP level was significantly (p<0.05) decreased following sildenafil injection as shown in Tables 2 and 3. Sildenafil-injected (3 mg/ml) rabbits had decreased the means of PAI-1 and mean tPA, as early as 1-day post-injection, with a significant decrease PAP first observed 3 days after injection that continued into both rabbits 2 and 3.

Table 1: Fibrinolytic and clotting parameters in rabbits Group-1 injected by sildenafil (1.7 mg/ml)

Variables	Mean±SD		p
	Group-1	Control	
PT Sec.	14.0±2.3	11.0±2.3	0.400
APTT Sec.	32.0±2.1	26.0±2.2	0.020*
FIB (g/l)	3.6±1.90	3.1±1.50	0.700
PAI-1 (ng/ml)	0.44±0.10	0.45±0.02	0.800
F1+2 (ng/ml)	75.9±17.7	73.9±19.3	0.910
tPA (ng/ml)	9.1±3.70	10.2±3.80	0.780
PAP (ng/ml)	96.6±5.30	91.6±3.30	0.240
DD (ng/ml)	346.9±22.1	342.5±24.1	0.830

*p<0.05 is statistically significant, Sec.: Seconds. PT: Prothrombin time, APTT: Activated partial thromboplastin time, FIB: Fibrinogen, PAI-1: Plasminogen activator inhibitor-1, F1+2: Fragment 1+2, tPA: Tissues plasminogen activator, PAP: Plasmin antiplasmin, DD: D-dimer, SD: Standard deviation

Table 2: Fibrinolytic and clotting parameters in rabbits' Group-2 injected by sildenafil (2.85 mg/ml)

Variables	Mean±SD		p
	Group-2	Control	
PT Sec.	14.0±2.2	11.0±2.3	0.200
APTT Sec.	28.0±2.3	26.0±2.2	0.300
FIB (g/l)	3.2±1.8	3.1±1.5	0.900
PAI-1 (ng/ml)	0.42±0.2	0.45±0.02	0.800
F1+2 (ng/ml)	74.8±23.5	73.9±19.3	0.900
tPA (ng/ml)	5.2±1.3	10.2±3.8	0.090
PAP (ng/ml)	55.8±3.1	91.6±3.3	0.000*
DD (ng/ml)	361.4±43.5	342.5±24.1	0.500

*p<0.05 is statistically significant. PT: Prothrombin time, APTT: Activated partial thromboplastin time, FIB: Fibrinogen, PAI-1: Plasminogen activator inhibitor-1, F1+2: Fragment 1+2, tPA: Tissues plasminogen activator, PAP: Plasmin antiplasmin, DD: D-dimer, SD: Standard deviation

Table 3: Fibrinolytic and clotting parameters in rabbits - 3 injected by sildenafil (3 mg/ml)

Variables	Mean±SD		p
	Group-3	Control	
PT Sec.	12.0±2.3	11.0±2.3	0.600
APTT Sec.	30.0±2.1	26.0±2.2	0.080
FIB (g/l)	3.6±1.9	3.1±1.5	0.700
PAI-1 (ng/ml)	0.40±0.10	0.45±0.02	0.440
F1+2 (ng/ml)	78.9±17.7	73.9±19.3	0.750
tPA (ng/ml)	8.1±6.70	10.2±3.8	0.660
PAP (ng/ml)	51.6±5.3	91.6±3.3	0.000*
DD (ng/ml)	371.9±22.1	342.5±24.1	0.190

*p<0.05 is statistically significant. PT: Prothrombin time, APTT: Activated partial thromboplastin time, FIB: Fibrinogen, PAI-1: Plasminogen activator inhibitor-1, F1+2: Fragment 1+2, tPA: Tissues plasminogen activator, PAP: Plasmin antiplasmin, DD: D-dimer, SD: Standard deviation

This study has shown that systemic administration of sildenafil injected has a significant difference (p<0.05) in reduces altered fibrinolytic and clotting parameters and had a controlling effect on function which did not achieve significance in a group of 12 rabbits. To maintain the fibrinolytic and clotting parameters within the therapeutic level we have to reduce of sildenafil dose for 2–3 days period; hence, acute clinical signs and symptoms required emergency [15]. PAI-1, tPA, and PAP were essential laboratory screening for monitoring of sildenafil since decreased of such more than the therapeutic level reflects a bleeding tendency. In the present study, systemic administration of sildenafil on rabbits injected with sildenafil showed that significant differences in reducing some parameter levels to nearly to the control level. These results agreed with those who used sildenafil to control of with diminished skin blood flow without with fibrinolysis change. Without fibrinolysis change. In addition to the cases of hereditary coagulation defect as haemophiliac patients factor VIII deficiency. The use of sildenafil administered either as hemorrhage reduction in patients with coagulation disorders [16].

Numerous preceding research has advised that the high negative predictive value of DD testing in outpatients with suspected thrombosis may be used as part of a diagnostic set of rules [17,18]. The value of mixing medical estimation of possibility with imaging assessments has been confirmed in different research [19]. We and others have proven the capability for the use of those checking out to simplify analysis further and showed the use of sildenafil within the treatment of erectile disorder and pulmonary arterial high blood pressure in which it relaxes the arterial wall and reduced pulmonary arterial resistance and pressure [20,21]. However, none of these strategies have previously been as compared in a randomized trial We have to demonstrate PAI-1, tPA, and PAP testing to rule out sildenafil overdose advantages from the other clinical conditions. The regular of some effects in rabbits might also need for a repeated take a look with high quality. This method has the extra advantage of increasing the percentage of rabbits may have an effective result on the repeated test. In the end, our effects are probably to be valid due to the fact the look at challenge was randomized, and the consequences had been determined by blinded to the rabbit assignments after sildenafil overdose.

Our observation was confirmed by previous medical study for the diagnosis of thrombosis, in which categorized rabbits into high, moderate, and low and others that not going to have thrombosis [22]. The addition to the scoring gadget of one factor for a preceding diagnosis of thrombosis lets to be used in previous thrombosis set. Therefore, we had excluded from earlier studies and evaluated the affected rabbits depending on the given results of previous test; our technique might be to be safe in sildenafil overdose; however, validation studies are required [23]. The use of these assays may be taken into consideration a challenge. However, the assay has some accuracy. We consequently consider our diagnostic strategy has to work regardless, of which the assay is used, provided that the assay selected has a sensitivity and specificity [24]. In comparison with previous studies that examined the relation of fibrinolytic and clotting parameters that related to sildenafil overdose, this study has the benefit of fibrinolytic imbalance and a statistical model that better addresses known confounders [25] which was in agreement with Eguchi *et al.* who demonstrated that endothelial cell mitochondria has decisive role on thrombus formation process [26,27].

CONCLUSION

In rabbits with active, major bleeding, plasma, in combination intravenous sildenafil overdose should be given and quickly reduces the dose in which may be discontinued temporarily or permanently. As during the initiation of sildenafil, the fibrinolytic activity in the period immediately after sildenafil is discontinued may not accurately reflect the degree of impairment of coagulation. A single intravenous dose of 2.5 mg/2 Kg sildenafil given preoperatively was effective in

reducing post-operative fibrinolytic and clotting parameters. Sildenafil overdose remained the mainstay of a rise of thrombotic. Better techniques to initiate and manage this drug must reduce the chance of each thrombosis and hemorrhage, at the same time as minimizing the want for laboratory tracking using PAI-1, tPA, and PAP examinations. Effective techniques for the treatment of bleeding ought to simplify the care of sildenafil overdose, reduce the long-time period morbidity resulting from hemorrhage and probably risky.

AUTHOR'S CONTRIBUTIONS

All the authors have contributed equally.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

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