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Effects of combination of tramadol and sildenafil citrate (Viagra) on hepatic and renal function parameters in Male Albino Rats

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Abstract

Erectile dysfunction, premature ejaculation and pleasure seeking have increased the use of sildenafil citrate, tramadol and their combination by youths who believe these drugs could positively impact their sexual life. Sildenafil citrate and tramadol are metabolized in the liver, and their by-products excreted through the kidneys, making these organs possible targets for toxicity.

Aim: This study evaluates the effects of combination of tramadol and sildenafil citrate on hepatic and renal function parameters in male albino rats.

Methodology: A total of forty-nine (49) male albino rats weighing between 150 to 180g were used for the study. Sildenafil citrate and tramadol were administered to the rats by means of oral gavage for 28 days. The liver enzymes alanine aminotransferase (ALT) and aspartate aminotransferase (AST) were determined using the Reitman-Frankel method. Alkaline phosphatase (ALP) was determined using the Colorimetric endpoint method. Total protein (TP) was determined using the biuret method. Albumin (ALB) was determined using the bromocresol green method. Sodium (Na+), potassium (K+) and chloride (CL-) were determined using ion selective electrode (ISE) method. Bicarbonate (HCO_3^-) was determined using titrimetric method. Urea was determined using Urease bertholet method. Creatinine was determined using Jaffe-Slot method.

Results: There were no significant differences (P>.05) in the liver enzymes ALT, AST and ALP in all the treatment groups compared to the negative control. Total protein (TP) and albumin (ALB) also showed no significant differences (P>.05) in the groups, compared to the negative control. there were no significant differences (P>.05) in the electrolyte levels in the treatment groups, compared to the negative control. Urea and creatinine levels were also not significantly different (P>.05) in the treatment groups, compared to the negative control.

Conclusion: Administration of sildenafil citrate singularly and in combination with tramadol, at recommended and double doses for 28 days, did not impact the liver and was also non-toxic to the kidneys.

Keywords: Sildenafil citrate (Viagra); Tramadol; Erectile dysfunction; Premature ejaculation; Liver and Renal function

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1. Introduction

Erectile dysfunction (ED) is defined as the persistent inability to achieve and or maintain penile erection, sufficient for sexual intercourse or for a satisfactory sexual activity. ED is caused by a variety of factors that interact to bring about its development. These factors range from hormonal, neurogenic, vascular, psychogenic, anatomic and iatrogenic causes etc [1,2]. There is an increase in the incidence and prevalence of erectile dysfunction worldwide, as reported by studies in the United Kingdom, United states of America and Europe. Africa and particularly Nigeria is not left out, as studies have also reported high prevalence rates in these regions with one particular study reporting the general prevalence of ED to be about 60% in the South-western part of Nigeria [3,4,5]. Erectile dysfunction is the most common form of sexual dysfunction in men, only after premature ejaculation.

The International Society for Sexual Medicine (ISSM) defines premature ejaculation (PE) as ejaculation that always or nearly always occurs prior to or within one minute of vaginal penetration, and the inability to delay ejaculation on all or nearly all vaginal penetrations, and negative personal consequences, such as distress, bother, frustration and/or the avoidance of sexual intimacy [6]. PE can be classified as lifelong or acquired, and it can negatively impact the quality of life not only of the male but also his female partner as well causing interpersonal difficulties, sexual dissatisfaction, and distress. Various studies have reported high prevalence of premature ejaculation with prevalence varying in literature from 2 to 27.5% [7,8]. Disorders of sexual interest/desire, erectile dysfunction and ejaculation are the most common sexual dysfunctions detected in men. It has led to the increased use and abuse of sex stimulants, medications for ED and some other medications perceived to delay ejaculation [9].

Sildenafil citrate (Viagra) a phosphodiesterase type 5 (PDE5) inhibitor, was originally developed for the treatment of angina pectoris, but has subsequently been extensively used clinically in the treatment of erectile dysfunction. Sexual stimulation causes the release of nitric oxide (NO) in the corpus cavernosum of the penile tissue. For a sustained erection, there has to be a sufficient amount of nitric oxide in the corpus cavernosum. The released nitric oxide activates the enzyme, guanylate cyclase which results in an increased level of cyclic guanosine monophosphate (cGMP), producing smooth muscle relaxation in the corpus cavernosum and allowing inflow of blood into the penile tissue. Sildenafil citrate enhances the actions of NO by inhibiting the enzyme phosphodiesterase type 5 (PDE5), which is responsible for degradation of cGMP in the corpus cavernosum, leading to accumulation of cGMP. With this mechanism, it increases the effects of endogenous nitric oxide, leading to penile smooth muscle relaxation, thereby creating arterial dilatation, which leads to the expansion of the corpus cavernosum [10,11,12].

Also, after sexual stimulation, NO concentration is significantly increased and contributes to the conversion of guanosine triphosphate (GTP) to cGMP. cGMP decreases intracellular calcium ions in the cavernosal smooth muscles, leading to smooth-muscle relaxation. Once relaxed, the smooth muscles collapse the veins, which reduces the drainage of arterial blood out of the penile tissue, thus sustaining an erection [13]. It is worthy to note that Sildenafil at recommended doses has no effect in the absence of sexual stimulation, it only amplifies the effect of sexual stimulation by retarding the degradation of cGMP.

Tramadol is a synthetic, centrally acting opioid agonist that also acts as a serotonin and norepinephrine reuptake inhibitor. It is used for the treatment of moderate to severe pain. Its mechanism of analgesic action is rather complex. The analgesic action of tramadol and other clinical effects are believed to be via both opioid and non-opioid mechanisms. Tramadol exerts its action through binding to the μ-opioid receptor, however, its action is less than that of morphine. It also inhibits the neuronal reuptake of norepinephrine and serotonin as do the antidepressant drugs such as amitriptyline and desimpramine [14,15]. Tramadol has become the most prescribe opioid worldwide. The easy and wide availability of tramadol as a pain killer for intermediate pain and many other forms of chronic pains could be basic factors of its wide spread and abuse. Additionally, its role in the treatment of premature ejaculation could be one of the most apparent reasons there is an increase in its use and abuse, mostly by youths who believe that tramadol has a positive impact on their sexual functions [16,17].

Out of curiosity, recreational use, and pleasure seeking, some with the aim of drug addiction, youths mostly without ED and ejaculatory problems combine sildenafil citrate and tramadol because sildenafil prolongs erection and tramadol delays ejaculation. This is done without thought to the possible side effects and pharmacokinetic interactions [18,19,20]. Sildenafil citrate and tramadol are metabolized in the liver through the cytochrome p450 enzyme system. Also, products of metabolism are excreted through the kidneys, thus these organs could be targets of toxicity. This study evaluates the effects of combination of tramadol and sildenafil citrate on hepatic and renal function parameters in male albino rats.

2. Materials and methods

2.1. Experimental Animals

A total of forty-nine (49) male Albino rats weighing between 150 to 180 g were used for the study. The rats were housed in standard cages at regulated room temperature, with controlled 12-hour light-dark cycles, and allowed access to feed and water *ad libitum*. The rats were allowed to acclimatize for two (2) weeks prior to the commencement of study.

2.2. Drugs

The drugs used for the study were Tramadol hydrochloride and Sildenafil citrate. Tramadol is manufactured by Vadis Pharm Ltd, Plot Rd 14 Trans-Ekulu, Enugu, Nigeria. Sildenafil citrate/Viagra (Vega 100) is manufactured by Hab Pharmaceuticals & Research Ltd, India. As standard drugs, the doses were translated from the human dose.

2.3. Dose Calculation

2.3.1. Tramadol Hydrochloride

The administered rat dose was extrapolated from the human daily dose [21] as shown below:

Human daily dose is 2 capsules (50 mg each) daily, which is 100 mg/day.

$$\text{Rat dose (mg/kg)} = \text{Human daily dose} \times 0.018 \times 5$$

$$= 100 \times 0.018 \times 5$$

$$= 9 \text{ mg/kg/day}$$

2.3.2. Sidenafil Citrate (Viagra)

The administered rat dose was extrapolated from the human daily dose [21] as shown below:

Human daily dose is 1 tablet (100mg) daily, which is 100 mg/day.

$$\text{Rat dose (mg/kg)} = \text{Human daily dose} \times 0.018 \times 5$$

$$= 100 \times 0.018 \times 5$$

$$= 9 \text{ mg/kg/day}$$

2.4. Experimental Design

The rats were weighed and grouped into seven (7) groups of six (7) rats each. Treatments (drugs) were administered daily according to the groupings by means of oral gavage for 28 days.

- Group 1: Negative control group.
- Group 2: Administered 9 mg/Kg of Tramadol.
- Group 3: Administered 9 mg/Kg of Viagra.
- Group 4: Administered double dose (18 mg/Kg) of Tramadol,
- Group 5: Administered double dose (18 mg/Kg) of Viagra.
- Group 6: Administered 9 mg/Kg of Tramadol and 9 mg/Kg of Viagra.
- Group 7: Administered double dose (18 mg/Kg) of Tramadol and double dose (18 mg/Kg) of Viagra.

On the 29th day, the rats were fasted for 6 hours anaesthetized and later sacrificed. Blood was collected from each rat by means of cardiac puncture. All the animal experiments were conducted according to the ethical norms approved by the Institutional Ethical Committee.

2.5. Reagents and Biochemical Analyses

All reagents were commercially purchased and the manufacturer's standard operating procedures strictly followed. Quality control (QC) samples were run together with the biochemical analysis. The liver enzymes alanine aminotransferase (ALT) and aspartate aminotransferase (AST) were determined using the Reitman-Frankel method [22], as modified by Randox laboratories limited (UK). Alkaline phosphatase (ALP) was determined using the Colorimetric endpoint method [23] as modified by Randox laboratories limited (UK). Total protein (TP) was determined using the biuret method [24], as modified by Randox laboratories limited (UK). Albumin (ALB) was determined using the bromocresol green method [25], as modified by Randox laboratories limited (UK). The electrolytes, sodium (Na^+), potassium (K^+) and chloride (CL^-) were determined using ion selective electrode (ISE) method [26]. Bicarbonate (HCO_3^-) was determined using titrimetric method [27]. Urea was determined using Urease bertholet method [28], as modified by Randox laboratories limited (UK). Creatinine was determined using the Jaffe-Slot method [29], as modified by Randox laboratories limited (UK).

2.6. Statistical Analysis

Data was analysed using Graph Pad Prism version 8.0.2. Differences between groups were compared using one way analysis of variance (ANOVA), followed by Tukey's multiple comparison test. Results were considered statistically significant at 95% confidence interval ($p \leq 0.05$). Values are expressed as Mean ± SD.

3. Results and discussion

Table 1 Liver Function Parameters of the Rats after Treatment

Groups	ALT (IU/L)	AST (IU/L)	ALP (IU/L)	Total Protein (g/L)	Albumin (g/L)
Group 1 (Neg. Contrl)	35.20 ± 4.09	114.8 ± 12.23	67.33 ± 9.04	41.33 ± 4.43	30.33 ± 2.52
Group 2 (Tra nd)	34.00 ± 2.53	115.8 ± 8.01	66.17 ± 11.62	44.50 ± 2.59	30.00 ± 2.28
Group 3 (Tra dd)	33.17 ± 4.45	104.5 ± 11.40	75.17 ± 11.82	40.33 ± 5.57	31.17 ± 3.55
Group 4 (Via nd)	36.17 ± 5.78	117.30 ± 10.68	73.33 ± 15.64	37.00 ± 6.69	28.50 ± 1.87
Group 5 (Via dd)	36.17 ± 2.86	104.5 ± 8.57	66.83 ± 6.97	32.33 ± 5.89	29.00 ± 3.63
Group 6 (Tra+Via nd)	35.17 ± 4.90	104.0 ± 10.01	72.00 ± 8.60	35.83 ± 5.08	29.33 ± 2.20
Group 7 (Tra+Via dd)	34.83 ± 3.13	108.65 ± 13.31	67.83 ± 8.80	36.83 ± 5.57	29.00 ± 1.90
P-value	0.3958	0.1780	0.5095	0.0504	0.4029
F-value	1.077	1.605	0.8963	2.395	1.066
Remark	NS	NS	NS	NS	NS

Tra – Tramadol, Via – Viagra, nd – Normal dose, dd – Double dose, NS – not significant.

Table 1 shows results of liver function parameters after treatment. It shows there were no significant differences ($P>.05$) in the liver enzymes ALT, AST and ALP in all the treatment groups compared to the negative control. Total protein (TP) and albumin (ALB) also showed no significant differences ($P>.05$) in the groups, compared to the negative control. The results indicate there was no hepatoxicity following the normal and double dose administration of sildenafil citrate and tramadol, singularly and combined. The results are in agreement with the works of Ibama et al. [30], in which the administration of sildenafil and revive capsules were seen not to be hepatotoxic. Sildenafil citrate has been reported to reverse biochemical/histopathological changes in the liver due to jaundice and offer hepatoprotective action against drug-induced hepatotoxicity [31,32]. Some other studies have however reported hepatotoxic effects of increasing doses of sildenafil and the combination of sildenafil and tramadol in experimental models [1,33].

Table 2 Renal Function Parameters of the Rats after Treatment

Groups	Na^+ (mmol/L)	K^+ (mmol/L)	Cl^- (mmol/L)	HCO_3^- (mmol/L)	Urea (mmol/L)	Creatinine (µmol/L)
Group 1 (Neg. Contrl)	133.7 ± 1.4	4.4 ± 0.4	102.2 ± 1.4	15.67 ± 2.52	3.73 ± 1.12	67.67 ± 6.43
Group 2 (Tra nd)	134.3 ± 0.8	4.5 ± 0.6	101.8 ± 2.3	15.50 ± 2.26	3.52 ± 1.67	63.83 ± 4.92
Group 3 (Tra dd)	132.3 ± 2.3	4.2 ± 0.2	99.8 ± 1.6	15.17 ± 2.64	2.58 ± 0.39	67.17 ± 8.31
Group 4 (Via nd)	134.0 ± 2.5	4.5 ± 0.5	104.7 ± 3.1	14.50 ± 1.05	2.35 ± 0.44	61.83 ± 8.16
Group 5 (Via dd)	131.7 ± 1.5	4.9 ± 0.6	100.00 ± 2.2	14.17 ± 1.47	2.87 ± 0.50	56.83 ± 7.96
Group 6 (Tra+Via nd)	131.0 ± 1.7	4.1 ± 0.2	98.67 ± 2.3	16.67 ± 1.75	2.50 ± 0.50	66.67 ± 5.85
Group 7 (Tra+Via dd)	133.0 ± 3.3	4.5 ± 0.5	103.3 ± 1.8	14.00 ± 0.89	2.68 ± 0.37	67.17 ± 6.89
P-value	0.0718	0.0944	0.0865	0.1993	0.1011	0.6787
F-value	2.151	1.985	3.112	1.533	1.960	0.6644
Remark	NS	NS	NS	NS	NS	NS

Tra – Tramadol, Via – Viagra, nd – Normal dose, dd – Double dose, NS – not significant.

Table 2 shows the renal function parameters of the rats after treatment. It shows there were no significant differences (P>.05) in the electrolyte levels in the treatment groups, compared to the negative control. Urea and creatinine levels were also not significantly different (P>.05) in the treatment groups, compared to the negative control. The results indicate there was no nephrotoxicity following the normal and double dose administration of sildenafil citrate and tramadol, singularly and combined. Similar studies have reported that the administration of sildenafil citrate at the extrapolated human dose did not impact renal function and was not nephrotoxic [30,34]. Baran et al. [35] reported sildenafil was nephroprotective and hepatoprotective in cadmium-induced multiple organ damage. They attributed the effect to its antioxidant properties, as it significantly decreased thiobarbituric acid reactive substances (TBARs) while increasing glutathione (GSH) and total thiol (T-SH) levels in the tissues evaluated. Other studies have pointed out the risk of increased hepatic and renal damage due to long-term use of tramadol and its combination with sildenafil [15,33].

4. Conclusion

The administration of sildenafil citrate singularly and in combination with tramadol, at recommended and double doses for 28 days, did not impact the liver and was also non-toxic to the kidneys. However, care should be taken and prescriptions adhered to, as abuse of these drugs at higher doses could be injurious to health.

Compliance with ethical standards

Conflict of interest

Authors have declared that no competing interests exist.

Statement of ethical approval

All animal experiments were carried out following ethical norms approved by the Institutional Ethical Committee.

Authors' Contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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