Histological Interfaces of Liver, Kidney and Cerebrum in Male Rats exposed to Fluoxetine

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1. INTRODUCTION

Depression is a condition of unfavorable mood and dislikes to do activities which lead to abnormal behavior and aggressive feeling. Depressed mood is a normal occurrence in response to adversity in all individuals and it is very common among persons who had a trouble and disease (Johnstone et al., 2007). The diagnoses such as Beck Depression Inventory and Children’s Depression Inventory have been used for assessment of depression and severity of its symptoms (Biros et al., 2008, Jang et al., 2016). In general, depressed mood may not need a professional treatment, but in most cases, the patients take treatments, so that the production and using of antidepressant such as selective serotonin reuptake inhibitors are the most frequently prescribed drugs to treat moody disease among
Fluoxetine is a selective serotonin reuptake inhibitor and as a first line drug which is used as a treatment of depression and many neuropsychiatric disorders (Cipriani et al., 2005, Wernicke, 2004). Serotonin in the central nervous system act as neurotransmitter included in variety of physiological and behavioral functions and considered to have an important role in the control of pain (Bardin, 2011).

Depression and drug using such as fluoxetine gradually increases in all ages especially among adults and teenagers in Kurdistan region-Iraq. Fluoxetine is easily available by the patients; it metabolizes in the liver and excreted in the urine. This study was planned to determine the histological effects of fluoxetine on liver, kidney and cerebrum.

2. MATERIALS AND METHODS

2.1. Experimental Animals

Sixteen adult Wister male albino rats (8-10 weeks old) and weighing (200-270 gm) were conducted in this study. They were housed in plastic rat cages (56 x 39 x 19 cm) in groups of eight rats per cage in a room with controlled temperature of (22 ± 1 °C), 12 hours light and 12 hours dark by using an automated light-switching device, in the animal house of Biology department, Faculty of Science, Soran University; under supervision and approval of local scientific committee and animal care rules. Rats were fed with standard laboratory chow and allowed drinking water *ad libitum.*

2.2. Experimental design

Rats were divided randomly into two groups; group 1: control group (n=8), group 2: fluoxetine-exposed rats in which rats were orally administrated with 10 mg/kg body weight/day (n=8).

2.3. Anesthesia, dissection and removal of the organs

At the end of the experiment, all rats were anesthetized with intraperitoneal injection of a mixture of Ketamine Hydrochloride (80 mg / Kg) and Xylazine (12 mg / Kg), then liver, kidneys and cerebrum were removed, cut into smaller pieces (approximately 0.5cm in thickness) in petri dish which contained a fixative (Bouin’s fluid) and then transferred into Bouin’s fluid for fixation.

2.4. Histological preparation (Paraffin method)

Fixed tissues (liver, kidney and cerebrum) were removed from Bouin’s fluid and dehydrated by a serial concentration of ethanol in ascending manner, infiltrated with paraffin wax after the clearing. Paraffin wax also used for making tissue blocks. The tissue blocks were cut into four to six micrometer thick paraffin sections by using rotary microtome (Bright, MIC) and stained with hematoxylin and eosin (H&E) (Bancroft et al., 1977). Finally, light microscope (digital binocular compound microscope 40x-2000x, built-in 3MP USB camera) was used to examine and photo taken.

3. RESULTS AND DISCUSSION

Daily oral administration of fluoxetine for one month caused several histological alterations in liver, kidney and cerebrum of rats.

3.1. Effect of fluoxetine on liver

Histological sections of liver control rats showed normal hepatic architecture in which
many healthy hepatocytes were polyhedral in shape appeared, exhibited centrally round or ovoid shape nucleus and well-known plasma membrane. Blood sinusoids appeared normally and located between hepatocytes. Central vein were normal (Fig. 3.1 and Fig. 3.2). The liver sections of fluoxetine treated rats showed that most of hepatocytes would be appeared as normal and there is no obvious architecture change except that a few of the hepatocytes degenerated, a little inflammatory infiltration of leukocytes and congested blood vessel would be observed (Fig. 3.3 and Fig. 3.4). Histopathological examination of the liver revealed hepatic injury after fluoxetine treatment (Yilmaz et al., 2016). Furthermore, hepatocellular hydropic vacuolar degeneration, portal area and lobular inflammation were observed as a result of rat exposure to fluoxetine which agree with our results (Özden et al., 2005).

3.2 Effect of fluoxetine on kidney

The paraffin sections of the cortical region showed well-designated glomeruli and kidney tubules both proximal convoluted tubules and distal convoluted tubules in the kidney of control group rats (Fig. 3.5). The medullary region of the control kidney rats also showed a normal appearance of tubules as well as their epithelial cells (Fig. 3.6). In contrast, fluoxetine caused alterations in kidney rats of treated group, the cortex region showed shrinking of glomeruli, hemorrhage and dead cells (Fig. 3.7) while in the medullary region, thickened the wall of kidney tubules and degeneration of some kidney tubule epithelial cells were observed (Fig. 3.8). At doses of 3 and 10 mg/kg, the significant reductions in renal nerve activity were observed especially 15 minutes after the intravenous injection of fluoxetine (Tiradentes et al., 2014). The hyponatremia arose during fluoxetine antidepressant therapy (ten Holt et al., 1996). Also, the sodium ions level decreased, Potassium unchanged and antidiuretic hormone remained unchanged, whereas the AQP2 protein abundance and water absorption in the inner medullary collecting duct were increased (Moyses et al., 2008).

3.3. Effect of fluoxetine on cerebrum

As shown in figure 3.9, the histological slides showed that the cerebrum of control rats have normal appearance of their structure such as layers of grey matter and normal neuronal cells, while dead pyramidal cells in second and third layers of grey matter were observed in the sections through the cerebrum of fluoxetine-exposed rats (Fig. 8.10). It has been suggested that fluoxetine cause increasing the concentration of serotonin in synaptic cleft and vasodilation of small cerebral arteries such as branches of the anterior cerebral arteries which induced by calcium channel openers (Ungvari et al., 2000). In addition, the electrophysiological studies have further demonstrated that fluoxetine inhibits different types of calcium channels in the neurons (Deak et al., 2000). Fluoxetine at 0.03 mM enhanced nicotine- and choline-induced relaxations in which nicotine induced norepinephrine release from cerebral perivascular sympathetic nerves but vasorelaxation was blocked by higher concentration of fluoxetine (>0.3 mM) that is mean, the high concentration of fluoxetine cause decrease neurogenic vasodilation while low concentration of fluoxetine cause increase neurogenic vasodilation (Chen et al., 2012). Furthermore, fluoxetine alters the levels and composition of brain GABA(A) receptors and reduces the responsiveness of GABA(A)-R to GABA-mimetic drugs such as pentobarbital (Matsumoto et al., 2007).
Figure 3.1: Photomicrograph from liver a control rat showing the normal central vein (CV), normal hepatocytes and blood sinusoids. H&E. 100X.

Figure 3.2: A magnified photomicrograph from liver control rat section showing normal appearance of hepatocytes (black arrow) which polyhedral in shape, round or ovoid shape nucleus and well-defined plasma membrane. Blood sinusoids (S) and central vein (CV) are normal. H&E. 400X.
Figure 3.3: Photomicrograph from liver treated rat with fluoxetine showing that the hepatocytes will appear as normal and there is no architecture change. A little inflammatory infiltration of leukocytes (black arrow) and congested blood vessel (white arrow) will present due to fluoxetine drug. H&E. 100X.

Figure 3.4: A magnified histological section of liver treated rat with fluoxetine showing congested blood vessels (white arrow) and leukocyte infiltration inflammation (black arrow). Most of hepatocytes show normal architecture and a few of them degenerated. H&E. 400X.
Figure 3.5: Section through the cortex of the kidney of control rat showing normal glomeruli (black arrow) and kidney tubules (white arrow). H&E. 400X.

Figure 3.6: Medullary region in the kidney of control rat having a normal appearance of tubules (T) H&E. 400X.
Figure 3.7: Photomicrograph from kidney cortex of treated rat with fluoxetine showing shrunken tuft of glomeruli (black arrow), hemorrhagic area (H) and degenerated cell area (D). H&E. 400X.

Figure 3.8: Section through the kidney of fluoxetine treated rat showing the kidney tubules in the medullary region which they had thickened wall (black arrow) and some of their cells were degenerated (white arrow). H&E. 400X.
Figure 3.9: Sections through the cerebrum of control rats. A) Grey matter of cerebrum shows different layers with normal structure. H&E. 100X. B) Normal appearance of grey matter in which normal pyramidal cells (black arrow) and glial cells (white arrow) were observed. H&E. 400X. C) Oil immersion magnification power showing normal pyramidal cells (black arrow) and glial cells (white arrow) in cerebral cortex layer. H&E. 1000X.
4. CONCLUSIONS

The present work was designed to determine the histological effect of fluoxetine on liver, kidney and cerebrum of male rats. The light microscopic examination showed that orally administration of fluoxetine for a period of one month had a bantam histological effect of liver, kidney and cerebrum of male rats.

Figure 3.10: Photomicrographs from cerebrum treated rats with fluoxetine. A) Grey matter of cerebrum which shows layers. H&E. 100X. B) Dead cells (black arrow) were observed due to fluoxetine in second and third layers of grey matter. H&E. 400X. C) High power magnification shows clearly dead cells (black arrow). H&E. 1000X.
Conflict of Interest
There is no conflict of interest.

REFERENCES


