Targeting Mitochondrial Biogenesis in Rat Brain for Treatment of Depression

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Abstract

Objectives: To evaluate brain mitochondrial role in depression pathogenesis through mitochondrial Dynamin-Related Protein (Drp-1), Fission 1 Protein (Fis-1), Brain Derived Neurotrophic Factor (BDNF) and Tyrosine Kinase B (Trk-B) receptor stimulation using three different antidepressants.

Methods: Sixty four healthy adult male Sprague-Dawley rats weighing 180 ± 20 gm, were used. We established an animal model of stress-induced depressive behavior named learned helplessness (LH). LH rats were treated with fluoxetine FLX (20 mg/kg), imipramine IMP (20 mg/kg) or citalopram CTL (20 mg/kg), i.p. for 2 weeks.

Key findings: Order of decreasing number of LH rats was CTL>IMP>FLX. Brain mitochondrial SDH, MDH, IDH, MAO and SOD enzymes, Total Antioxidant Status (TAS), percentage ATP production, BDNF, Drp-1, Trk-B and Fis-1 proteins of depressed (DEP) rats significantly declined. Brain lipid peroxides were highly elevated in DEP-rats. All these levels were back to normalcy after intake of antidepressants and protein expression of BDNF, Drp-1, Trk-B, and Fis-1 were enhanced. Direct positive correlations were recorded between TAS and neurotrophins proteins among DEP-rats.

Conclusion: The use of FLX, IMP, and CTL alleviated depression induced in rats via antioxidant mechanism and neurotrophins expression modulation in rat brain mitochondria.

Keywords: Antidepressants; Brain; Dynamin-related protein (Drp-1); Fission-1 protein (Fis-1); Brain-derived neurotrophic factor (BDNF); Tyrosine kinase-B (Trk-B) receptor

Introduction

Targeting mitochondrial mechanisms are implicated for studying psychiatric disorders [1]. The Brain-Derived Neurotrophic Factor (BDNF), Dynamin-Related Protein-1 (Drp-1), Tyrosine Kinase Receptor-B Protein (Trk-B), and Fission-1 Protein (Fis-1) proteins are neurotrophins that share common target gene of antidepressant treatment via regulation of synaptic plasticity [2]. Antidepressant effect of BDNF is due to the discharge of BDNF into midbrain of depressed animals, through binding with its receptor, Trk-B protein [3].

Mitochondria function is remodeled by fission of individual mitochondria and the fusion of different mitochondria [4]. The Drp-1 is present in the cytoplasm and includes GTPase and GTPase effector domains. Fis-1 is existed in outer mitochondrial membrane and composed of 6 antiparallel helices protein with 4 central helices containing 2 tandem tetratricopeptide repeats motifs [4]. Patients with depression exhibited decreased mitochondrial ATP production rate associated with a decrease in mitochondrial proteins and increased mtDNA mutations [5].

We were interested in studying mitochondria involvement in pathogenesis and treatment of depression through investigating the molecular link between depression, mitochondrial activity, and mitochondrial biogenesis and thereby correlating outcomes to effects of antidepressant drugs towards a molecular link between neurotrophic factors and changes in mitochondrial biogenesis in rat pre-and post-antidepressant treatments.

Materials and Methods

ATP percentage production was estimated using bioluminescent somatic cell assay kit (Sigma Aldrich®-USA). Antibodies and protease inhibitor cocktail were purchased from Santa Cruz Biotechnology® (USA) and Roche® (Switzerland); respectively. β-actin gene and anti-actin antibodies were purchased from TIBMOLBIOLs® (Germany). Protein assay kit and semi-dry gel transfer system (Bio-Rad® laboratories- USA) were used. The BDNF quantitative EIA kit was supplied by Quantikine® (R and D systems, USA). The Drp-1, Trk-B receptor and Fis-1 quantitative EIA kits were purchased from MyBiosource® (USA). Total Antioxidant Status (TAS) kit was purchased from Randox® (UK). This study was conducted in accordance with local research ethics committee of Taibah University, Saudi Arabia under number TUCDREC/20110068 and regulations stated in the declaration of Helsinki.

To induce Learned Helplessness (LH) depression model; rats were exposed to inescapable electrical shocks and placed in one side of clear plexiglass restraining tubes shuttle box supplied with electrified floor grid (MedAssociates, USA) divided into 2 equal rooms by rigid plastic divider [6]. Restriction brackets made of wire racks coated with plastic that set in place 2 cm above rat dorsal surface to movement though limiting frequency of jumping when rat placed inside the room. The shock was delivered from constant-current shock generators (1.0 mA) to electrodes provided with electrode paste to the rat's tail. Each shock had duration of 15 sec with interval periods between 30 and 90 sec over a period of 3 hrs. The inescapable shock session comprised 100 5-secs, A 1.0 mA shocks delivered on 1-min interval schedule. At the start of each shuttle box session, rats were exposed to a 5 min habituation period in the same chamber where Inescapable Shock (IS) or Escapable Shock (ES) was applied, followed by 30 escape trials in which gate separating the two halves of the shuttle box opened 5 sec prior to shock onset. Randomized foot shocks were transmitted at an intensity of 0.6 mA for 30-sec duration of escape latency.

Sixty-four healthy adult male Sprague-Dawley rats weighing 180 ± 20 gm were housed singly in polyethylene cages and transferred to clear plexiglass restraining tubes during the experiment. Rats were divided into 8 groups (n=8). Group 1: Control rats received 0.2 ml 0.9% saline (i.p.) for 2 weeks; Group 2: Rats received 0.2 ml 0.9% saline (i.p.) for 2 weeks and exposed to chronic inescapable shock in clear plexiglass restraining tubes for 1 hr/day over 2 weeks and then tested for LH model. These rats are nominated as depressed (DEP)-rats; Group 3, 4, 5: Rats received 20 mg/kg/day fluoxetine (FLX), imipramine (IMP) or citalopram (CTL); respectively in normal saline (i.p.) for 2 weeks without stress and Group 6, 7, 8: DEP-rats received FLX, IMP or CTL; respectively and tested for LH

model. At end, rats were under food deprivation overnight prior to sacrifice and were decapitated 24 hours after last dose and brain was isolated, get mitochondrial homogenate for biochemical analysis, %ATP production, Lipid Peroxides (LPOs) expressed as Thiobarbituric Acid Reactive Substance (TBARS), TAS, BDNF, Drp-1, Fis-1 and Trk-B receptor proteins. Protein expression of brain BDNF, Drp-1, Fis-1 and Trk-B for different groups were estimated.

The gray matter was homogenized according to protocol [7]. Protein concentration was ~20-45 ng/ml and stored at 4°C for only 4 days to ensure mitochondria activity. Estimation of rat mitochondrial brain enzymes; Succinate Dehydrogenase SDH (nmol formazan formed/min/mg protein) [8], Malate Dehydrogenase (MDH), Isocitrate Dehydrogenase (IDH) (nmol NADH oxidized/min/mg protein) [9,10] and monoamine oxidase MAO (nmol benzaldehyde formed/min/mg protein) [11], were measured.

Estimation of rat brain mitochondrial percentage ATP production was carried out [12]. %ATP production=ATP sample (moles) × ATP standard (moles)/light emitted by both sample and standard-light emitted by the sample.

Measurement of rat brain mitochondrial LPOs (nmol TBARS/mg brain mitochondrial protein) was done [13]. Superoxide dismutase SOD (U/mg mitochondrial protein) [14] and brain mitochondrial TAS was measured [15].

Rat brain mitochondrial BDNF (pg/ml), Drp-1 (ng/ml), Trk-B (pg/ml) and Fis-1 (ng/ml) proteins were measured according to manufacturer protocol. Protein expression of BDNF, Drp-1, Fis-1, and Trk-B were determined using Western blotting analysis. The apparent molecular weights of ~13, 82, 17 and 100 KDa, for BDNF, Drp-1, Fis-1, and Trk-B; respectively, were estimated. Actin was used as a control. Blots were probed with a β-actin antibody (1:300) as an internal control.

Statistical significance determined by ANOVA test (P=0.05 or lower were considered statistically significant) using GraphPad Prism 5 software (USA). Linear regression analysis was made between different parameters.

Results and Discussion

We generated LH depression model in rats. Rats were treated with CTL, IMP or FLX and exposed to non-escapable shock showed increase mean escape latencies. Mean escape latency time for 5 FR-1 (fixed ratio-1) trials and 25 FR-2 trials were showed in **Tables 1 and 2**. FR-2 mean latencies decreased significantly in rats received non-escapable shock after treatment with IMP, FLX or CTL **(Table 1)**. Analysis of FR-1 means escape latencies of tested animals showed insignificant differences among treated rats, suggesting that treatment with FLX, IMP or CTL had no effect on mean escape latency times **(Table 1)**. The FR-2 means were used to screen rats whether LH or NLH after they received tail shock stress. Rat received an electric shock and elicited mean FR-2 escape latency more than 2 standard deviations (SDs) higher than control FR-2 escape latency average was considered as helpless. NLH rats had a mean FR-2 escape latency records that were less than 1 SD from

the average FR-2 means of control **(Table 2)**. Rats elicited FR-2 mean escape latency records that lied between 1 and 2 SDs greater than control were omitted from statistical analysis of achieved data due to the difficulty to classify rats as LH or NLH **(Table 3)**. The order of decreasing number of LH was:- CTL>IMP>FLX **(Table 3)**. The rats received FLX and IMP were considered helpless indicated that two-week treatment was not enough to ameliorate helpless behaviour **(Table 3)**. Similarly, other investigator reported that not all animals that undergo non-escapable shock associated with high mean escape latencies became LH [16].

Table 1: Learned helplessness (LH) depression model-Mean FR-1 and FR-2 latencies (seconds) of rats (n=8) exposed to non-escapable tail chock.

Treatment	Average FR-1 mean (sec)	SD	Average FR-2 mean (sec)	SD
Saline	11	8	24.2	7.1
FLX	9.8	7.4	10.8*	7.4
IMP	9.3	7.2	12.3*	8.1
CTL	9.6	7.5	15.6**	7.8

Note: *$p<0.05$, **$p<0.01$, significant change difference in average FR-1 or FR-2 means compared with control

Table 2: Learned helplessness (LH) depression model- Mean FR-1 and FR-2 times of tested rats (n=8) for determination of LH and non-learned helplessness (NLH) behaviour.

Treatment	Average FR-1 mean (sec)	Average FR-2 mean (sec)	SD	Classification	
				LH	NLH
Saline	4.1	7.1	0.4	>7.5	<6.9
FLX	3.1	6.5	0.7	>8.5	<7.0
IMP	2.6*	5.9	0.5	>9.2	<8.5
CTL	2.2**	5.4	0.3	>7.7	<6.8

Note: *$p<0.05$, **$p<0.01$, significant change difference in average FR-1 or FR-2 means compared with control

Table 3: Learned helplessness (LH) depression model-LH and NLH numbers for each tested rat (n=8) received a non-escapable shock and treated with saline, fluoxetine (FLX), imipramine (IMP) or citalopram (CTL).

| Group treatment	No. of LH rats	No. of NLH rats	No. of rats between 1 and 2 SD	Total rats number
Saline	12	1	3	16
FLX	8	10	1	19
IMP	6	8	1	15
CTL	4	6	1	11

SDH showed a significant decline by -22.4% in case of DEP-rats. CTL DEP-rats showed a significant increase in SDH by +27.7%. Neither intake of FLX nor IMP to DEP-rats produced a significant change in SDH **(Figure 1A)**. MDH showed a significant decline by -42.7% in case of DEP-rats. FLX, IMP or CTL intake to DEP-rats showed significant increase in MDH by +32.9%, +40.2% and +68.4%, respectively **(Figure 1B)**. IDH showed a significant decline by -28.4% in case of DEP-rats. IMP or CTL intake to DEP-rats showed a significant increase in IDH by +20.7%, +34.9%, respectively. FLX intake to the DEP-rats produced an insignificant change in IDH **(Figure 1C)**. The NDH showed a significant decline by -39.1% in case of DEP-rats; p<0.0001. CTL DEP-rats showed a significant increase in NDH by +50%. Neither intake of FLX nor IMP to DEP-rats produced significant changes in NDH **(Figure 1D)**. NDH showed a significant decline by -59% for DEP-rats. CTL DEP-rats showed a significant increase in MAO by +110%. Neither intake of FLX or IMP to DEP-rats produced any significant change in MAO **(Figure 1E)**. These findings confirmed the fact that, psychiatric symptoms have been recorded in subjects with mitochondrial diseases [17]. CTL DEP-rats showed a significant increase in SDH, MDH, IDH, NDH, and MAO. IMP DEP-rats showed a significant increase in MDH and IDH. Besides, FLX DEP-rats showed an only a significant increase in MDH.

The percentage ATP production showed significant decrement by -40%, -35% and -33%, in P+M (pyruvate+malate), G+M (glutamate+malate) and S (succinate) substrates; respectively in case of DEP-rats. Intake FLX or IMP to DEP-rats showed a significant increase in brain %ATP production level by +16.6%

and +14.8%; respectively using P+M substrate. Intake of CTL to DEP-rats resulted in increase in % ATP production by +63.3%, +30.7% and +22.4%, in P+M, G+M and S groups; respectively **(Figure 2A)**. The intake of IMP and FLX enhanced ATP synthase activity [18], and thereby increased ATP production. The order in enhancing %ATP production levels after intake of antidepressants to DEP-rats was: FLX<IMP<CTL.
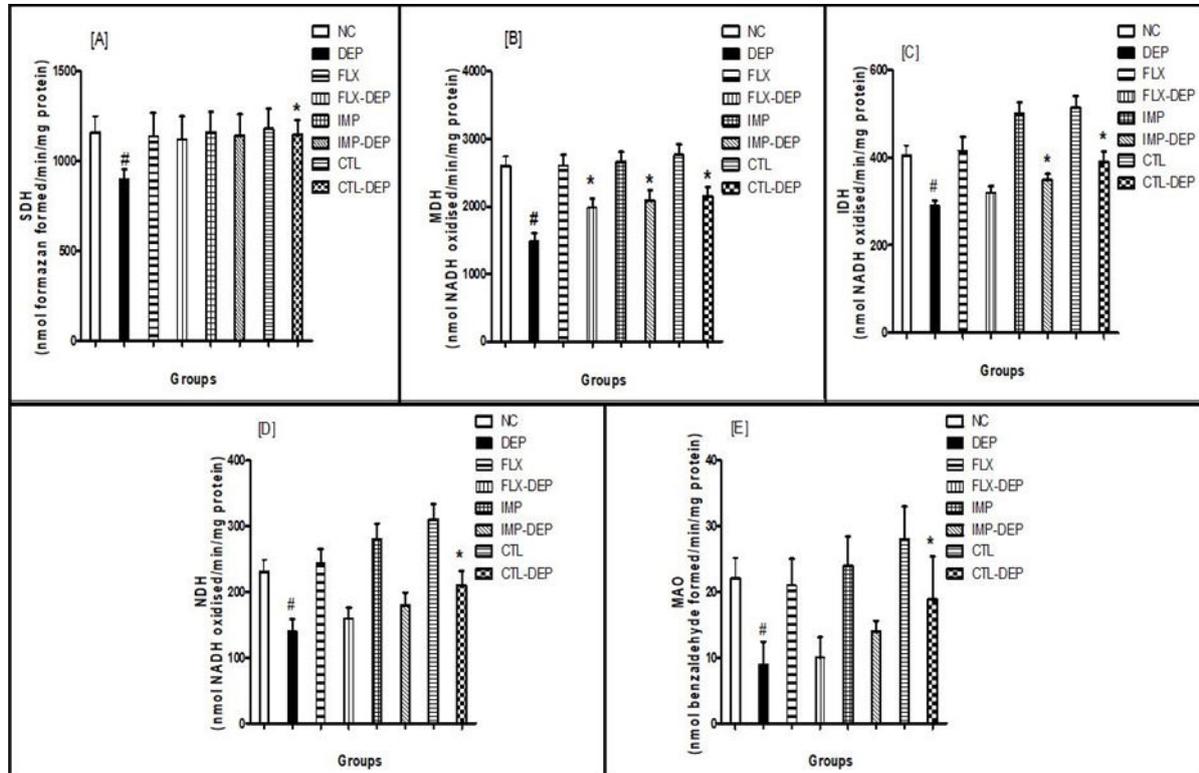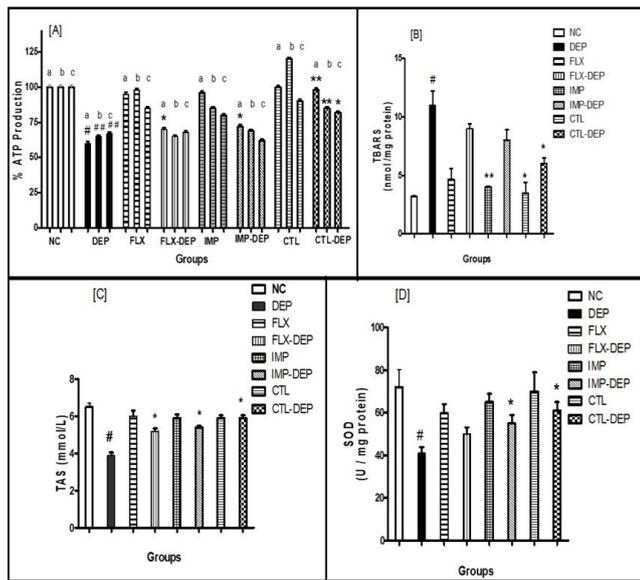

Figure 1: Effect of intake of fluoxetine (FLX), imipramine (IMP) or citalopram (CTL) to DEP-rats, on brain mitochondrial (A) succinate dehydrogenase (SDH); (B) malate dehydrogenase (MDH); (C) isocitrate dehydrogenase (IDH); (D) nicotinamide dehydrogenase (NDH); (E) monoamine oxidase (MAO) enzymes levels. Group 1: normal control (NC); Group 2: depressed rats (DEP); Group 3: FLX rats; Group 4: FLX-DEP rats; Group 5: IMP rats; Group 6: IMP-DEP rats; Group 7: CTL rats and Group 8: CTL-DEP rats. Data were presented as mean ± SE. A number of rats (n)=8 rats/group. *Significant difference from DEP-rats, p<0.01. #Significant difference from NC rats, p<0.001

LPOs level elicited significant increment by +343.7% among DEP-rats. Administration of FLX, IMP or CTL to DEP-rats showed a significant decline in TBARS by -18.2%, -27.3% and -45.5%, respectively **(Figure 2B)**, that was in congruence with the fact that, oxidative damage may be linked to depression [19]. LPOs increment in DEP-rats reflects enrollment of oxidative damage and disruption of mitochondria [20]. LPOs increase among DEP-rats subsequently initiates reactive oxygen species production, because the mitochondrial membrane is rich in polyunsaturated fatty acids and their configuration is facilitated by SOD [21]. Intake of antidepressants declines LPOs [22]. The order in decreasing LPOs after intake antidepressants to DEP-rats was: FLX<IMP<CTL.

The SOD showed a significant decrease by -43% in the case of DEP-rats. Intake of IMP or CTL to DEP-rats showed a significant increase in SOD by +34% and +48.8%. Intake of FLX to DEP-rats produced no significant change in SOD **(Figure 2D)**. It was reported that SOD was declined among depression patients and thereby strengthens oxidative damage of mitochondrial membrane due to the accumulation of superoxide radicals,

which in turn inhibit antioxidant enzymes [23]. Administration of either IMP or CTL to DEP-rats showed significant increment in brain SOD.

The TAS showed a significant decline by -40% in case of DEP-rats. However, TAS (mmol/L) elicited significant increment by +33.3%, +38.5% and +51.3%, for FLX-DEP, IMP-DEP and CTL-DEP, respectively **(Figure 2C)**. TAS elicited significant increment in FLX-DEP, IMP-DEP and CTL-DEP rats. It was reported that low levels of antioxidants and high LPOs were prominent in depression [24]. The order in decreasing SOD levels due to the intake of antidepressants to DEP-rats was: IMP<CTL; however, SOD in FLX-DEP rats was insignificantly changed. On another hand, declining order of TAS level was: FLX<IMP<CTL.
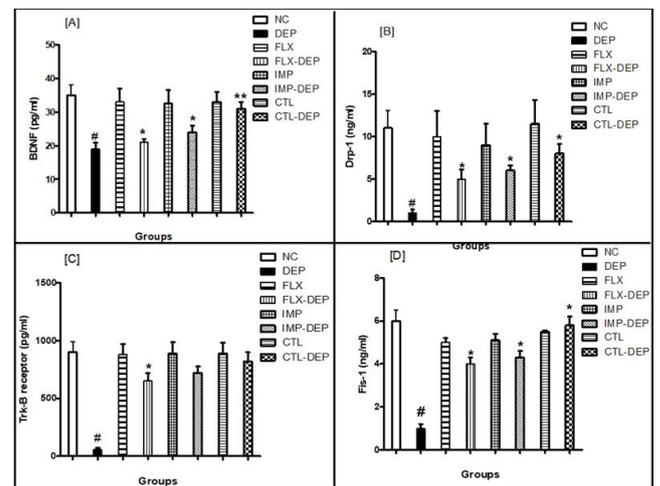
Figure 2: Effect of fluoxetine (FLX), imipramine (IMP) or citalopram (CTL) to DEP-rats, on (A) percentage of ATP production using (a) pyruvate+malate (b) glutamate+malate or (c) succinate; (B) thiobarbituric acid reactive substance (TBARS); (C) total antioxidant status (TAS); (D) SOD levels in rat brain mitochondria. Group 1: normal control (NC), Group 2: depressed rats (DEP), Group 3: FLX rats, Group 4: FLX-DEP rats, Group 5: IMP rats, Group 6: IMP-DEP rats, Group 7: CTL rats and Group 8: CTL-DEP rats. Data were presented as mean ± SE. The result is a duplicate/group. * A significant difference from DEP-rats, $p<0.05$. ** Significant difference from DEP-rats, $p<0.01$. # Significant difference from NC rats, $p<0.05$. ** A significant difference from DEP-rats, $p<0.05$. ## Significant difference from NC rats, $p<0.01$.

The DEP-rats showed a significant decline in BDNF level by -45.7%. BDNF level elicited significant increment by +15%, +26%, and +63%, for FLX-DEP, IMP-DEP and CTL-DEP; respectively **(Figure 3A)**. The low BDNF levels are linked with depression [25]. Meta-Analysis research studied BDNF during depression episodes concluded that BDNF in depression was significantly lower than control. Other mechanism showed that BDNF could be declined before the start of a depression attack [26]. On contrary, we showed that BDNF elicited significant increment for FLX-DEP, IMP-DEP and CTL-DEP rats. Similarly, elevation of BDNF after use of antidepressants was reported [27]. However, other study declared that impairment BDNF expression does not lead to depression, but affect the efficacy of antidepressants [28].

The Drp-1 showed a highly significant decline by -90% in the case of DEP-rats. Drp-1 elicited significant increment by 4, 5 and 7 folds, for FLX-DEP, IMP-DEP, and CTL-DEP; respectively **(Figure 3B)**. Trk-B receptor showed a significant decline by -93% in the case of DEP-rats. However, Trk-B receptor elicited significant increment by 10 folds in the case of FLX-DEP **(Figure 3C)**. Fis-1 significantly declined by 5 folds in the case of DEP-rats. Fis-1 elicited significant increment by 4, 3.3 and 4.8 folds, in the case of FLX-DEP, IMP-DEP, and CTL-DEP; respectively **(Figure 3D)**. The results obtained from our study showed significant increment in
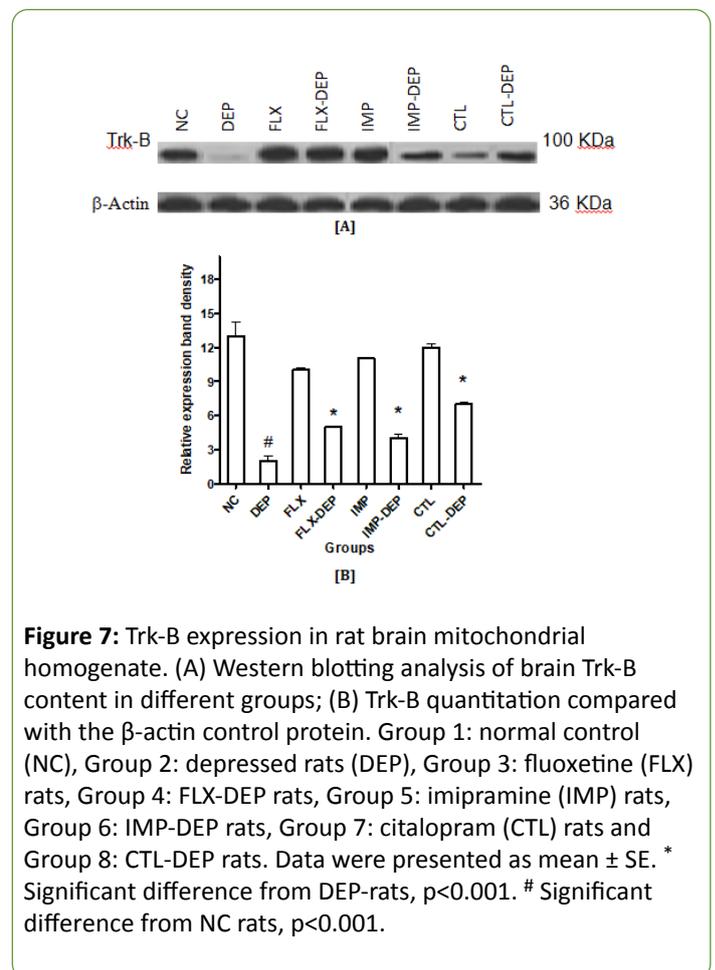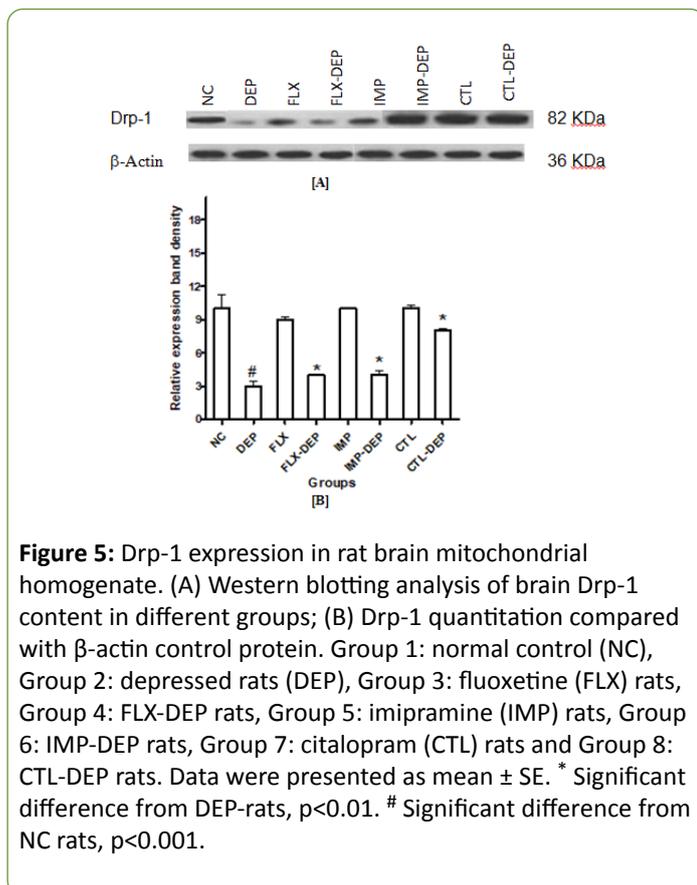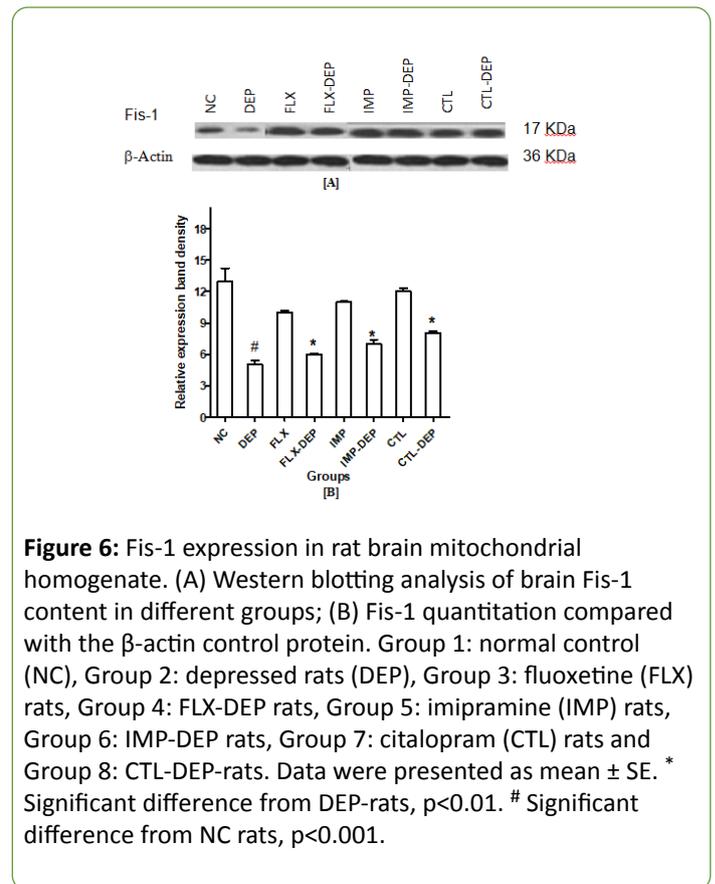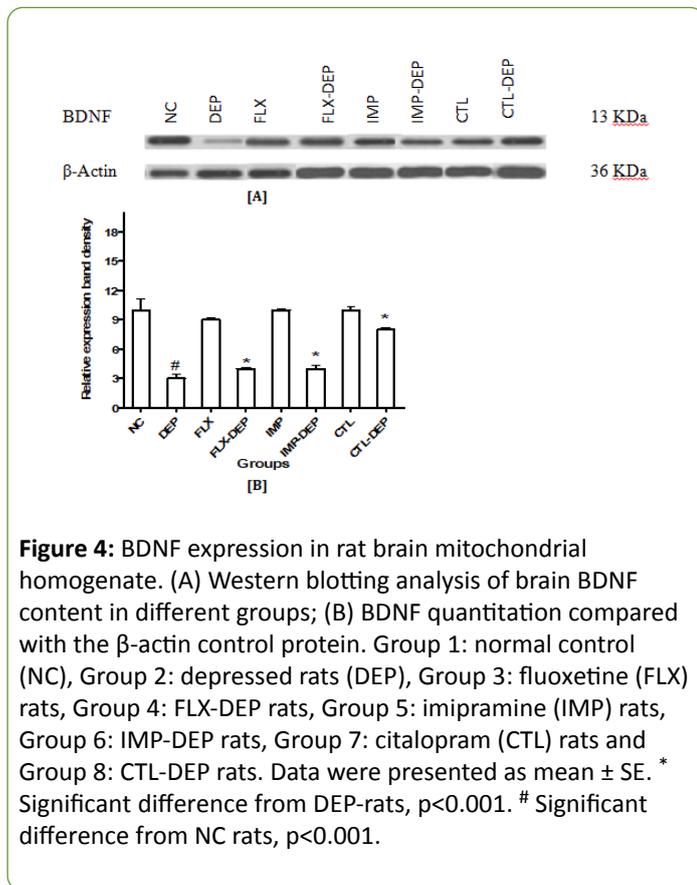
Drp-1 and Fis-1 in FLX-DEP, IMP-DEP and CTL-DEP groups, however, Trk-B was significantly increased in the case of FLX-DEP rats. In agreement with our results, depression resulted in a significant decline in BDNF, Drp-1, Fis-1, and Trk-B. Mitochondrial fission is mediated by Drp-1 and Fis-1 proteins to regulate apoptosis through the removal of injured mitochondria [28].

Figure 3: Effect of intake of fluoxetine (FLX), imipramine (IMP) or citalopram (CTL) to depressed (DEP) rats, on brain mitochondrial (A) Brain derived neurotrophic factor (BDNF); (B) Dynamin-Related Protein-1 (Drp-1); (C) Tyrosine Kinase B Receptor (Trk-B); (D) Fission-1 Protein (Fis-1) proteins levels. Group 1: normal control (NC), Group 2: depressed rats (DEP), Group 3: fluoxetine (FLX) rats, Group 4: FLX-DEP rats, Group 5: imipramine (IMP) rats, Group 6: IMP-DEP rats, Group 7: citalopram (CTL) rats and Group 8: CTL-DEP rats. Data were presented as mean ± SE. A number of rats (n)=8 rats/group. * Significant difference from DEP-rats, $p<0.05$. ** Significant difference from DEP rats, $p<0.001$. # Significant difference from NC rats, $p<0.001$.

In vitro expression of mitochondrial fission, genes lead to mitochondrial apoptosis, whereas expression of fusion genes protects cells from apoptosis [28]. Induction of depression in rats resulted in a significant decline in BDNF expression by -75% **(Figure 4A and 4B)**. FLX, IMP or CTL intake significantly increased BDNF expression by +67%, +100% and +333%; respectively **(Figure 4B)**. DEP-rats produced a non-significant decline in protein expression of Drp-1 by -70% **(Figure 5A and 5B)**. Intake of FLX, IMP or CTL significantly increased Drp-1expression by +33%, +33% and +62.5%; respectively **(Figure 5B)**. DEP-rats resulted in a significant reduction in protein expression of Fis-1 in rat brain by -60.5% **(Figure 6A and 6B)**. Intake of FLX, IMP or CTL significantly increased Fis-1 expression by +20%, +40% and +60%; respectively **(Figure 6B)**. DEP-rats resulted in a non-significant decline in protein expression of Trk-B by 15 folds **(Figure 7B)**. Intake of FLX, IMP or CTL resulted in non-significant changes in rats Trk-B expression. Intake of FLX, IMP or CTL significantly increased Trk-B expression by 2.5, 2 and 3.5 folds; respectively **(Figure 7A and 7B)**. Reduced levels of Drp-1 and

Fis-1 proteins among DEP-rats were in accordance with another study [29].

Figure 4: BDNF expression in rat brain mitochondrial homogenate. (A) Western blotting analysis of brain BDNF content in different groups; (B) BDNF quantitation compared with the β-actin control protein. Group 1: normal control (NC), Group 2: depressed rats (DEP), Group 3: fluoxetine (FLX) rats, Group 4: FLX-DEP rats, Group 5: imipramine (IMP) rats, Group 6: IMP-DEP rats, Group 7: citalopram (CTL) rats and Group 8: CTL-DEP rats. Data were presented as mean ± SE. * Significant difference from DEP-rats, $p < 0.001$. # Significant difference from NC rats, $p < 0.001$.

Figure 5: Drp-1 expression in rat brain mitochondrial homogenate. (A) Western blotting analysis of brain Drp-1 content in different groups; (B) Drp-1 quantitation compared with β-actin control protein. Group 1: normal control (NC), Group 2: depressed rats (DEP), Group 3: fluoxetine (FLX) rats, Group 4: FLX-DEP rats, Group 5: imipramine (IMP) rats, Group 6: IMP-DEP rats, Group 7: citalopram (CTL) rats and Group 8: CTL-DEP rats. Data were presented as mean ± SE. * Significant difference from DEP-rats, $p < 0.01$. # Significant difference from NC rats, $p < 0.001$.

Figure 6: Fis-1 expression in rat brain mitochondrial homogenate. (A) Western blotting analysis of brain Fis-1 content in different groups; (B) Fis-1 quantitation compared with the β-actin control protein. Group 1: normal control (NC), Group 2: depressed rats (DEP), Group 3: fluoxetine (FLX) rats, Group 4: FLX-DEP rats, Group 5: imipramine (IMP) rats, Group 6: IMP-DEP rats, Group 7: citalopram (CTL) rats and Group 8: CTL-DEP-rats. Data were presented as mean ± SE. * Significant difference from DEP-rats, $p < 0.01$. # Significant difference from NC rats, $p < 0.001$.

Figure 7: Trk-B expression in rat brain mitochondrial homogenate. (A) Western blotting analysis of brain Trk-B content in different groups; (B) Trk-B quantitation compared with the β-actin control protein. Group 1: normal control (NC), Group 2: depressed rats (DEP), Group 3: fluoxetine (FLX) rats, Group 4: FLX-DEP rats, Group 5: imipramine (IMP) rats, Group 6: IMP-DEP rats, Group 7: citalopram (CTL) rats and Group 8: CTL-DEP rats. Data were presented as mean ± SE. * Significant difference from DEP-rats, $p < 0.001$. # Significant difference from NC rats, $p < 0.001$.

Figure 8, showed linear regression analysis of TAS and: (A) BDNF and Trk-B in DEP-rats (r^2=+0.887 and +0.8277; respectively). (B) Drp-1 and Fis-1 in DEP-rats (r^2=+0.5317 and +0.8101; respectively). (C) BDNF and Trk-B in DEP-FLX rats (r^2= +0.8498 and +0.5823; respectively). (D) Drp-1 and Fis-1 in DEP-FLX rats (r^2= +0.8412 +0.9002; respectively). (E) BDNF and Trk-B in DEP-IMP rats (non-significant). (F) Drp-1 and Fis-1 in DEP-IMP rats (r^2 = +0.9297 and +0.932; respectively). (G) BDNF and Trk-B in DEP-CTL rats (non-significant). (H) Drp-1 and Fis-1 in DEP-CTL rats (r^2=+0.7553 and +0.5833; respectively). Strong correlations were showed between TAS and above parameters in all groups except in case of Trk-B in DEP-IMP and DEP-CTL rats. Our data elucidated correlation between antioxidant status and activation of mitochondrial biogenesis during use of antidepressants in DEP-rats.
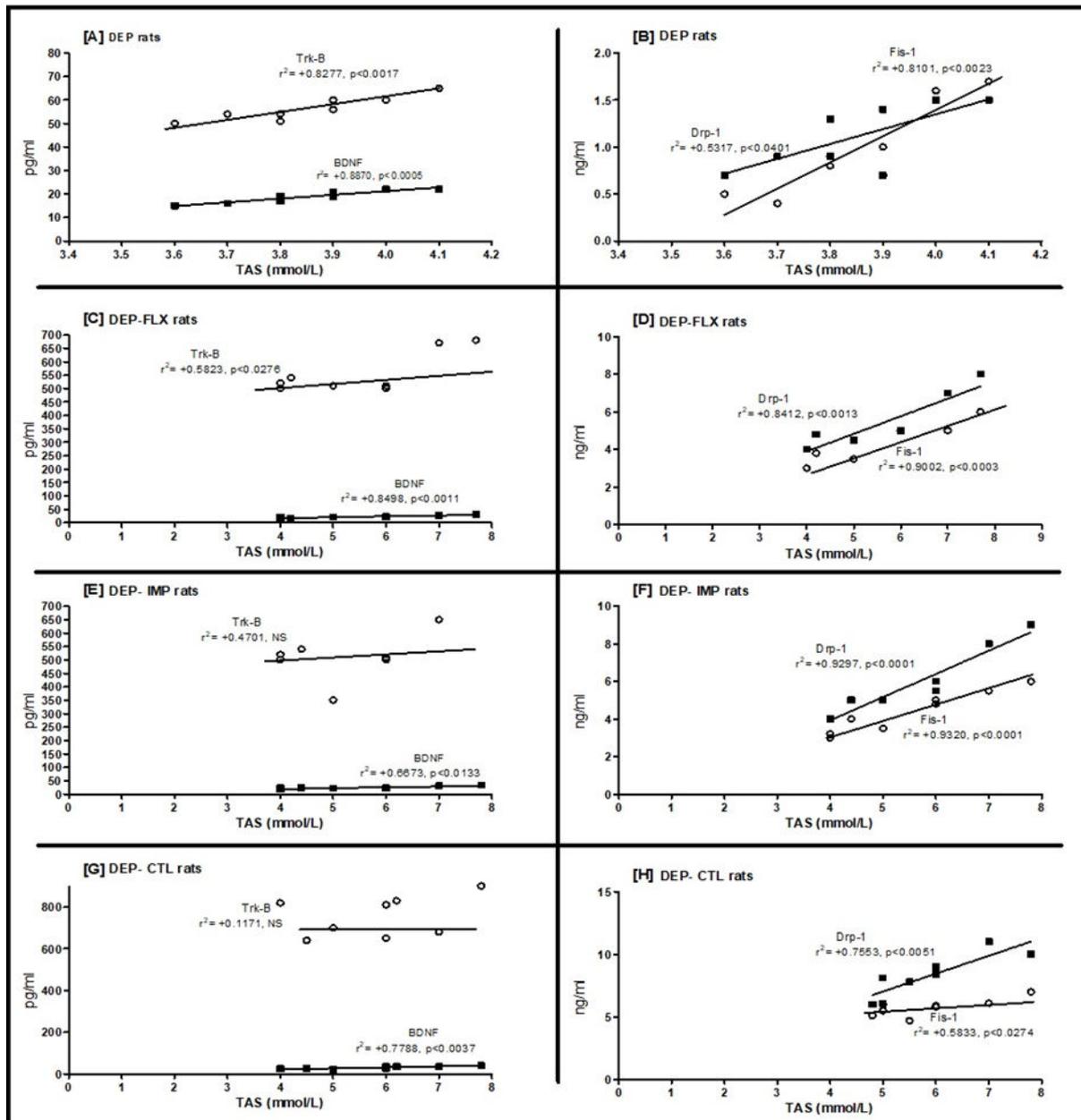
Figure 8: Linear regression analysis of TAS and: (A) BDNF and Trk-B in DEP-rats ; (B) Drp-1 and Fis-1 in DEP-rats; (C) BDNF and Trk-B in DEP-FLX rats; (D) Drp-1 and Fis-1 in DEP-FLX rats; (E) BDNF and Trk-B in DEP-IMP rats; (F) Drp-1 and Fis-1 in DEP-IMP rats; (G) BDNF and Trk-B in DEP-CTL rats; (H) Drp-1 and Fis-1 in DEP-CTL rats.

In conclusion, FLX, IMP or CTL alleviates depression in rats possibly via antioxidant mechanism and modulation of neurotrophins. We recommend studying the use of different antidepressants to gain insight density and distribution of mitochondria in brain regions.

Acknowledgment

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