

## Acute Effect of Sidr Leaves Extract on some Neurotransmitter Contents in Different Brain Areas of Male Albino Rats

Abeer M. Waggas

Department of Zoology, Faculty of Girls Education, Scientific Departments, Saudi Arabia

### Abstract

The acute i.p. injection of Sidr (*Zizyphus spina-christi*) leaves extract (100 mg / kg body wt) caused a significant increase in epinephrine (E), norepinephrine (NE), dopamine (DA), serotonin (5-HT), 5-hydroxyindoleacetic acid (5-HIAA) and a significant decrease in gamma-aminobutyric acid (GABA) content in different brain areas (cerebellum, pons plus medulla oblongata, striatum, cerebral cortex, hypothalamus, midbrain and hippocampus) of male albino rats. The increase in E, NE, DA, 5-HT, and 5-HIAA content in the different CNS areas of albino rat may be due to the inhibition of  $Ca^{2+}$ /calmodulin binding which plays an important role in the release of these neurotransmitters, at the same time GABA inhibits the release of these neurotransmitters by increasing the permeability to Cl ions and such effect may be due to the presence of peptide and cyclopeptide alkaloids in the extract. *Zizyphus spina-christi* leaves may potentially be safe for use as a sedative drug.

**Key words :** Zizyphus, E, NE, DA, 5-HT, 5-HIAA, GABA, Brain, Rat.

### Introduction

The plant *Zizyphus spina-christi* Willd is of the Rhamnaceae family, and it grows wild in northern Africa, Australia and tropical America (Irvine, 1961; Taeckholm, 1974; Hong, 1987). *Zizyphus spina-christi* (ZSS) is a subtropical plant known in Saudi Arabia as 'Sidr'. It is characterized by thorny branches used as a hedge to form defensive enclosure. The fruit has sweet edible pulp (Nawwar *et al.*, 1984; Grieve, 1998). Medicinally, the leaves are applied locally to sores (Dalziel, 1937) as wound powder and antiseptics (Fleurentin and Pelt, 1982) and was reported to exhibit hypoglycemic activity against streptozotocin diabetic rats (Anand *et al.*, 1989; Glombitza *et al.*, 1994; Abdel-Zaher *et al.*, 2005) and antibacterial efficacy against Gram-positive strains (Ali *et al.*, 2001; Suksamrarn *et al.*, 2006). While its root bark is used in folk medicine as remedy against pain (Adzu *et al.*, 2002; Adzu *et al.*, 2003). In Saudi Arabia folk medicine, the leaves are used to heal wounds, treat some skin diseases, some inflammatory conditions, sores,

against ringworm, fever, sex disease and ulcer (Blatter, 1978; Shahat *et al.*, 2001).

Since *Zizyphus spina-christi* is a wild tree commonly available in Saudi Arabia and its leaves are used in folk medicine for treatment, it is therefore deemed interesting to examine the effect of acute administration of *Zizyphus spina-christi* leaves extract on epinephrine (E), norepinephrine (NE), dopamine (DA), serotonin (5-HT), 5-hydroxyindoleacetic acid (5-HIAA) and gamma-aminobutyric acid (GABA) contents in different brain areas of male albino rats.

### Material and methods

The leaves of *Zizyphus spina-christi* were collected from trees growing in Jeddah (Saudi Arabia) in August 2005.

#### 1. preparation of the plant extract

Aqueous extract: The leaves were carefully removed, washed, and cut into pieces. They were weighed (100g)

out in a beaker and cold distilled water poured into them to give a final volume of 200 ml as reported previously (Adzu *et al.*, 2001). This mixture was grinded using an electric grinder, and shacked for 24 h at room temperature, then filtered through filter paper, and use as such.

## 2. Animals

The experimental animals used in this study were adult male albino rats, *Rattus rattus* (100 - 120 g). They were supplied with food and water *ad libitum* under standard conditions of light, humidity and temperature (22-25°C). The animals were randomly divided into two groups.

The first group (n=6) was treated with saline vehicle and killed at the beginning of the experiment and served as a control. The second group (n=36) was injected (i.p.) with 100mg/kg of *Zizyphus Spina-christi* leaves extract (Adzu *et al.*, 2001) and six rats were decapitated after 1, 2, 4, 8, 12 and 24 hr post injection.

The rat was killed by sudden decapitation at the designed times. The brain was rapidly and carefully excised and then dissected on dry ice glass plate according to the method of Glowinski and Iversen (1966) into the following regions: cerebellum, pons plus medulla oblongata, striatum, cerebral cortex, hypothalamus, midbrain and hippocampus. The brain tissues were wiped dry with a filter paper, weighed,

wrapped in plastic films and then in aluminum foil and quickly frozen in dry ice pending analysis.

E, NE, DA and 5-HT were extracted and estimated according to the method of Chang (1964) modified by Ciarlone (1978). 5-HIAA was estimated according to the method described by Miller *et al.*, (1970). GABA was estimated according to the method of Sutton and Simmodes (1973). The fluorescence was measured in Jenway 6200 fluorometer.

## 3. Statistical analysis

The data are presented as mean  $\pm$  S.E. Statistical analyses between control and treated animals were performed using paired student 't' (Hill, 1971).

## Result

The single i.p. injection of 100 mg/kg of *Z. spina-christi* leaves extract significantly increased the E content in cerebellum, pons + medulla oblongata, midbrain and hippocampus at all time intervals; in striatum and cerebral cortex at 1 to 12 h; and in hypothalamus at 1 and 2 h following the injection (Table 1).

There was a significant increase in NE content in cerebellum, pons + medulla oblongata, hypothalamus, midbrain and hippocampus at all time intervals ;in striatum at 1 to 12 h; and in cerebral cortex at 1 to 8 h following the injection (Table 2).

**Table 1** :Effect of acute administration of *Zizyphus spina-christi* leaves extract (100mg/kg , i.p.) on epinephrine ( E)content in different brain areas of albino rat.

Time of decapitation	Cerebellum $\mu\text{g}\pm\text{S.E}$	Pons+medulla oblongata $\mu\text{g}\pm\text{S.E}$	Striatum $\mu\text{g}\pm\text{S.E}$	Cerebral cortex $\mu\text{g}\pm\text{S.E}$	Hypothalamus $\mu\text{g}\pm\text{S.E}$	Midbrain $\mu\text{g}\pm\text{S.E}$	Hippocampus $\mu\text{g}\pm\text{S.E}$	
Control	360.14 $\pm$ 5.58	471.76 $\pm$ 12.55	778.03 $\pm$ 11.71	127.14 $\pm$ 1.88	826.97 $\pm$ 12.05	462.65 $\pm$ 18.55	676.05 $\pm$ 9.41	
Treatment	1 hr %	422.54 $\pm$ 12.09 17.32*	529.79 $\pm$ 13.45 12.30*	915.14 $\pm$ 11.51 17.62*	140.80 $\pm$ 2.14 10.74*	925.26 $\pm$ 9.23 10.62*	529.07 $\pm$ 11.54 14.35*	786.03 $\pm$ 9.01 16.26*
	2 hr %	587.96 $\pm$ 11.98 63.25*	653.57 $\pm$ 10.24 38.53*	1149.99 $\pm$ 32.63 47.80*	155.81 $\pm$ 2.32 33.56*	1052.46 $\pm$ 13.40 27.26*	692.43 $\pm$ 7.22 49.66*	905.55 $\pm$ 7.79 33.94*
	4 hr %	432.01 $\pm$ 14.92 19.95*	552.17 $\pm$ 14.88 17.04*	949.92 $\pm$ 11.41 22.09*	135.08 $\pm$ 3.06 21.97*	867.74 $\pm$ 14.18 4.69	554.94 $\pm$ 17.23 19.94*	773.74 $\pm$ 12.21 14.45*
	8 hr %	419.60 $\pm$ 15.67 16.51*	570.25 $\pm$ 13.20 20.87*	898.94 $\pm$ 11.94 15.54*	145.15 $\pm$ 1.72 14.16*	882.86 $\pm$ 8.64 6.75	522.01 $\pm$ 15.75 19.31*	798.21 $\pm$ 13.87 18.06*
	12 hr %	412.35 $\pm$ 12.00 14.49*	521.79 $\pm$ 6.93 10.60*	877.61 $\pm$ 11.74 14.08*	144.45 $\pm$ 2.07 11.98*	879.27 $\pm$ 13.02 6.32	521.92 $\pm$ 14.15 12.81*	744.70 $\pm$ 8.92 10.15*
	24 hr %	405.42 $\pm$ 11.61 12.57*	581.16 $\pm$ 9.16 11.37*	819.25 $\pm$ 9.67 5.29	130.95 $\pm$ 0.96 2.99	821.12 $\pm$ 9.75 -0.70	513.80 $\pm$ 9.44 11.05*	777.29 $\pm$ 12.52 14.97*

Statistical analyses were performed between control (C=6) and treated ( T=6) animals by using paired t' test.

% : percentage of change from control. \* significant at p <0.01 .

**Table 2 :** Effect of acute administration of *Zizyphus spina-christi* leaves extract (100mg/kg , i.p.) on norepinephrine ( NE ) content in different brain areas of albino rat.

Time of decapitation		Cerebellum $\mu\text{g}\pm\text{S.E}$	Pons+medulla oblongata $\mu\text{g}\pm\text{S.E}$	Striatum $\mu\text{g}\pm\text{S.E}$	Cerebral cortex $\mu\text{g}\pm\text{S.E}$	Hypothalamus $\mu\text{g}\pm\text{S.E}$	Midbrain $\mu\text{g}\pm\text{S.E}$	Hippocampus $\mu\text{g}\pm\text{S.E}$
Control		330.04 $\pm$ 7.85	448.88 $\pm$ 4.34	787.17 $\pm$ 11.24	124.54 $\pm$ 1.62	784.19 $\pm$ 9.38	461.44 $\pm$ 10.91	642.94 $\pm$ 12.34
Treatment	1 hr %	444.36 $\pm$ 10.74 34.63*	566.98 $\pm$ 6.52 26.30*	932.03 $\pm$ 9.53 18.40*	141.65 $\pm$ 1.84 13.73*	870.57 $\pm$ 11.61 11.01*	537.35 $\pm$ 13.98 16.45*	766.87 $\pm$ 10.42 19.27*
	2 hr %	639.59 $\pm$ 9.94 53.19*	645.44 $\pm$ 11.70 43.78*	1118.56 $\pm$ 23.01 42.09*	168.31 $\pm$ 2.60 35.14*	1053.80 $\pm$ 13.71 34.38*	699.05 $\pm$ 8.93 51.49*	876.24 $\pm$ 8.08 36.28*
	4 hr %	409.14 $\pm$ 8.22 23.96*	511.26 $\pm$ 9.39 13.89*	910.99 $\pm$ 11.71 15.72*	137.14 $\pm$ 1.90 10.11*	913.73 $\pm$ 7.71 16.51*	538.52 $\pm$ 14.67 16.70*	723.99 $\pm$ 12.00 12.60*
	8 hr %	391.99 $\pm$ 14.79 18.77*	501.10 $\pm$ 16.13 11.63*	879.40 $\pm$ 12.92 11.71*	138.37 $\pm$ 1.38 11.10*	886.41 $\pm$ 20.63 13.03*	520.931 $\pm$ 17.72 12.89*	725.91 $\pm$ 14.05 12.90*
	12 hr %	383.53 $\pm$ 15.05 16.20*	522.07 $\pm$ 13.93 14.97*	944.50 $\pm$ 12.23 19.98*	131.16 $\pm$ 2.07 5.31	904.28 $\pm$ 9.24 15.31*	522.16 $\pm$ 11.46 13.15*	722.47 $\pm$ 11.81 12.36*
	24 hr %	377.77 $\pm$ 15.06 14.46*	503.16 $\pm$ 15.27 12.09*	847.36 $\pm$ 16.02 7.64	128.67 $\pm$ 0.82 3.31	920.37 $\pm$ 10.27 17.36*	509.44 $\pm$ 11.89 10.40*	762.78 $\pm$ 12.81 18.63*

Statistical analyses were performed between control (C=6) and treated (T=6) animals by using paired *t'* test.

% : percentage of change from control. \* significant at  $p < 0.01$  .

**Table 3 :** Effect of acute administration of *Zizyphus spina-christi* leaves extract (100mg/kg , i.p.) on dopamine ( DA ) content in different brain areas of albino rat .

Time of decapitation		Cerebellum $\mu\text{g}\pm\text{S.E}$	Pons+medulla oblongata $\mu\text{g}\pm\text{S.E}$	Striatum $\mu\text{g}\pm\text{S.E}$	Cerebral cortex $\mu\text{g}\pm\text{S.E}$	Hypothalamus $\mu\text{g}\pm\text{S.E}$	Midbrain $\mu\text{g}\pm\text{S.E}$	Hippocampus $\mu\text{g}\pm\text{S.E}$
Control		348.43 $\pm$ 5.14	457.92 $\pm$ 12.43	778.43 $\pm$ 13.20	125.71 $\pm$ 1.41	773.02 $\pm$ 12.26	449.46 $\pm$ 11.90	637.40 $\pm$ 12.90
Treatment	1 hr %	451.86 $\pm$ 13.11 29.68*	523.24 $\pm$ 12.86 14.26*	924.69 $\pm$ 11.91 18.78*	142.56 $\pm$ 1.68 13.40*	853.71 $\pm$ 12.39 10.43*	517.18 $\pm$ 9.74 15.06*	729.37 $\pm$ 10.39 14.42*
	2 hr %	510.62 $\pm$ 10.14 46.54*	555.22 $\pm$ 16.04 21.24*	1051.19 $\pm$ 13.94 35.03*	141.95 $\pm$ 2.19 18.48*	947.08 $\pm$ 10.60 22.51*	565.74 $\pm$ 14.33 25.87*	766.87 $\pm$ 12.15 20.31*
	4 hr %	444.50 $\pm$ 12.77 27.57*	551.89 $\pm$ 14.78 20.52*	952.66 $\pm$ 13.15 22.38*	146.79 $\pm$ 2.32 16.76*	889.38 $\pm$ 21.79 15.05*	549.31 $\pm$ 16.73 22.21*	727.69 $\pm$ 13.01 14.16*
	8 hr %	409.04 $\pm$ 14.47 17.39*	522.46 $\pm$ 17.15 14.09*	861.78 $\pm$ 11.38 10.70*	139.78 $\pm$ 1.67 11.19*	863.25 $\pm$ 12.12 11.67*	540.14 $\pm$ 11.41 20.17*	728.36 $\pm$ 15.25 12.33*
	12 hr %	417.91 $\pm$ 12.49 19.94*	510.04 $\pm$ 12.50 11.38*	835.48 $\pm$ 12.32 7.32	136.87 $\pm$ 1.59 8.87	809.81 $\pm$ 16.45 4.50	524.28 $\pm$ 11.87 16.64*	738.21 $\pm$ 11.29 15.81*
	24 hr %	408.71 $\pm$ 15.48 17.30*	519.62 $\pm$ 12.31 13.47*	788.20 $\pm$ 8.61 1.25	129.33 $\pm$ 1.25 2.87	777.73 $\pm$ 13.26 0.60	505.45 $\pm$ 12.40 12.45*	724.34 $\pm$ 13.35 13.63*

Statistical analyses were performed between control (C=6) and treated (T=6) animals by using paired *t'* test.

% : percentage of change from control. \* significant at  $p < 0.01$  .

Data in Table 3 show that the injection of 100 mg/kg of *Z. spina-christi* leaves extract caused a significant increase in the DA content in cerebellum, pons + medulla oblongata, midbrain and hippocampus at all time intervals; and in striatum, cerebral cortex and hypothalamus at 1 to 8 h following the injection.

Moreover, the 100 mg/kg injection significantly

increased the 5-TH content in cerebellum, pons+medulla oblongata, midbrain and hippocampus at all time intervals; in striatum at 1 to 8 h; and in cerebral cortex and hypothalamus at 1 to 4 h following the injection (Table 4).

Furthermore, the 100 mg/kg injection significantly increased the 5-HIAA content in pons+medulla

**Table 4 :** Effect of acute administration of *Zizyphus spina-christi* leaves extract (100mg/kg , i.p.) on serotonin ( 5-TH )content in different brain areas of albino rat .

Time of decapitation		Cerebellum μ/g±S.E	Pons+medulla oblongata μ/g±S.E	Striatum μ/g±S.E	Cerebral cortex μ/g±S.E	Hypothalamus μ/g±S.E	Midbrain μ/g±S.E	Hippocampus μ/g±S.E
<b>Control</b>		319.87±5.30	458.29±15.74	794.27±8.62	125.88±0.98	754.59±7.91	453.07±13.41	667.61±4.23
<b>Treatment</b>	<b>1 hr %</b>	420.74±8.83 31.53*	541.13±8.98 18.07*	884.55±10.89 11.36*	186.53±2.15 10.84*	847.32±9.01 12.28*	509.71±17.74 12.50*	735.57±12.75 10.17*
	<b>2 hr %</b>	500.04±8.06 56.32*	579.87±14.20 26.52*	954.86±15.26 20.21*	140.49±2.16 11.60*	883.31±8.13 17.05*	554.94±10.25 22.48*	784.09±10.36 17.44*
	<b>4 hr %</b>	440.69±10.68 37.77*	535.77±13.37 16.90*	912.22±13.89 14.85*	143.64±1.03 14.10*	864.73±6.52 14.59*	527.28±11.61 16.37*	764.39±8.15 14.49*
	<b>8 hr %</b>	357.88±9.54 11.88*	517.19±11.54 12.85*	882.22±10.04 11.07*	133.47±1.47 6.02	805.76±10.00 6.78	506.75±9.66 11.84*	737.49±11.93 10.46*
	<b>12 hr %</b>	361.85±8.04 13.12*	548.16±13.30 19.60*	774.45±12.81 -2.49	124.52±1.44 -1.08	768.60±17.50 1.85	545.92±14.82 20.49*	741.20±11.03 11.02*
	<b>24 hr %</b>	366.16±7.14 14.47*	515.08±11.70 12.39*	808.92±10.60 1.84	127.45±1.13 1.24	781.64±11.54 3.58	515.64±14.48 13.81*	741.68±9.72 11.09*

Statistical analyses were performed between control (C=6) and treated ( T=6) animals by using paired *t'* test.  
% : percentage of change from control. \* significant at p <0.01 .

**Table 5 :** Effect of acute administration of *Zizyphus spina-christi* leaves extract (100mg/kg , i.p.) on 5-hydroxyindoleacetic acid (5-HIAA)content in different brain areas of albino rat .

Time of decapitation		Cerebellum μ/g±S.E	Pons+medulla oblongata μ/g±S.E	Striatum μ/g±S.E	Cerebral cortex μ/g±S.E	Hypothalamus μ/g±S.E	Midbrain μ/g±S.E	Hippocampus μ/g±S.E
<b>Control</b>		349.20±9.28	458.76±7.06	768.92±14.90	127.65±2.17	759.18±11.44	476.49±8.80	649.12±11.00
<b>Treatment</b>	<b>1 hr %</b>	432.47±9.02 23.84*	527.29±9.94 14.93*	849.85±10.37 10.52*	148.40±1.88 16.25*	858.71±7.30 13.11*	558.59±9.06 17.23*	739.81±9.87 13.97*
	<b>2 hr %</b>	487.07±11.27 39.48*	582.29±12.48 26.92*	901.38±13.32 17.22*	155.05±1.56 21.46*	869.26±11.30 14.49*	578.09±10.73 21.32*	785.80±9.48 21.05*
	<b>4 hr %</b>	416.59±9.87 19.29*	513.48±11.81 11.92*	848.83±10.78 10.39*	141.49±1.93 10.84*	849.13±20.40 11.84*	527.11±9.90 10.62*	737.42±10.71 13.60*
	<b>8 hr %</b>	432.22±11.43 23.77*	560.35±13.32 22.14*	856.15±11.36 11.34*	126.50±1.36 -0.90	821.16±12.74 8.16	537.64±11.34 12.83*	753.78±10.43 16.12*
	<b>12 hr %</b>	351.22±9.92 0.57	555.98±11.75 21.19*	783.45±10.71 1.88	128.54±1.09 0.69	777.56±12.69 2.42	547.64±12.14 14.93*	734.22±8.47 13.11*
	<b>24 hr %</b>	353.30±11.08 1.17	515.57±13.46 12.38*	754.57±12.09 -1.86	131.84±1.04 3.28	791.20±8.18 4.21	531.19±12.99 11.47*	741.24±13.01 14.19*

Statistical analyses were performed between control (C=6) and treated ( T=6) animals by using paired *t'* test.  
% : percentage of change from control. \* significant at p <0.01 .

oblongata, midbrain and hippocampus at all time intervals; in cerebellum and striatum at 1 to 8 h; and in cerebral cortex and hypothalamus at 1 to 4 h following the injection (Table 5).

However, the 100 mg/kg injection significantly decreased the GABA content in cerebellum, pons +

medulla oblongata, cerebral cortex, midbrain and hippocampus at all time intervals ; and in striatum and hypothalamus at 1 to 8 h following the injection (Table 6).

Data in Table 1-5 show that the maximum increase in E, NE, DA, 5-TH and 5-HIAA contents in all areas

**Table 6** : Effect of acute administration of *Zizyphus spina-christi* leaves extract (100mg/kg , i.p.) on gamma-aminobutyric acid (GABA) content in different brain areas of albino rat .

Time of decapitation	Cerebellum μ/g±S.E	Pons+medulla oblongata μ/g±S.E	Striatum μ/g±S.E	Cerebral cortex μ/g±S.E	Hypothalamus μ/g±S.E	Midbrain μ/g±S.E	Hippocampus μ/g±S.E	
Control	± 355.25 13.77	9.58 ± 460.76	± 842.12 13.95	1.97 ± 129.55	11.02 ± 845.98	± 461.30 13.60	7.33 633.55±	
Treatment	1 hr %	321.25±12.01 -9.57	401.55±12.12 -12.85*	805.92±16.61 -4.29	115.31±1.31 -10.21*	808.31±12.31 -4.45	383.56±17.33 -16.85*	591.31±18.71 -6.66
	2 hr %	276.32±9.12 -22.21*	359.21±11.67 -22.03*	727.25±21.31 -13.64*	105.32±1.31 -18.70*	736.52±18.71 -12.93*	325.31±16.22 -29.47*	505.26±18.44 -20.24*
	4 hr %	253.12±14.13 -28.74*	325.87±9.31 -29.27*	714.56±18.71 -15.14*	101.25±0.83 -21.84*	715.64±26.13 -15.40*	329.25±13.31 -28.62*	515.74±13.17 -18.59*
	8 hr %	274.31±8.17 -22.78*	349.88±13.31 -24.06*	748.31±17.03 -11.13*	110.31±1.11 -14.85*	749.11±18.97 -11.45*	349.23±17.33 -24.29*	520.31±19.22 -17.87*
	12 hr %	255.41±10.21 -28.10*	356.21±10.91 -22.69*	777.44±15.12 -7.68	111.31±1.01 -14.07*	825.91±17.72 -2.37	341.31±12.71 -26.01*	545.61±17.35 -13.88*
	24 hr %	235.19±13.31 -33.79*	334.01±7.03 -27.50*	825.31±17.31 -1.99	112.39±1.31 -13.24*	872.32±19.21 3.11	333.31±11.34 -27.74*	569.56±13.42 -10.10*

Statistical analyses were performed between control (C=6) and treated (T=6) animals by using paired *t*' test.  
% : percentage of change from control. \* significant at  $p < 0.01$  .

of the CNS examined was observed at 2 h following the 100 mg/kg injection. Moreover, data in Table 6 show that the maximum reduction in GABA content was observed at 2 h in midbrain and hippocampus and at 4 h in the rest of the examined areas of the CNS.

## Discussion

*Zizyphus spina-christi* (ZSS) has been used as an anti-convulsant and a hypnotic material in oriental countries due to its CNS inhibitory activity ( Yu-ching , 1983; Han *et al.*, 1986; Hung, 1999; Shou *et al.*, 2002; Zhang *et al.*, 2003; Park *et al.*, 2004).

Adzu *et al.*, (2002) studied the effect of *Zizyphus spina-christi* aqueous extract (100, 200 mg/kg, i. p) on the central nervous system in mice. They observed that the aqueous extract of *Zizyphus spina-christi* root bark may have some sedative activity. That was evident from the marked inhibition of the exploratory behavior, spontaneous motor activity (SMA) and prolonged sleeping time in mice. Those results suggest that the extract contained some constituents that depress the central nervous system. Such findings correlate with observation of Morishita *et al.*, (1987) on the aqueous extract of *Zizyphus* seeds.

The effects of various sedative cyclopeptides and peptide alkaloids from *Zizyphus* species on calmodulin-dependent  $Ca^{2+}$ -ATPase, protein kinase II and phosphodiesterase were investigated by Ikram and Tomlinson, 1976; Klee *et al.*, 1980; Shah *et al.*, 1986; Han *et al.*, 1990; Ebdel-Galil and El-jissry, 1991; Cheng *et al.*, 2000; Hawang *et al.*, 2001; Shahat *et al.*, 2001 Tripathi *et al.*, 2001.

Calmodulin is an ubiquitous cytosolic protein that plays a critical role in regulating cellular function by altering the activity of a large number of ion channels and activity of several calcium-dependent enzyme such as adenylyl cyclase, protein kinase II,  $Ca^{2+}$ -ATPase and phosphodiesterase (Klee *et al.*, 1980).

From the present result; it is clear that the acute injection of 100mg/kg (i.p)of *Zizyphus spina-christi* leaves extract caused a significant increase in epinephrine (E), norepinephrine (NE), dopamine (DA), serotonin (5-HT) and 5-hydroxyindoleacetic acid (5-HIAA); and a significant decrease in gamma-aminobutyric acid (GABA) content in different brain areas at different time intervals. The increase observed may be due to the important role of GABA that inhibits the release of these neurotransmitters. Previous studies indicated that administration of the *Zizyphus spina-christi* extract

has an effects on calmodulin-dependent  $Ca^{2+}$  (Han *et al.*, 2005). The role of  $Ca^{2+}$  / calmodulin in regulating neurotransmitter-mediated inhibition and stimulation brain areas (Fujisawa *et al.*, 1984; Cooper *et al.*, 1988; Dzhura *et al.*, 2000 ; Haeseleer *et al.*, 2002; Nadif Kasri *et al.*, 2003). So, *Zizyphus* could act as a competitive inhibitor of the link between calcium and calmodulin. which plays an important role in the release of these neurotransmitters.

Calcium regulation of ion channel activity is mediated by the ubiquitous calcium sensor protein, calmodulin (Lavitan, 2004). Calmodulin is tethered constitutively to the intracellular carboxyl-terminal tail domain of this channel in a calcium –independent manner. When the channel is opened by membrane depolarization, calcium ions rush in through the open channel and bind to the tethered calmodulin. The  $Ca^{2+}$ , by binding with calmodulin, causes the vesicles filled with neurotransmitters to migrate towards the presynaptic membrane, the neurotransmitter is released into the synaptic cleft and binds with receptor channel membranes that are located in both presynaptic and postsynaptic membranes (Dzhura *et al.*, 2000; Nadif Kasri *et al.*, 2004).

GABA is the chief inhibitory neurotransmitter in CNS (Roberts and Frankel, 1950;. Patel *et al.*, 2005) and it is particularly abundant in presynaptic region which contains neurotransmitter storage vesicles (Liu *et al.*, 1989; Storm-Mathisen,1992).

GABA plays an important role in the release of the neurotransmitter. It initiates the action of the natural inhibitory transmitter by increasing the permeability to Cl ions, thus decreasing the propability of release of quanta of excitatory transmitter (Roberts, 1974).

From the previous studies as well as from present results, it could be concluded that the acute injection of 100mg/kg of of *Zizyphus spina-christi* extract caused an increase in neurotransmitter (E, NE, DA, 5-HT, 5-HIAA) contents which may be in part, due to inhibition of calmodulin-dependent calcium – ATPase and phosphodiesterase by cyclopeptides and peptide alkaloids (Han *et al.*, 2005). These agents share the ability to depress excitable tissue at all levels of the CNS, leading to a decrease in the amount of transmitter released by the nerve impulse, as well as to general depression of postsynaptic responsiveness and ion

movement (Levin and Weiss, 1979; Bloom, 2001), as a result the content of neurotransmitters is increased. At the same time the *Zizyphus spina-christi* extract also caused inhibition of  $Ca^{2+}$  / calmodulin binding (Han *et al.*, 2005) which decreases the neurotransmitter release, as a result the content of neurotransmitters is increased. Also, GABA increasing the permeability to Cl ions thus decreasing the propability of release of quanta of excitatory transmitter.

From the present results, it is clear that the acute administration of *Zizyphus spina-christi* leaves extract caused a significant increase in neurotransmitter contents in most of the tested brain areas at different time intervals; cerebellum which is responsible for the voluntary movement; pons + medulla oblongata which is responsible of essential reflexive acts; striatum which is a brain region responsible for motor activity; cerebral cortex which is responsible for motor; hypothalamus which is responsible for appetite, body temperature, water balance, sleep, and blood pressure; midbrain which is responsible for the regulation of sleep, wakefulness and level of arousal as well as for the coordination of eye movements; and hippocampus which is responsible for memory (Swanson, 1999 and Bloom, 2001). Fujisawa *et al.* (1984) demonstrated that calmodulin –dependent protein kinase may play a number of roles in the functioning of the cerebral cortex, brainstem and cerebellum. The previous studies suggested that *Zizyphus* has inhibitory effects on hippocampus and probably acts through its anti-calmodulin action (Zhang *et al.*, 2003). The increase in 5-HIAA content may be due to the increase in 5-HT content.

In conclusion, the acute administration of *Zizyphus spina-christi* leaves extract produced sedative effect which may be due to inhibition of calmodulin-dependent calcium–ATPase and phosphodiesterase, also, inhibition of  $Ca^{2+}$  / calmodulin binding which decrease in neurotransmitter release; at the same time GABA inhibits the release of these neurotransmitters. As a result the content of neurotransmitters is increased. this may be due to the presence of peptide and cyclopeptide alkaloids. *Zizyphus spina-christi* leaves may potentially be safe for use as sedative drug.

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## التأثير الحاد لمستخلص أوراق نبات السدر على المحتوى الكلي لبعض الموصلات العصبية في المناطق المختلفة من مخ ذكور الجرذان البيضاء .

عبير محمد وقاص

قسم علم الحيوان - كلية التربية للبنات - الأقسام العلمية - المملكة العربية السعودية .

### الملخص

أدى الحقن الحاد داخل التجويف البطني بمستخلص أوراق السدر ( ١٠٠ مج/كجم من وزن الجسم ) إلى زيادة معنوية في محتوى الإبينفرين والنورإبينفرين والدوبامين والسيرتونين و ٥-هيدروكسي إندول حامض الخليك وإنخفاض معنوي في محتوى جاما أمينو حامض البيوتريك في مناطق الدماغ المختلفة ( المخيخ، القنطرة والنخاع المستطيل، الجسم المخطط، القشرة المخية، تحت المهاد البصري، المخ المتوسط، قرين آمون ) لذكور الجرذان البيضاء .

الزيادة في محتوى الإبينفرين والنورإبينفرين والدوبامين والسيرتونين و ٥-هيدروكسي إندول حامض الخليك في مناطق الجهاز العصبي المركزي المختلفة للجرذ الأبيض قد تكون نتيجة لتثبيط ارتباط الكالسيوم بالمولدوليين مما أدى إلى ارتفاع محتوى الموصلات العصبية داخل مناطق الدماغ المختلفة، وفي نفس الوقت عملت الجابا على دخول أيونات الكلور والتي تلعب دوراً هاماً في تحرير الموصلات العصبية وهذا ربما يكون ناتجاً من وجود البيبتيدات والبيبتيدات القاعدية الحلقية في المستخلص . ومن الممكن استخدام مستخلص أوراق نبات السدر كعقار مهدئ آمن .