CHAPTER IV

Results

Immunostaining for NMDAR1

The experiments demonstrated the negative and positive control sections stained with polyclonal rabbit NMDAR1 antibody. Figure 23 shows the negative control section (A), which was treated without primary antibody, and positive control sections, which were treated with primary antibody, including hippocampal formation (B), frontal cortex (C) and striatum (D). In western immunoblot detection of NMDAR1 protein, positive control with NMDAR1 antibody for rat liver and celebellum tissue lysates represented an immunoreactive band corresponding to approximately 116 kDa. Negative control for NMDAR1 antibody showed absent immunoreactivity band at approximately 116 kDa in rat testis and skeletal muscle tissue (Figure 24). These findings correspond with integrated optical density (IOD) of NMDAR1 immunoreactivity (NMDAR1-IR) bands which showed a high level of IOD in liver and cerebellum, whereas no IOD values were detected in testis and skeletal muscle (Figure 24). The expression of NMDAR1 showed similar results with previous reports of NMDAR1 distribution in rat tissues (Beke et al., 1995; Naoaki etal., 2004).

Immunostaining for EAAT3

The immunostaining sections represented negative control and positive control sections stained with polyclonal goat EAAT3 antibody. The negative control for EAAT3 demonstrated in section (A) and positive control for EAAT3 in hippocampal formation, frontal cortex and striatum showed in section (B), (C), (D), respectively (Figure 25). The positive immunoreactivity of EAAT3 proteins at approximately 70 kDa performed by western blotting technique were showed in rat cerebellum, testis, and liver tissue, whereas it was not found in rat skeletal muscle. In densitometric analysis, the liver tissue showed IOD values at approximately 70 kDa than that of the cerebellum and testis, and no IOD was detected in skeletal muscle (Figure 26). The expression of EAAT3 showed similar results with previous reports of

EAAT3 distribution in rat tissues (Rothstein et al., 1994; Hinoi et al., 2003; Takarada et al., 2004).

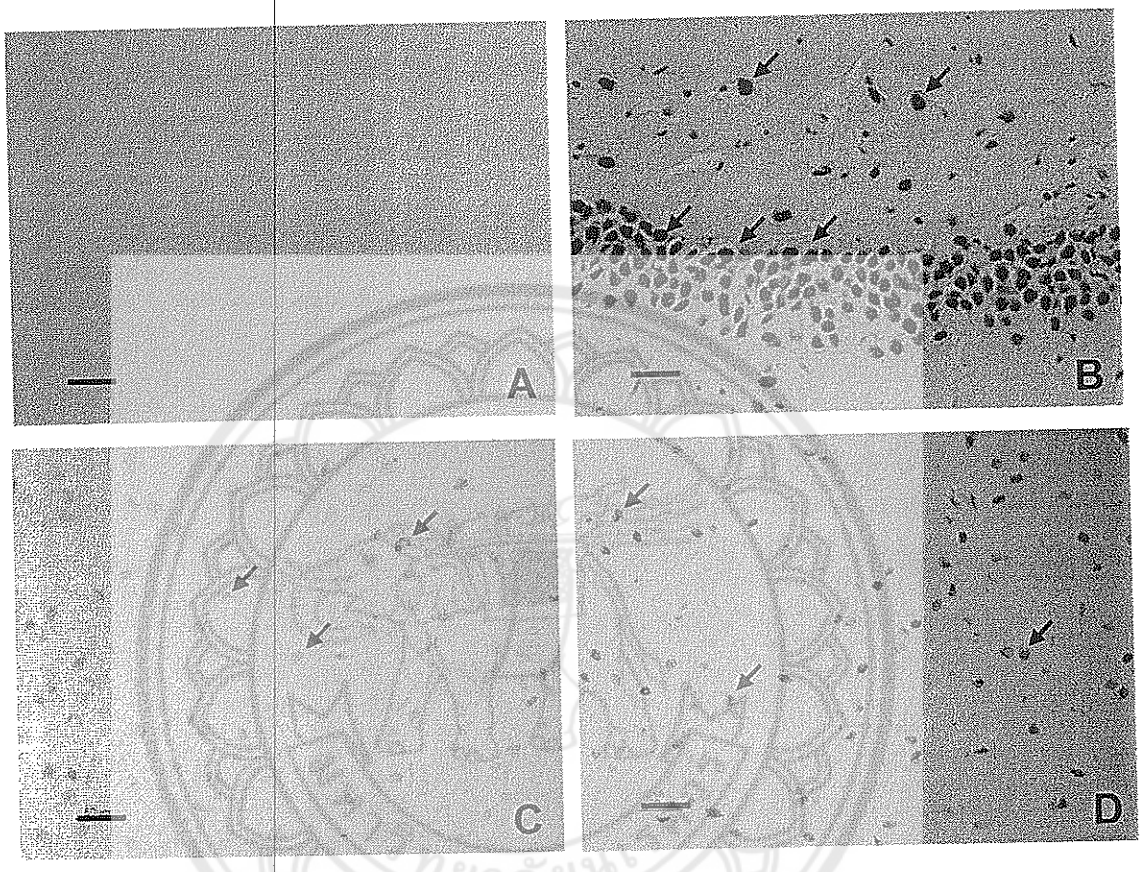

Figure 22 Immunostaining for NMDAR1 immunoreactive neurons in rat brain sections. Negative control (A) Immunohistochemistry of EAAT3-positive neurons reveals dark staining in cells within in hippocampal formation (B) frontal cortex (C) striatum (D). Scale bar = 50 μm

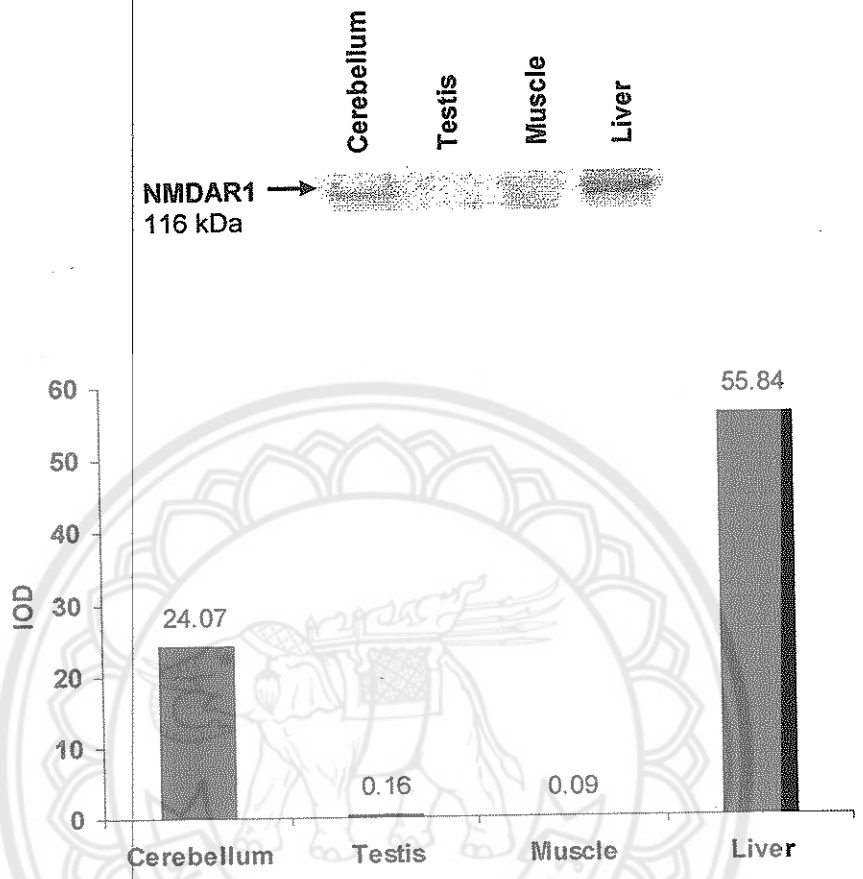

Figure 23 Demonstrating of NMDAR1 in rat tissue homogenates by immunoblot. Immunodetection of NMDAR1 showed a 116 kDa band in cerebellum and liver (positive control), while no immunoreactive band was detected in testis and skeletal muscle (negative control). Bar graphs represent optical density of NMDAR1 in rat cerebellum and liver

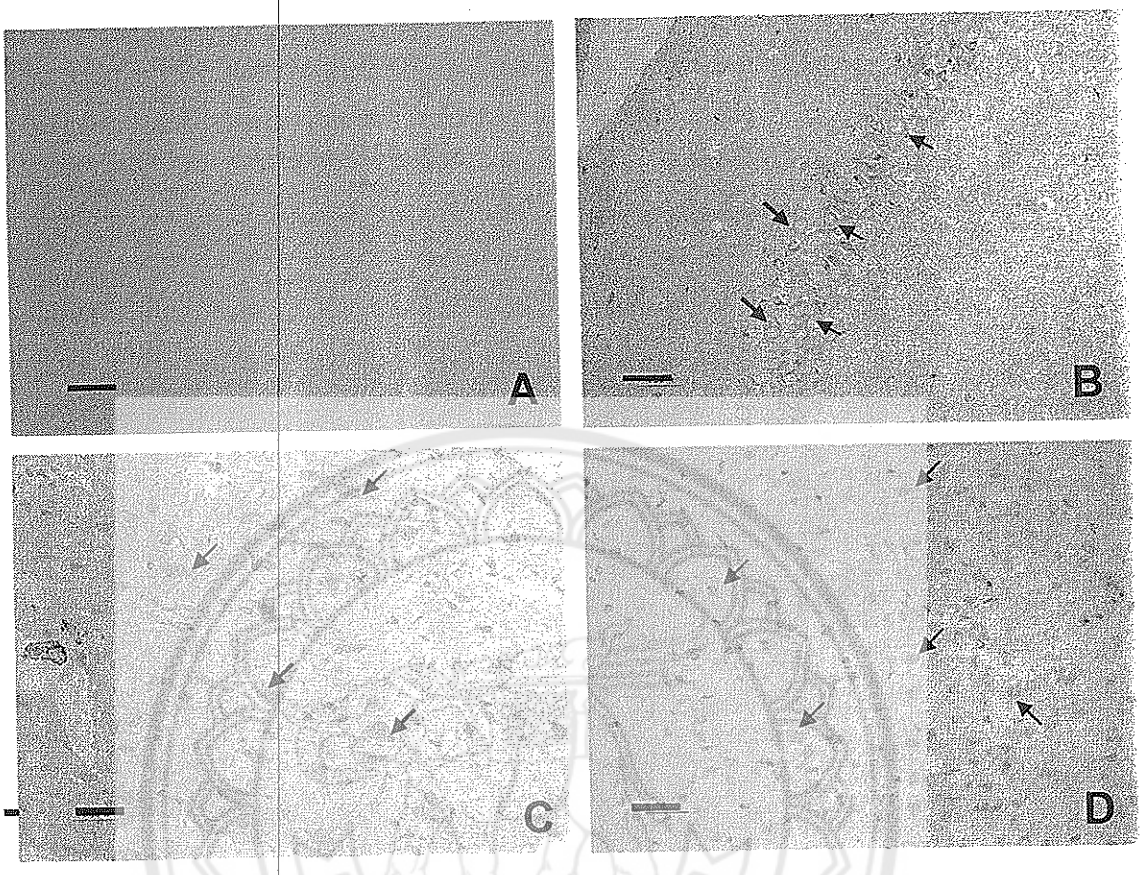

Figure 24 Immunostaining for EAAT3 immunoreactive neurons in rat brain sections. Negative control (A) Immunohistochemistry of EAAT3-positive neurons reveals dark staining in cells within in hippocampal formation (B) frontal cortex (C) striatum (D). Scale bar = 50 µm

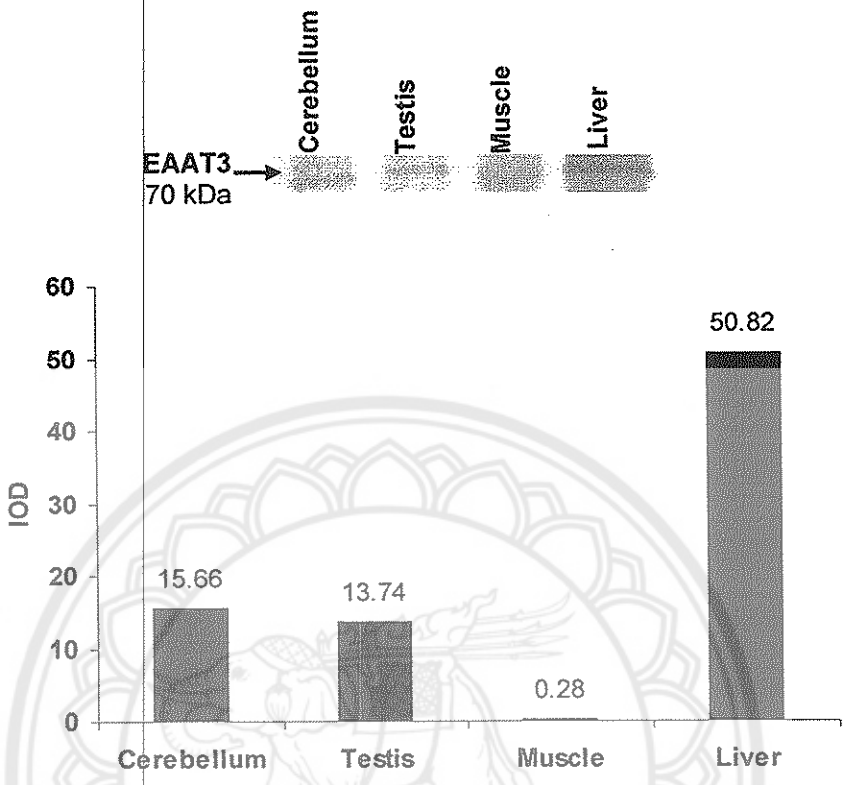

Figure 25 Demonstrating of EAAT3 in rat tissue homogenates by immunoblot. Immunodetection of EAAT3 showed a 70 kDa band in cerebellum, testis and liver (positive control), while no immunoreactive band was detected in skeletal muscle (negative control). Bar graphs represent optical density of EAAT3 in rat cerebellum, testis and liver.

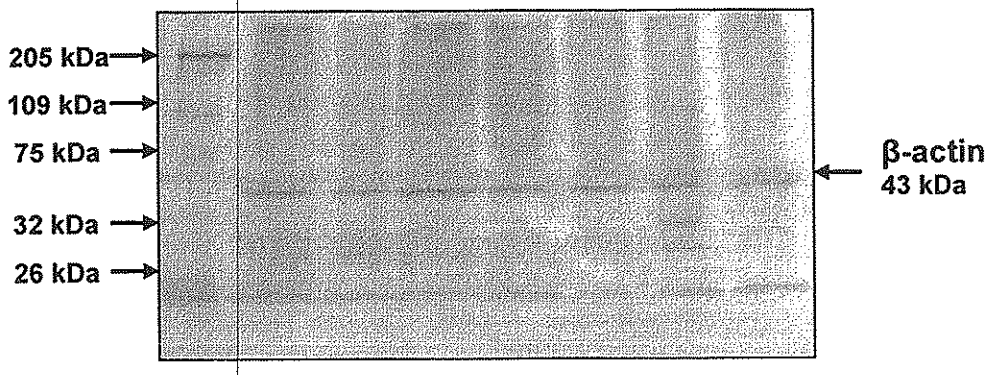

Figure 26 Western immunoblot detection indicating the β-actin protein of ~43 kDa in rat brains

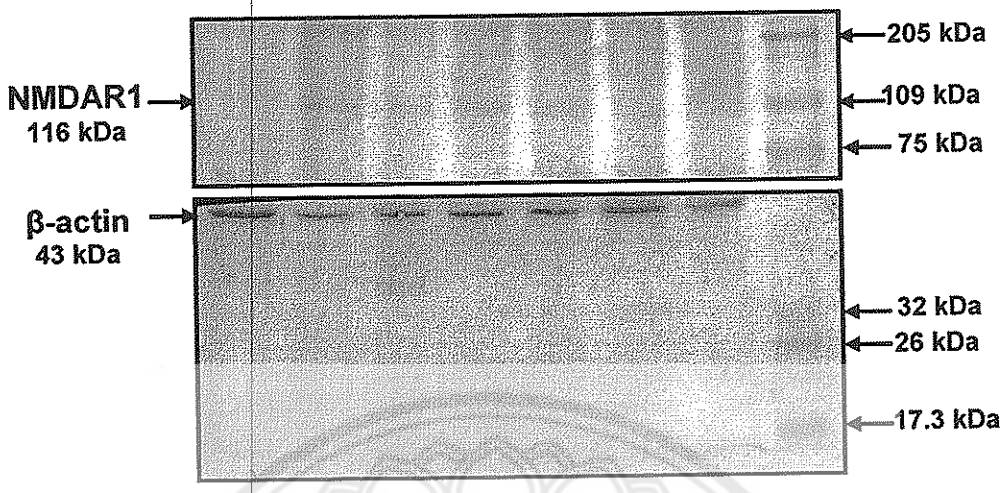

Figure 27 Western immunoblot detection indicating the NMDAR1 protein of ~116 kDa (upper row) and β-actin protein of ~43 kDa (lower row) in rat brains

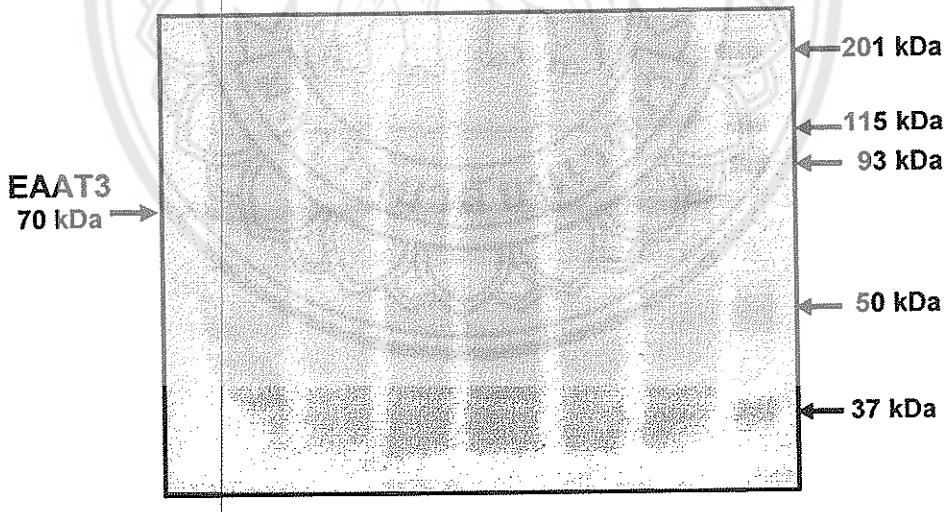

Figure 28 Western immunoblot detection indicating the EAAT3 protein of ~70 kDa in rat brains

Expression of glutamate NMDAR1 receptor

Immunodetection of NMDAR1 consistently demonstrated an immunoreactive band corresponding to approximately 116 kDa and β-actin immunoreactive band was approximately shown 43 kDa (Figure 28). Densitometric analysis was performed to quantify the alteration of NMDAR1 expression relative to β-actin immunoreactivity. ANOVA demonstrated a significant difference of NMDAR1-IR in striatum (F= 5.728, p=0.015) (Table 9 and Figure 32) and frontal cortex (F=3.616, p=0.054) (Table 6 and Figure 35), but not in hippocampal formation (F=0.260, p=0.774) (Table 3 and Figure 38).

Expression of NMDAR1 in hippocampal formation

Post hoc Dunnett test and t-test demonstrated no significant differences of NMDAR1-IR in hippocampal formation either in acute (Post hoc; p=0.839 Table 3 and Figure 32), t-test; p=0.588 Table 1 and Figure 30) or chronic (Post hoc; p=0.711 (Table 3 and Figure 32), t-test; p=0.499 (Table 2 and Figure 31)) methamphetamine groups compared with control group.

Expression of NMDAR1 in frontal cortex

There was a significant increase of NMDAR1-IR about 508% over control was also observed in the frontal cortex in chronic methamphetamine (Post hoc; p=0.036 (Table 6 and Figure 35), t-test; p=0.083 (Table 5 and Figure 34)). However, there was no significant difference of NMDAR-IR in acute methamphetamine group (Post hoc; p=0.580 (Table 6 and Figure 35), t-test; p=0.168 (Table 4 and Figure 33)).

Expression of NMDAR1 in striatum

There were significant increases in NMDAR1-IR above control in the striatum in both acute (Post hoc; p=0.025 (Table 9 and Figure 38), t-test; p=0.016 (Table 7 and Figure 36)) and chronic (Post hoc; p=0.023 (Table 9 and Figure 38), t-test; p=0.004 (Table 8 and Figure 37)) methamphetamine administrations. An elevation of NMDAR-IR in acute and chronic methamphetamine groups showed about 196% and 198% of control group, respectively.

Table 1 Effects of acute methamphetamine administration on NMDAR1 expression in rat hippocampal formation. Data are integrated optical density of NMDAR1-IR band and normalized to β-actin levels. Values are Mean ± SEM

N	Hippocampal formation	NMDAR1-IR	NMDAR1-IR (% of control)	Level of significance (t-test)
6	Control	0.5701 ± 0.1123	100.00 ±19.70	p=0.588
6	Acute	0.6744 ± 0.1486	118.30 ± 22.03	

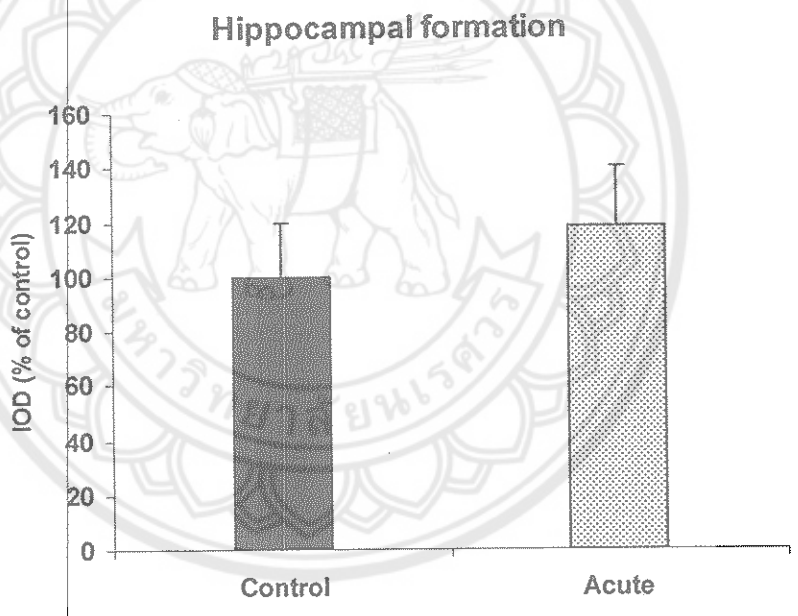

Figure 29 NMDAR1 expression of acute methamphetamine administration in rat hippocampal formation. Data are integrated optical density of NMDAR1-IR band, normalized to β-actin levels and expressed as percentage of the control group. n=6 for control and acute groups. Values represent Mean ± SEM

Table 2 Effects of chronic methamphetamine administration on NMDAR1 expression in hippocampal formation. Data are integrated optical density of NMDAR1-IR band and normalized to β-actin levels. Values are Mean ± SEM

N	Hippocampal formation	NMDAR1-IR	NMDAR1-IR (%Control)	Level of significance (t-test)
6	Control	0.5701 ± 0.1123	100.00 ± 19.70	p=0.499
6	Chronic	0.7174 ± 0.1776	125.84 ± 24.75	

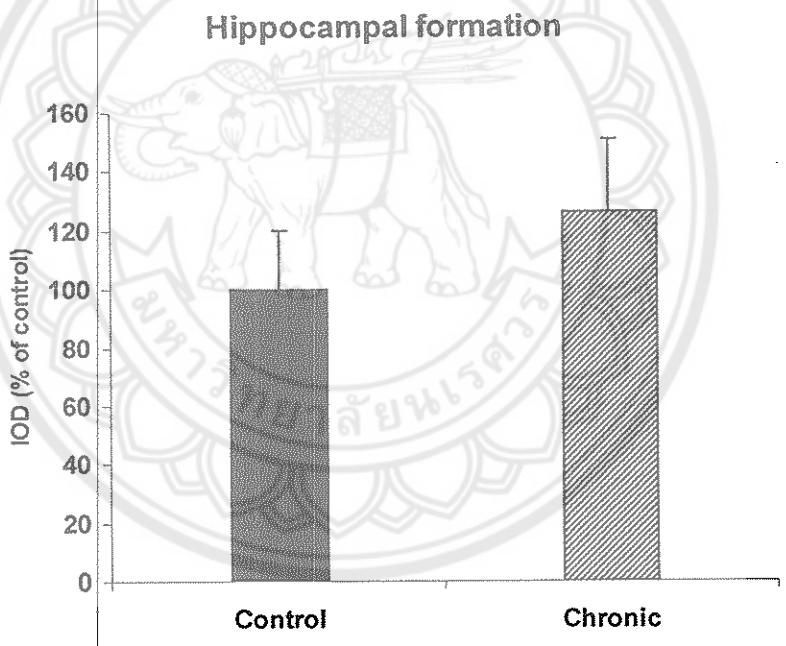

Figure 30 NMDAR1 expression of chronic methamphetamine administration in rat hippocampal formation. Data are integrated optical density of NMDAR1-IR band, normalized to β-actin levels and expressed as percentage of the control group. n=6 for control and acute groups. Values represent Mean ± SEM

Table 3 Effects of methamphetamine administration on NMDAR1 expression in rat hippocampal formation. Data are integrated optical density of NMDAR1-IR band. Values are Mean ± SEM

N	HIP	NMDAR1	NMDAR1 (% of control)	Level of significance	
				Dunnett post hoc test	ANOVA
6	Control	0.5701 ± 0.1123	100.00 ± 19.70		p=0.774
6	Acute	0.6744 ± 0.1486	118.30 ± 26.06	p=0.839	
6	Chronic	0.7174 ± 0.1776	125.84 ± 31.15	p=0.711	

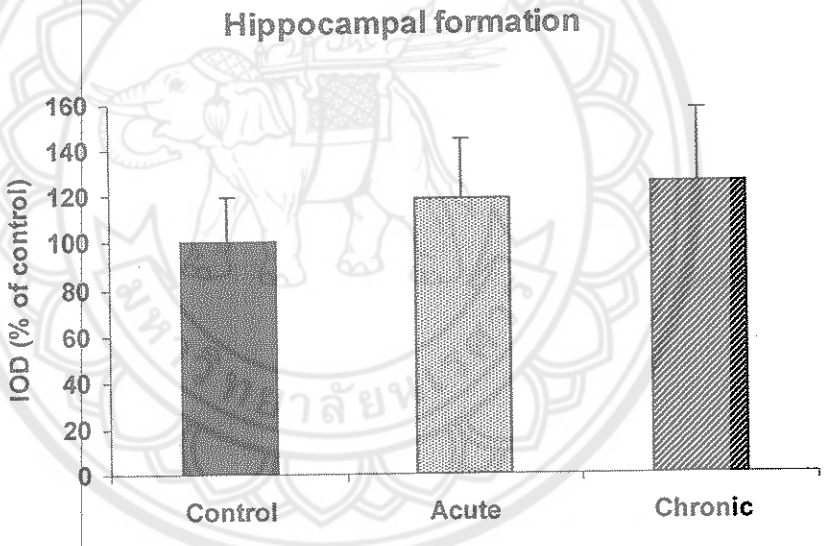

Figure 31 NMDAR1 expression after methamphetamine administration in rat hippocampal formation. Data are integrated optical density of NMDAR1-IR band, normalized to β-actin levels and expressed as percentage of the control group. Values represent Mean ± SEM (n=6)

Table 4 Effects of acute methamphetamine administration on NMDAR1 expression in rat frontal cortex. Data are integrated optical density of NMDAR1-IR band and normalized to β-actin levels. Values are Mean ± SEM. #p=0.168 in comparison with control group by t-test

N	Frontal cortex	NMDAR1	NMDAR1 (% of control)	Level of significance (t-test)
5	Control	0.7078 ± 0.2053	100.00 ± 29.00	p=0.168#
7	Acute	1.6212 ± 0.4924	229.05 ± 30.37	

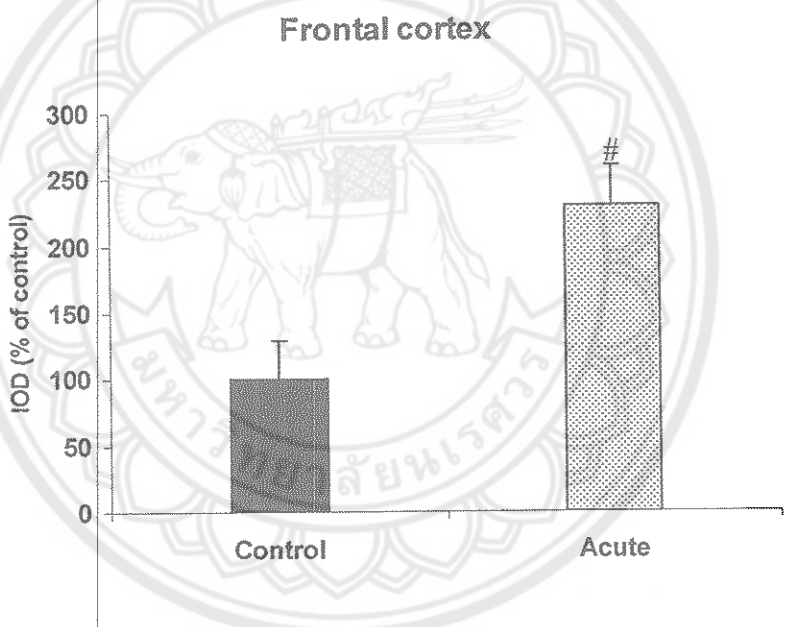

Figure 32 NMDAR1 expression of acute methamphetamine administration in rat frontal cortex. Data are integrated optical density of NMDAR1-IR band, normalized to β-actin levels and expressed as percentage of the control group. n=5 for control group and n=7 for acute group. Values represent Mean ± SEM. #p=0.168 in comparison with control group by t-test

Table 5 Effects of chronic methamphetamine administration on NMDAR1 expression in rat frontal cortex. Data are integrated optical density of NMDAR1-IR band and normalized to β-actin levels. Values are Mean ± SEM. #p=0.083 in comparison with control group by t-test

N	Frontal cortex	NMDAR1	NMDAR1 (% of control)	Level of significance (t-test)
5	Control	0.7078 ± 0.2053	100.00 ± 29.00	p=0.083#
5	Chronic	3.5964 ± 1.2570	508.11 ± 34.95	

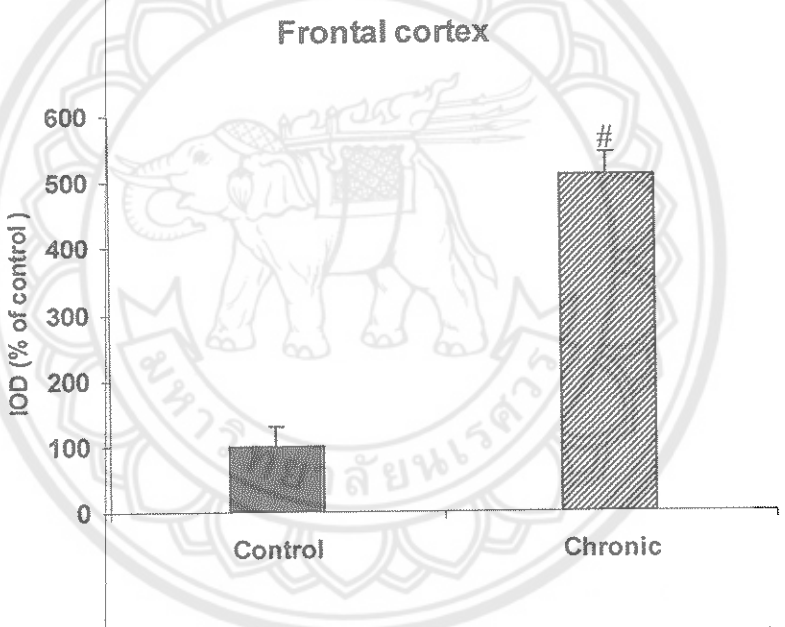

Figure 33 NMDAR1 expression of chronic methamphetamine administration in rat frontal cortex. Data are integrated optical density of NMDAR1-IR band, normalized to β-actin levels and expressed as percentage of the control group. n=5 for control and chronic groups. Values represent Mean ± SEM. #p=0.083 in comparison with control group by t-test

Table 6 Effects of methamphetamine administration on NMDAR1 expression in rat frontal cortex. Data are integrated optical density of NMDAR1-IR band. Values are Mean ± SEM. *p <0.05 in comparison with control group by one-way ANOVA with Dunnett post hoc test

N	Frontal cortex	NMDAR1	NMDAR1 (% of control)	Level of significance	
				Dunnett post hoc test	ANOVA
5	Control	0.7078 ± 0.2053	100.00 ± 29.00		**p=0.054***
7	Acute	1.6212 ± 0.4924	118.30 ± 69.57	p=0.580	
5	Chronic	3.5964 ± 1.2570	125.84 ±177.59	**p=0.036***	

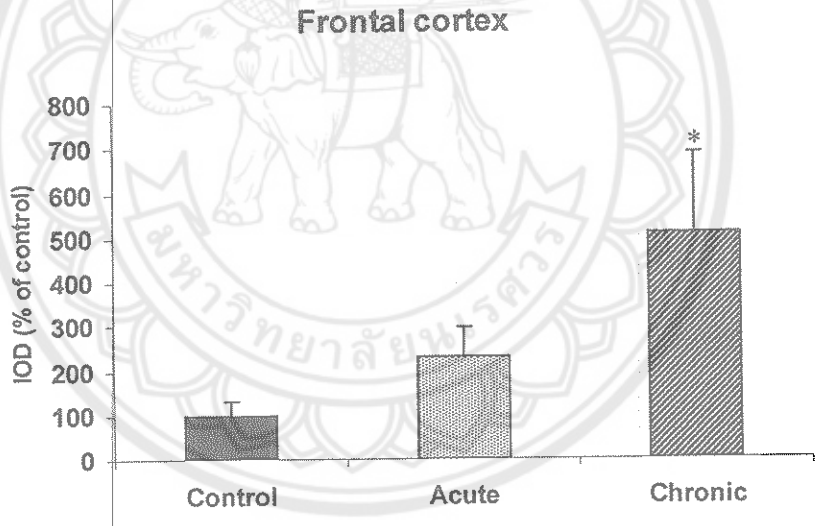

Figure 34 NMDAR1 expression after methamphetamine administration in rat frontal cortex. Data are integrated optical density of NMDAR1-IR band, normalized to β-actin levels and expressed as percentage of the control group Values represent Mean ± SEM (n=5-7). *p<0.05 in comparison with control group by one-way ANOVA with Dunnett post hoc test

Table 7 Effects of acute methamphetamine administration on NMDAR1 expression in rat striatum. Data are integrated optical density of NMDAR1-IR band and normalized to β-actin levels. Values are Mean ± SEM. *p<0.05 in comparison with control group by t-test

N	Striatum	NMDAR1	NMDAR1 (% of control)	Level of significance (t-test)
7	Control	1.1560 ± 0.1164	100.00 ± 10.07	p=**0.016***
5	Acute	2.2679 ± 0.4294	196.19 ± 18.93	

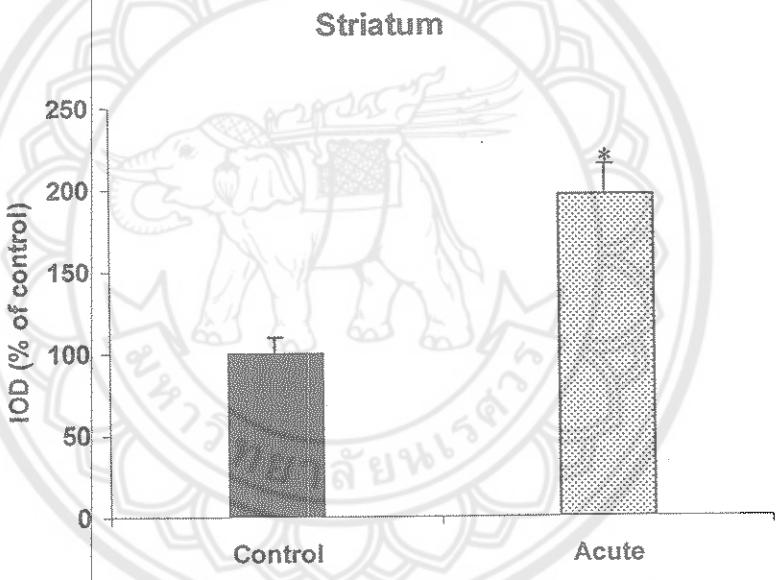

Figure 35 NMDAR1 expression of methamphetamine administration in rat striatum. Data are integrated optical density of NMDAR1-IR band, normalized to β-actin levels and expressed as percentage of the control group. n=7 for control group and n= 5 for chronic group. Values represent Mean ± SEM. *p<0.05 in comparison with control group by t-test

Table 8 Effects of chronic methamphetamine administration on NMDAR1 expression in rat striatum. Data are integrated optical density of NMDAR1-IR band and normalized to β-actin levels. Values are Mean ± SEM. **p<0.01 in comparison with control group by t-test

N	Striatum	NMDAR1	NMDAR1 (% of control)	Level of significance (t-test)
7	Control	1.1560 ± 0.1164	100.00 ± 10.07	p=0.004**
5	Chronic	2.2883 ± 0.3226	197.95 ± 14.10	

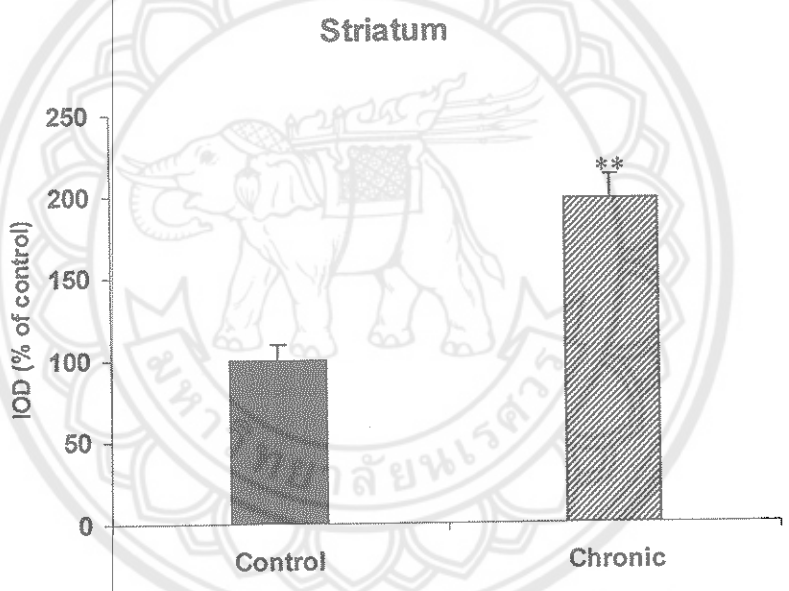

Figure 36 NMDAR1 expression of chronic methamphetamine administration in rat striatum. Data are integrated optical density of NMDAR1-IR band, normalized to β-actin levels and expressed as percentage of the control group. Values represent Mean ± SEM. n=7 for control group and n= 5 for chronic group. **p<0.01 in comparison with control group by t-test

Table 9 Effects of methamphetamine administration on NMDAR1 expression in rat striatum. Data are integrated optical density of NMDAR1-IR band. Values are Mean ± SEM. *p <0.05 in comparison with control group by one-way ANOVA with Dunnett post hoc test

N	Striatum	NMDAR1	NMDAR1 (% of control)	Level of significance	
				Dunnett post hoc test	ANOVA
7	Control	1.1560 ± 0.1164	100.00 ± 10.07		**p=0.015***
5	Acute	2.2679 ± 0.4294	196.19 ± 37.14	**p=0.025***	
5	Chronic	2.2883 ± 0.3226	197.95 ± 27.91	**p=0.023***	

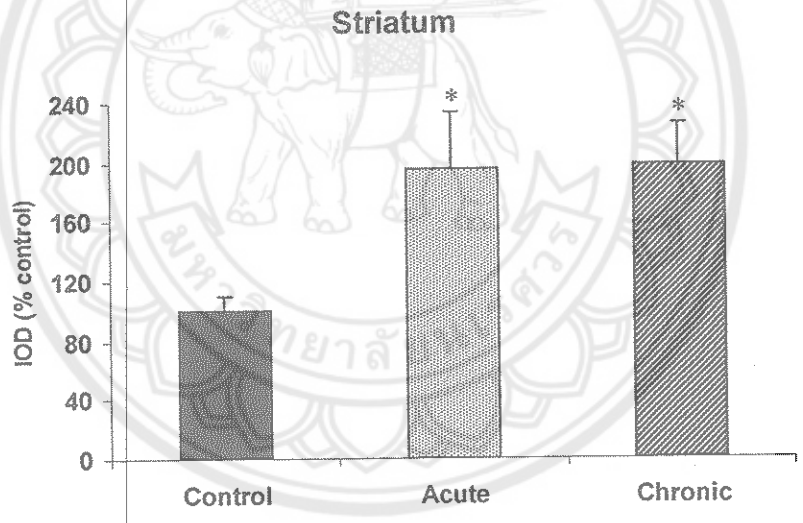

Figure 37 NMDAR1 expression after methamphetamine administration in rat striatum. Data are integrated optical density of NMDAR1-IR band, normalized to β-actin levels and expressed as percentage of the control group. Values represent Mean ± SEM (n=5-7). *p<0.05 in comparison with control group by one-way ANOVA with Dunnett post hoc test

Expression of neuronal glutamate EAAT3 transporter

Immunodetection of EAAT3 consistently demonstrated an immunoreactive band corresponding to approximately 70 kDa. ANOVA showed a significant effect of treatment group on EAAT3 immunoreactivity (EAAT3-IR) in hippocampal formation F=8.992, p=0.04) (Table 12 and Figure 41), striatum (F=9.060, p=0.005) (Table 15 and Figure 47) and frontal cortex (F=2.530, p=0.115) (Table 18 and Figure 44).

Expression of EAAT3 in hippocampal formation

There was a significant difference in NMDAR1 expression between control and chronic methamphetamine group in hippocampal formation (Post hoc; p=0.006 (Table 12 and Figure 41), t-test; p=0.009 (Table 11 and Figure 40)), and reached about 149% of the control group. On the other hand, there was no significant difference in NMDAR1 expression between control and acute methamphetamine group in hippocampal formation (Post hoc; p=1.000 (Table 12 and Figure 41), t-test; p=0.995 (Table 10 and Figure 39))

Expression of EAAT3 in frontal cortex

No significant change in EAAT3-IR was observed in acute methamphetamine group in frontal cortex (Post hoc; p=0.237 (Table 15 and Figure 44), t-test; p=0.205 (Table 13 and Figure 42)), whereas a trend towards decrease of EAAT3-IR was found in chronic methamphetamine group but this did not reach significance (Post hoc; p=0.070 (Table 15 and Figure 44), t-test; p=0.061(Table 14 and Figure 43)).

Expression of EAAT in striatum

A significant decrease in EAAT3-IR was observed in striatum in both acute (Post hoc; p=0.004 (Table 18 and Figure 47), t-test; p=0.002 (Table 16 and Figure 45)) and chronic (Post hoc; p=0.03 (Table 18 and Figure 47), t-test; p=0.039 (Table 17 and Figure 46)) methamphetamine groups. A reduction of EAAT3-IR in acute and chronic methamphetamine groups showed approximately 48% and 64% of control group, respectively.

Table 10 Effects of acute methamphetamine administration on EAAT3 expression in rat hippocampal formation. Data are integrated optical density of EAAT3-IR band. Values are Mean ± SEM.

N	HIP	EAAT3	EAAT3 (% of control)	Level of significance (t-test)
5	Control	31.5940 ± 2.2614	100.00 ± 7.16	p=0.995
5	Acute	31.6160 ± 2.3163	100.07 ± 7.33	

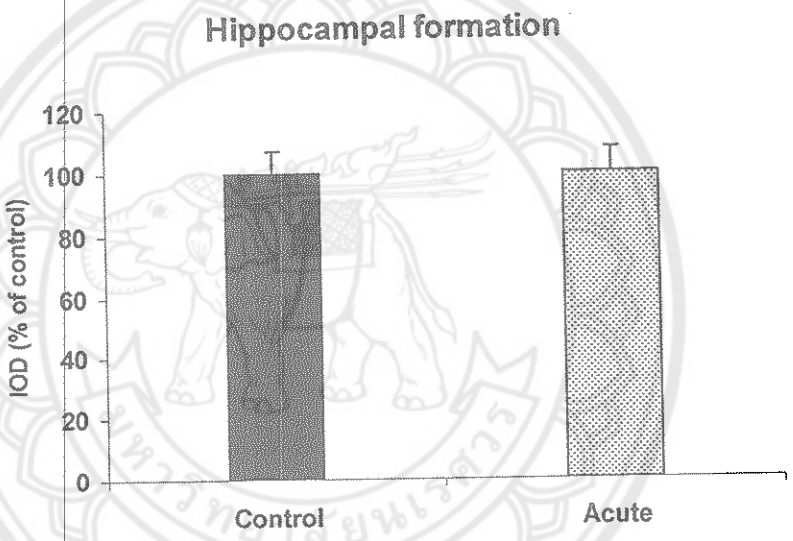

Figure 38 EAAT3 expression of chronic methamphetamine administration in rat hippocampal formation. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. n=5 for control and acute groups. Values represent Mean ± SEM

Table 11 Effects of chronic methamphetamine administration on EAAT3 expression in rat hippocampal formation. Data are integrated optical density of EAAT3-IR band. Values are Mean ± SEM. **p<0.01 in comparison with control group by t-test

N	Hippocampal formation	EAAT3	EAAT3 (% of control)	Level of significance (t-test)
5	Control	31.5940 ± 2.2614	100.00 ± 7.16	p=0.009**
6	Chronic	47.2067 ± 3.8742	149.42 ± 8.21	

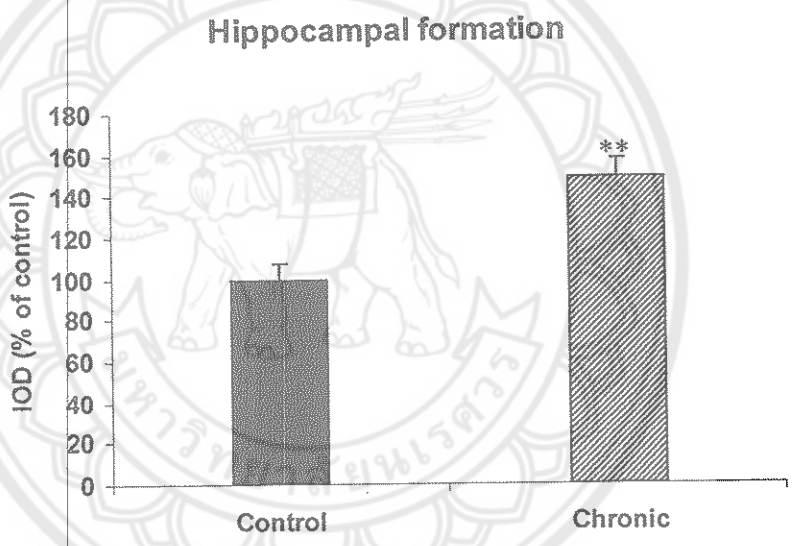

Figure 39 EAAT3 expression of chronic methamphetamine administration in rat hippocampal formation. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. n=5 for control group and n=6 for chronic group. Values represent Mean ± SEM. **p<0.01 in comparison with control group by t-test

Table 12 Effects of methamphetamine administration on EAAT3 expression in rat hippocampal formation. Data are integrated optical density of EAAT3-IR band. Values are Mean ± SEM. **p<0.01 in comparison with control group by one-way ANOVA with Dunnett post hoc test

N	HIP	EAAT3	EAAT3 (% of control)	Level of significance	
				Dunnett post hoc test	ANOVA
5	Control	31.5940 ± 2.2614	100.00 ± 7.16		**p=0.004****
5	Acute	31.6160 ± 2.3163	100.07 ± 7.33	p=1.000	
6	Chronic	47.2067 ± 3.8742	149.42 ± 12.26	**p=0.006****	

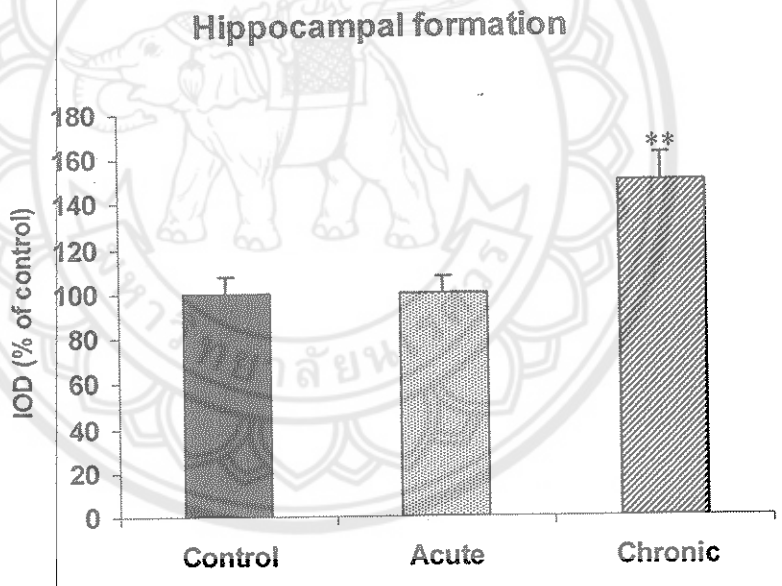

Figure 40 EAAT3 expression after methamphetamine administration in rat hippocampal formation. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. Values represent Mean ± S.E.M (n=5-6). **p<0.01 in comparison with control group by one-way ANOVA with Dunnett post hoc test

Table 13 Effects of acute methamphetamine administration on EAAT3 expression in rat frontal cortex. Data are integrated optical density of EAAT3-IR band. Values are Mean ± SEM

N	Frontal cortex	EAAT3	EAAT3 (% of control)	Level of significance (t-test)
5	Control	45.8275 ± 4.0712	100.00 ± 8.88	p=0.205
5	Acute	39.8486 ± 2.3923	86.95 ± 6.00	

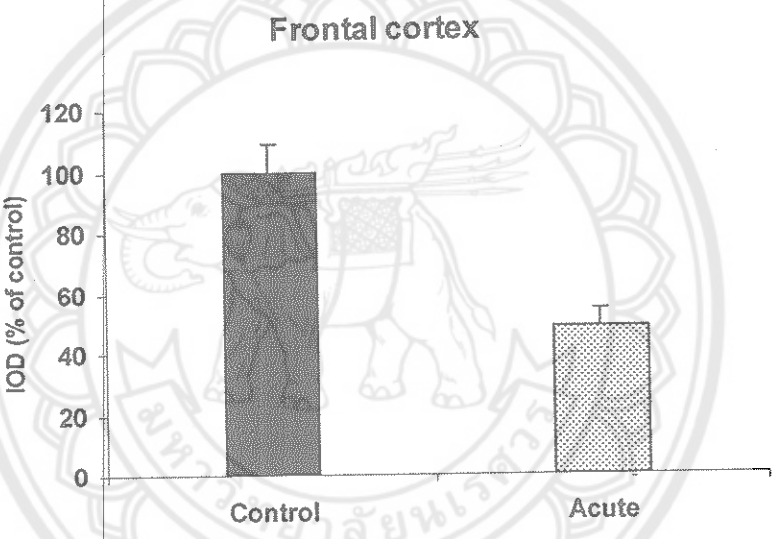

Figure 41 EAAT3 expression of acute methamphetamine administration in rat frontal cortex. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. n=5 for control and acute groups. Values represent Mean ± SEM (n=4-7)

Table 14 Effects of chronic methamphetamine administration on EAAT3 expression in frontal cortex. Data are integrated optical density of EAAT3-IR band. Values are Mean ± SEM. #p=0.061 in comparison with control group by t-test

N	Frontal cortex	EAAT3	EAAT3 (% of control)	Level of significance (t-test)
6	Control	45.8275 ± 4.0712	100.00 ± 8.88	p=0.061#
4	Chronic	36.2283 ± 2.4027	79.05 ± 6.63	

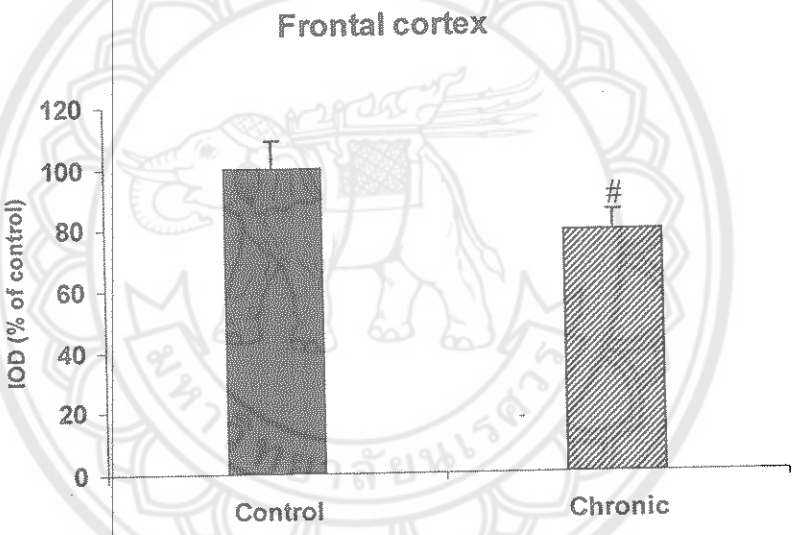

Figure 42 EAAT3 expression of acute methamphetamine administration in rat frontal cortex. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. n=6 for control group and n=4 for chronic group. Values represent Mean ± SEM. #p=0.061 in comparison with control by t-test

Table 15 Effects of methamphetamine administration on EAAT3 expression in rat frontal cortex. Data are integrated optical density of EAAT3-IR band. Values are Mean ± SEM. #p=0.070 in comparison with control group by one-way ANOVA with Dunnett post hoc test

N	Frontal cortex	EAAT3	EAAT3 (% of control)	Level of significance	
				Dunnett post hoc test	ANOVA
4	Control	45.8275 ± 4.0712	100.00 ± 8.88		p=0.115
7	Acute	39.8486 ± 2.3923	86.95 ± 5.22	p=0.273	
6	Chronic	36.2283 ± 2.4027	79.05 ± 5.24	p=0.070#	

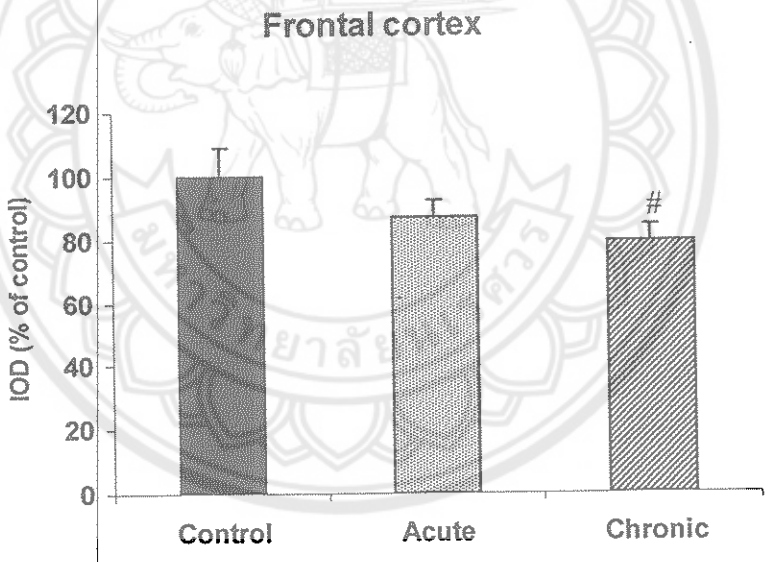

Figure 43 EAAT3 expression after methamphetamine administration in rat frontal cortex. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. Values represent Mean ± SEM (n=4-7). #p=0.07 in comparison with control by one-way ANOVA with Dunnett post hoc test

Table 16 Effects of acute methamphetamine administration on EAAT3 expression in rat striatum. Data are integrated optical density of EAAT3-IR band. Values are Mean ± SEM. **p<0.01 in comparison with control group by t-test

N	Striatum	EAAT3	EAAT3 (% of control)	Level of significance (t-test)
6	Control	32.0350 ± 2.9019	100.00 ± 9.06	p=0.002**
4	Acute	15.4854 ± 1.0070	48.34 ± 6.50	

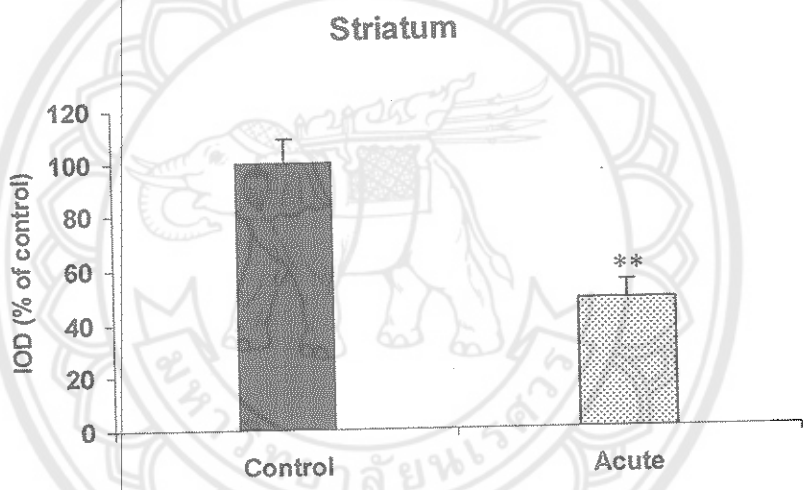

Figure 44 EAAT3 expression of acute methamphetamine administration in rat striatum. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. n=6 for control group and n=4 for acute group. Values represent Mean ± SEM. **p<0.01 in comparison with control group by t-test

Table 17 Effects of chronic methamphetamine administration on EAAT3 expression in rat striatum. Data are integrated optical density of EAAT3-IR band. Values are Mean ± SEM. *p<0.05 in comparison with control group by t-test

N	Striatum	EAAT3	EAAT3 (% of control)	Level of significance (t-test)
6	Control	32.0350 ± 2.9019	100.00 ± 9.06	p=0.039*
4	Chronic	20.3867 ± 3.8527	63.64 ± 18.90	

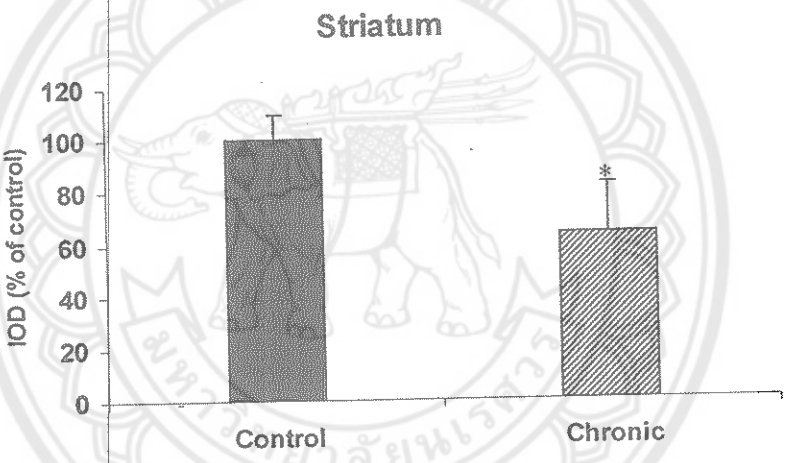

Figure 45 EAAT3 expression of chronic methamphetamine administration in rat striatum. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. n=6 for control group and n=4 for chronic group. Values represent Mean ± SEM. *p<0.05 in comparison with control group by t-test

Table 18 Effects of methamphetamine administration on EAAT3 expression in rat striatum. Data are integrated optical density of EAAT3-IR band. Values are Mean ± SEM. *p<0.05; **p<0.01 in comparison with control group by one-way ANOVA with Dunnett post hoc test

N	Striatum	EAAT3	EAAT3 (% of control)	Level of significance	
				Dunnett post hoc test	ANOVA
6	Control	32.0350 ± 2.9019	100.00 ± 9.06		**p=0.005****
4	Acute	15.4854 ± 1.0070	52.61 ± 3.42	**p=0.004****	
4	Chronic	20.3867 ± 3.8527	69.26 ± 13.09	**p=0.030***	

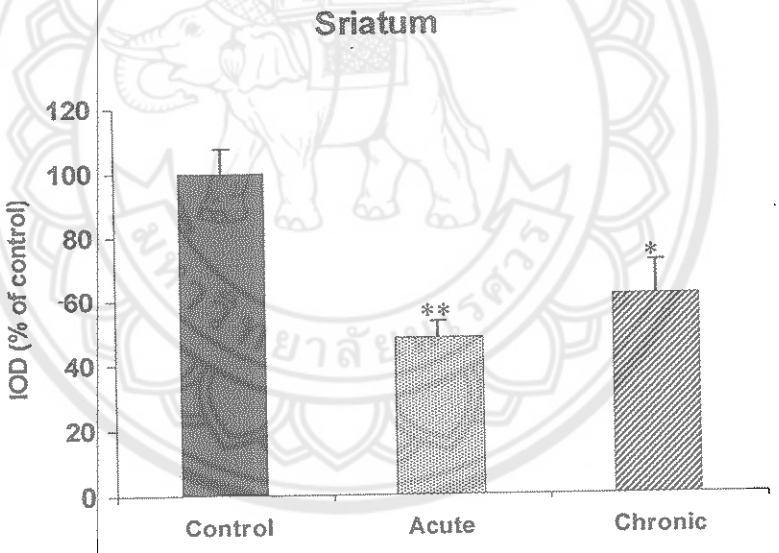

Figure 46 EAAT3 expression after methamphetamine administration in rat striatum. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. Values represent Mean ± SEM (n=4-6). *p<0.05; **p<0.01 in comparison with control group by one-way ANOVA with Dunnett post hoc test

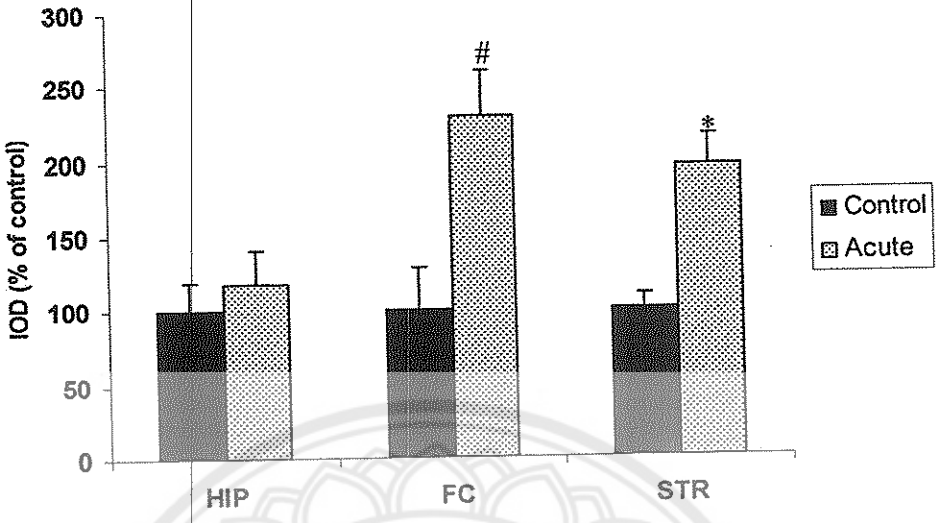

Figure 47 Effects of acute methamphetamine administration on expression of NMDAR1 in rat hippocampal formation, frontal cortex and striatum. Data are integrated optical density of NMDAR1-IR band, expressed as percentage of the control group. Values represent Mean ± SEM (n=5-7). *p<0.05; #p=0.168 in comparison with control group by t-test

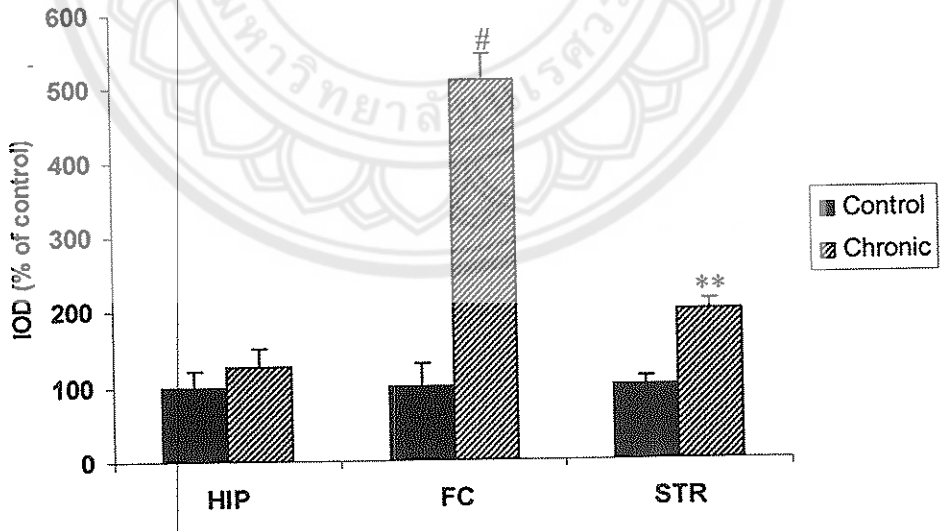

Figure 48 Effects of chronic methamphetamine administration on expression of NMDAR1 in rat hippocampal formation, frontal cortex and striatum. Data are integrated optical density of NMDAR1-IR band, expressed as percentage of the control group. Values represent Mean ± SEM (n=5-7). **p<0.01; #p=0.083 in comparison with control group t-test

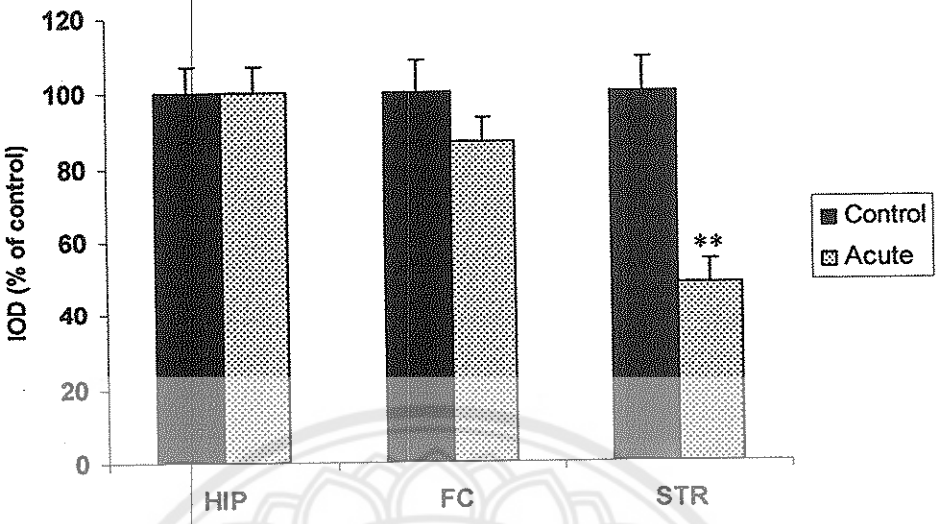

Figure 49 Effects of acute methamphetamine administration on expression of EAAT3 in rat hippocampal formation, frontal cortex and striatum. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. Values represent Mean ± SEM (n=4-7). $^{**}p<0.01$ in comparison with control group by t-test

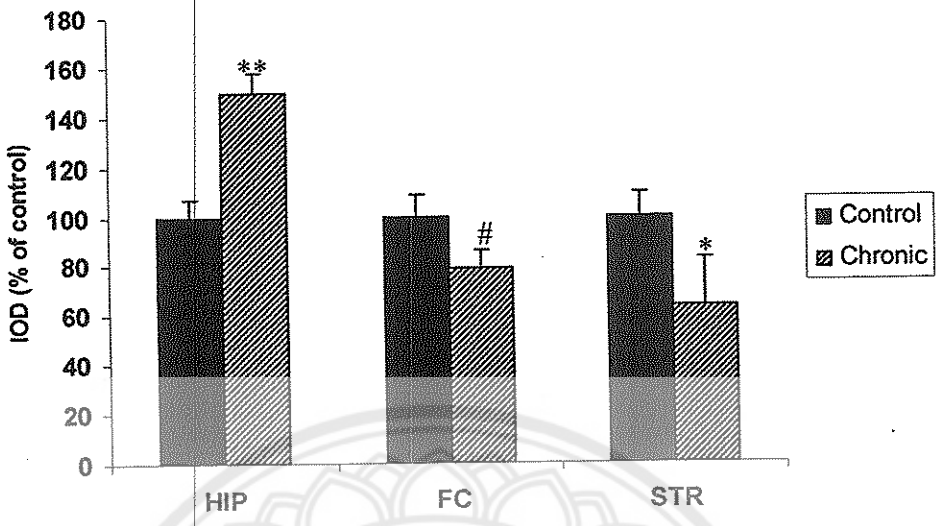

Figure 50 Effects of chronic methamphetamine administration on expression of EAAT3 in rat hippocampal formation, frontal cortex and striatum. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. Values represent Mean ± SEM (n=4-6). *p<0.05; **p<0.01; #p=0.061 in comparison with control group by t-test

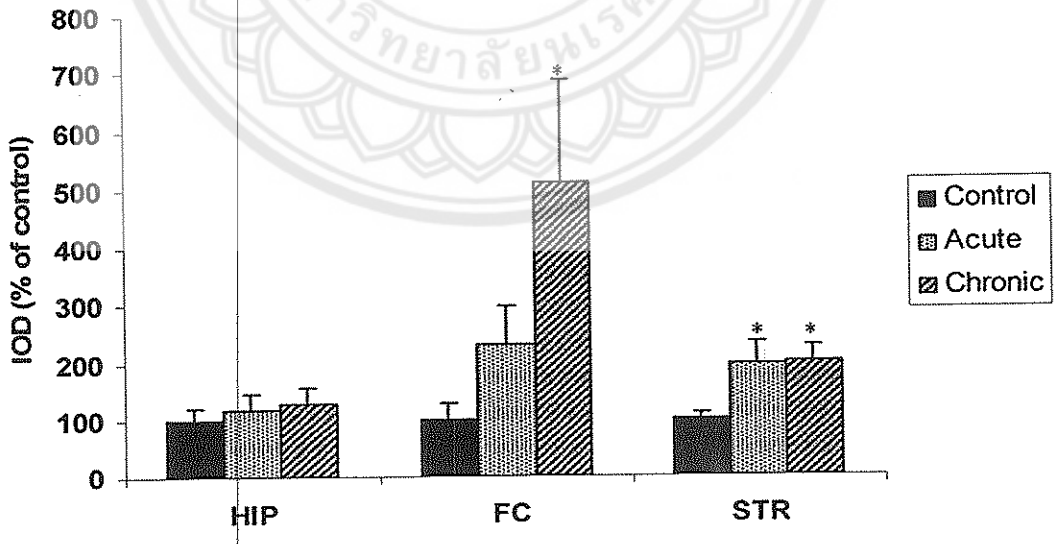

Figure 51 Expression of NMDAR1 in rat hippocampal formation, frontal cortex and striatum. Data are integrated optical density of NMDAR1-IR band. Values represent Mean ± SEM (n=5-7). *p<0.05 in comparison with control group by one-way ANOVA with Dunnett post hoc test

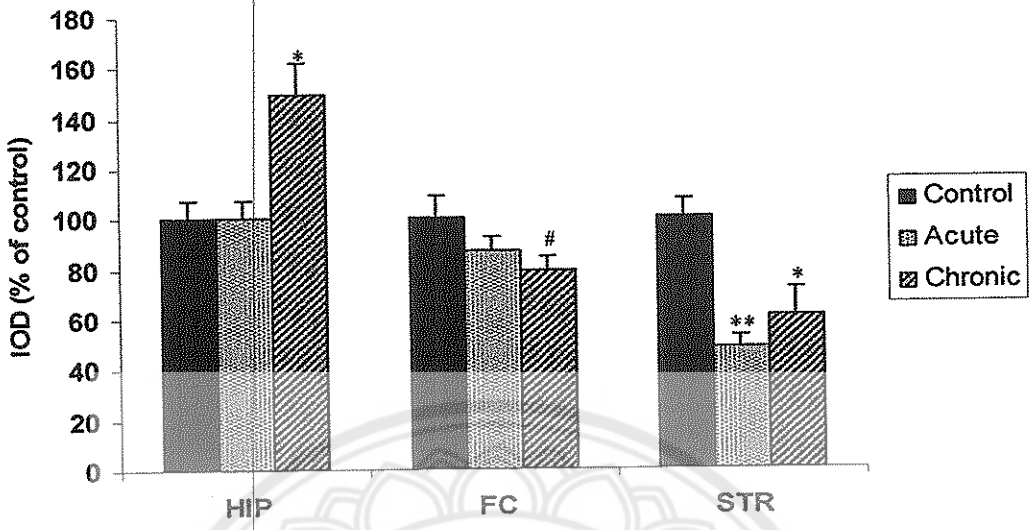

Figure 52 Expression of EAAT3 in rat hippocampal formation, frontal cortex and striatum. Data are integrated optical density of EAAT3-IR band, expressed as percentage of the control group. Values represent Mean ± SEM (n=4-7). *p<0.05; **p<0.01 #p=0.07 in comparison with control by one-way ANOVA with Dunnett post hoc test

The correlation of the effects of methamphetamine on NMDAR1 and EAAT3 expressions in rat brain areas

There was no significant correlation between NMDAR1 and EAAT3 expression in either acute or chronic methamphetamine group in hippocampal formation, frontal cortex and striatum, although a trend toward significant correlation between NMDAR and EAAT3 expressions was observed in acute methamphetamine group in striatum (p=0.120, r=0.880 (Table 19)). However, a negative correlation between NMDAR1 and EAAT3 expression was seen in chronic methamphetamine group in striatum (p=0.139, r=-0.861(Table 19)), but this failed to show a significance.

Table 19 **The correlation of the effects of methamphetamine on NMDAR1 and EAAT3 expressions in hippocampal formation, frontal cortex and striatum. Data are Pearson correlation coefficient (R), level of significance (P) and number of subjects (N)**

Regions	Acute		Chronic	
	r	p	r	p
Hippocampal formation (n=6-7)	0.520	0.290	0.487	0.328
Frontal cortex (n=5-7)	0.425	0.575	0.321	0.535
Striatum (n=4-5)	0.880	0.120	-0.861	0.139