Sıçanlarda Rapamisinin Maternal Agresyon Üzerine Etkisi

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Effect of Rapamycin on Maternal Aggression in Rats [1] [2]

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Abstract

Rapamycin which is an inhibitor of mammalian target of rapamycin (mTOR), has effects as antineoplastic, retarding aging, anti-inflammatory and neuroprotective. Aim of this study is to investigate the effects of rapamycin on maternal aggression in rats. In this study 63 Wistar female rats were used. The animals were divided into 3 groups: the solvent (DMSO) group, the 5 mg/kg rapamycin group, and the 10 mg/kg rapamycin group. For behavioral testing the resident-intruder paradigm was used. The groups were compared in terms of the latency to the first aggressive behavior, the number of attacks, the total duration of aggressive behaviors and the intensity of attacks. When the groups were compared in terms of the latency to the first aggressive behavior, it was found that 5 and 10 mg/kg rapamycin groups were significantly prolonged latencies compared to the control group. When were evaluated the number of aggressive behaviors, total duration of aggressive behaviors and average severity of attacks it was found that 5 mg/kg rapamycin group's values were significantly lower than the control groups. These results show that acute administration of rapamycin, especially in 5 mg/kg dose of rapamycin prolongs the latency of maternal aggression, and decreased the number of attacks, the intensity of attacks and the total duration of aggressive behaviors in rats. Therefore rapamycin may have potential for use as a sedative drug, however it is necessary to conduct further studies.

Keywords: *Maternal aggression, Rapamycin, mTOR, Rat*

Sıçanlarda Rapamisinin Maternal Agresyon Üzerine Etkisi

Özet

Memelideki rapamisin hedefinin (mTOR) bir inhibitörü olan rapamisin antineoplastik, yaşlanmayı geciktirici, anti-inflammatuar ve nöroprotektif etkilere sahiptir. Bu çalışmadaki amacımız sıçanlarda rapamisinin maternal agresyon üzerine etkisini araştırmaktır. Çalışmada 63 adet dişi Wistar sıçan kullanıldı. Hayvanlar çözücü (DMSO) grubu, 5 mg/kg rapamisin grubu ve 10 mg/kg rapamisin grubuna ayrıldı. Davranış testi için ev sahibi-yabancı paradigması kullanıldı. Gruplar; ilk agresif davranışın başlama zamanı, toplam atak sayısı, agresif davranışın toplam süresi ve atak şiddeti açısından karşılaştırıldı. Gruplar ilk agresif davranış başlama zamanı bakımından karşılaştırıldığında 5 ve 10 mg/kg rapamisin gruplarının kontrol grubuna göre başlama zamanını anlamlı düzeyde uzattığı bulundu. Toplam agresif davranış sayısı, agresif davranışın toplam süresi ve ortalama atak şiddeti değerlendirildiğinde 5 mg/kg rapamisin grubunun değerleri kontrol grubuna göre anlamlı düzeyde düşük bulundu. Bu sonuçlar akut rapamisin uygulamasının, özellikle 5 mg/kg dozda, sıçanlarda maternal agresyonun başlama zamanını uzattığını, toplam atak sayısı, atak şiddeti ve agresyonda geçen toplam süreyi kısalttığını göstermektedir. Fakat rapamisinin sedatif bir ilaç olarak kullanılabilme potansiyeline sahip olabilmesi için daha ileri çalışmalar gerekmektedir.

Anahtar sözcükler: *Maternal agresyon, Rapamisin, mTOR, Sıçan*

INTRODUCTION

Many animals may show aggression to protect their children and defend their habitats from other animals (intruders), or do harm to other animals, for preying or outflank the mating. There are many types of aggression in animals. One of the important aggression types is maternal aggression in animals. Many of the lactating female mammals display an aggressive behavior, which is called maternal aggression, to protect their pups from intruders in their living area [1,2]. In lactating females aggression is temporarily increased, and this aggressive behavior is a remarkable feature during the first two or three weeks after delivery. After the third weeks of delivery,

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aggressive behavior in lactating females decreases and later disappears even if the lactation continues [1]. Despite alterations in many hormones and neurotransmitters (i.e. oxytocin, serotonin, dopamine, GABA…etc) levels within the brain during lactating period, underlying mechanisms of maternal aggression still remains unclear [3,4].

In modern medicine rapamycin, which is a macrolide antibiotic is used as an immunosuppressive serine-threonine kinase inhibitor. Rapamycin at low doses has immunosuppressive effects, and at higher doses it shows antifibroblast, antiproliferative and antineovascularization effects. Rapamycin (Sirolimus) binds to intracellular receptor FKBP12 to form immunophilin complex in the cell, and this immunophilin complex binds directly to the FKBP12-Rapamycin Binding (FRB) domain of mTOR for inhibiting mTOR's activity [5].

Mammalian target of rapamycin (mTOR) is a serine/threonine protein kinase (PI3K), and it is a member of the phosphatidylinositol 3-kinase (PI3K) related kinase family. mTOR consists two multi-protein complexes defined as distinct protein binding partners with Rapamycin (Sirolimus). The first complex is mTORC1, which is known mTOR's rapamycin sensitive complex, and the other one is mTORC2, which is largely insensitive to rapamycin [6]. In studies about the mTOR's effect on the nervous system by using the mTOR inhibitors like rapamycin, it has been shown that mTOR is very important for the development of the central nervous system's cell survival, differentiation, axonal development, synaptogenesis; in adult synaptic plasticity, such as long-term potentiation which plays an important role in the process of learning and memory in hippocampus [7-10]. mTOR which is inhibited by rapamycin, is known to be effective in various cellular and molecular processes like neurotransmitter, receptor and ion channel expression, neuronal death, apoptosis and neuronal excitability in the CNS.

Changes in maternal behavior have been observed in mice, rats and other laboratory animals [2,11]. Differences in maternal aggression have been observed between the species, strains and even subtype [11]. The effects of various substances on maternal aggression in different animal species or different species or strains of laboratory animals has not been studied yet. Experimental evidence indicates that most mother rats, mice or hamsters that have recently given birth do not display maternal aggression [12,13]. In a study by Gammie and Nelson [13], it was found that 43% of the lactating animals did not show aggressive behavior. Similarly in another study 47% of the animals did not show any aggressive behavior [14]. It has been suggested in some of the studies that, maternal aggressive behavior occurs in response by gender of the intruding animal [2,12]. For this reason we used female virgin rats as intruders in our studies. In this study, all experimental animals were randomly divided into different groups regardless of whether they had exhibited maternal aggression to female intruders naturally or not. Animals were compared with each other with regard to selected drug effects. In these groups, it was included mother rats which had not displayed aggressive behavior under all test conditions and in natural settings.

In the present study, our aim was to investigate the acute effects of 5 mg/kg and 10 mg/kg of rapamycin on maternal aggression in rats.

MATERIAL and METHODS

Animals and Housing

All the experiments were performed in accordance with the Declaration of Helsinki and the local Bioethical Standards of Animal Experiments. The experimental protocol was approved by the Animal Ethics Committee at Abant Izzet Baysal University (2013-37). The animals which were used in the study were provided by Abant Izzet Baysal University, Experimental Animals Research Center. Sixty-three Wistar female rats (90 days old, body weight 225±25), which were used for the experiment, were kept in the laboratory conditions; 23°C room temperature, 60±5% humidity and 12:12 light-dark cycle, in optimal values, and with access to food and water ad libitum. Animals were housed in standard polycarbonate cages; the floor of the cages was covered with wood shavings, which were used as bedding and nesting material. Each female rat (70 days old) was housed with a stud male rat in the same cage until the female rats get pregnant. Vaginal smear tests were performed in order to confirm that the animal was pregnant. After the beginning of pregnancy; male rats were removed from the cages.

Drugs and Doses

In this study, the effects of rapamycin on maternal aggression, in the doses of 5 mg/kg and 10 mg/kg were investigated. Rapamycin was purchased from LC Labs (Woburn, MA, USA), and 5 mg/kg and 10 mg/kg rapamycin which were solved in 99% dimetylsulfoxide (DMSO; LobaChemie, India) used as subtances and 1 mL/kg saline was used as control.

The animals were randomly divided into three groups: 5 mg/kg rapamycin (n=16), 10 mg/kg rapamycin (n=16) and DMSO (n=16). As treatment control, 1 mL/kg intraperitoneal (i.p) saline was given to lactating rats. Basal aggression levels were measured in three groups on Day 2 and Day 3. A dose of 5 mg/kg or 10 mg/kg rapamycin, which can be considered as effective dose in pharmacological studies were applied to animals on Day 2 and Day 3 [15].

Rapamycin Treatment

Before administration, rapamycin was first dissolved in DMSO. DMSO defined as vehicle for carrying rapamycin. To determine effects of rapamycin on maternal aggression we used lactating rats both as control and test groups.

We chose the crossover experimental design as optimal method for the study. In the crossover experimental design, each group receives a series of treatments over time. Treatment in a predetermined sequence is given to the each experimental group. The time points at which substance was administered are usually called periods. In a crossover design, each experimental group serves as its own control. Thus, a crossover design should give smaller standard errors for comparisons between treatment groups than a design where treatment groups are assigned to different subjects [16]. Moreover crossover experimental design is used in studies to eliminate the effect of sequence of substance administration.

According to experimental evidence appropriate aggressive behaviors may vary from species to species and even individual to individual [14]. Therefore in our study, it has been used each substance group as its own control instead of using different control group. 5 mg/kg rapamycin, 10 mg/kg rapamycin, DMSO or saline i.p. were given to the lactating rats (each group n=16) for two consecutive days (Day 2 and Day 3), before done the series of behavioral tests with using cross-over experimental design.

In the experiments each group (n=16) was divided into two subgroups (n=8), and then saline, substance and DMSO were administered as following;

For 5 mg/kg rapamycin group (n=16); substance was administered to lactating rats on day 2, and on the same subgroup saline was administered on Day 3 (n=8). For the other subgroup, (n=8) saline was administered on day 2 and substance was administered on Day 3.

For 10 mg/kg rapamycin group (n=16); substance was administered to lactating rats on day 2, and for the same subgroup, saline was administered on Day 3 (n=8). In the other subgroup (n=8) saline was administered on day 2 and substance was administered on Day 3.

For the DMSO group (n=16); DMSO was administered to lactating rats on day 2, and in the same subgroup saline was administered on Day 3 (n=8). In the case of the other subgroup (n=8) saline was administered on day 2 and substance was administered on Day 3.

During the behavioral tests, each lactating rat was recorded with camera for 20 min, and these recordings were watched and evaluated by two trained observer.

The Behavioral Test

There are many paradigms for testing animals' aggressive behavior. One of these paradigms used in the maternal aggression studies is resident-intruder paradigm. The resident-intruder paradigm is a well-validated model of maternal aggression. The date of birth was considered as postpartum Day 0. Animals were tested on postpartum Day 2 and Day 3 after birth, because mother rats display high aggressive behavior towards female intruders mainly in the first lactation week. Aggressive behavior has usually decreased by the second lactation week [17].

On Day 2 and Day 3, the substances were given to the resident rats intraperitoneally 30 minutes before the experiment. The pups were removed from the home cage 5 min before the aggression test to prevent any harmful situation [14,18,19]. Thirty min after - administration of subtances to the resident rat, an intruder female rat was placed into the home cage and then the aggressive behaviors of the resident rat were observed and recorded with video camera for 20 min. To prevent intruder's odorant becoming permeated into the cages or resident animals becoming familiar with the intruders, the cages were cleaned after the each test and a different female rat was used in each test as an intruder. All intruders were young female virgins and their body sizes were not bigger than lactating mothers.

Aggressive behaviors displayed by lactating rats towards the intruder rats were observed and recorded according to the following parameters [12,14]:

a) The latency to the first aggressive behavior: The time of first aggressive attack that directed towards the intruder. If there was not any aggressive behavior, total test time (1200 s) was used as data of the latency for the first aggressive behavior.

b) The number of attacks: The number of attacks which were exhibited towards the intruder rat.

c) The total duration of aggressive behaviors: The total time of aggressive behaviors which were exhibited towards the intruder rat.

d) The intensity of attacks: The intensity of attack exhibited by the resident rat to the intruder which was scored as follows;

(0); no aggressive manifestations;

(1); scattered mild aggressive posture or attack towards the intruder, no vocalizations

(2); scattered upright aggressive posture, violent attack or boxing with the intruder, low vocalizations, but no biting or continuous fighting

(3); continuous fighting or attempts to bite the intruder rat by resident, loud vocalizations.

The lactating rats which did not exhibit any aggressive behavior, were accepted as non-aggressive rats, and they were removed from the study.

Statistical Analysis

In this study, data was collected on the following variables; the latency to first aggressive behavior (sec), the number of attacks, total duration of aggressive behaviors (sec) and the intensity of attacks. Descriptive values of these

variables were computed as mean±SD. Values obtained from the study have not shown normal distribution according to Kolmogorov-Smirnov test. After logarithmic transformation was applied, values have shown normal distribution. Levene test was used for homogeneity of variable of parameters. A crossover experimental design which is taking into account effects of sequence was used for data analysis. During the experimental procedure, experiments were done on Day 2 and Day 3 after birth, and all lactating rats were divided to the groups randomly. Least Significant Difference method with Bonferroni correction as a Post hoc test was used for detecting significant differences. Related crossover experimental design was given at the *Table 1*. If the P value obtained from calculations was smaller than 0.05, the results would be accepted as statistically significant. NCSS (version 11) packaged software was used in calculations.

RESULTS

All of the 63 female rats were used for the experiment, 48 of the mother rats (76%) displayed maternal aggressive behavior in at least one test day, and 15 of the mother rats (24%) have did not show any maternal aggressive behavior in both test days. These non-aggressive rats were removed from further experiments and were not included in further statistical analysis. Because we thought that they did not contribute to the aims of our study which were related to the effects of substances on maternal aggression.

After logarithmic transformation was applied, values have shown normal distribution. Variances of groups were found homogeneous for latency and number of attack, but variances were found nearly homogeneous for total duration of aggressive behaviors and the intensity of attacks.

Crossover analysis of variance was used for the comparisons of the groups in terms of latency to first aggressive behavior, the number of attacks, total duration of aggressive behaviors and the intensity of attacks. Results of analysis showed that there was no significant effect of sequence of administration of the subtances, so crossover model was found as applicable in comparison of substance with saline. Differences between groups were evaluated based on these results and obtained following results.

Effects of Rapamycin on the Latency to First Aggressive Behavior

Following the administration of 5 and 10 mg/kg rapamycin, the latency to first aggressive behavior was significantly increased compared to control (P values; 0.028 and 0.024 respectively). However there was no statistically significant difference between the dose groups (5 and 10 mg/kg), DMSO and other test days' values in terms of the latency to the first aggressive behavior (P>0.05) *(Table 2, Fig. 1A)*.

Effects of Rapamycin on the Total Attack Number

It was found that rapamycin reduced the total number of aggressive attacks. At the dose of 5 mg/kg of rapamycin, the average total number of attack number was significantly low compared to control (P=0.014). However there was no statistically significant difference between the dose groups (5 and 10 mg/kg), DMSO and other test days' values in terms of the total attack number (P>0.05) *(Table 2, Fig. 1B)*.

Effects of Rapamycin on the Total Duration of Aggressive Behaviors

The effect of rapamycin of the total duration of aggressive behavior was found significantly. The total duration of aggressive behaviors for the group administered 5 mg/kg rapamycin was significantly shorter than the total duration of aggressive behavior in the control group (P=0.033). However, there was no statistically significant difference between the dose groups (5 and 10 mg/kg), DMSO and other test days' values in terms of the total attack number (P>0, 05) *(Table 2, Fig. 1C)*.

Effects of Rapamycin on the Intensity of Attack

As a result of the assessments made in terms of the effects of rapamycin on the intensity of attack, it has been found that the group given 5 mg/kg rapamycin showed lower total number of attacks than the control group (P=0.0125). However there was no statistically significant difference between the dose groups (5 and 10 mg/kg),

Table 1. *Crossover experimental design*
Tablo 1.*Çapraz tasarım deney düzeni*

Groups	Non-aggressive (N)	Aggressive (N)	Administered Substance	
			Day 2	Day 3
5 mg/kg rapamycin	4	8	5 mg/kg rapamycin	1 mL/kg saline
	3	8	1 mL/kg saline	5 mg/kg rapamycin
10 mg/kg rapamycin	2	8	10 mg/kg rapamycin	1 mL/kg saline
	1	8	1 mL/kg saline	10 mg/kg rapamycin
DMSO	2	8	1 mL/kg DMSO	1 mL/kg saline
	3	8	1 mL/kg saline	1 mL/kg DMSO

Table 2. *The effects of saline (control), DMSO, and 5 mg/kg and 10 mg/kg doses of rapamycin on latency of maternal aggression, total attack number, total duration of aggressive behaviors and intensity of attack*

Tablo 2. *Salin (kontrol), DMSO, 5 mg/kg ve 10 mg/kg rapamisin dozlarının maternal agresyonun başlama latensi, toplam atak sayısı, agresyonda geçen toplam süre ve atak şideti üzerine etkileri*

Parameters	Group	N	Mean+	Median	SD	Log-M	Log-SD	P
Latency	DMSO	8	447.75	146.00	497.357	2.310	0.610	0.036
	DMSO control	8	280.44	82.50	407.505	2.065	0.575	
	5 mg/kg rapamycin	8	595.44	359.50	514.963	2.536*	0.526	
	5 mg/kg control	8	303.38	113.00	412.361	2.146	0.563	
	10 mg/kg rapamycin	8	572.94	420.00	463.564	2.575Δ	0.456	
	10 mg/kg control	8	456.25	222.50	430.655	2.461	0.452	
Total Attack Number	DMSO	8	3.63	3.00	3.324	0.596	0.308	0.045
	DMSO control	8	6.31	4.00	6.954	0.669	0.435	
	5 mg/kg rapamycin	8	2.88	2.00	4.334	0.518*	0.349	
	5 mg/kg control	8	6.19	4.00	7.423	0.709	0.330	
	10 mg/kg rapamycin	8	3.38	2.00	3.735	0.597	0.291	
	10 mg/kg control	8	3.56	3.00	3.226	0.520	0.360	
Total Duration of Aggressive Behaviors	DMSO	8	14.75	6.00	22.335	1.040	0.481	0.048
	DMSO control	8	38.00	8.00	70.301	1.174	0.653	
	5 mg/kg rapamycin	8	13.63	3.00	24.503	0.971*	0.601	
	5 mg/kg control	8	19.69	11.00	29.129	1.147	0.391	
	10 mg/kg rapamycin	8	8.13	4.50	8.793	0.962	0.342	
	10 mg/kg control	8	8.13	7.00	7.553	0.8700	0.393	
Intensity of Attack	DMSO	8	5.75	4.00	5.825	0.952	0.390	0.043
	DMSO control	8	11.63	4.00	15.684	0.806	0.572	
	5 mg/kg rapamycin	8	5.44	2.00	10.308	0.674*	0.482	
	5 mg/kg control	8	12.00	6.50	18.889	0.921	0.398	
	10 mg/kg rapamycin	8	4.69	3.00	4.729	0.754	0.283	
	10 mg/kg control	8	5.25	3.50	5.994	0.620	0.447	

+ Original data mean, ***SD;*** *standard deviation,* ***Log-M;*** *logarithmic mean,* ***Log-SD;*** *logarithmic standard deviation, * statistically significant differences compared to 5 mg/kg control group, Δ statistically significant differences compared to 10 mg/kg control group*

DMSO and other test days' in terms of the total attack number (P>0.05) *(Table 2, Fig. 1D)*.

Effects of Pup Number on the Maternal Aggression

When examined relationship between number of pups and maternal aggression, it was determined that the latency to first aggressive behavior decreased significantly when increase in number of pups (P=0.028). In addition, the number of attacks showed increases significantly when number of pups get higher (P=0.049). However, a significant difference was not observed between increasing the number of pups, the total duration of aggressive behavior and severity of attack (P values; 0.090 and 0.094 respectively).

DISCUSSION

In the present study, the effects of i.p administration of 5 mg/kg and 10 mg/kg rapamycin on maternal aggressive behavior which occurred in lactating rats were investigated. Maternal aggression was evaluated by using resident intruder paradigm in this study.

In our study, latency to first aggressive behavior was observed between 3 and 7 min on average. In terms of latency, the longest time was found in the 5 mg/kg and 10 mg/kg rapamycin dose group. In terms of the total duration of aggressive behaviors, the total attack number and intensity of attack, significant decrease was observed only in 5 mg/kg dose group. On the other hand significant changes were observed in 10 mg/kg rapamycin dose group, however it was not significant statistically. DMSO which was used as a solvent did not have any effect on maternal aggression.

In a number of studies, it has been reported that maternal aggressions often seen in lactating rats do not occur in every individual [14]. Gammie and Nelson [13] used the house mouse in their study, and they also reported that the proportion of animals exhibiting aggressive behavior is 57% (n=14). On the other hand in a study in

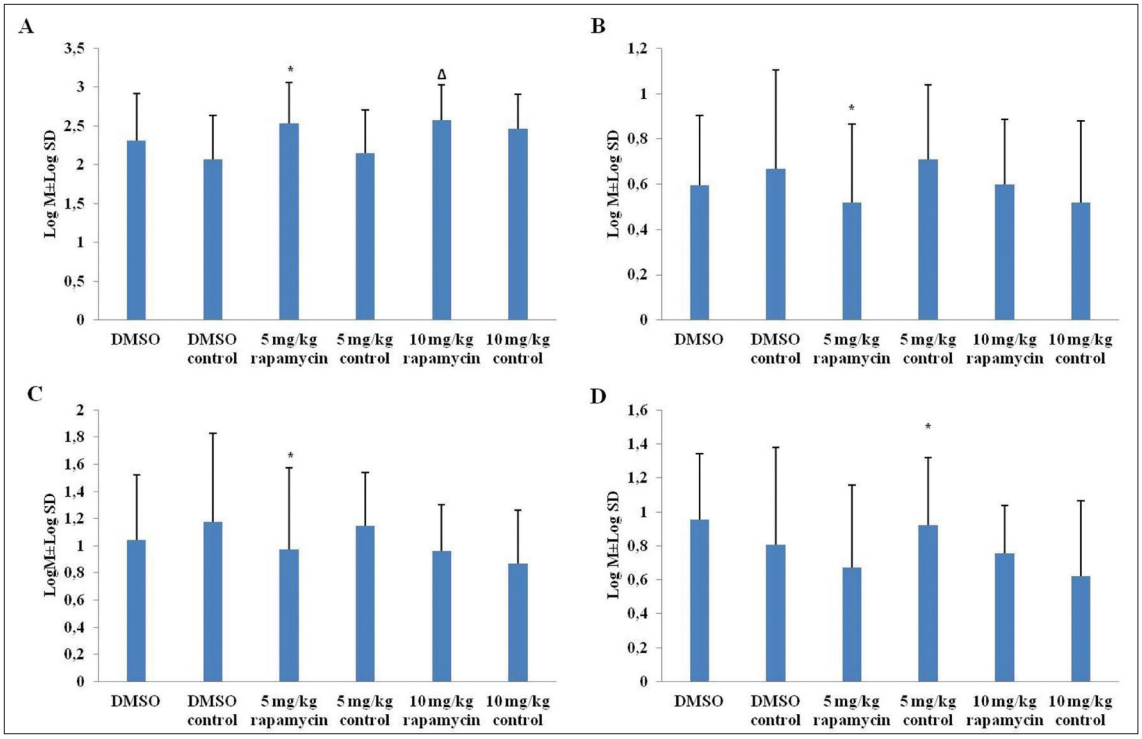

Fig 1. The effects of saline (control), DMSO, and 5 mg/kg and 10 mg/kg doses of rapamycin on latency of maternal aggression (A), total attack number (B), total duration of aggressive behaviors (C) and intensity of attack (D). * statistically significant differences compared to 5 mg/kg control group, Δ statistically significant differences compared to 10 mg/kg control group

Şekil 1. Salin (kontrol), DMSO, 5 mg/kg ve 10 mg/kg rapamisin dozlarının maternal agresyonun başlama latensi (A), toplam atak sayısı (B), agresyonda geçen toplam sure (C) ve atak şideti (D) üzerine etkileri. *5 mg/kg kontrol grubu ile karşılaştırıldığında istatistiksel olarak anlamlı, Δ 10 mg/kg kontrol grubu ile karşılaştırıldığında istatistiksel olarak anlamlı

which the wistar rats were used, it was reported that the proportion of animals exhibiting aggressive behavior was 75% [20]. Ankarali et al.[14] reported that the proportion of animals exhibiting aggressive behavior was 53%. In our study, the proportion of the study subjects displaying maternal aggression was 76% (48 of the 63 animals). In some studies, it has been reported that mothers with birth experience show more aggressive behavior [21]. In our study some of the lactating rats (n=15) did not display maternal aggressive behavior, and the reason that lactating rats did not exhibit aggressive behavior may due to laboratory environment, or mother rats inexperience on birth and puppy care. Lactating rats aggressive behaviors toward intruders depends on the intruders' age and hormonal status [22].

This study is the first study about rapamycin's effect on the time to the onset of first episode of aggressive behavior (latency), the total number of attacks, the intensity of attacks and the total duration of aggressive behaviors. There is only one previous study on effects of rapamycin on aggression seen in pilocarpine animal model for epilepsy. In the study which is done by Huang et al.[15], the sex of the animals was not specified, and resident-intruder paradigm was used. Huang et al.[15] reported that rapamycin reduced aggression in pilocarpine induced epilepsy in an animal epilepsy model. Our study findings that rapamycin administration reduced aggressiveness were compatible with the literature. However, the time to first episode of maternal aggressive behavior, the total number of attacks, the total duration of aggressive behavior and the intensity of attacks could not compared due to lack of studies in literature.

Psychiatric disorders are common in persons with epilepsy [15,23]. Aggression is one of several psychiatric disorders that have long been observed in epileptic patients including those with temporal lobe epilepsy [24], cortical dysplasia [25] and tuberous sclerosis [15,26]. The relationship between aggression and epilepsy has been shown in several animal models, including models of pilocarpine [27] and domoic acid [28]. The underlying molecular mechanisms of association between aggression and epilepsy remain unknown.

Furthermore, in a study related to aggression and epilepsy, not only epileptic seizure has occurred in animals but also aggression level of the animals has been observed increase substantially in pilocarpine induced epilepsy model. Huang et al.[15] has reported that pilocarpine induced status epilepticus is accompanied by an increase aggressive behavior in rats, and this increase in aggression was by 5 mg/kg rapamycin. These data support our results which demonstrated that 5 mg/kg dose of rapamycin reduced the maternal aggression. Rapamycin is a potential mTOR inhibitor, and has been shown to reduce aggressive-

ness. This suggests that there is a relationship between mTOR hyperactivation and mechanism of aggression.

Generally, it is assumed that maternal aggression may be triggered by changes in level of hormones on last period of pregnancy or after delivery [29]. Although different results have been obtained in many studies, changes levels of estrogen, oxytocin and prolactin are thought to be responsible for observed maternal aggression.

There are very few studies regarding the role of GABAergic activity on maternal aggression. Increased GABA release may be important for maternal aggression in rodents. Because GABA agonists administered to virgin female rats treated like lactating mother rats, however GABA antagonists administered lactating mother rats' maternal aggression have decreased [30]. GABAergic activity is necessary for maternal aggression in rats. It has been demonstrated that maternal aggression is generated in specific areas of the brain. When bicuculline, which is a receptor antagonist of $GABA_A$, is infused into hypothalamus or medial amygdala reduces aggressive attacks [31], and infusion bicuculline into the central periaquaductal grey matter inhibits maternal aggression [32]. Although Bjork et al.[33] have reported that there is positive relationship between plasma level of GABA and maternal aggression based on their family history of psychiatric patients, how a connection between plasma GABA levels and nerve tissue is not yet established.

Weston et al.[34] found that mTOR regulates growth of GABAergic neurons and synaptic transmission in their study. However they have also reported that use of rapamycin inhibits these regulations. By suppressing mTOR activity, rapamycin inhibits GABAergic activity, and consequently leads to reduction in maternal aggression. This supports the findings of our study.

Although mTOR's molecular mechanism play role in regulation of maternal aggression, these mechanisms have not been fully studied yet. It is known that mTOR pathway is involved in many processes like the regulation of axonal growth, synaptogenesis, receptor and channel expression. All of these processes may cause to increased excitability in the brain, and this increased excitability may lead to maternal aggression [35].

Results from our previous and present studies indicate that rapamycin has dual antiaggression properties [36]. Again, some antiaggressive drugs such as diazepam and buspirone also possess dual activities [37]. It will be interesting to see if other antiaggressive drugs have a similar effect in this model in the future.

Consequently, it is thought that as a result of suppression or reduction of all specified process (mTOR pathways and its process) by rapamycin regulates maternal aggressive behavior. We expect from the future studies that the role of rapamycin on the maternal aggression will be clarified. Rapamycin remains a prospective agent for treatment of maternal aggression.

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Conflict of Interest

The authors declare that there are no conflicts of interest. All authors approved the final manuscript.

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