Dr. Sema KURNAZ ÖZBEK (Histoloji)

Investigation of melatonin protectiveness against adverse effect of chemotherapy (bep regimen) on testicular tissue and sperm by applying various methods

Objective: The purpose of this study is to evaluate side effects of BEP regimen on testicular tissue, sperm structure and sperm parameters which is commonly used in cancer therapy and to investigate the protective effects on male fertility in the implementation of the antioxidant properties of melatonin as a factor for the reduction or prevention of these effects.

Method: A total of 72 male Wistar albino rats, aged 13-15 weeks, were included in the study. Each experimental animal group were divided into six groups and composed of twelve rats. Rats are assigned to the following experimental groups (G): (G1) control group, (G2) control group of melatonin (ethanol+saline), (G3) BEP treated group, (G4) BEP treated group (containing a 9-week recovery period), (G5) BEP+Melatonin treated group, (G6) BEP+Melatonin treated group (containing a 9-week recovery period). At the end of the experimental period, blood samples were taken from the animals by intracardiac infusion and after decapitation, testes and epididymides were removed by opening the abdomen of the animals and their weights will be measured and stored. Left testes were fixed in Bouin solution and examined by histochemical, immunocytochemical and electron microscopy methods. Right testes were allocated for biochemical measurements and western blotting studies. Sperm samples taken from the caudal region of the epididymis were examined under a phase-contrast microscope. Sperm counts were performed and sperm were evaluated for motility. In addition, the integrity of the sperm DNA was examined and morphological structure of sperm was evaluated by performing toluidine blue staining.

Results: Testicular structure and biochemical parameters were normally observed in rats of Saline and Ethanol + Saline group ($p<0,05$). Vacuolysis, immature germ cells, giant cells and basal lamina thickening was found in the seminiferous tubules of the BEP group. At the same time, apoptotic cells were seen more when compared to other groups. Biochemically, antioxidant levels and testosterone levels were low and oxidant levels, FSH (Follicle Stimulating Hormone) and LH (Luteinizing hormone) levels were higher in testicular tissue and blood samples. All values of BEP + Melatonin and BEP + Melatonin + Recovery groups were almost the same as the control group ($p<0,05$).

Conclusions: The BEP regimen causes oxidative stress in the testis, resulting in structural defects, and melatonin improves the oxidative damage of the BEP regimen in the testis by the action of antioxidants.

Dr. Özgür Doğa ÖZSOY (Biyokimya)

Chronic administration of alcohol in rats to evaluate the effects on brain tissue oxidation and neurotrophin levels

Objective: Chronic alcohol use disrupts the functioning of many organs/ systems in the body, especially in the liver and brain, and in some cases creates permanent damage. The aim of this study is to investigate the effect of ethanol oxidative stress on the levels of BDNF, NT-3, NT-4, AOPP, 8-OHDG and IL-1β in brain tissue.

Method: Six months old Wistar Albino rats were used in the study. There are 3 groups in total, namely Saline group (SG), chronic alcohol group (CAG) and chronic sucrose group (CSG). Our work lasted 8 weeks. SG: These animals have not received any treatment. CAG: These animals were given 5 g/kg of 20% alcohol by gavage for 5 days per week. CSG: These animals were given 5% sucrose by gavage for 5 days per week. The amount given is calculated as the calorie equivalent of alcohol. The parameters of our study were also analyzed using ELISA kits.

Results: There was no statistically significant difference between groups in cortex, brain stem and hippocampus tissues and serum BDNF levels. There was a significant increase in cortex NT-3 levels in KSG compared to SG and CAG, respectively (p= 0,009, p= 0,049). There was a significant decrease in KSG NT-3 levels in brain stem compared to SG and CAG, respectively (p= 0,003, p= 0,016). There was a significant decrease in KSG NT-4 levels in brain stem compared to SG and CAG, respectively ($p<0.0001$, p= 0.002). At AOPP levels, there was a significant decrease in serum in CSG according to SG (p= 0.029). At the 8-OHDG level, which is a DNA damage indicator, there was a significant decrease in serum in CSG according to SG (p= 0.009). When the levels of IL-1β in the hippocampal tissue were examined, there was a significant decrease in CSG relative to SG (p = 0.002).

Conclusions: Although ethanol has no direct effect on the brain and even peripheral tissues, it can be said that the duration of exposure to ethanol may have been short. Interestingly, it is possible to say that sucrose, which is thought to be the equivalent of calories, is especially affected in the tissues of the cortex and brain stem and in the peripheral tissues in the light of serum results.

Dr. Sabriye KARADENİZLİ (Fizyoloji)

Investigating the impact of endoplasmic reticulum stress (ers) on unfolded protein response (upr) in absance epilectic rats

Objective: Endoplasmic Reticulum(ER) is associated with many cellular functions, from post-transcriptional modifications to the correct folding of proteins. Disturbances in intracellular calcium balance disrupts the physiological balance of ER. Although studies have shown that the pathology of neurodegenerative diseases is associated with ER stress, studies showing a relationship between epilepsy and ER stress are very limited. The aim of our study was to investigate the effects of ER stress induced by low and high doses of Thapsigargin(Tg) on behavioral learning, epileptic activity, and molecular levels in cortex and thalamus tissues of Wistar Albino Glaxo/Rijswick (WAG / Rij) rats.

Methods: 6-8 month old WAG/Rij male rats were used in this study. Separate groups were created for epileptic activity, behavioral-learning tests, and molecular studies. Electrodes for EEG recordings and cannulas for applying Tg, SF(Saline) and DMSO(Dimethyl sulfoxide) intracerebroventricularly, were placed on skull. EEG recors were taken to assess epileptic activity. Locomotor activity, passive avoidance and water-maze test were performed for behavior-learning experiments. RT-PCR technique was used to examine the expression of PERK, XBP-1, ATF6, CACNA1H mRNA and western blot technique to examine GRP78 and ERp57 protein changes.

Results: 20 ng and 200 ng Tg administration were not effective on locomotor activity and passive avoidance, 200 ng Tg administration showed a significant decrease in water-maze test compared to naive and 20 ng Tg group. It was found that the activity of SWD in 20 ng Tg administration was higher than SF(Saline), DMSO and 200 ng Tg groups at 24th hour and had remained to be high in 48th hour. In the group treated with 20 ng Tg increased GRP78 protein amount, Eif2ak3, CACNA1H and XBP-1 mRNA expression in the talamus were observed. An increase in expression of ATF6 mRNA in the thalamus tissue was observed with 200 ng Tg administration.

Conclusions: Increased level of GRP78 protein amount EIF2AK3, XBP-1 and CACNA1H mRNA expression in low-dose ER stress conditions may exert an enhancing effect on SWD activity by triggering survival processes of thalamic neurons. Increased ATF 6 mRNA expression in high-dose ER stress conditions may have reduced SWD activity by triggering pro-apoptotic processes.

Dr. Tuğçe DEMİRTAŞ ŞAHİN (Farmakoloji)

Investigation of the effects of stem cell therapy and conventional treatment on cardiovascular changes in genetic absence epileptic wag/rij rats

Objective: In this study we investigated the efficacy of neural stem cell (NSC) and chronic ethosuximide treatment in absence epilepsy and their effects on the potentially observable cardiovascular changes in WAG/Rij rats with genetic absence epilepsy.

Method: We divided 2 month old Wistar and WAG/Rij rats into four groups: Control, NSC, Sham and Ethosuximide. NSCs taken from fetal medial ganglionic eminence (MGE) were transplanted to the NSC groups and we waited for 3 months for cell differentiation. We determined the cell differentiation into neurons, astrocytes, oligodendrocytes and GABAergic neuronsin vitro. The ethosuximide groups received chronic ethosuximide treatment for 3 months. At the end of this period; the number, mean duration and total duration of spike wave discharges (SWD) were evaluated using EEG recordings. Each group was then divided into two subgroups. In the first group mean arterial blood pressure (MAP) and heart rate (HR) measurements were performed. In the second group, in vitro isolated organ studies were conducted on the thoracic aortas of animals.

Results: MGE-derived NSCs were found to differentiate into astrocytes, neurons, oligodendrocytes and GABAergic neurons in vitro. Ethosuximide and NSC treatment significantly reduced the number, mean duration and total duration of SWDs in WAG/Rij rats compared to control and sham groups. In WAG/Rij control, NSC and sham groups, MAP was significantly higher than Wistar control. HR in Wistar ethosuximide group was significantly lower than Wistar control group. KCl contraction responses increased in the Wistar ethosuximide and decreased in the WAG/Rij control group compared to Wistar control group. Carbachol relaxation responses significantly increased in the WAG/Rij control and decreased

in the Wistar ethosuximide group compared to Wistar control group. There was no significant difference in SNP responses.

Conclusions: NSC treatment is a potential alternative to conventional antiepileptic drug therapy in absence epilepsy. Cardiovascular function changes have been observed in the WAG/Rij rats with absence epilepsy and as a result of ethosuximide treatment.

Dr. Sertan ARKAN (Fizyoloji)

The investigation of calcium binding proteins on cortical neurons of genetic absence epileptic rats

Objective: Absence epilepsy is a kind of epilepsy, which is classified idiopathic generalized epilepsies. Experimental studies on absence epilepsy are performed on genetically absence epilepsy rat models. One of these models is WAG/Rij rats, that are in-bred rats. Recent studies have shown the existence of hyperexcitable focus in somatosensory cortex of these rats. Cellular and molecular reasons of the formation of this hyper excitable cortical focus remains unknown. However, there is some evidence that GABAergic system deficits might contribute to development of this hyperexcitable focus.

Methods: In this study, we investigate the changes of Parvalbumin, Calretinin and Calbindin proteins, which are neuronal marker, and have intracellular calcium binding properties buffers during epileptogenesis. In our study, we research the cellular density and regional distribution of these calcium binding proteins (CaBPs) in somatosensory cortices of rats by using immunofluorescence (IF) staining and western blot (WB) methods.

Results: PV expression of the SPo1 regions of WAG/Rij rats, including fronto parietal cortex shows a significant decrease, that is depend on strain, compared to Wistar rats. Even though there is a tendency to decrease with age, there is no statistical significance in Wistar rats.

Conclusions: The most striking result of our study related with CaBPs; the amount of PV decrease (WB) in the cortical areas of epileptic animals by depending on strain, and there is tendency to decrease the number of PV positive neuron (IF). The results of both immunoblotting and immunofluorescence staining show that the inhibitor GABAergic conduction is reduced in the somatosensory cortices of epileptic WAG/Rij rats, and that this reduction is limited to the PV containing neurons.

Doğanhan Kadir ER (Mikrobiyoloji) (Devam Ediyor)

Investigation of the Ability to Form Biofilm and Biofilm Susceptibility of Uropathogenic *Escherichia coli*

Escherichia coli (*E. coli*) is the most common cause of urinary tract infections (UTIs). *E. coli* was divided to four main groups including A, B1, B2 and D with phylogenetic classification. A clonal group, sequence type 131 (ST131) *E. coli* which was identified recently and involved with B2 phylogroup has become a clonal group frequently encountered in UTI. It is known that ST131 *E. coli* has high virulence and resistance ability. The hypervirulent ST131 *E. coli* makes it difficult to treat infections and can cause relapse. ST131 *E. coli*' s biofilm formation ability is one of the important factor that reducing the treatment success.

Routine *in vitro* antimicrobial susceptibility tests used today, do not accurately identify the antimicrobial susceptibility of the biofilm-formed organisms. There is no data available in the literature on biofilm susceptibility of highly resistant ST131 *E. coli*. The aim of this study is to determine to various antimicrobial agents susceptibilities of biofilm-formed ST131 and non-ST131 isolates.

The phylogenetic groups of the isolates were previously determined by PCR-based molecular methods. A total of 78 -including 39 ST131 and 39 non-ST131- *E. coli* were selected from previously identified isolates. Six antibiotics in different groups (amoxicillin/clavulonic acid, gentamicin, nitrofurantoin, ceftriaxone, ciprofloxacin, trimethoprim/ sulfamethoxazole) which are commonly prescribed, will be used in the study. The minimum inhibitory concentrations of microorganisms against antimicrobials will be determined. Then, biofilm is formed on the peg lids with a device (Calgary biofilm device) immersed in 96-well plates and the minimum biofilm eradication concentration will be defined. ST131 and non-ST131 biofilm-associated resistance profiles will be investigated in the context of available data.

In the light of the obtained data, the basic information about hypervirulent ST131 *E. coli* biofilm-associated-resistance will be gathered. Also, biofilm associated resistance, which is one of the most important cause of failure in the treatment of the patients infected with ST131 *E. coli*, will be better understood.

Esra ACAR (Biyokimya) (Devam Ediyor)

The Effect of 25-Hydroxy Vitamin D3 Level on Thioredoxin System and its Relationship with DNA Oxidation in Bladder Cancer

Oxidative stress is the main mechanism in pathogenesis of bladder cancer in many reasons such as smoking, chemical exposure, radiation, eating habits. The Thioredoxin-interacting protein has a major role in the antioxidant defense system. Some studies in recent years have shown that TXNIP expression increases in and around hypoxic areas in different cancer models. Experimental, clinical and epidemiological findings suggest that vitamin D may provide protection against cancer as well as cancer progression in different types of cancer. There is limited data on the relationship between 25 Hydroxy Vitamin D3 and bladder cancer risk and prognosis. Moreover, the mechanisms by which it can act on bladder carcinogenesis are not fully understood.

This study will include tumor tissue and serum samples obtained from transurethral resection (TUR-BC) from patients who applied to Kocaeli University Faculty of Medicine Urology Department between 2017-2018. thioredoxin interactive protein, 8-oxoguanine DNA glycosylase and vitamin D receptor genes level will be investigated by RT-PCR. And serum levels of TRX, 8OHdG, and 25OH vitamin D will be investigated by ELISA. In addition, selenium levels will be investigated by Inductive Coupled Plasma-Optical Emission Spectroscopy (ICP-OES).

The aim of this study was to investigate the protective effect of 25-Hydroxy Vitamin D3 levels in bladder cancer with high exposure to oxidative stress. In addition, the role of thioredoxin system and DNA oxidation levels are compared in the pathogenesis of bladder cancer.