A stereological volume assessment of the corpus callosum in children with cerebral palsy


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ARTICLE INFO

Cerebral Palsy is a disorder caused by many different etiological factors in the prenatal and perinatal period. As with many congenital abnormalities, the abnormalities of the Corpus Callosum also accompany the abnormalities of patients with cerebral palsy. Structural changes as well as the decrease in volume of the corpus callosum of patients with cerebral palsy have been reported in the literature. The aim of this study was to examine the volumes of the Corpus Callosum of patients with cerebral palsy who did not have distinctive abnormalities in magnetic resonance images (MRI). MRI of 22 patients (10 male and 12 female) with cerebral palsy in the radiology clinic were selected from 150 patients with cerebral palsy MRI examinations during the years 2009-2010. Patients who presented with just a headache to the clinic and with normal MRI were used as the control group which comprised of 14 males and 15 females. The volumes of the Corpus Callosum of these patients were estimated using Cavalieri Method-a stereological method. In our study the mean volumes of the Corpus Callosum of the patients with cerebral palsy were significantly low when compared with the control group (p=0.003). When both the male and female cerebral palsy patients were evaluated separately a statistically significant decrease was observed as compared to the control group (p<0.05). As a result the decrease in volumes of the Corpus Callosum of patients with cerebral palsy can be considered significant and this finding can be thought to contribute to the early diagnosis of cerebral palsy.


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A case of acute extradural hematoma: Stereological analysis

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ARTICLE INFO

ABSTRACT

In 1-2% of head injury cases were diagnosed as extradural hematoma (EDH) which is preventable cause of mortality. Acute bilateral form is also uncommon. In the present case we evaluated a case with acute bilateral extradural hematoma and intracranial volumes on cranial CT using stereological morphometrical analysis comparing with age and gender matched healthy subject. A 36-year-old male case presented with acute biparietal extradural hematoma after a traffic accident having a severe head injury was suspected initially and administration of surgical management was considered using Cavalier’s principle. The presentation was only loss of consciousness and the Glasgow coma scale score was 4; an emergency brain CT showed biparietal extradural hematomas and right frontoparietal cranial fracture. Biparietal extradural hematomas evacuated with biparietal craniectomy for brain oedema early but the patient didn’t improve and died 5 days after surgery because of respiratory problems in intensive care unit according to the multi-trauma. In conclusion severe head injury with with a low GCS (<9) although has a poor outcome for this group, early diagnosis and surgery can improve recovery but still a clinical challenge. The stereological method is simple, cheap and unbiased.


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The effect of constitutive ErbB2 receptor overexpression on adult mouse median nerve regeneration: A stereological and biomolecular approach*

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ARTICLE INFO

ABSTRACT

Schwann cells have a central role in nerve repair. NRG1/ErbB pathway is a key axonal signal that regulates Schwann cell proliferation, migration and myelination. In this study we investigate the effect of constitutive ErbB2 receptor overexpression on adult mouse median nerve in physiological and regenerative conditions using a stereological and biomolecular approach. With a design-based quantitative morphology we first evaluated the number of myelinated fibers, the fiber density, the diameters of axons and fibers and the myelin thickness in high resolution light microscopy. Moreover, we evaluated the number of Schwann cells (both myelinating and non-myelinating) as well as the number of myelinated axons in electron microscopy. Stereological assessment of healthy nerves showed that peripheral nerves develop normally in ErbB2-overexpressing animals (BALB-neuT) with no difference in number and size of myelinated fibers compared to wild-type mice (BALB/c). No differences in Schwann cells number between BALB/c and BALB-neuT were seen in electron microscopy. On the other hand, after a nerve crush injury, the motor recovery was faster in BALB-neuT compared to BALB/c mice and the number of regenerated myelinated fibers was higher with a thinner axon and fiber diameter in BALB-neuT mice. Moreover, BALB-neuT showed more Schwann cells nuclei profiles. Real time PCR analysis, performed two days after injury, revealed a decreased expression of all the ErbB receptors and of the transmembrane (type III) NRG1 isoforms both in BALB/c and BALB-neuT mice. By contrast, the level of the soluble NRG1 isoforms (type I/II, alpha and beta) increased and, intriguingly, the expression level in BALB-neuT mice was significantly higher than in BALB/c animals. Altogether, these results suggest that constitutional ErbB2 receptor over-expression does not influence the physiological development of peripheral nerves, while it improves nerve regeneration following traumatic injury, possibly strengthening the up-regulation of soluble NRG1 isoforms.

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The effect of kindling on pyramidal neuron number in hippocampus

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The aim of present work was to investigate the effect of kindling, induced by subconvulsive doses of pentylenetetrazole (PTZ) on neuron number of hippocampus of rat. The experiments were carried out on adult male and female Wistar albino rats. Using unbiased stereology, we calculated approximately total pyramidal neuron numbers in the CA1, CA2, CA3 areas and whole hippocampus after PTZ induced kindling. The numbers of pyramidal cells (area CA1-3) in the hippocampus were estimated and analyzed by the optical fractionator, a stereological counting technique. Groups were compared with the independent samples t-test. Stereological analysis showed that, in kindled rats, there was a significant decrease in the total number of pyramidal neurons in the hippocampus, compared to control groups (p<0.05). The pyramidal neuron numbers in CA1 were significantly lower in both male and female kindled rats compared to the controls (p<0.05). A significant difference was found in CA2 of only female rats (p<0.05). No significant difference was found in the neuron numbers of CA3 in male and female rats (p>0.05). Our study shows that total neuronal number in the hippocampus is reduced after kindling which suggests that kindling may cause neurotoxicity.


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Comparison of cerebral hemispheres volumes and comparison of ratios of white and gray matters volumes in those hemispheres of paw-preference determined rats

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ARTICLE INFO

This study is performed by using 12 female Wistar rats (weighing 200-260 g), which below to KTU Surgical Research Center, at KTU Medical Faculty Department of Anatomy. We attempted to search differences of brain hemisphere volumes also the differences of white and gray matter ratios in these volumes. Paw preference of the rats is determined by food extension test. The obtained material is histologically examined and then sectioned according to the stereological sampling rules. By stereological techniques, volumes of both hemispheres and ratios of white and gray matters are determined. For statistical analysis, paired t-test for parametric test, Man-Whitney U test and Wilcoxon test for non-parametric tests are used by SPSS 13.0. By our measurements, we observed that: Those rats which have the preference of left paw had higher volumes of right hemisphere’s white matter (p=0.046; p<0.05). Ratios of volumes of the left hemispheres’ gray matters to the volume of the same hemisphere are higher for those rats which have the right paw preference (p=0.041, p<0.05). Ratios of volumes of the left hemispheres’ white matters to the volume of the same hemisphere are higher for those rats which have the left paw preference (p=0.015, p<0.05). Ratios of volumes of the right hemispheres’ gray matters to the volume of the same hemisphere are higher for those rats which have the right paw preference (p=0.015, p<0.05). Dominant hemispheres which are defined by examining motor functions are dominant for motor actions. Therefore, in addition to comparison of the whole hemispheres’ white and gray matter volumes, also comparison of specific motor and somatic areas’ white and gray matter volumes and comparison of numbers of neurons in those areas should be performed with further studies.


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Anti-inflammatory effects of inhaler salbutamol on sepsis models of inflammation

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ARTICLE INFO

ABSTRACT

One of the beta-adrenergic receptors (beta-ARs) subtypes is β-2 adrenergic receptors have been implicated in the control of various metabolic processes by catecholamines. We hypothesized that salbutamol would prevent inflammatory and stimulating β-2 adrenergic receptors and during the acute inflammation and DNA damages process in rats. The rats were divided into five groups, each containing six rats: Control, cecal ligation and puncture (CLP), cecal ligation and puncture (CLP)+salbutamol inhaler (1-mg/kg), cecal ligation and puncture (CLP)+salbutamol inhaler (2-mg/kg) and sham operated+ salbutamol inhaler (2-mg/kg). Twenty four hours after the surgery, the rats decapitated and then ovary tissues were removed for histopathologic and stereologic analysis. The results of this study demonstrated, salbutamol dose- dependently decreased the polymorphonuclear cell infiltration and 8 OHDG immune positive cell number in sepsis induced rats ovary. Salbutamol can decrease acute and chronic inflammation, possibly through the stimulation of β-2 adrenergic receptors. These findings suggested here the roles of β-2adrenergic receptors in inflammatory process.

The aim of this study was to evaluate beneficial effects of CAPE on rat aorta in streptozotocin (STZ)-induced diabetes in Wistar Albino rats. In this study twenty-eight male rats were randomly divided into four groups as follows: 1. Group: Control, 2. Group: Diabetic (DM), 3. Group: Diabetic+CAPE (DM+CAPE), 4. Group: Caffeic acid phenetyl ester (CAPE). Diabetes was induced by STZ injection (55 mg/kg). CAPE was administered intraperitoneally at the dose of 10 µg/kg for 20 days. Blood glucose levels and weights of all rats were measured at the beginning and at the end of the study. After 21 days of study, rats were sacrificed under ketamin/xylazine anesthesia, aorta were removed and were applied to routine tissue fixation process and embedded in paraffin. Sections stained with Hematoxylin- Eosin (H&E). Sections were examined using a Leica DFC280 light microscope. In control group’s aorta, tunica intima, media and adventitia showed normal histological appearance. CAPE group was similar to control group. Tunica media thickness were measured. In DM group, light microscopic examination of sections showed extensive thickening of media (504.59±63.9µm) and disorganization in media. Tunica media thickness were found to decreased in DM+CAPE (454.84±53.5µm), compared with DM group (p<0.05). In DM group, increase of connective tissue and inflammation were seen in tunica adventitia layer. CAPE significantly reduced diabetes-induced morphological changes. It was concluded that CAPE is effective in reducing aorta damage induced by STZ.
We aimed to study the protective effect of resveratrol and melatonin on carbon tetrachloride induced ileum injury. In this study, 40 male rats were randomly divided into five groups each containing 8 rats. 1. Group: Saline, 0.5 ml, 2. Group: Olive oil, 0.5 ml, 3. Group: Olive oil-CCl₄, 1 ml/kg/day (1/1), 4. Group: Olive oil-CCl₄, 1 ml/kg+Resveratrol 10 mg/kg, 5. Group: Olive oil-CCl₄, 1 ml/kg+Melatonin 20 mg/kg. In this investigation, applications were injected with intraperitoneal for four days and on fifth day ileum samples were collected. Routine tissue fixation process applied and embedded in paraffin. Blocks were cut at 5 µm and sections stained with hematoxylin-eosin. Each section was observed under 20X magnification and 5 villi were selected randomly for histological analysis. Sections were examined using a Leica DFC-280 light microscope and villi diameter, height and numbers were measured. Control and olive oil groups showed a normal histological ileum architecture. In group 3, numbers, height and diameters of villi were reduced. Melatonin and resveratrol injection reduced number and diameters of villi but heights of villi were elongated. These results suggest that resveratrol and melatonin are effective in ileum damage induced by CCl₄.
Comparison of four different techniques used for the estimation of the brain volume using MRI


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ARTICLE INFO

ABSTRACT

There are several software freely available to estimate the brain volume from MR images using stereology such as EasyMeasure, Analyze etc. The purpose of this study was to compare brain volume estimations, obtained through using the Cavalieri principle as a combination of point-counting technique and different software: EasyMeasure, ImageJ and Stereo Investigator. We assumed to be the best estimate of the true brain volume using the Cavalieri principle as a combination of point-counting technique and it was defined as the reference volume. To test our hypothesis, the volume of the brain was estimated in a total of 30 healthy subjects (17 males and 13 females) using a 3-T scanner. We found that brain volumes obtained by four different methods were not statistically different. Importantly, we noted that the use of EasyMeasure software for time efficient estimate of brain volume takes about a few minutes. Based on our results, it is concluded that estimation of the brain volume using these techniques may be useful parameter for the assessment of the cortical atrophy.


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Comparison of two methods for the estimation of the lateral ventricles volume using fetal magnetic resonance imaging

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ARTICLE INFO

ABSTRACT

The ventricular system of the brain consists of two lateral ventricles and the midline third and fourth ventricles. The lateral ventricles can be measured and used to define ventriculomegaly using different imaging techniques. In this study, lateral ventricles were evaluated by ultrasonography (USG) and magnetic resonance imaging (MRI) in 14 cases. We used two different methods for estimating the lateral ventricles volume in MR imaging. Cavalieri principle, a first method, was used to measure lateral ventricles volume with fetal MR axial images. We used Image J software for estimated volume as second method. We compared the estimated values obtained from two methods with USG, MR images width of lateral ventricles measurements. We estimated the mean lateral ventricles volumes obtained with the Cavalieri principle and Image J software method to be 2.52±1.31 cm³, 2.49±1.35 cm³ respectively. There is no statistically significant difference between the two methods. (p=0.827, p>0.05). Our study has demonstrated a good correlation between fetal lateral ventricles volume with width of the right–left lateral ventricles measured by USG and MRI. Fetal lateral ventricles volume may be easily estimated directly using the Cavalieri principle. These parameters can potentially provide useful information for assessing fetal brain and lateral ventricles development.

Repeated controlled ovarian hyperstimulation (COH) treatment effects on the number of primordial and primary follicle number: A stereological study

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ARTICLE INFO

**ABSTRACT**

COH is a widely used treatment for the multiple ovarian follicle growth for oocyte collection, in in vitro fertilization (IVF) cycles. Despite the increasing success rate of IVF, most infertile couples need more than one cycle of COH treatment to achieve pregnancy. The aim of this study was to investigate the effects of repeated COH treatments on primordial (PrF), primary (PF) and non-growing follicle (NGF) pool and ovarian structure. In this study 21 young (5week) and 21 aged (6 month) Wistar albino female rats seperated randomly to control, 2cycle and 8cycle COH treatment subgroups. Control groups injected only with %0.9 NaCl, where 2cycle and 8cycle subgroups recived sequentially 2 and 8 times COH protocol that contains injection of 15IU/0.2 ml/rat rFSH and following 48 hours 5IU/0.1 ml/rat HCG injection for ovulation. At the end of the experiment rats were killed and ovaries were fixed in Bouin’s fluid. In the H-E stained sections PrF and PF numbers were counted with an unbiased stereological optical disector method. Both in young and aged rats between control, 2cycle and 8cycle COH treatment subgroups there were no statistically significant differences of non-growing follicle (NGF) numbers (the sum of PrF and PF numbers) that represents the ovarian reserve. The PF numbers found to be increased in 2cycle COH subgroups of aged and young rats compared with controls, were in accordance with the literature that has been shown that inactive follicles were activated by gonadotrophins. However, 8cycle COH subgroups have less PF than 2cycle COH subgroups. We suggested that this numerical decrease is thought to be enhanced due to damage caused by the exogenous hormones used in COH treatments reported in literature. In conclusion the results of this study indicates that giving a resting period between consecutive cycles will enhance retrieval of healthy follicle numbers and oocyte quality in a subsequent cycle.


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The effects of cape on streptozotocin-induced diabetic liver injury in rats

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ABSTRACT

This study was designed to investigate the improving effects of CAPE on the histological alterations in liver in streptozotocin (STZ)-induced diabetic rat model. Male Wistar rats were divided into 4 groups: (1) Control group, (2) CAPE-treated nondiabetic group, (3) Diabetic group and (4) CAPE-treated diabetic group. Diabetes was induced by STZ injection (55 mg/kg). CAPE was administered intraperitoneally at the dose of 10 µg/kg for 20 days. Blood glucose levels and the weights of all rats were measured at the beginning and at the end of the study. After 21 days of study, rats were sacrificed under ketamin/xylazine anesthesia, liver were removed and samples were applied to routine tissue fixation process and embedded in parafin wax. Liver damage was examined by using hematoxylin and eosin, periodic acid-Schiff and Toluidin blue staining methods were applied to serial sections obtained from parafin blocks and preparations were examined on Leica DFC-280 light microscop. In diabetic group, blood glucose levels were increased and the body weights were decreased compared to the control group. The liver structure was normal and similar both in the untreated or CAPE administered control rats. In the STZ group, major histological alterations were observed such as mononuclear cell infiltration, hemorrhage. The glycogen storage in hepatocytes was observed as decreased by PAS staining in the STZ group as compared to controls. Moreover, number of mast cells in portal area was increased had a statistical significance compared to the control group. CAPE treatment did not completely ameliorate these lesions and milder degenerative alterations as loss of the glycogen content was still present. However, histopathological changes were not as extensive as in the STZ group. In this study, as a result of all these morphological and biochemical findings, it is concluded that CAPE has a protective effect against the hepatotoxicity produced by STZ-diabetic rats.


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Effect of CAPE on testicular damage in streptozotocin-induced diabetes rats

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In the present study, we aimed to demonstrate the effect of CAPE on testicular damage in male rats with streptozotocin (STZ)-induced diabetes. Male Wistar rats were divided into 4 groups: (1) Control group, (2) CAPE-treated nondiabetic group, (3) Diabetic group and (4) CAPE-treated diabetic group. Diabetes was induced by STZ injection (55 mg/kg). CAPE was administered intraperitoneally at the dose of 10 µg/kg for 20 days. Blood glucose levels and the weights of all rats were measured at the beginning and at the end of the study. After 21 days of study, rats were sacrificed under ketamin/xylazine anesthesia, testis were removed and samples were applied to routine tissue fixation process and embedded in paraffin wax. Testicular damage was examined by using hematoxylin and eosin staining methods applied to serial sections obtained from paraffin block. Preparations were examined on Leica DFC-280 light microscope and seminiferous tubule diameter and thickness of and germinal epithelial were measured. In diabetic group, blood glucose levels were increased and the body weights were decreased compared to the control group. Diabetic rats showed a reduction in seminiferous tubule diameter and thickening of germinal epithelial and degenerated germ cells. CAPE significantly attenuated the diabetes-induced morphological changes. These results suggest that intraperitoneal administration of CAPE for 20 days is a potentially beneficial agent to reduce testicular damage in adult diabetic rats, probably by decreasing oxidative stress.

Effect of chronic organophosphate exposure on the pyramidal cells of the hippocampus in the adult male rat

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ARTICLE INFO

Organophosphate compounds are highly toxic chemicals with widespread applications, despite rising concerns over their ability to damage the developing brain. We therefore intended to investigate the chronic toxic effect of organophosphate compounds on pyramidal cells numbers in the CA region of the male rat hippocampus using the optical fractionator method. Eighteen male adult Wistar albino rats, with initial weights of 270–300g, were used. Rats were randomly divided into three equal groups. The fenthion group (n=6) was administered 30 mg/1 ml saline/1 kg body weight fenthion by gavage per day, and the sham group (n=6), was given 1 ml saline by gavage per day. Nothing was administered to the control group (n=6). Animals lived freely in their cages under normal laboratory conditions without stress. At the end of the 30th day, all rats were sacrificed and their brains were removed. The left hemispheres were blocked and cut into serial sections of 30-μm thickness in the coronal plane using a rotary microtome. The sections were mounted onto gelatinized glass slides and stained with 0.1% cresyl violet. The total number of pyramidal cells in the CA of the hippocampus was estimated using the optical fractionator technique. The results showed that pyramidal cell numbers in the fenthion group rats were lower than those in the control and sham groups. Comparing the three groups, a statistically significant decrease in pyramidal cell numbers was observed in the fenthion group (p<0.05). There was no statistically significant difference between the control and sham groups’ pyramidal cell numbers in the hippocampus. Chronic fenthion intoxication significantly reduces total pyramidal neuron numbers in the hippocampus and increases apoptosis.


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Effect of different periods of ischemia on the hippocampal volume and total number of neurons in the chicks

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ARTICLE INFO

Previous studies suggest that cerebral ischemia may lead neuronal death and threatens human life and brain functions. Main purpose of our study is to investigate the possible changes in neuron number after different periods of cerebral ischemia in domestic chicks. Animals were divided into 5 groups as cerebral ischemia for 15 minutes (Group I), surgical stress for 15 minutes (Group II), cerebral ischemia for 30 minutes (Group III), surgical stress for 30 minutes (Group IV) and pure control group (Group V). Surgical intervention and the model of ischemia were applied on the second day postnatally. For this purpose, fifteen minutes of ischemia was applied to the Group I (n=5) and thirty minutes of ischemia was applied to the Group III (n=5) by the occlusion of the internal carotid arteries. Internal carotid arteries were isolated by cervical surgery without applying ischemia to the chicks for 15 minutes in the Group II (n=5) and for 30 minutes in the Group IV (n=5). No surgical intervention was performed on the chicks in the Group V (n=5). Afterwards, animals were perfused and the brains of them were removed at the end of postoperative day of 10th. The brains were embedded in paraffin and cut into serial sections with at thickness of 40 μm. Optical fractionator technique was used for cell counting on these sections that were stained with cresyl fast violet. There was no significant difference found not only in neuron number but also the volume of hippocampus among groups (p>0.05). No significant difference among groups suggest that the possible acute neuronal damage resulted from surgical operation may disappear after one week. Additionally these findings may indicate that such interventions are not sufficient to induce a marked neuronal death in domestic chicks.


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Poster

Effects of salbutamol on sepsis-induced models of inflammation in rat’s ovary


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ABSTRACT

β-2 adrenergic receptors possibly have a role in modulation of inflammatory process. In addition, salbutamol is a β-2 adrenergic receptor agonist, which may have beneficial anti-inflammatory and analgesic effects. Therefore, in this study, we investigate the effect of salbutamol on the sepsis-induced inflammatory activity and DNA damages in rat’s ovary. We used thirty female rats and the rats were divided into 5 groups; control group, cecal ligation and puncture applied (CLP), CLP+ low-dose salbutamol given group (1-mg/kg), CLP+ high-dose salbutamol given group (2-mg/kg), sham operated group+salbutamol (i.v.) (2-mg/kg). Polymicrobial sepsis was induced through cecal ligation and a two-hole puncture in CLP groups. Twenty-four hours after the surgery, the rats were decapitated and then ovary tissues were removed for histopathologic and stereologic analysis. Both doses of salbutamol effectively diminished polymorphonuclear cell infiltration and 8-OHDG immune-positive cell in sepsis induced rat’s ovary. As a result Salbutamol can decrease the inflammation, possibly through the stimulation of β-2 adrenergic receptors. This anti-inflammatory effect may be of significance in treatment of sepsis mediated infertility.


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Estimation of the brain surface area using invariator with MR imaging: A pilot study

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ARTICLE INFO

ABSTRACT

Although there are lots of stereology studies focusing on the volume estimation of brain using segmentation and stereological methods, there are a few studies on the measurement of the surface area of the brain using different methods in the literature. In this study, an applied measurement of brain surface area on MR images is presented using invariator. Importantly, we observed that this method has an advantage for application on a pivotal section using a two dimensional device. Based on our findings, we suggest that surface area of brain using this technique may provide important information in some neurological and neuropsychiatric disorders.

Evaluation of the effect of mobile phone exposure on the testis morphology in rats: A stereological study

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ARTICLE INFO

Abstract

The electromagnetic energy emitted from mobile phone (MP) reveals thermal, non-thermal, genetic and cancerogenic effects on the tissues. In the present study, we proposed to evaluate the effect of electromagnetic waves emitted from MP on the rat testis morphology using stereological techniques. A total of thirty-two (n=32) male Wistar albino rats 8-10 weeks old represented this study’s material. Animals were randomly divided into four groups as mobile phone speech (S, n=8, 2 h/day), mobile phone stand by (ST by, n=8, 12 h/day), sham (Sh, n=8, 12h/day) and control (C, n=8). The electromagnetic field was produced by a mobile phone and it had 900 MHz carrier frequency, 217 Hz modulation frequency, 250 mW maximum averaged power, 2 W maximum peak power. The experiment duration was 10 weeks for all groups. Testis volume was determined by the help of a computer-assisted stereological analyses device that uses a special software called Stereo Investigator (Version 8.0, MicroBrightField, USA). The volume of testes were evaluated by Cavalieri’s point counting method. A decline was observed in the testis weights of the both ST by and S groups than the Sh and C groups. The mean weight of testes of ST by was less than weights of Sh and C groups (p<0.05). The volume of testis of ST by and S groups were less than Sh and C groups. The testis volume of ST by and S groups were less than the testis volume of C group (p<0.05). There was also statistically difference between testis volume of ST by and Sh groups (p<0.05). The results of this study showed that the electromagnetic waves exposed from mobile phone negatively affect the rat testis weight and volume.


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Stereological evaluation of post-traumatic peripheral nerve regenerating fibers and denervated-related muscle fibers after VEGF gene therapy

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VEGF
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ABSTRACT

After peripheral nerve injury it is necessary to improve post-traumatic nerve repair and to prevent the fiber atrophy of the denervated related skeletal muscle. In this study we applied the gene therapy based on Adeno-Associated Virus (AAV) to determine the over-expression of vascular endothelial growth factor (VEGF) in both regenerating peripheral nerve and in the denervated skeletal muscle. Rat median nerve defect was reconstructed by a vein segment filled with muscle fibers previously injected with either AAV2-VEGF or AAV2-LacZ, and the morphological outcome of nerve regeneration was assessed three months after surgery. A second experimental group was used to study the prevention of muscle atrophy evaluating the effect of VEGF over-expression on denervated muscles. One semi-thin section from each nerve and from each muscle was randomly selected and used for the quantitative analysis. We used a design-based quantitative morphology to evaluated some of the most important geometrical parameters that can be used for the assessment of nerve regeneration: Number of fibers, density of fibers, axon and fiber diameter and myelin thickness. For the stereological evaluation of muscle fibers size we measured the cross sectional area of each muscle fiber inside sampling fields randomly selected. Results showed that over-expression of VEGF in the muscle-vein-combined guide interfered with the normal muscle degeneration inside the scaffold leading to a worse nerve regeneration. On the other hand, a positive effect was observed in the muscle treated with AAV2-VEGF that showed a significantly lower progression of atrophy in comparison to muscles treated with AAV2-LacZ. In conclusion, while local delivery of VEGF by AAV2-VEGF-injected muscle fibers did not represent a rational approach to promote axon regeneration along a nerve guide, AAV2-VEGF in a denervated skeletal muscle significantly prevents denervation-related atrophy thus creating a promising new strategy for improving the outcome of posttraumatic neuromuscular recovery. This research was supported by grants MOVAG from the Compagnia di San Paolo and the European Community’s Seventh Framework Programme (FP7-HEALTH-2011) under grant agreement n°278612”.


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Investigation of development of the brain and ventricles in the postnatal period of rats by stereological methods

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ARTICLE INFO

ABSTRACT

Determination of the brain volumes of rats in the postnatal period as well as delineating the effects of neuronal proliferation and the structures of ventricular-subventricular zones by means of sexes and developmental periods at cellular level during the development of brain and ventricles were aimed. Sprague Dawley rats aged 1, 7, 14, 30, 60, 90, 120, 150 and 180 days were used in this study. The brains were fixed with formaldehyde and volumes were calculated stereologically. Furthermore, after routine tissue tracking the sections were stained with hematoxyline-eosin, then ependymal and subependymal cells were classified according to their shape and cellular density in subventricular region was calculated. In the sections obtained from the anterior part of third ventricle and the inferior horn of lateral ventricles, proliferation index of ependymal, subependymal, glial, neuronal cells were examined after staining with ki67. Brain volumes showed an increase among the groups parallel to the age when calculated with point counting grid and brain volume of male rats was found to be larger than females. In light microscopic examination, cubic ciliated and stratified cells were more common in 1, 7, and 14 day-old rats, however in older groups one layered cubic ciliated cells were widespread and proliferation rate decreased. On the other hand, proliferation index was found significantly high in sections passing through the inferior horn of lateral ventricle. The brain volume was demonstrated to increase parallel to age in postnatal rats in our study. On the other hand, cellular density in ventricular and subventricular regions was found to decrease in contrast with the age and cellular differentiation was shown to continue, in accordance with other studies in the literature.


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Investigation of the relation between the projection area of corpus callosum and ventricular volumes with the motor and cognitive functions in children with periventricular leukomalasia

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ARTICLE INFO

ABSTRACT

In this study we aimed at investigating the change in ventricular volumes as well as the area of corpus callosum by using stereological methods in children with periventricular leukomalasia, and correlate these changes with the clinical picture. Cranial MRIs of 24 children with periventricular leukomalasia and 24 healthy ones with the age range of 1-11 years who applied to our pediatric neurology clinic between the years of 2009 and 2010 were evaluated. Projection area of corpus callosum and ventricular volumes were calculated by applying a stereological method on the MR images. Motor functions were evaluated according to gross motor function measurements and cognitive functions were evaluated according to Wechsler Intelligence scale. Mean projection area of corpus callosum in children with periventricular leukomalasia was found statistically significantly lower than the control group, however their ventricular volumes were seen to be increased. Furthermore, the correlations between the thinning of the corpus callosum and both motor and cognitive functions were 70% and 43%, respectively. It was seen in our study that there was a strong correlation between the ventricular volumes and projection area of corpus callosum on one side and motor and cognitive functions on the other. Motor and cognitive impairment in patients in the study group as well as the dilatation of the lateral ventricles and thinning of the corpus callosum were in accordance with the literature. Since the increase in volume of lateral ventricles will cause a decrease in cerebral volume, it may coexist with severe impairment in motor and cognitive functions.


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Poster

Estimating the volume and volume fraction of the nasal structures in the Roe Deer (Capreolus capreolus) using computer tomography images

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Volume
Volume fraction

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ABSTRACT

The volumetric analysis of the nasal cavity of the roe deer has not evaluated yet. Therefore, the normal volume values of the nasal structures may be the basic information for the experimental studies on the mentioned species. The aim of the present study was to determine the accuracy of volume and volume fraction estimates of the conchae, nasal septum and nasal cavity for the first time. In this preliminary study, the nasal cavities of 2 roe deer were scanned using computed tomography. The images sampled randomly at a 1/5 sampling fraction. Sections with an interval of 2.5 mm were obtained. The section surface areas of the nasal cavity, nasal septum and conchae were estimated using the Cavalieri principle. The values were determined as 46.87 cm³ for the nasal cavity volume, 20.40 cm³ for the conchae volume, 19.20 cm³ for the nasal space volume, 7.26 cm³ for the septum volume, 43.60% for the concha fraction, 40.83% for the nasal space fraction and 17.56% for the nasal septum fraction. These findings showed the composition of the nasal structures would contribute to diagnosis of the problems with the nasal cavity and the anatomic literature.


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Volumetric and volume fractional comparison of the nasal structures of the stork (Ciconia ciconia) and seagull (Larus fuscus) using computed tomography images

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ARTICLE INFO

ABSTRACT

In birds the rostral part of the nasal cavity is colder than the caudal part. This prevents the transformation of the moisture in the air to water. This is critical importance in the water balance of birds in desert habitats and migrating birds. In this study, structures of the nasal cavity of 14 seagulls and 7 storks were examined and volume and volume fractions of the nasal cavity and nasal structures i.e., conchae and septum were estimated. Heads of animals were scanned using computed tomography. The images sampled randomly at a 1/5 sampling fraction. We obtained 11 to 16 sections with an interval of 2.5 mm from the Stork and 16 to 20 sections from the seagull. The section surface areas of the nasal cavity, nasal septum and conchae were estimated using the planimetry. The sectional surface areas were used for the estimation of the volume and volume fractions using the Cavalieri principle. Results obtained using the CT images were compared with the both species. The total volume of the nasal cavity, concha volume, nasal space volume, nasal septum volume, concha fraction, nasal space fraction nasal septum fraction were 4.36 cm³, 1.32 cm³, 2.41 cm³, 0.63 cm³, 30.24%, 55.22%, 14.54%, in storks and 3.89 cm³, 1.27 cm³, 2.09 cm³, 0.54 cm³, 33.04%, 53.23%, 13.73%, in seagulls. As a result, there is no a significant difference in between investigated species.

This study was supported Project Management Office of the Ondokuz Mayıs University. Project No: PYO.VET.1401.11.026.
Protective effects of melatonin on ischemic rat kidney and stereological results

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ARTICLE INFO

Abstract

Ischemic acute kidney injury is a clinical syndrome with high morbidity and mortality. It is known to contribute kidney dysfunction in a variety of clinical situation such as kidney transplantation and cardiac surgery. Furthermore, Renal ischemia reperfusion (I/R) injury is of great clinical importance because of its frequent occurrence and may cause acute renal failure. Recently studies have provided the evidences that ROS are of greater pathophysiological importance in I/R-induced renal injury. Melatonin, an antioxidant, was shown as a powerful free radical scavenger and antioxidant. Furthermore, Exogenous melatonin has been reported to preserve renal function and to reduce lipid peroxidation and histological kidney injury. In the present study, to evaluate the effects of melatonin in renal ischemia reperfusion (I/R) injury, we examined the mean numerical density, total number, height and volume of glomeruli and volume of Bowman’s space of the rats using stereological methods. Fifteen male Wistar rats were divided into three groups of five rats each: (1) sham-operated (control), (2) I/R group, (3) Melatonin+I/R group. Control and I/R group rats received normal saline intraperitoneally (i.p.). The Melatonin+I/R group received melatonin (10 mg/kg, i.p.). Right nephrectomy was performed through dorsolateral incisions on all rats and the left renal vessels were occluded for 60 min, followed by 24 h reperfusion. All animals were anesthetized and killed by decapitation. Tissues were processed to paraffin wax by routine protocols. Tissue sections were stained using standard protocols. Stereological examinations were made using a stereo investigator programme and light microscopy. Optical fractionator method was used to count the cardiac myosites. Statistical analysis was produced by Mann-Whitney U test. In the I/R group, shrinkage mean volume, shrinkage mean area, shrinkage mean diameter and the space mean diameter was found to be increased compared to the control significantly. The values of these four parameters in the Melatonin+I/R group did not differ compared to control. According to the our stereologic findings, it may be suggested that I/R injury causes to disturbances the inner glomerular structures without affects the outer glomerular structures and melatonin may reverses this pathologies induced by I/R in rat kidney.

Keywords:
Renal ischemia reperfusion (I/R) Melatonin Kidney Stereology

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Nitric oxide (NO), an endogenous vasoactive molecule, has an important role in the regulation of vascular tone. Renal medullary and cortical blood flows, tubulovascular and tubuloglomerular feedback processes and glomerular filtration are nitric oxide-dependent processes. In the case of the decreased NO bioavailability, there are increased susceptibility to hypertension via mechanism in which includes reduced vascular relaxation and lower renal pressure natriuresis response. The aim of this study was to investigate the renal tubular stereological changes caused by N(G)-nitro-L-arginine methyl ester (L-NAME) induced nitric oxide synthase (NOS) inhibition and to evaluate the effects of propolis that is suggested to have antioxidant activity. Wistar male rats were divided into four groups equally (n=7): (1) Control, (2) propolis, (3) L-NAME and (4) L-NAME+propolis groups. Control rats received normal saline intraperitoneally (i.p.) for 15 days. Propolis (200 mg/kg, orally) rats received intraperitoneally (i.p.). L-NAME rats received NOS inhibitor L-NAME (40 mg/kg, i.p.) for 15 days. The L-NAME+propolis group received both L-NAME (40 mg/kg, i.p.) for 15 days and propolis (200 mg/kg, orally) for the last 5 days. Stereological examinations were made using a stereo investigator programme and light microscopy. Optical fractionator method was used to count the renal tubular cells. Data are expressed as median and range. The values obtained at the end of the stereological study of renal tubular tissue was expressed as median and range. There were significant difference between L-NAME (134.83 (116-138)) and control groups (217.74 (198-232)) in terms of the number of tubular cells (P<0.01). In addition, the numbers of L-NAME+Propolis (205.94 (155-242)) were significantly found to increased compared to the L-NAME groups (P<0.01) and there was no significant difference among L-NAME+Propolis, Propolis (215.00 (191-229)) and control groups. According to the present stereologic results, it may be suggested that the reduction of the number of cells per unit area in renal tubular structure can be completely reversed by propolis treatment.


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The effects of propolis on the L-NAME-induced renal tubular changes and stereological results

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Nitric oxide
L-NAME
Propolis
Stereology

ABSTRACT

Nitric oxide (NO), an endogenous vasoactive molecule, has an important role in the regulation of vascular tone. Renal medullary and cortical blood flows, tubulovascular and tubuloglomerular feedback processes and glomerular filtration are nitric oxide-dependent processes. In the case of the decreased NO bioavailability, there are increased susceptibility to hypertension via mechanism in which includes reduced vascular relaxation and lower renal pressure natriuresis response. The aim of this study was to investigate the renal tubular stereological changes caused by N (G)-nitro-L-arginine methyl ester (L-NAME) induced nitric oxide synthase (NOS) inhibition and to evaluate the effects of propolis that is suggested to have antioxidant activity. Wistar male rats were divided into four groups equally (n=7): (1) Control, (2) propolis, (3) L-NAME and (4) L-NAME+propolis groups. Control rats received normal saline intraperitoneally (i.p.) for 15 days. Propolis (200 mg/kg, orally) rats received intraperitoneally (i.p.). L-NAME rats received NOS inhibitor L-NAME (40 mg/kg, i.p.) for 15 days. The L-NAME+propolis group received both L-NAME (40 mg/kg, i.p.) for 15 days and propolis (200 mg/kg, orally) for the last 5 days. Stereological examinations were made using a stereo investigator programme and light microscopy. Optical fractionator method was used to count the renal tubular cells. Data are expressed as median and range. The values obtained at the end of the stereological study of renal tubular tissue was expressed as median and range. There were significant difference between L-NAME (134,83 (116-138)) and control groups (217,74 (198-232)) in terms of the number of tubular cells (P<0,01). In addition, the numbers of L-NAME+Propolis (205,94 (155-242)) were significantly found to increased compared to the L-NAME groups (P=0,01) and there was no significant difference among L-NAME+Propolis, Propolis (215,00 (191-229)) and control groups. According to the present stereologic results, it may be suggested that the reduction of the number of cells per unit area in renal tubular structure can be completely reversed by propolis treatment.


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Protective effects of Azadirachta indica on cerebral pyramidal cells in experimental cerebral malaria model: Immunohistochemical-morphometric study.

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ARTICLE INFO

ABSTRACT

A major complication of malaria is cerebral malaria. Alternative drugs to treat this complication are needed due to increasing drug resistance. We investigated the neuronal protective effect of an ethanolic extract of the leaves from a traditionally used medicinal plant, “Azadirachta indica” (Neem) in a mouse model of experimental induced Cerebral malaria. Swiss albino mice were intraperitoneally infected with 10^6 “Plasmodium berghei” ANKA, a rodent malaria parasite. Ethanolic extracts of Neem at 300, 500 and 1000 mg/kg were administered intraperitoneally daily for five days from the day parasitaemia reached 5% of the initial parasite inoculation. Intraperitoneal chloroquine and artemether were used as standard drug treatment controls. Histological sections of brain were prepared and examined. During this study, immunohistochemistry for apoptotic cell markers and the inflammatory triggers of apoptosis Fas, FasL, TNF-α and nitric oxide synthase (NOS) as well as morphometry were performed and analyzed. In this study, a difference in the volumetric density of apoptotic pyramidal cells between the untreated control group and Neem treatments was observed. In addition, differential apoptosis in pyramidal cells and the differences in markers of the inflammatory triggers of apoptosis were observed in the Neem treatment groups compared to the disease control. These results suggest that Neem may have some neuronal protective effect. This effect may warrant further investigation.


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Poster

Stereological evaluation of substantial enlargement of the lateral ventricles on the cases with Huntington Disease

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Huntington’s disease (HD) is an autosomal dominantly transmitted, progressive neuro-degenerative disease which presents itself with motor, psychiatric and cognitive dysfunction. HD causes atrophy associated with neuronal loss and gliosis especially in caudate nuclei and putamen and these results with the substantial enlargement of the lateral ventricles. Thus we aimed to evaluate lateral ventricles volumes of the case with HD comparing with a healthy gender and age matched case. The case with HD was a forty-year old male, referred to the Neurology Division, University of Afyon Kocatepe because of involuntary movements and cognitive deficits. The gender and age matched a control subject with normal radiological volumetrical evaluation who applied Department of Neurology School of Medicine at Afyon Kocatepe University was selected for comparing. The MRI imaging examination of the lateral ventricles included T1 weighted gradient-echo MR images acquired in the sagittal plane 10 slices with 5mm slice thickness. Stereological volumetric evaluation was accomplished by the Cavalieri’s principle for both the patient and the healthy subjects. The present case report may contribute to the volumetric knowledge of ventricle changes in cases with a HD.


Keywords:
Huntington’s disease (HD)
Cavalieri’s principle
MRI
Lateral ventricles
Volume
Stereological investigation of the effects of prenatally administered thiazolidinediones on the number of pyramidal cells in adult rat (Wistar albino) hippocampus

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Keywords:
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PPAR-γ
Adult rat

Rosiglitazone belongs to the thiazolidinedione (TZD) class of drugs indicated for the treatment of diabetes mellitus type 2 and it works as an insulin sensitizer by binding to the peroxisome proliferator activated receptor gamma (PPAR-γ). In this study, the effect of prenatally administered PPAR-γ agonist rosiglitazone on the number of pyramidal cell in hippocampus was investigated using histochemical and stereological techniques. A total of eighteen female rats (Wistar albino) were divided into 3 groups as control (C), low-dose rosiglitazone (LDR) and high-dose rosiglitazone (HDR). Pregnant female rats in the LDR group received 2mg/kg dose of rosiglitazone via gavage daily for the first 16 days of pregnancy whereas rats in HDR group received 5mg/kg. Baby-rats were divided into three age groups as 4-week (4W) and 12-week (12W). After histological process, stereological analyses were performed with systematic random sampling using the optical fractionation method. Experimental observations revealed no morphological abnormalities in juvenile rats in both control and experimental groups and significant differences between the numbers of offspring from rats. In the light microscopic examination of rats, general histological structure and morphology of pyramidal cells of the hippocampal brain tissues of all groups were consistent with those described in the literature. As a result of the stereological counts, no difference was found in the number of pyramidal cell in the hippocampus of 4-week and 12-week rats in control and LDR group. However, it was observed that the number of pyramidal cells in the hippocampus of 4-week and 12-week rats in HDR group was greater than those in control and LDR group. The results obtained can be interpreted that rosiglitazone exerts dose-dependent biphasic effect and exhibit neuron protective and anti-apoptotic activity against neurons at high doses.

Stereological investigation of the effects of prenatally administered thiazolidinediones on the number of pyramidal cells in new born rat (Wistar albino) hippocampus

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ARTICLE INFO

Rosiglitazone belongs to the thiazolidinedione (TZD) class of drugs indicated for the treatment of diabetes mellitus type 2 and it works as an insulin sensitizer by binding to the peroxisome proliferator activated receptor gamma (PPAR-γ). In this study, the effect of prenatally administered PPAR-γ agonist rosiglitazone on the number of pyramidal cell in hippocampus was investigated using histochemical and stereological techniques. A total of eighteen female rats (Wistar albino) were divided into 3 groups as control (C), low-dose rosiglitazone (LDR) and high-dose rosiglitazone (HDR). Pregnant female rats in the LDR group received 2mg/kg dose of rosiglitazone via gavage daily for the first 16 days of pregnancy whereas rats in HDR group received 5mg/kg. New born (NB) rats were sacrificed on the day of birth and their brains were dissected. After histological process, stereological analyses were performed with systematic random sampling using the optical fractionation method. Experimental observations revealed no morphological abnormalities in juvenile rats in both control and experimental groups and significant differences between the numbers of offspring from rats. In the light microscopic examination of rats, general histological structure and morphology of pyramidal cells of the hippocampal brain tissues of all groups were consistent with those described in the literature. As a result of the stereological counts, when newborn groups were compared, the number of pyramidal cells in HDR NB group was less than those in LDR NB and CNB groups. The results obtained can be interpreted that rosiglitazone exerts dose-dependent biphasic effect and causes cell death by inducing apoptosis of neural stem cells or prevents differentiation of neural stem cells to neurons.


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Stereological evaluation of the volume of newborns brain in magnetic resonance images

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ARTICLE INFO

ABSTRACT

Brain development after birth is considered to be critical period in neurodevelopmental disorder. This study aimed to evaluate the brain volume by the use of a fluid displacement principle and stereological method and after than to compare these approaches each other in newborns brain. This study was performed on five newborn cadavers mean weighing 2220±1056 g with no signs of neuropathology. The mean (±SD) age of the subjects was 39.7 (±1.5) weeks. The volume of the brain was determined on magnetic resonance (MR) images using the point-counting approach of stereological methods and by the use of fluid displacement technique. The mean (±SD) brain volume by fluid displacement and point counting methods were 246±79.4, 256.6±71.1, respectively. There were no differences between two technique (p>0.05). Stereological estimation may be accepted a beneficial and new tool for neurological evaluation in vivo research of the newborn brain.

A case of nontraumatic bilateral chronic subdural hematoma: Stereological analysis

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ARTICLE INFO

ABSTRACT

Chronic subdural hematoma is a common disease in neurosurgery, generally occur in elderly patients. Head trauma seen only below half of these patients and the other risks are alcohol abuse, seizures, over drainage of CSF and coagulopathies. In the present case we evaluated a case with nontraumatic bilateral frontotemporoparietal chronic subdural hematoma and intracranial volumes on cranial CT. A 77 year old male case presented with nontraumatic bilateral frontotemporoparietal chronic subdural hematoma according to the antiaggregation therapy for pulmonary embolism after having a cardiopulmonary resuscitation. The patient has a headache, confusion and urinary incontinence for two weeks with an examination of quadriparesis. The hematoma and intracranial volumes were measured on cranial CT by using stereological morphometrical analysis comparing with age and gender matched healthy subject. Cranial CT scanning revealed bilateral frontotemporoparietal chronic subdural hematoma with a width of 2 cm. Because of overdose usage of antiaggregation therapy a high INR level: 7 was measured at the presentation. After aggregation normalized, bifrontoparietal two burr-hole craniostomy with drain used for bilateral frontotemporoparietal chronic subdural hematoma evacuation was performed. After surgery the patient had a good recovery and a better neurological examination but still had the risks of other chronic comorbidities such as pulmonary failure. The volumes were measured by two bilinded investigator using Cavalier’s principle. As a conclusion chronic subdural hematoma is a risky disease for elderly patients because the mortality rate depends on presenting neurological condition and other comorbidities.


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The effect of the sulfite on granule cell layer volume in rat dentate gyrus

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ARTICLE INFO

ABSTRACT

Sulfite is a toxic molecule that can cause cellular toxicity by reacting with a variety of cellular components. The aim of this study was to investigate the possible toxic effects of sulfite on granule cell layer volume of the rat dentate gyrus. For this purpose, male albino rats were divided into a Control group and a Sulfite group (25 mg/kg). Sulfite was administered to the animals via drinking water for eight weeks. At the end of the experimental period, brains were removed by craniotomy. Frozen brains were cut by a cryostat. Sections collected via systematic random were stained haematoxylin and eosin. Granule cell layer volume estimated in dentate gyrus by using the Cavalieri method-a stereological method. Results showed that sulfite treatment caused a significant decrease in the volume of granule cell layer of the dentate gyrus in the Sulfite group (mean±S.D.=0.526 mm³±0.003) compared with the Control group (mean±S.D.=0.678 mm³±0.005) (p<0.05, Mann Whitney U test). It was concluded that exogenous administration of sulfite causes volume loss of granule cell layer of the rat dentate gyrus.


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The protective effects of exercise on the ovariectomized rats lungs against menopause-induced changes

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ARTICLE INFO

ABSTRACT

Estrogen secretion is decreased during menopause. In female, biological and psychological some similar changes of aging is shown in menopause. This experiment was aimed to determine whether exercise training has a protective role against the harmful effects aging in ovariectomized rats. In our study, there are four groups. Each group (n=6), including a total of six animals were used 24 Wistar-Albino female rats. Design of the control groups (6 months), ovariectomized groups (9 months), exercise groups (6 months) and ovariectomized-exercise groups (9 months) is respective. Exercise and ovariectomized-exercise (after surgery 12 weeks) groups rats were applied to treadmill-running program, at 15 m/min speed and 15° incline once every two days during 30 days. Pulmonary alveolar area ratio were calculated the measurement-point scale by stereological techniques. Control groups, ovariectomized groups, exercise groups and ovariectomized-exercise groups lung alveolar area ratio of 82.36±1.16, 127.71±1.81, 227.89±3.22, 327.86±4.63 μm², are respectively. There were significant differences in all group about lung alveolar area ratio (p<0.05). Pulmonary alveolar area ratio at least of control groups is more than which exercised group and ovariectomized group. The alveolar area ratio of the lung alveoli exercise to be increases was seen in ovariectomized-exercise groups. As result, exercise and menopause cause changes in lung alveoli and type I - II pulmonary epithelial cells, and may lead to beginning of many process in lung tissue.


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Acetaminophen (APAP), an analgesic and antipyretic, is toxic in overdose to the different organs. The objective of present study was to evaluate the testicular toxicity of acetaminophen administration in Wistar albino rats. 40 Wistar albino male rats were randomly divided into five groups each containing 8 rats. Group 1: Saline, Group 2: Ethanol (4%), Group 3: Resveratrol (RSV) (10 mg/kg/ip/single dose), Group 4: APAP (1000 mg/kg/ip/single dose) and, Group 5: APAP+RSV. After 24 hours testis tissues were removed. The tissue samples were fixed in 10% formalin and were embedded in paraffin. Blocks were cut at 5 µm, mounted on slides stained with Hematoxylin-eosin. The diameter of testis seminiferous tubule and thickness of germinal epithelium were measured. Each slide was observed under 20X magnification and 20 tubules were selected randomly. Tissues were examined using a Leica DFC280 light microscope. All data are expressed as arithmetic mean±SE. Kruskal-Wallis and Conover tests were used for comparison of data. p<0.05 was regarded as significant. The mean diameter of seminiferous tubules and thickness of germinal epithelium were 296.88±1.69 and 47.22±0.44, 286.43±2.51 and 42.96±0.32, 313.84±2.25 and 46.74±0.42 in saline, ethanol and RSV groups, respectively. Statistically significant decrease in diameter of seminiferous tubules (281.17±2.47) and thickness of germinal epithelium (34.85±0.53) were found in APAP group, when compared to group 1 and group 3 (p<0.0001, for all). The mean diameter of seminiferous tubules (320.34±2.30) and thickness of germinal epithelium (44.26±0.45) were statistically increased in APAP+RSV group, when compared to APAP group (p<0.0001). We conclude that acetaminophen-induced testicular damage was improved by resveratrol administration.


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Volume estimation of the subcortical structures in Parkinson’s disease using Cavalieri technique

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ARTICLE INFO

ABSTRACT

Parkinson’s disease (PD) is a neurodegenerative disorder that shares common symptoms such as tremor, rigidity and bradykinesia. Atrophy is regarded a sensitive marker of neurodegenerative pathology. The aim of the current study was to compare the volumes of subcortical brain structures between healthy subjects and patients with PD using stereological (point-counting) method. Twenty-four patients with PD and 23 age matched healthy subjects who are free of any psychiatric, neurological or cognitive impairment were included in our study. MR scans, obtained from subjects, were analyzed by using stereological (point-counting) method. Patients with PD had significantly smaller caudate nucleus and lentiform nucleus volumes than controls (p < 0.05). However no significant differences was determined between patients and control subjects for the volume of thalamus (p>0.05). Magnetic Resonance Imaging (MRI) is considered helpful to facilitate the diagnosis in vivo of patients with PD, revealing atrophy of specific brain regions such as caudate nucleus and lentiform nucleus. We think that these findings may help for the evaluation of patients with PD and further studies are required with larger samples in order to support these data.

Volume estimation of the thalami using the image J

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ABSTRACT

The thalami are two large oblong masses of gray matter, each portion situated on either side of third ventricle, and lying between the diverging portions of the corpora striata. The thalami is of central interest in many disorders of the nervous system. Analysis of volume or shape using MRI techniques may provide important information with respect to the involvement of the thalamus in neurological and neuropsychiatric disorders. In the current study, we estimated the volume of thalami in normal subjects using ImageJ software. We carried out the present study on 30 subjects (13 males, 9 females). All participants were healthy volunteers with no history of major psychiatric, neurological or cognitive impairment. MR scans, obtained from subjects, were analyzed by using ImageJ software. The mean values of thalamus volumes for the men and women were 8.64±1.61 and 7.51±2.87 respectively. A significant differences was determined between male and female subjects. Volume estimation of thalami were determined using planimetry methods and may be important in several clinical conditions. Further studies are required with larger samples in order to support these data.

ARTICLE INFO

The aim of this study is to search the effects of melatonin (MLT) that is being used to suppress the oxidative stress on the follicle growth of ovarian in rats, which are fed with high fat diet. For this purpose, 36 Wistar albino type female rats were divided into 4 groups (n=6) obese (O) and obese melatonin (OMLT) groups were fed with high fat diet which consists of 40% fat for 9 weeks. Other groups that are non obese control (NC) and non obese melatonin (NMLT) groups were fed with regular commercial diet. By the end of 9th weeks, their obesity levels in these animals were evaluated by measuring their body mass index (BMI). For 42 days, MLT has been injected (n=6) to the OMLT group and the NMLT group which was on regular diet. No MLT injection was applied to the control groups. By the end of the 42 days, the ovaries were removed and the tissues were prepared for histological and stereological analyses. The cortex, medulla, granulosa, theca and antrum volumes were estimated by Cavalieri principle. The numbers of follicles were calculated by physical dissector counting technique. Both primordial and primer follicle numbers has been decreased in the O group in comparison to the other groups (p<0.001). Also the volume of cortex has decreased in O group when compared to NC, NMLT and OMLT groups (p<0.001). The volume of medulla has decreased in O group when compared to NC, NMLT and OMLT groups (p<0.001). This study has shown that melatonin may show a useful effect on the follicle growth and ovarian structure. At this point, to see real effect of MLT on the ovarian structures of obese animals, further studies is required.


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Effects of indole-3-Carbinol on testis structure of high-fat diet-induced obese rats

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ARTICLE INFO

ABSTRACT

In recent years, there is an increase in the number of obesity conditions as a result of increasing amounts of uptake of high-energy foods, decreasing physical activities, and a sedentary lifestyle. Oxidative stress caused by obesity is closely related to various diseases such as cardiovascular diseases, hypertension, diabetes, cancers, and infertility. Indole-3-carbinol, which is a phytochemical substance found only in plants of Cruciferae family, has a suppressive effect on the oxidative stress. The aim of this study is to investigate the effects of indole-3-carbinol on the obesity related oxidative stress in a testicle. In this study, 24 Wistar albino male rats were separated into 4 groups randomly (n=6). While the obese control (OC) and the obese indole (OI) groups, which will generate an obesity model, were fed on a special diet containing 40% fat, non-obese control (NC) and non-obese indole (NI) groups were fed on standard commercial provender for nine weeks. At the end of ninth week, obesity conditions were evaluated by calculating body mass indexes (BMI). Intraperitoneal injections were carried out simultaneously to the OI group and the NI group for six weeks. After six-week treatment, testes were removed under an anaesthetized operation. Tissues were prepared for histological and stereological studies. The average volume of seminiferous tubule and testicular volume estimated using Cavalieri methods. The numbers of spermatogonia, spermatocytes, and spermatids were estimated by using optical fractionator technique. Statistical analyses demonstrated that the number of spermatocyte in OC group was lower than the NC group (p<0.001). The number of spermatocytes in both N and O groups were lower than the NI and OI groups (p<0.001). The average volume of seminiferous tubule in the both N and O groups were lower than the NI and OI groups (p<0.05). A decreased volume of testis in the both N and O groups were observed in comparison of the NI and OI groups (p<0.05). The results of this study show that indole-3-carbinol application may have a useful effect on the number of spermatocyte and spermatids which are decreased by obesity. Hence, infertility may be treated by consuming of Cruciferae family plants.
Exercise effects on menopause-induced changes in adrenal glands of ovariectomized rats, a sterological study

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Abstract

The physiological changes associated with estrogen deprivation in menopause have a significant impact on organ functions. Physiological menopause model by induced ovariectomy in animal similar human menopause via estrogen deprivation. To determine the effects of exercise and menopause on adrenal gland in rat. In this experiment, five subgroups were arranged as follows: Young rats (group 1), old rats (group 2), ovariectomized rats (group 3), young (group 4) and exercised (group 5) rats, ovariectomized and exercised rats. Rats were induced to jog on a 20-degree inclined treadmill for 10 minutes, thrice a day, for 15 days. At the end of the experiment adrenal glands of animals were removed and placed in formalin. Tissue samples from these adrenal glands were prepared with tissue preparation device. One out of every ten microtome slices were sampled and stained. These samples were analyzed stereological and histopathological methods. Different histopathological changes were observed in medulla and cortex of adrenal glands in all groups. Addition to histopathological changes, chromaffine cytoplasmic and nucleus volumes obtained from the stereo-investigator and nucleator analysis are reported as: (1410,894-92,195), (723,252-113,454), (1125,318-108,352), (1276,256-77,725), (1293,805-74,088) (μm³). According to stereological analyses, there were no significant differences between group-2 and group-3, and group-4 and group-5 about nucleus volume. In other groups, there were significant differences in all group about nucleus and cytoplasmic volume (p<0.05). As result exercise aging, exercise and menopause cause changes in chromaffine cell and may lead to activation of many process in all body via hormonal changes.


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Periperal nerve regeneration research: A stereological approach

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Stereological approach

ARTICLE ABSTRACT

Quantitative assessment of nerve fibers is a very important element for evaluating the outcome of experimental microsurgery and tissue engineering for the repair of severed peripheral nerves. Histomorphometrical data on the number, density, and size of nerve fibers can provide us with answers to many important questions, such as whether a new microsurgical technique or tissue engineering approach is superior to a traditional technique. Good answers to these questions depend on using an appropriate counting/measuring method to estimate differences. In this presentation, the main critical issues in nerve fiber quantitative morphology are addressed. The importance of respecting the equal opportunity rule, i.e., the basic paradigm of random sampling, is emphasized, together with the explanation of how sampling errors, in the selection of histologic fields and of the nerve fibers inside them, can produce a bias in quantitative estimates. Finally, some practical suggestions on how to cope with the most common sampling errors are provided, in order to help researchers obtain reliable quantitative data on peripheral nerve fibers.


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Oral Presentation

Changes in neocortical neuron number—what happens in the human brain in disease and during aging?

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ARTICLE INFO

Stereology has been applied to studies of the normal brain, including aspects of the neurodevelopment of the central nervous system from the early gestational period to time of birth. The accumulation of data from quantitative stereological postmortem studies of the human brain has significantly increased our knowledge about normal and disease-related structural changes of the central nervous system. We and others have reported quantitative data concerning the primary structures of the normal human brain such as the total number of neurons and glial cells in neocortex, as well as the total length of myelinated fibers in the brain, identifying approximately 150,000 kilometers myelinated nerve fibers in the adult brain, with significant variation as a function of age and gender. Recently, a new stereological method, the proportionator, has been introduced. An example of the application of this new method to human cerebellum is presented.


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Stereological methods, precision and applications in biomedicine: State of the art and future research

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Precision
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Volume estimator

ABSTRACT

Since stereology was founded as a discipline in the early 1960’s, a range of methods have been proposed to estimate geometrical parameters of biological structures, such as volume, surface area, length, number of cells or particles in a compartment, mean particle size, curvature, etc. The aim of this talk is twofold. In the first part of this talk, some of the most commonly applied stereological estimators in the areas of biology and medicine will be explored. Relevant information regarding their level of precision and the approaches that are currently available to estimate their coefficient of error based on a data sample will be discussed. The quantification of local spatial arrangements of biological structures (e.g., nerve fibers, neurons, capillaries) is becoming a crucial component in many biomedical studies. For instance, information such as the spatial distribution of fibers or how neuron cells are spatially arranged with respect to glial cells in healthy and malignant tissue are examples of the open problems to be tackled. Despite the clear need for methodology to quantify and characterize properties of spatial structures, little has been done in this area. The second part of this talk will focus on estimators of spatial distributions, known as ‘second-order’ estimators, sampling designs behind this type of estimators, their precision, biomedical applications and open problems.

Oral Presentation

Stereological estimation of cardiac functional parameters on MR images

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ARTICLE INFO  ABSTRACT

Cardiovascular diseases are one of the main causes of death worldwide. Cardiac imaging with various modalities plays a major role in the diagnosis and therapeutic management of these disorders. Magnetic resonance (MR) imaging is currently considered as the reference non-invasive modality for cardiac function assessment. Cardiac function can be quantified through the estimation of the left and right ventricular end-diastolic volume, end-systolic volume, ejection fraction and mass. The aforementioned parameters are currently estimated with the aid of segmentation methods that enable the manual or semi-automatic definition of endocardial and epicardial boundaries on a series of short-axis cine MR images. The efficiency and applicability of these methods will be discussed in this presentation. The computer-assisted stereological method may be also combined with MR imaging data for estimating cardiac function parameters. This method involves the random placement of a systematic grid of test points over each cardiac MR image. Cardiac volume estimations are based on the simple process of counting points falling within the left or right ventricle and they are independent upon the requirement of any contour extraction from MR images. The optimization of stereological volume estimations through the systematic slice sampling procedure and the determination of the proper spacing between test points of the grid will be presented. The high accuracy, good reproducibility and reduced time expenditure of the optimized stereological method will be demonstrated using a group of twenty-six consecutive patients with known or suspected coronary artery disease. The stereological approach may be considered as an efficient and reliable method for cardiac function analysis from MR images in every day clinical practice without the need for image segmentation.

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The effects of electromagnetic field on the nervous system

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The biological effects of electromagnetic field (EMF) exposure from mobile phones have growing concern among scientists since there are some reports showing increased risk for human health, especially in the use of mobile phones for a long period. In our previous studies we investigated prenatally the effects of 900 megahertz (MHz) EMF on nervous system. In the first study, the effects of prenatal exposure of EMF on the granule cell number in the dentate gyrus of 4-week-old rats were searched. In this experiment we used a control group and an EMF exposed group that exposed to an EMF of up to 900 MHz for 60 min/day between the first and last days of gestation. In the second study, it was investigated the number of pyramidal cells in the cornu ammonis (CA) of the 16-week-old female rat hippocampus following postnatal exposure to a 900 MHz. In this study, only EMF exposed group were exposed to 900 MHz EMF (1 h/day for 28 days) in an exposure tube. The control group was not put in to tube but the sham exposed group was. In the third study, a control group and an experimental group of pregnant rats that were exposed to an electromagnetic field were used. For obtaining electromagnetic field exposed offspring, the pregnant rats were exposed to 900 MHz electromagnetic fields during the 1-19th gestation days. The effects EMF exposure on the number of Purkinje cells in the cerebellum of 16 weeks old female rats were investigated. In the last study, a control group, sham exposed group and an electromagnetic field exposed group were used. At the end of the four experiments the number of cells was estimated using stereological techniques. Histopathological evaluations were also made. Results of these studies have showed that long duration exposure to 900 MHz EMF leads to decreases of Purkinje cell numbers in the cerebellum, pyramidal cell numbers in the CA, granule cells on dentate gyrus.


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Oral Presentation

Variables affecting the estimated volumes on radiological images

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Stereology

The combination of stereological methods with the radiological images is used commonly to obtain volumetric data about the size of organs. However, there are lots of variables affecting the estimated volume values that should be considered. In the present study the factors contributing to the estimations are summarized and solutions are suggested. The number, thickness, orientation, resolution and windowing of the sections may be affective on the estimated values. Moreover, matching the scale with point counting grids, threshold values in automated planimetry and inter-observer variations may also affect the data. The usage of sufficient number of slices, sections with 3–4mm thicknesses, orientation that allows the best delineation of the structures, high-resolution images at least 256×256 field of view values and same windowing levels may decrease the bias. Calipers can be used to match the scale of film with the distance between points, using same threshold values and having values obtained by the same observer may also decrease the variations. As a conclusion, stereological techniques are unbiased techniques that provide reproducible and reliable quantitative data. However, the factors contributing to the estimated values should be standardized to strengthen the obtained data.


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Oral Presentation

Significance of microscopy in modern scientific researches

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ABSTRACT

As qualitative microscopic studies are still necessary in biological tissue analysis we should use proper microscopic technics at the beginning of our investigations. Unfortunately, this is not enough to estimate the effects of any agent in various biological tissues of interest. Nowadays in modern scientific researches, using the quantitative stereological methods such as disector and fractionator are essential and imperative to reveal effects of any agent, and these methods can be applied from organelle to whole organ of any biological organism. So, specimen preparation become a crucial part of a quantitative investigation because of influences of fixation, dehydration, and embedding of the tissues on particle size and shape. In addition to above, counting and measuring technics are also important for obtaining accurate data from the biological tissues in our work. These new stereological methods yield three-dimensional information from 2-D images. Particle number, surface area, real volume and mean particle size are frequently used extrapolating structural quantities in the stereological studies. Since tissue shrinkage, section thickness, partial size/shape influence counts of interested particles during microscopy, in our studies these parameters must be taken into consideration to prevent wrong interpretation.


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Oral Presentation

Some stereological problems and practical solutions: An alternative method to measure step length

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<th>ARTICLE INFO</th>
<th>ABSTRACT</th>
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<td><strong>Correspondence to:</strong></td>
<td><strong>Stereologists recommend lots of methods evaluating the biological materials for scientists. Every new method means new tools, new materials, new budget and additional time to learn the techniques for the researcher. It seems that the stereological methods provide opportunity for the companies to establish new market. So that, researchers want to use more easy and low-cost stereologic methods. It is aimed in this presentation to share our experiences deal with measuring of step length during application of optical fractionator method or other stereological applications need length measuring. We designed a new simple and low-cost morphometric measurement method. A light microscope connected to a screen (or even a simple drawing tube) is needed for application this method. The screen should be calibrated before measuring for ever magnification of the microscope. A microscopic scale (e.g. micrometric discs, Thoma Glass) with a known unit distance can be used for screen calibration. We used Thoma Glass and metric system in this study. The unit distance of the microscopic scale (Thoma scale) was lined on an acetate film sheet superimposed on microscopic view of the scale transferred to the screen. This procedure was repeated for every microscopic magnification. Then, the lined acetate film sheets were marked to indicate magnification, and used for measuring the distance the microscopic views transferred to the screen. Our team developed the SHTEREOM software, which included this calibration method. The software helps the researchers to count, estimate and save the data easily. On the other hand every researcher can calibrate his/her microscope and use the method without any additional software or device.</strong></td>
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The use of stereological methods in different medical disciplines

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ABSTRACT

Stereology is used to make interpretations from 2D sections that we obtain from 3D structures actually. The usage of stereological methods by medicine has risen recently. Stereological methods are beneficial in studies by both preclinical and clinical departments. The studies in preclinical fields are mostly concentrated in experimental studies (i.e. microscopic studies) and experimental models. These include studies such as counting of cells and neurons, estimating surface area, volume estimation, volume density and numerical density etc. On the other hand clinical studies include volume estimation, volume fraction and estimating areas etc. Using the methods on computed tomography and magnetic resonance imaging scenes is helpful to get new knowledge in normal and pathological conditions. In this presentation the fields of usage of stereological methods in medicine and which methods are used by which disciplines will be tried to manifest generally. Stereological methods are used not only by histology, pathology, anatomy but also by neurology, radiology, neurosurgery and even dermatology, and day after day the usage of stereology is rising in medicine. In conclusion, stereology helps clinical practices and researches regarding all kinds of medical disciplines to improve.

Oral Presentation

The effects of thirty-minute ischemia on the number of purkinje and granular cells in the chick cerebellum

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Keywords: Ischemia, Purkinje cell, Granular cell, Chick, Optical fractionator

Ischemia frequently appears in clinical practice and threatens human life and brain functions from the fetal period to old age. Our aim in this study was to investigate the effects of ischemia on the number of granular and Purkinje cells in the chick brain. Fifteen chicks were randomly divided into three equal groups; an ischemia group (ISG), a surgical stress alone group (SSG) and a pure control group (PCG). Surgical intervention and the model of ischemia were applied on the second day postnatally. For this purpose, internal carotid arteries were isolated by cervical surgery without applying ischemia to the chicks in the SSG (n=5). Thirty minutes of ischemia was applied on the in the ISG (n=5) by the occlusion of the internal carotid arteries. No surgical intervention was performed on the chicks in the PCG (n=5). Chicks lived freely under normal laboratory conditions without stress throughout experiment. On the 10th postnatal day, chicks were sacrificed and their brains were removed. The cerebella were blocked and cut into serial sections of 25-μm thickness in the sagittal plane using a rotary microtome. The sections were mounted onto gelatinized glass slides and stained with toluidine blue for stereological analyses. Total numbers of cerebellar Purkinje and granular cells were estimated using the optical fractionator technique. Ischemia applied for 30 minutes in the chick brain led to a decrease in the number of granular cells. However, there was no increase or decrease in the number of Purkinje cells. Ischemia applied for 30 minutes leads to a decrease in the number of granular cells.


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Oral Presentation

Fractals, self-similarity and stereology: Understanding the whole by analyzing its parts

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Keywords: Fractal geometry, Stereology, Self-similarity, Morphometry

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ABSTRACT

Morphogenesis is a complex procedure largely governed by nonlinear and chaotic processes in living structures. Scientists dealing with morphometric methods are generally confused by complexity and versatility of natural structures. Design based stereology is one of the major advances in morphometry and enabled us to understand many quantitative features of living and non-living forms with great accuracy, and in an unbiased manner. Despite these advances, and the improvements in different imaging techniques, we still have a poor understanding about the natural forms and the interrelations of the forms belonging to different scales in nature. Fractal geometry may help us in that context. Fractals are computer-generated images and bear a great resemblance to the natural forms that we see in our investigations. This resemblance is not just an appearance; it may show us some new ways to predict the unpredictable, at least, to a degree. Self-similarity, which is a common feature in both fractal geometrical shapes and natural forms, might be a good starting point for generating a new methodology in morphometry. Fractal analysis techniques are already in use in different areas of science and morphometry, but there is still a large gap between stereology and fractal analysis studies. In this presentation, large-scale application of fractal geometry and fractal shape analysis will be discussed, together with the possible applications in life sciences and stereology.

Oral Presentation

Stereology of testis

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Nucleator

ABSTRACT

All the known stereological techniques are used successfully for the estimation of cell numbers in the testis. However, who wants to work with testis should choose the methods very carefully. Because testis has densely-packed different cells, the successful result depends on good preparation of the sections and the available equipments. For direct estimation of cell number optical dissector is the most preferred methods. The major advantage of the optical dissector to the physical dissector is that it avoids the issue of aligning physically separate sections and suitable for densely-packed germ cells. Beside its advantage, optical dissector needs a very good fixation and thick sections which is a new technique for many labs. It also needs a set of special equipments for counting cells. For the non specialist labs want to work on the testis, indirect cell counting methods can be more easily used. Numerical density can also be estimated indirectly by dividing volume fraction of a particular groups of cell/nuclei by the individual volume. There are different methods for estimating nuclear volume such as point-sampled intercept, selector, nucleator and rotator. Among them, we prefer to use the nucleator in our lab. It should be remembered that other data except cell numbers can be easily obtained and used for the understanding of the treatments effecting on testis. As long as the data is obtained with unbiased systematic random sampling.


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MRI volumetry of the temporal lobe in Sudan; comparative study between epileptics and matching control


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ARTICLE INFO

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Estrogen, which is a steroid hormone secrete from ovary with large amount and less amount from adrenal gland for non-pregnant woman and from placenta for pregnant woman. Estrogen not only for fertilization and running the secondary consumer profile but also has a major role on the tissues like; brain, bone, liver, kidney and cardiovascular system. Lack of estrogen which is advancing by menopause can cause many important health problems. Estrogen is suggested to be one of the most important regulators of neuronal function, including neuronal proliferation, survival and plasticity. There is a broad consensus that the loss of ovarian hormones is associated with neurodegeneration in the hippocampus that leads to cognitive symptoms such as anxieties, difficulty in concentrating, overreacting to minor upsets, quickly being irritated and forgetfulness on approximately 70-80% of all women around the world. It is shown that, estrogen effects the rate of synapse formation by activating specific genes by way of the receptors which are found in astrocyte and microglia in central nervous system (CNS). For this study, a total of 24 female sprague dawley rats (12 weeks old, 180-200 g) which were subjected to bilateral ovariectomy were included. After ovariectomy, the rats were housed for 123 days in a standard laboratory. At the end of the 123 days, the rats were euthanized and the brain sections were removed. After removing the brains, all samples were rapidly fixed in 10% buffered formalin for 24-48 h for histological examination. All the procedures of light and electron microscopic examinations were applied to brain tissues. In this study by the way of stereological, the number of hippocampal neurons was calculated on the microscopic slides by using disruption principle and the optical dissector combination. The neurons which fell into the optical dissector on brain slices were marked. Then hippocampal volumes were calculated by Cavalieri method. The last step of calculation, total neuron numbers were calculated in hippocampus on each brain section. Finally, In the terms of histopathological examination, the regular structure of almost all axon extensions was lost in the examination of electron and light microscopic figures. The majority of these extensions had a saw tooth-like appearance in longitudinal section profiles. Especially in transfer section profiles of myelinated axons, some morphological changes were shown which may be matched up with light microscopic findings. Deficiency of estrogen will initially affect microtubule organization. When this organization breaks down, it will physically cause the distribution of the normal structure of axonal plasmalemma. This in turn will lead to the distribution of physical organizations of estrogen and other different types of receptors which are placed in both the membrane and microtubules in the axon.


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An evaluation of neuroprotective effects of melatonin, against adverse effects of prenatal exposure to a non-steroidal anti inflammatory drug, during the peripheral nerve development

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Neuroprotective effect of melatonin was investigated to prevent or decrease the impairment of the fetal peripheral nerve system due to maternal consumption of diclofenac sodium (DS). Eighty four pregnant rats were divided to 7 groups as control group (Group 1), saline administered group (Group 2), DS administered group (Group 3), DS with low dose melatonin administered group (Group 4), DS with high dose melatonin administered group (Group 5), low dose melatonin administered group (Group 6) and high dose melatonin administered group (Group 7). After pregnancy period, six male newborn rats that reached to 20 weeks of age from each group were sacrificed. Sciatic nerves of these rats were harvested and the mean axon numbers, diameter and myelin thicknesses were estimated with stereological techniques. Mean myelinated axon numbers are counted. According to these results the number of myelinated axon numbers in Group 3 is significantly lower compared to groups 1, 2, 5, and 7 (p<0.05). Mean myelinated axon cross section area is measured. According to these results the number of myelinated axon area in Group 3 is significantly lower compared to groups 1, 2, 5, and 7 (p<0.05). Also in the Group 4, myelinated axon cross section area is significantly lower compared to groups 1 and 7 (p<0.05). Mean myelin sheet thickness are measured. According to these results there were no differences between the groups regarding mean myelin sheet thickness. Current study proves that the prenatal exposure to DS decreases axon number and axon area and these effects can be reversed with melatonin prophylaxis. The mechanisms for such effect are believed to be by the apoptotic effect and inhibition of differentiation of DS on embryonic neuronal stem cells is either prevented from the beginning or reversed afterwards.


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Oral Presentation

Effects of Diabetes Mellitus on the rat liver during the postmenopausal period

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ARTICLE INFO

Diabetes results in severe complications in humans, such as liver failure. Estrogen and its derivatives are medically acceptable, powerful antioxidant agents that can enable liver and other important organs to defend themselves against oxidative related injury. Estrogen deficiency, which occurs in the postmenopausal period and in individuals with diabetes, may play a significant role in the progression of liver failure. The present study investigated whether diabetes worsened the onset of liver injury/damage during the ovariectomized (OVX)-induced postmenopausal period in rats. For this aim, rats were divided into four groups: Control (Group I), diabetic (Group II), ovariectomy (Group III) and ovariectomy plus diabetes (Group IV). After the diabetes and ovariectomized experiments, quantitative histopathological and immunohistochemical changes in liver were detected using ligh microscopy and modern stereological systems. Histopathological examinations pointed that there were many necrotic and apoptotic hepatocytes in the lobules of Group II. Additionally, there were a larger number of necrotic cells in Group III than Group II. In contrast to Group II, there were also apoptotic cells in the portal areas in Group III. Moreover, evidence of liver injury was higher in the sections of Group IV than all other groups. In biochemical results, there were statistically important differences between all the groups (P<0.001) in the terms of catalase (CAT), glutathione peroxidase (GSH) and myelo peroxidase (MPx) activity. In addition, the amount of lipid peroxidation (LPO) was significantly different between experimental groups. In stereological results, there were significant differences between Groups I and II and Groups II and IV. The present study provided a new insight into the pernicious effects of ovariectomy on liver injury, following the onset of diabetes. Indeed, the present study found that increases in liver oxidative activity in OVX-rats following the onset of diabetes correlates with elevated MPx, LPO and histopathological changes in rat liver.
Sepsis, an inflammatory disorder, is characterized by the neutrophils accumulation, related with the increased generation of reactive oxygen and nitrogen species as well as pro-inflammatory cytokines. It also causes damages to multiple organs such as liver, kidney, heart. Lithium is the common used agent for treatment of several psychiatric disorders and effects physiologic process in many cells. We designed to investigate dose-dependent lithium treatment on ovarian damage induced by sepsis with using immunohistochemical and stereologic methods. Thirty female Sprague Dawley rats (180-200 g) were used in this study. The rats were divided into five groups; control, cecal ligation and puncture (CLP), lithium-treated-CLP (10-mg/kg), and lithium-treated CLP (20-mg/kg), lithium-sham-operated group (20-mg/kg). Polymicrobial sepsis was induced through ceacal ligation and a two-hole puncture in CLP groups. Twenty-four hours after the surgery, the rats were decapitated and then ovary tissues were removed for histopathologic and stereologic analysis. In this study, lithium dose-dependently decreased the polymorphonuclear cell infiltration and 8-OHdG immune-positive cell number in sepsis induced rats ovary. In conclusion, this study shows that there is a remarkable decrease in neutrophil infiltration and 8-OHdG immune-positivity in sepsis induced-rats ovary. Our results suggest that dose-dependent lithium may be of therapeutic agents for sepsis-associated ovarian inflammation.

Oral Presentation

Estimation of motor neuron number in the spinal cord

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ARTICLE INFO

Spinal cord is long structure. It runs through the vertebral canal. It has cervical, thoracic, lumbar and sacral regions and it is not uniform in diameter. It is thicker in cervical and lumbar regions and thinner in the thoracic region. In cross sections the outer part is called the white matter and the inner part is called the gray matter where central canal traverses in the center. Gray matter is divided to dorsal and ventral portions. The motor neurons are situated in the ventral portion of the gray matter. Amyotrophic lateral sclerosis (ALS) is a fatal neurodegenerative disorder resulting in progressive degeneration of motor neurons. The SOD1 (G93A) genetic mouse model is a valuable animal model to study the disease course of progressive motor neuron loss comparable to human ALS. Our aim was to understand the effect of exercise to the motor neuron number for both wild type and G93A mouse. We estimated motor neuron number using stereology (optical fractinator) in the cryo cut sections of the cord.


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Estimation of the volume and volume fraction of brain and brain structures on radiological images

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\textbf{ARTICLE INFO}

\textbf{ABSTRACT}

Size changes of the brain is monitorized for clinical or research purposes. Stereological methods provide some techniques to obtain quantitative information about the size of the brain or size relation of its components within the whole. In the present study the Cavalieri principle of stereological methods used to estimate the volume of brain and its components. Using the Cavalieri principle, the volume of any object could be estimated from a set of slices through the object, provided that they are parallel, separated by a known distance. The cut surface areas of the sections are assessed and the multiplication of the total cut surface area with the mean of the section thickness provides an estimation of the volume of the examined object. The point-counting and planimetry are two methods for the assessment of sectional cut surface areas in the Cavalieri principle. Sometimes, the volume of brain could not provide comparative information among the groups. Scientists have documented several factors that contribute to the size of brain. Factors related to brain growth, such as gender and physical size, are thought to influence the maximal size of an individual’s brain. Comparing solely the brain volumes or its components between two groups will not provide reliable data. At this point the volume fraction method of stereological approaches proposes the solution. The volume fraction is simply expressed as the fraction of component within the reference volume. Point-counting and planimetry could be used digitally or they can be applied on the printed films. There are many studies describing the techniques and its applications. However, mostly they contain advanced information that is not suitable to be digested by newcomers. In the present study we gave simple information on the application of both techniques. We also discussed the factors affecting the volume estimations on radiological images.


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Oral Presentation

Exercise-induced changes on liver cells density in ovariectomized rats


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ARTICLE INFO

The menopause is a mild life event, characterized by estrogenic deficiency and associated with bio-psycho and social changes which impair quality of life. In addition, exercise can be described as any bodily activity that enhances or maintains physical fitness and overall life quality. This experiment was conducted to determine if exercise training has a protective role against the deleterious effects of aging in ovariectomized rats. In this study, thirty-six Sprague Dawley rats were used. The rats were divided into six subgroups. Subgroups consisted of young rats, old rats, ovariectomized rats, young exercise-trained rats, old exercise-trained rats and ovariectomized exercise-trained rats. Control rats and ovariectomized rats 12 weeks after surgery were subjected to a 4 week treadmill-running program. In exercise groups, the rats were subjected to treadmill exercise during which time each rat walked on a motor-driven treadmill for 15 m min⁻¹ speed and 15° incline once every 2 days for a period of 10 days over three courses for 5, 10 and 15 mins per day, totally for 30 days. In the stereological analysis of this study, the numerical density of the control groups were found significantly higher compared to ovariectomized and/or training groups (P<0.05). On the other hand, the liver cell numbers of ovariectomized exercise-trained group was found to be lesser than other trained and ovariectomized groups. In addition, the cell density of ovariectomized group estimated as a little higher than other groups except for control group (P<0.05). The stereological results revealed that ovariectomy and training can induce the decrease of liver cell density in ovariectomized rats.


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Oral Presentation

A new hypothesis about neuronal degeneration appeared after a rat model of menopause

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Estrogen, which is a steroid hormone secreted from the ovary with a large amount and less amount from the adrenal gland for non-pregnant women and from the placenta for pregnant women. Estrogen not only plays a role in fertilization and running the secondary consumer profile but also has a major role in the tissues like brain, bone, liver, kidney and cardiovascular system. Lack of estrogen which is advancing by menopause can cause many important health problems. Estrogen is suggested to be one of the most important regulators of neuronal function, including neuronal proliferation, survival and plasticity. There is a broad consensus that the loss of ovarian hormones is associated with neurodegeneration in the hippocampus that leads to cognitive symptoms such as anxiety, difficulty in concentrating, overreacting to minor upsets, quickly becoming irritated and forgetfulness in approximately 70-80% of all women around the world. It is shown that estrogen effects the rate of synapse formation by activating specific genes by way of the receptors which are found in astrocyte and microglia in the central nervous system (CNS). For this study, a total of 24 female Sprague Dawley rats (12 weeks old, 180-200 g) which were subjected to bilateral ovariectomy were included. After ovariectomy, the rats were housed for 123 days in a standard laboratory. At the end of the 123 days, the rats were euthanized and the brain sections were removed. After removing the brains, all samples were rapidly fixed in 10% buffered formalin for 24-48 h for histological examination. All the procedures of light and electron microscopic examinations were applied to brain tissues. In this study by the way of stereological, the number of hippocampal neurons was calculated on the microscopic slides by using disruption principle and the optical dissector combination. The neurons which fell into the optical dissector on brain slices were marked. Then hippocampal volumes were calculated by Cavalieri method. The last step of calculation, total neuron numbers were calculated in hippocampus on each brain section. Finally, in the terms of histopathological examination, the regular structure of almost all axon extensions was lost in the examination of electron and light microscopic figures. The majority of these extensions had a saw tooth-like appearance in longitudinal section profiles. Especially in transfer section profiles of myelinated axons, some morphological changes were shown which may be matched up with light microscopic findings. Deficiency of estrogen will initially affect microtubule organization. When this organization breaks down, it will physically cause the distribution of the normal structure of axonal plasmalemma. This in turn will lead to the distribution of physical organizations of estrogen and other different types of receptors which are placed in both the membrane and microtubules in the axon.

Amiodarone ameliorates sepsis-induced ovarian inflammation in female rats by a neutrophil-dependent mechanism

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ARTICLE INFO

Amiodarone is a widely used antiarrhythmic agent and a multiple ion channel (Ca, Na, K) blocking drug. Recent studies show that the anti-inflammatory effect of amiodarone on acute and chronic sepsis via antioxidant mechanisms in many organs. The aim of this study is to firstly investigate the role of amiodarone on polymorphonuclear leukocyte infiltration and DNA oxidation in ovarian tissue. Thirty female Sprague Dawley rats were divided into 5 groups; healthy control, cecal ligation and puncture (CLP), CLP+amiodarone (25 mg/kg), CLP+amiodarone (50 mg/kg), and sham operated+amiodarone (50 mg/kg). A cecal ligation polymicrobial peritonitic sepsis model was applied to sepsis group rats. Polymicrobial sepsis was induced through cecal ligation and a two-hole puncture. All animals were sacrificed and then ovaries were removed for stereologic analysis. Numerical densities of the PMNL and 8-OHDG immune-positive cell were estimated in ovarian sections via fractionator probe. Our results showed that amiodarone ameliorated the sepsis induced neutrophil infiltration and decreased DNA oxidation (8-OHGD) immune-positive cell counts in rat’s ovary. The results indicate that amiodarone dose-dependently treatment showed protective effects on sepsis induced rats ovary.


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Kapsam:

Deneyesel ve Klinik Tip Dergisi * (DKTD, p ISSN 1309-4483, e ISSN 1309-5129), deneyesel ve klinik tip bilimleri alanlarında sinir bilimleri, pediatri, diabeti, kadın doğum ve enfeksiyon hastalıkları gibi bilim dalları başta olmak üzere tüm dallarındaki çalışmalarını yayımlanmaktadır. Dergi, deneyesel ve klinik araştırma makalelerine öncelikle tanımakla birlikte, derleme yazıları, olgu sunumları, editöre mektuplar ve toplantılar dahil tüm dallardaki özgün çalışmalarını yayımlamaktadır.

Makale Sunumu:

Deneyesel ve Klinik Tip Dergisi dergisi açıkça dergiye sunulması için öncelik tanımaktadır. Dergi deneysel ve klinik araştırma makalelerine öncelik tanımakla birlikte, derleme yazıları, olgu sunumları, editöre mektuplar ve toplantılar dahil tüm dallardaki özgün çalışmalarını yayımlamaktadır.

Deneysel ve Klinik Tıp Dergisi

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