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TÜBİTAK

1st INTERNATIONAL CELL DEATH RESEARCH CONGRESS - TURKEY*

(2nd CONGRESS OF CELL DEATH RESEARCH SOCIETY-TURKEY)

4-7 May 2016
Dokuz Eylül University
School of Medicine
IZMIR - TURKEY

ABSTRACT BOOK

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(2nd CONGRESS OF CELL DEATH RESEARCH SOCIETY-TURKEY)

4-7 May 2016
Dokuz Eylül University, School of Medicine, IZMIR - TURKEY

Dear Colleagues,

On behalf of the Cell Death Research Society of Turkey (HÖAD, Hücre Ölümü Araştırma Derneği) it is a great honor and pleasure to invite you to the 1st International Cell Death Research Congress, which will be held on 04-07 of May 2016 in Izmir, TURKEY.

The congress is scientific refereed. The main topics of the congress will be the focusing on the importance and role of different types of cell death in oxidative stress, immunity, stem cell therapies, genome signatures, therapeutic approaches and polyphenols as anti-cancer agent in diseases.

The congress will feature plenary, key and short lectures besides poster presentations. Our main goal is to gather the scientists from universities, research centers from all over the world and offer to all a great and attractive congress joining different knowledge together on cell death.

A number of the internationally distinguished speakers with the expertise in the fields are invited. They will introduce the cutting-edge research and the future perspectives relevant to the subjects covered in the present meeting. Besides, oral and poster presentations are included for more scientific discussions.

Two years ago, the first congress of the society was international participating and over than 300 researchers from all over world mostly US and Europe including the 46 speakers were attended. Therefore, we are expecting a similar or higher participation on the date of May 2016, which is very nice time of the season in İzmir, Turkey.

Congress will be held in Dokuz Eylul University School of Medicine in Izmir - one of the preeminent and biggest universities in Turkey. Izmir region itself, near the Aegean Sea, is famous for its rich culture, history and outstanding scenery.

We would like to take this opportunity to invite all researchers and accompanying parties to attend this meeting and to enjoy the rich scientific program and the beauty of Turkey and Turkish hospitality.

Sincerely yours,
On behalf of the Organizing Committee, I would appreciate your interest and hope to welcome you in Izmir.

Professor Semra KOÇTÜRK, Ph.D
President, Cell Death Research Society-Turkey

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	Topics of The Congress:
Session 1 :	Cellular Mechanisms
Session 2 :	Oxidative stress and cell death
Session 3 :	Cancer and cell death
Session 4 :	Stem cell, Pluripotency and cell death
Session 5 :	Polyphenols in cell death
Session 6 :	Immunity and cell death
Session 7 :	Therapeutic approaches to cell death

Scientific Programme

1st INTERNATIONAL CELL DEATH RESEARCH CONGRESS - TURKEY
(2nd CONGRESS OF CELL DEATH RESEARCH SOCIETY-TURKEY)
04-07 MAY 2016
TURKEY-IZMIR

MAY 4th 2016 (WEDNESDAY)	
08:30-17:30	**Registration**
09:00-10:30	**Opening Ceremony:** ***Semra KOCTURK (President of CDRS-Turkey)** ***Pinar TUNCEL (Vice Dean of DEU School of Medicine)** ***Murat OZGOREN (Chairman of DEPARK Executive Board)** ***Mehmet FUZUN (Rector of DEU)** *Music Recital - REY Quartet, Performed by; Zeynep Simhe ACUNAZ (Violin) Ceyda OZDEMIR (Violin) Iris ICELLIOGLU (Viola) Berk KAVAK (Violoncello) *Visual show: AQUA graphs. Lights Written on Water**, Presented by Alp CAN**
10:30-11:00	**Coffee Break**
11:00-12:00	**Opening Lecture**
	Chair: Mauro PIACENTINI
	Role Of RIP Kinases In Regulating Cell Death and Survival In Vitro and In Vivo, **Peter VANDENABEELE (President of ECDO)**
12:00-13:00	**LUNCH**
13:00-13:30	**Company Presentation - FLUIDIGM:**
	The Polaris System: Integrating Cell and Molecular Analysis, **Gregory GONZALEZ**
13:30-15:15	**Session 1 : Cellular Mechanisms**
	Chairs: Peter VANDENABEELE, Devrim GOZUACIK
13:30-14:15	Type 2 Transglutaminase: A Key Regulator Of Proteostasis Under Cellular Stressful Conditions, **Mauro PIACENTINI (Keynote Lecture)**
14:15-15:00	Autophagy At The Intersection Between Cell Survival and Cell Death: Roles In Inflammation and Lysosomal Homeostasis, **Vojo DERETIC (Keynote Lecture)**
15:00-15:30	Novel Regulators Of Autophagy, **Devrim GOZUACIK**
15:30-16:00	**Coffee Break**
16:00-17:00	**Open Discussion with Editorial Members of Journals**
	Moderator: Kemal S. Korkmaz
	Mauro PIACENTINI
	Boris ZHIVOTOVSKY
	Francesco CECCONI
	Peter VANDANABEELE
	Yahya LALELI
	Ekrem GUREL

17:00-19:00	**Open Discussion Problems, Suggestions for Solutions in Health Research in Turkey (Session will be held in Turkish)**	
	Moderator: Devrim GOZUACIK	
	Sevim AYDIN - TÜBİTAK-SBAG Başkanı	
	Murat OZGOREN - DEPARK Yönetim Kurulu Başkanı	
	Esra ERDAL - IBG İzmir Müdür Yardımcısı	
19:30-22:30	**OPENING COCKTAIL –AQUA MARIN**	
MAY 5th 2016 (THURSDAY)		
09:00-10:45	**Session 2: Oxidative Stress and Cell Death**	
	Chairs: Ferhan SAGIN, Hilal KOCDOR	
09:00-09:45	Mitochondrial Substrates: A Tool To Combat Cancer, **Boris ZHIVOTOVSKY (Keynote lecture)**	
09:45-10:15	AMBRA1 Negative Control At The Crossroad Among Autophagy, Cell Proliferation and Cell Death, **Francesco CECCONI**	
10:15-10:45	In Vitro and In Vivo Characterization Of Cell Survival Genes Using Destabilized Cas9, **Serif SENTURK**	
10:45-11:15	**Coffee Break**	
11:15-12:15	**Oral Presentations**	
	Session-2 Conference Hall Chair: Semra KOCTURK	**Session-1 Classroom- B (Downstairs) Chair: Saime BATIREL**
	OP-1 Bilge Debelec BUTUNER	OP-1 Mimoune BEREHAB
	OP-2 Mehmet Eray ALCIGIR	OP-2 Ozlem ORAL
	OP-3 Pinar ERKEKOGLU	OP-3 Gulce Sari KAPLAN
	OP-4 Ozge CAGLAR	OP-4 Ulvi AHMADOV
		OP-5 Elgin Turkoz ULUER
12:15-13:30	**LUNCH**	
13:30-15:15	**Session 3: Cancer and Cell Death**	
	Chairs: Fahri Saatcioglu, Kemal S. Korkmaz	
13:30-14:15	STAMPing Proliferation and Cell Death In Prostate Cancer, **Fahri SAATCIOGLU (Keynote Lecture)**	
14:15-14:45	ETS Transcription Factors - Critical Regulators Of Brain Tumor Initiating Cell Proliferation vs Neurodegeneration?, **Isil AKSAN KURNAZ**	
14:45-15:15	Diminished Cyclin Dependent Kinase Activity and mTOR Are Critical In The Cell Death Decision Through Affecting STAT Signalling Differently In Prostate Cancer Cells, **Elif Damla ARISAN**	
15:15-15:45	**Coffee Break**	
15:45-17:35	**Session 4: Stem Cell, Pluripotency and Cell Death**	
	Chairs: Alp CAN, H. Seda VATANSEVER	
15:45-16:15	The Bright and The Dark Sides Of Reprogramming To Pluripotency, **Andras NAGY (Keynote Lecture)**	
16:15-16:45	Stem Cell Therapy Approaches To Ischemic Cardiomyopathy, **Alp CAN**	
16:45-17:15	**Oral Presentations**	
	Session-3 Conference Hall Chair: Kemal S. KORKMAZ	**Session-4 Classroom- B (Downstairs) Chair: Gulinnaz ERCAN**
	OP-1 Betul KARADEMIR	OP-1 Feyzan Ozdal KURT
	OP-2 Tuna ONAL	OP-2 Hilal KABADAYI
	OP-3 Aysun EKINCI	OP-3 Remziye KENDIRCI
17:15-18:30	**POSTER SESSION - Refreshment**	
18:30-19:30	**GENERAL ASSEMBLY OF CELL DEATH RESEARCH SOCIETY OF TURKEY**	

MAY 6th 2016 (FRIDAY)		
09:00-10:45	**Session 5 : Polyphenols in Cell Death**	
	Chairs: Bulent OZPOLAT, Semra KOCTURK	
09:00-09:45	Targeting Inflammatory and Apoptotic Pathways By Agents Designed By Mother Nature For Prevention and Treatment Of Cancer, **Bharat AGGARWAL (Keynote lecture)**	
09:45-10:15	Molecular Mechanisms Of Flavonoid Quercetin In Enhancing Apoptosis In Chronic Lymphocytic Leukemia, **Gian Luigi RUSSO**	
10:15-10:45	Development Of Novel Targeted Therapies For Solid Tumors, **Bulent OZPOLAT**	
10:45-11:15	**Coffee Break**	
11:15-12:15	**Oral Presentations**	
	Session-5 Conference Hall Chair: Zekiye S. ALTUN	**Session-3-4-5-6 Classroom- B (Downstairs) Chair: Betul KARADEMIR**
	OP-1 Mehmet Fatih SEYHAN	OP-1 Serap CELEBI
	OP-2 Ayşe Mine YILMAZ	OP-2 Mehmet Kadir ERDOGAN
	OP-3 Abdullah Tuncay DEMIRYUREK	OP-3 Levent ELMAS
	OP-4 Gunes OZEN	OP-4 Saime BATIREL
	OP-5 Ibrahim BOZGEYIK	OP-5 Ceylan HEPOKUR
	OP-6 Ayfer KARLITEPE	
12:15-13:30	**LUNCH**	
13:30-15:15	**Session 6 : Immunity and Cell Death**	
	Chairs: Nesrin OZOREN, Ayten NALBANT	
13:30-14:15	Mer Receptor Tyrosine Kinase and Macrophage Phagocytosis Of Apoptotic Cells, **Ian DRANSFIELD**	
14:15-14:45	Unequal Cell Death In The Differentiation Of T Helper Cell Subsets, **Ayten NALBANT**	
14:45-15:15	**Oral Presentations**	
	Session-6 Conference Hall Chair: Isil KURNAZ	**Session-7 Classroom- B (Downstairs) Chair: Tuncay DEMIRYUREK**
	OP-1 Ayten NALBANT	OP-1 Hatice Mehtap KUTLU
	OP-2 Ferdiye TANER	OP-2 Zekiye S. ALTUN
	OP-3 Seminay GULER	OP-3 Melike OZGUL
15:15-15:45	**Coffee Break**	
15:45-17:30	**Session 7 : Therapeutic Approaches to Cell Death**	
	Chairs: Haval SHIRWAN, Pinar AKAN	
15:45-16:30	Monocytes: Killers or Saviors, **Gabriel LOPEZ-BERESTEIN**	
16:30-17:00	Apoptosis As A Powerful Means Of Immune Modulation For The Treatment Of Type 1 Diabetes, **Haval SHIRWAN**	
17:00-17:30	Abnormal Brain Ganglioside Accumulation Triggers Apoptosis In Early Onset Tay - Sachs Disease Mouse Model, **Volkan SEYRANTEPE**	
17:30-18:00	**AWARDS AND CLOSING CEREMONY**	

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Dokuz Eylül Üniversitesi İZMİR-1982

Sabancı Üniversitesi

Türk Biyokimya Derneği Turkish Biochemical Society 1975

LECTURES

S-1

REGULATION OF RIPKS IN CELL SURVIVAL AND CELL DEATH BY APOPTOSIS AND NECROPTOSIS, INSIGHTS AND THERAPEUTIC POTENTIAL

Peter Vandenabeele[1,2,3]

[1]VIB Inflammation Research Center, Technologiepark 927, Zwijnaarde-Ghent, 9052, Belgium
[2]Department of Biomedical Molecular Biology, Ghent University, Technologiepark 927, Zwijnaarde-Ghent, 9052, Belgium
[3]Methusalem program, Ghent University, Technologiepark 927, Zwijnaarde-Ghent, 9052, Belgium

Necroptosis was initially identified as a backup cell death program when apoptosis is blocked. However, it is now recognized as a cellular defense mechanism against infections and is presumed to be a detrimental factor in several pathologies driven by cell death. Necroptosis is a prototypic form of regulated necrosis that depends on activation of the necrosome, which is a protein complex in which receptor interacting protein kinase (RIPK) 3 is activated. The RIP homotypic interaction motif (RHIM) is the core domain that regulates activation of the necrosome. To date, three RHIM-containing proteins have been reported to activate the kinase activity of RIPK3 within the necrosome: RIPK1, Toll/IL-1 receptor domain-containing adaptor inducing IFN-β (TRIF), and DNA-dependent activator of interferon regulatory factors (DAI).

RIPK1 is a key molecule determining cellular fate downstream of several innate immune receptors. It is a serine/threonine kinase consisting of an N-terminal kinase domain linked by a largely unstructured intermediate domain to a C-terminal death domain. In the TNF signaling pathway, RIPK1 paradoxically promotes cell survival as well as cell death. These opposite cellular functions are mediated by 2 distinct faces of RIPK1. Upon binding of TNF to TNFR1, RIPK1 is recruited to the receptor complex I where it acts as a scaffold protein promoting cell survival, in part, by activating the canonical NF-kB pathway. Specific conditions can however activate RIPK1, and its kinase activity then regulates assembly of 2 possible cytosolic death-inducing complexes, namely complex IIb (RIPK1-FADD-Caspase-8) and the necrosome (RIPK1-RIPK3-MLKL). These complexes respectively drive TNF-mediated apoptosis or necroptosis. The precise molecular mechanism(s) controlling RIPK1 activation is (are) currently unknown. Similarly, how RIPK1 kinase activity contributes to both cell death processes still remains unclear. Despite this lack of understanding, it is evident that RIPK1 can play a dual role downstream of TNFR1 and that its kinase activity needs tight repression to avoid unnecessary damage to the organism.

Targeting necroptosis can occur at three levels: blocking RIPK1 and RIPK3 kinase activity, and blocking MLKL. Novel drugs and known drugs have been identified in cellular screening which block necroptosis. In vivo they are effective in blocking inflammatory, degenerative and infectious diseases. On the other hand induction of necroptosis has been found effective in inducing immunogenic cell death.

S-2

TYPE 2 TRANSGLUTAMINASE: A KEY REGULATOR OF PROTEOSTASIS UNDER CELLULAR STRESSFUL CONDITIONS

Mauro Piacentini

Department of Biology, University of Rome "Tor Vergata", Rome, Italy.
National Institute for Infectious Diseases, IRCCS "Lazzaro Spallanzani", Rome, Italy.

Eukaryotic cells are equipped with an efficient quality control system to selectively eliminate misfolded and damaged proteins, and organelles. Abnormal polypeptides that escape from proteasome-dependent degradation and aggregate in the cytosol can be transported via microtubules to inclusion bodies called 'aggresomes', where misfolded proteins are confined and degraded by autophagy. We showed that Type 2 transglutaminase (TG2) knockout mice display impaired autophagy and accumulate ubiquitinated protein aggregates upon starvation. Furthermore, p62-dependent peroxisome degradation is also impaired in the absence of TG2. We also demonstrate that, under cellular stressful conditions, TG2 physically interacts with p62 and they are localized in cytosolic protein aggregates, which are then recruited into autophagosomes, where TG2 is degraded. Interestingly, the enzyme's crosslinking activity is activated during autophagy and its inhibition leads to the accumulation of ubiquitinated proteins. Taken together, these data indicate that the TG2 transamidating activity has an important role in the assembly of protein aggregates, as well as in the clearance of damaged mitochondria by macroautophagy.

Recently, we have isolated and characterized exosomes derived from cells either expressing or not TG2, under stressful conditions (i.e. proteasome impairment or expressing a mutated form of huntingtin (mHtt) containing 84 polyglutamine repeats). Our results show that TG2 is present in the exosomes only upon proteasome blockade, a condition in which TG2 interacts with TSG101 and ALIX, two key proteins involved in exosome biogenesis. Interestingly, we found that TG2 favours the assembly of a protein complex including mHtt, ALIX, TSG101 and BAG3, a co-chaperone involved in the clearance of mHtt. The formation of this complex is paralleled by the selective recruitment of mHtt and BAG3 in the exosomes derived from TG2 proficient cells only. Overall, our data indicate that TG2 is an important player in the biogenesis of exosomes controlling the selectivity of their cargo under stressful cellular conditions. Taken together these data indicate that TG2 plays a key role in the regulation of proteostasis under stressful cellular conditions.

S-3

AUTOPHAGY AT THE INTERSECTION BETWEEN CELL SURVIVAL AND CELL DEATH: ROLES IN INFLAMMATION AND LYSOSOMAL HOMEOSTASIS

Vojo Deretic

Departments of Molecular Genetics and Microbiology, Cell Biology and Physiology and Neurology, University of New Mexico Health Sciences Center, USA

Autophagy is a fundamental biological process that fulfills general and specialized roles in cytoplasmic homeostasis, and is at the intersection between cell survival and cell death. This talk will cover the subsystems in autophagy and the recent progress in our understanding of how they come together in the contest of immunity and inflammation. We will also give an update on organizers of precision autophagy in the context of immune and other functions. Furthermore, the role of TRIM proteins in autophagy-based lysosomal homeostasis and their role in in lysosomal cell death will be presented.

S-4

NOVEL REGULATORS OF AUTOPHAGY*

Devrim Gözüaçık, MD PhD

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Autophagy is key biological event that occurs at low basal levels in all cell types from yeast to mammals under non-deprived conditions, performing homeostatic functions such as protein degradation and organelle (e.g. mitochondria) turnover. It is rapidly upregulated during cellular stress, providing cells with recycled intracellular building blocks and substrates for energy generation, hence allowing them to survive unfavorable conditions.Autophagy dysregulations play a critical role in the pathogenesis and progress of several human health problems, including neurodegenerative disorders (i.e. Alzheimer's, Parkinson's and Huntington's diseases), degenerative syndromes (i.e. Dystrophies and dystrophic syndromes), lysosomal storage disorders (i.e. Gaucher's disease), inflammation and cancer.

In Gozuacik Laboratory in Sabanci University, we mainly focus on the discovery of novel autophagy regulators: RNAs, proteins and pathways (basic research). Moreover in close collaboration with clinicians, we study implications of our findings in human disease formation (pathology and pathogenesis research) and diagnosis (marker research). In collaboration with chemists and pharmacologists, we search for means to modulate autophagy for treatment purposes (drug research). In this speech, results from our basic and medical studies on autophagy will be discussed.

*This work was supported by The Scientific and Technological Research Council of Turkey (TUBITAK) 1001 Grant numbers 112T272 and 110T405 and Sabanci University.

Selected References:

1) Arachiche A and **Gozuacik D^x**. Autophagy in health and disease. In the book: Toxicity and autophagy in neurodegenerative disorders. Jose Manuel Fuentes (Ed.). **Springer Publishing**. 2015. ISBN: 978-3-319-13938-8.
2) Tekirdag KA, Ozturk DG, **Gozuacik D^x**. Regulation of autophagy by miRNAs. In the book: Autophagy: Cancer, Other Pathologies, Inflammation, Immunity, and Infection. Hayat MA (Ed.). Elsevier Academic Press. 2015. ISBN: 9780128010327
3) Korkmaz G*, Tekirdag KA*, Ozturk DG, Kosar A, Sezerman OU and **Gozuacik D^x**. MIR376A is a regulator of starvation-induced autophagy. **PLoS ONE**, 2013, 8(12): e82556. doi:10.1371/journal.pone.0082556.
4) Tekirdag AK*, Korkmaz G*, Ozturk DG, Agami R, **Gozuacik D^x**. miR-181a regulates starvation- and rapamycin-induced autophagy through targeting of ATG5. **Autophagy**, 2013 March; 9(3): 1-12.
5) Oral O*, Oz-Arslan D*, Itah Z, Naghavi A, Deveci R, Karacali S, **Gozuacik D^x**. Cleavage of Atg3 protein by caspase-8 regulates autophagy during receptor-activated cell death. **Apoptosis**, 2012 Aug; 17(8):810-20.
6) Korkmaz G, le Sage C, Tekirdag AK, Agami R, **Gozuacik D^x**. miR-376b controls starvation and mTOR inhibition-related autophagy by targeting ATG4C and BECN1. **Autophagy**, 2012 February; 8 (2): 165-176.

S-5

MITOCHONDRIAL SUBSTRATES: A TOOL TO COMBAT CANCER

Boris Zhivotovsky

Institute of Environmental Medicine, Division of Toxicology, Karolinska Institutet, Stockholm, Sweden;
Lomonosov Moscow State University, Moscow, Russia

Mitochondria play an important role in regulation of various cell death modalities. Outer mitochondrial membrane permeabilization and release of several proteins, such as cytochrome c, SMAC, AIF, etc. from the intermembrane space of mitochondria are regarded as a “point of no return” in many models of apoptosis. Accumulating evidence suggested that mitochondria-generated reactive oxygen species are involved in this process. However, depending on their overall concentration at steady state levels, and efficiency of mitochondrial antioxidant enzymes cell death can be prevented.

Cancer cells demonstrate dramatically increased glycolysis even under air-saturated conditions (Warburg effect), whereas mitochondrial contribution to ATP supply is restrained. Consequently, drugs that can perturb glycolysis, might display beneficial therapeutic effects. Our recent observations revealed that among agents that can modulate tumor cell death are members of the Krebs cycle, succinate and citrate. The later can directly suppress glycolysis via inhibition of phosphofructokinase. Importantly, mutations of succinate dehydrogenase (SDH) characterize several tumors. Inhibition of SDH results in accumulation of succinate in cytosol and subsequent activation of hypoxia-inducible factor, which is responsible for upregulation of glycolytic pathway and mitochondrial silencing. We found that in addition to this pathway succinate might suppress apoptosis at mitochondrial level. A link between mitochondrial metabolic changes and cell death as well as how alteration of energy producing pathways can sensitize tumor cells to treatment will be discussed.

S-6

AMBRA1 NEGATIVE CONTROL AT THE CROSSROAD AMONG AUTOPHAGY, CELL PROLIFERATION AND CELL DEATH

Francesco Cecconi

Department of Biology, University of Rome Tor Vergata, Rome, Italy
Cell Stress and Survival Unit, Danish Cancer Society Research Centre, Copenhagen, Denmark

The activating molecule in Beclin 1-regulated autophagy (AMBRA1), also known as autophagy/beclin-1 regulator 1, is a highly intrinsically disordered and vertebrate-conserved adapter protein that is part of the autophagy signalling network. AMBRA1 is an important regulator of embryonic development, and both its mutation or inactivation have been shown to impact several pathologies of the nervous system, and to be involved in carcinogenesis. Recent studies have revealed that AMBRA1 can coordinate a cell response to starvation or other stresses by integrated functions that include translocation of the autophagosome core complex to the ER, regulative ubiquitylation and stabilization of the kinase ULK1, selective mitochondria removal and cell cycle down-regulation. On the other side, AMBRA1 itself appears to be targeted by a number of regulations, such as Cullin-dependent degradation, caspase cleavage and several modifications, ranging from phosphorylation to ubiquitylation.

Here we will discuss two relevant novel pathways of AMBRA1 down-regulation:
i) AMBRA1 targeting by miR7, an autophagy- and cell-cycle-related microRNA, in a signalling loop we have identified that involves the oncogene c-myc and the phosphatase PP2A;
ii) AMBRA1 proteolytic cleavage to generate a novel positive mediator of mitochondrial apoptosis. Indeed, the C-Terminal part of AMBRA1, generated by caspase cleavage upon apoptosis induction, is able to inhibit the anti-apoptotic factor BCL2 by a direct binding through its BH3-like domain.

Altogether, both mitochondrial AMBRA1-BCL2 networking and AMBRA1 regulation by miRNAs may represent novel targets in development of therapeutic approaches in human diseases.

S-7

IN VITRO AND IN VIVO CHARACTERIZATION OF CELL SURVIVAL GENES USING DESTABILIZED CAS9

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One of the problems limiting the use of current CRISPR systems is the constitutive endonuclease activity when Cas9 and its sgRNA are co-expressed. Here, we sought to improve upon existing CRISPR/Cas9 techniques to generate a system that (1) would provide potent, robust and temporally controlled gene editing, (2) be applicable to a broad spectrum of cell types and tissues, (3) facilitate high throughput manipulation and (4) be traceable. To this end, we exploited recently developed strategies in which a cell-permeable ligand is used in conjugation with a single genetically encoded destabilizing domain (DD) to regulate the expression of any protein of interest. By fusing the FKBP12-derived destabilizing domain to Cas9 (i.e., DD-Cas9) we demonstrated that this method of conditional regulation of protein stability could be exploited for rapid and reversible Cas9 expression in vitro. We validated the efficiency of this new platform by conditionally targeting a variety of genes controlling diverse biological processes. By targeting the RPA3 gene and EGFR in the “EGFR-addicted” cells, in particular, we demonstrated the ability of the system to identify genes that are essential for sustained cell growth and survival. The unique aspect of this method is the conditional regulation of Cas9 protein expression independently of its mRNA expression. When coupled with a conditional Cre allele (Cre-ER^{T2}), DD-Cas9 could be utilized to facilitate the analysis of genes that modulate disease onset and progression in a variety of pre-existing mouse models of human disease based on Cre-lox system. In summary, our data indicate that fusing Cas9 to a destabilizing domain provides a highly efficient and potent, easy scalable, robust and tunable new modality for temporal control of gene editing that can be applicable to a broad spectrum of in vitro and in vivo models.

S-8

STAMPING PROLIFERATION AND CELL DEATH IN PROSTATE CANCER

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The six transmembrane protein of prostate (STAMP) family, also known as six transmembrane epithelial antigen of prostate (STEAP), have been implicated in prostate cancer (PCa). STAMP1 and STAMP2 protein expression is increased in human PCa compared with benign prostate and they regulate central proliferative signaling pathways in PCa cells *in vitro* and *in vivo*. In addition, STAMP1 and STAMP2 expression inhibits cell death resulting in robust tumor growth. Consistent with these findings, drug-induced therapeutic silencing of STAMPs by systemic nanoliposomal-siRNA delivery profoundly inhibits tumor growth in preclinical mouse PCa models. These data suggest that STAMPs have a key role in determining life and death decisions in PCa and thus may serve as novel therapeutic targets.

S-9
ETS TRANSCRIPTION FACTORS - CRITICAL REGULATORS OF BRAIN TUMOR INITIATING CELL PROLIFERATION VS NEURODEGENERATION

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Object: Elk-1, a member of the ETS superfamily of transcription factors, has long been associated with regulation of immediate-early genes in response to mitogen-dependent MAPK pathway activation. However, its presence in post-mitotic neurons had been an intriguing fact. Work from our lab and others have since shown that Elk-1 is important for survival of neurons, and that knockdown of Elk-1 triggers cell death, yet the mechanism through which Elk-1 mediates this was unclear.
Material and Method: A combination of microarray, promoter analyses and luciferase assays, as well as real-time PCR and brain tumor initiating cell assays have been used to underpin the role of Elk-1 in neurodegeneration vs tumorigenesis.
Results: Microarray analysis of Elk-1 overexpression in SH-SY5Y cell lines have shown that a number of apoptosis and autophagy-related genes were regulated by Elk-1, in addition to components of hypoxia signaling pathway. More interestingly, however, quite a number of organogenesis- and stem cell-related genes were found to be regulated in response to Elk-1. To validate the results, we have carried out a series of real-time PCR and luciferase assays to show that these genes indeed are regulated by Elk-1 in various brain tumor cell lines, and that CD133+ cells express higher Elk-1 transcript.
Conclusion: In line with previous findings that Elk-1 is essential in human embryonic stem cells (hESCs) and that ERK2 MAPK cooccupies various promoters of cell-cycle and pluripotency-associated genes, our study shows that Elk-1 also regulates pluripotency-related promoters in brain tumor initiating cells. On the contrary, T417-phosphorylation of Elk-1 was reported to be associated with inclusion bodies in a number of neurodegenerative diseases. We believe that these two results are not contradictory – our hypothesis is that in the absence of proper survival signaling, or in the presence of pro-apoptotic ones, Elk-1 instead initiates the apoptotic pathway, hence acting as a choice-point between life or death.
Keywords: ETS, Elk-1, neurodegeneration, brain tumor initiating cell, tumorigenesis

S-10

DIMINISHED CYCLIN DEPENDENT KINASE ACTIVITY AND MTOR ARE CRITICAL IN THE CELL DEATH DECISION THROUGH AFFECTING STAT SIGNALLING DIFFERENTLY IN PROSTATE CANCER CELLS

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Prostate cancer is the second most frequently diagnosed cancer as well as the sixth leading cause of death in males with cancer worldwide. Androgens play a critical role in prostate cancer development. However prostate cancer cells may progress androgen-independently that causes higher mortality rates. Therefore, new therapeutic targets and clarification of their signaling pathways is critical in treatment of aggressive prostate cancer cases. One of the promising anti-cancer strategy is the inhibition of proliferating cancer cells via targeting cyclins and cyclin-dependent kinases (CDKs) complexes, which causes supression of cell survival signalling routes. New generation CDK inhibitors roscovitine (CYC202, seliciclib) or purvalanol inhibits specific CDK targets and thus prevents cell proliferation and induces apoptosis. In this study, purvalanol and roscovitine was used to expose the mechanism underlying mTOR-related apoptotic and/or autophagic response and to understand roles of mTOR depending on its signal cascades in the cell death processes via mTOR silenced androgen receptor (AR) negative PC3, DU145 and AR positive LNCaP prostate cancer cell lines. In PC3 and LNCaP cells, CDK inhibitors were used alone and with mTOR siRNA combination to scan and analyze differentiation upstream and downstream targets of mTOR using Pathscan ELISA Assay. CDK inhibitors purvalanol and roscovitine affects activation of mTOR and mTOR-related kinases in a CDK-independent manner. However purvalanol exerts more potent inhibitory function than roscovitine. In PC3 and LNCaP cell lines, mTOR deficiency causes blockage of apoptotic processes induced by CDK inhibitors. On the other hand, regulation of STAT1 and STAT3 proteins by mTOR seems to determine apoptotic effets of those drugs. Increased STAT3 Ser727 phosphorylation levels by CDK inhibitors leads to decrease STAT3-FoXO1 and CDK5 activity. Diminished CDK5 activity then causes AR-STAT3 dissociation. Therefore, especially differentiation in STAT3 expression and phosphorylation status play a vital role in the manner of regulation of cell survival and cell death pathways signalling. However, CDK inhibitors and their combination with mTOR siRNA leads to diverse STAT3 expression profiles in DU145 cell line. In conclusion, CDK inhibitors are promising drug candidates in the treatment of aggressive prostate cancer cells through modulating different molecular targets depends on the cell type.

S-11

THE BRIGHT AND THE DARK SIDES OF REPROGRAMMING TO PLURIPOTENCY

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Somatic cell reprogramming with a few defined transcription factors to pluripotency is a several weeks long process. The driving forces behind this phenomenon and the cascade of events are very poorly understood. It is however crucial to uncover the fine details of this process in order to comprehend the true property of these induced pluripotent stem cells (iPSCs) and so better tailor their future therapeutic and disease study use.

Several years ago, we developed a reprogramming method utilizing a *piggyBac* (PB) transposon-mediated delivery of the reprogramming transgenes. Beyond the ability of seamless removal of the transgenes once pluripotent stem cells have been generated, this system has additional unique features. For example, when combined with the doxycycline inducible transgene expression system, we found that these transgenes are very efficiently regulatable by adding or withdrawing doxycycline. *In vivo* differentiated somatic cells derived from these iPSCs can be reprogrammed to “secondary” iPSCs (2ºiPSc) by simply adding doxycycline to the culture medium. Somatic cell lines produced with this method frequently return to 2ºiPSc in a "population" manner, which allows us to study the cascade of molecular events during the entire process of reprogramming. This unexpected and unique property of the PB reprogramming system allowed the characterization of the three week reprogramming process at an almost daily resolution at multiple omics levels, leading to a better understanding the molecular events associated with generating pluripotent cells from somatic cells.

In parallel, we have investigated the genetic changes associated with the reprogramming process. We identified de novo generated copy number variations at the early phase of reprogramming, which created a high level of genetic mosaicism. Intriguingly, the genome damage load is decreasing when the cells are cultured for an intermediate period of time. Our studies led us to conclude that this phenomenon is due to selection against mutated cells.

S-12

STEM CELL THERAPY APPROACHES TO ISCHEMIC CARDIOMYOPATHY

Alp Can

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Over the past 15 years, numerous stem cell trials have been performed in patients with ischemic cardiomyopathy (ICM), using both autologous and allogeneic stem cells. Although many individual studies reported encouraging signals, these were all phase 1 or 2 studies with appropriately small numbers of patients, and their conclusions must therefore be considered preliminary. In an attempt to increase statistical robustness, a recent meta-analysis assessing the results of all randomized clinical trials of stem cell therapy for patients with acute myocardial infarction (AMI) was performed, demonstrating no net beneficial effects on outcomes, except for a small improvement in ejection fraction. Given these results, a reassessment of the rationale for the use of stem cells in cardiovascular disease is timely. Patients with ICM invariably have usually extensive areas of myocardial scar. ICM patients had areas of myocardial dysfunction due not to scar, but to dysfunctional viable myocardium (DVM). DVM provides a potential target for therapeutic interventions in ICM. If the dysfunctional tissue consists of viable rather than scarred myocardium, LV function can presumably be improved. The concept of DVM may also help direct which patients may benefit most from stem cell therapy. As the stage of HF that may be considered too late for stem cell therapy is unclear, the presence of DVM may help guide the identification of those patients with the most potential to benefit. The potential of any therapy, including stem cells, to improve outcomes in ICM is related not only to its effects on restoring function to DVM, but also to its capacity to improve processes that contribute to progressive deterioration of LV structure and function. On the basis of this conceptual framework, my aim is to explore the potential of stem cells to exert beneficial effects in ICM by considering the overlap between pathways believed to contribute to disease progression (other than atherosclerotic disease of the large coronary arteries) and the known activities of stem cells that could favorably influence these pathways. (Project No: 0741-STZ-2014)

S-13

TARGETING INFLAMMATORY AND APOPTOTIC PATHWAYS BY AGENTS DESIGNED BY MOTHER NATURE FOR PREVENTION AND TREATMENT OF CANCER

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Chronic infections, obesity, alcohol, tobacco, radiation, environmental pollutants, and high-calorie diet have been recognized as major risk factors for the most common types of cancer. All these risk factors are linked to cancer through inflammation and apoptosis. While acute inflammation that persists for short-term mediates host defense against infections, chronic inflammation that lasts for long-term can predispose the host to various chronic illnesses, including cancer. Linkage between cancer and inflammation is indicated by numerous lines of evidence; first, transcription factors NF-κB and STAT3, two major pathways for inflammation, are activated by most cancer risk factors; second, an inflammatory condition precedes most cancers; third, NF-κB and STAT3 are constitutively active in most cancers; fourth, hypoxia and acidic conditions found in solid tumors activate NF-κB; fifth, chemotherapeutic agents and gamma irradiation activate NF-κB and lead to chemoresistance and radioresistance; sixth, most gene products linked to inflammation, survival, proliferation, invasion, angiogenesis, and metastasis are regulated by NF-κB and STAT3; seventh, suppression of NF-κB and STAT3 inhibits the proliferation and invasion of tumors; and eighth, most chemopreventive agents mediate their effects through inhibition of NF-κB and STAT3 activation pathways. Thus suppression of these proinflammatory pathways may provide opportunities for both prevention and treatment of cancer. We will discuss the potential of nutraceuticals derived from spices and from traditional medicine in suppression of inflammatory pathways and their role in prevention and therapy of cancer.

Selected References:

1. NF-kappaB and cancer: how intimate is this relationship. Prasad S, Ravindran J, Aggarwal BB. Mol Cell Biochem. 2009 Oct 8.
2. Curcumin Inhibits COPD-Like Airway Inflammation and Lung Cancer Progression in Mice. Moghaddam SJ, Barta P, Mirabolfathinejad SG, Ammar-Aouchiche Z, Torres Garza N, Vo TT, Newman RA, Aggarwal BB, Evans CM, Tuvim MJ, Lotan R, Dickey BF. Carcinogenesis. 2009 Sep 30.
3. Curcumin modulates the radiosensitivity of colorectal cancer cells by suppressing constitutive and inducible NF-kappaB activity. Sandur SK, Deorukhkar A, Pandey MK, Pabón AM, Shentu S, Guha S, Aggarwal BB, Krishnan S. Int J Radiat Oncol Biol Phys. 2009 Oct 1;75(2):534-42.
4. Design of curcumin-loaded PLGA nanoparticles formulation with enhanced cellular uptake, and increased bioactivity in vitro and superior bioavailability in vivo. Anand P, Nair HB, Sung B, Kunnumakkara AB, Yadav VR, Tekmal RR, Aggarwal BB. Biochem Pharmacol. 2009 Sep 6.
5. Signal transducer and activator of transcription-3, inflammation, and cancer: how intimate is the relationship?. Aggarwal BB, Kunnumakkara AB, Harikumar KB, Gupta SR, Tharakan ST, Koca C, Dey S, Sung B. Ann N Y Acad Sci. 2009 Aug;1171:59-76.
6. Curcumin potentiates the antitumor effects of gemcitabine in an orthotopic model of human bladder cancer through suppression of proliferative and angiogenic biomarkers. Tharakan ST, Inamoto T, Sung B, Aggarwal BB, Kamat AM. Biochem Pharmacol. 2009
7. Inflammation, a silent killer in cancer is not so silent! Aggarwal BB. Curr Opin Pharmacol. 2009 Aug;9(4):347-50.
8. Inflammation and cancer: how friendly is the relationship for cancer patients? Aggarwal BB, Gehlot P. Curr Opin Pharmacol. 2009 Aug;9(4):351-69.
9. Zerumbone enhances TRAIL-induced apoptosis through the induction of death receptors in human colon cancer cells: Evidence for an essential role of reactive oxygen species. Yodkeeree S, Sung B, Limtrakul P, Aggarwal BB. Cancer Res. 2009 Aug 15;69(16):6581-9.

10. Curcumin sensitizes human colorectal cancer to capecitabine by modulation of cyclin D1, COX-2, MMP-9, VEGF and CXCR4 expression in an orthotopic mouse model. Kunnumakkara AB, Diagaradjane P, Anand P, Kuzhuvelil HB, Deorukhkar A, Gelovani J, Guha S, Krishnan S, Aggarwal BB. Int J Cancer. 2009 Nov 1;125(9):2187-97.
11. Curcumin and cancer cells: how many ways can curry kill tumor cells selectively? Ravindran J, Prasad S, Aggarwal BB. AAPS J. 2009 Sep;11(3):495-510.
12. Acetyl-11-keto-beta-boswellic acid inhibits prostate tumor growth by suppressing vascular endothelial growth factor receptor 2-mediated angiogenesis. Pang X, Yi Z, Zhang X, Sung B, Qu W, Lian X, Aggarwal BB, Liu M. Cancer Res. 2009 ;69(14):5893-900.
13. Fabrication and characterization of silk fibroin-derived curcumin nanoparticles for cancer therapy. Gupta V, Aseh A, Ríos CN, Aggarwal BB, Mathur AB. Int J Nanomedicine. 2009;4:115-22.
14. K-Ras promotes angiogenesis mediated by immortalized human pancreatic epithelial cells through mitogen-activated protein kinase signaling pathways. Matsuo Y, Campbell PM, Brekken RA, Sung B, Ouellette MM, Fleming JB, Aggarwal BB, Der CJ, Guha S. Mol Cancer Res. 2009 Jun;7(6):799-808.
15. Molecular targets of nutraceuticals derived from dietary spices: potential role in suppression of inflammation and tumorigenesis. Aggarwal BB, Van Kuiken ME, Iyer LH, Harikumar KB, Sung B. Exp Biol Med (Maywood). 2009 Aug;234(8):825-49.
16. Nuclear factor-kappa B links carcinogenic and chemopreventive agents. Ralhan R, Pandey MK, Aggarwal BB. Front Biosci (Schol Ed). 2009 Jun 1;1:45-60.
17. Models for prevention and treatment of cancer: problems vs promises.Aggarwal BB, Danda D, Gupta S, Gehlot P. Biochem Pharmacol. 2009 Nov 1;78(9):1083-94.
18. Curcumin circumvents chemoresistance in vitro and potentiates the effect of thalidomide and bortezomib against human multiple myeloma in nude mice model. Sung B, Kunnumakkara AB, Sethi G, Anand P, Guha S, Aggarwal BB. Mol Cancer Ther. 2009 Apr;8(4):959-70.
19. Zerumbone abolishes RANKL-induced NF-kappaB activation, inhibits osteoclastogenesis, and suppresses human breast cancer-induced bone loss in athymic nude mice. Sung B, Murakami A, Oyajobi BO, Aggarwal BB. Cancer Res. 2009 Feb 15;69(4):1477-84.
20. The guggul for chronic diseases: ancient medicine, modern targets. Shishodia S, Harikumar KB, Dass S, Ramawat KG, Aggarwal BB. Anticancer Res. 2008 Nov-Dec;28(6A):3647-64.
21. Withanolide sulfoxide from Aswagandha roots inhibits nuclear transcription factor-kappa-B, cyclooxygenase and tumor cell proliferation. Mulabagal V, Subbaraju GV, Rao CV, Sivaramakrishna C, Dewitt DL, Holmes D, Sung B, Aggarwal BB, Tsay HS, Nair MG. Phytother Res. 2009 Jul;23(7):987-92.Targeting inflammatory pathways for prevention and therapy of cancer: short-term friend, long-term foe. Aggarwal BB, Vijayalekshmi RV, Sung B. Clin Cancer Res. 2009 Jan 15;15(2):425-30.
22. Boswellic acid blocks signal transducers and activators of transcription 3 signaling, proliferation, and survival of multiple myeloma via the protein tyrosine phosphatase SHP-1. Kunnumakkara AB, Nair AS, Sung B, Pandey MK, Aggarwal BB. Mol Cancer Res. 2009 Jan;7(1):118-28.
23. Pharmacological basis for the role of curcumin in chronic diseases: an age-old spice with modern targets. Aggarwal BB, Sung B. Trends Pharmacol Sci. 2009 Feb;30(2):85-94.
24. Butein suppresses constitutive and inducible signal transducer and activator of transcription (STAT) 3 activation and STAT3-regulated gene products through the induction of a protein tyrosine phosphatase SHP-1.Pandey MK, Sung B, Ahn KS, Aggarwal BB. Mol Pharmacol. 2009 Mar;75(3):525-33.
25. Resveratrol addiction: to die or not to die. Shakibaei M, Harikumar KB, Aggarwal BB. Mol Nutr Food Res. 2009 Jan;53(1):115-28.
26. Zerumbone down-regulates chemokine receptor CXCR4 expression leading to inhibition of CXCL12-induced invasion of breast and pancreatic tumor cells. Sung B, Jhurani S, Ahn KS, Mastuo Y, Yi T, Guha S, Liu M, Aggarwal BB. Cancer Res. 2008 Nov 1;68(21):8938-44.
27. Aggarwal BB, Kunnumakkara AB, Harikumar KB, Tharakan ST, Sung B, Anand P. Potential of Spice-Derived Phytochemicals for Cancer Prevention. Planta Med. 2008 Jul 8.
28. Aggarwal BB, Harikumar KB. Potential therapeutic effects of curcumin, the anti-inflammatory agent, against neurodegenerative, cardiovascular, pulmonary, metabolic, autoimmune and neoplastic diseases. Int J Biochem Cell Biol. 2008 Jul 9. [Epub ahead of print]
29. Sethi G, Sung B, Aggarwal BB. TNF: a master switch for inflammation to cancer. Front Biosci. 2008 May 1;13:5094-107.
30. Aggarwal BB. The past, present and future of multi-targeted cancer treatment "Naturally": Food for thought. Cancer Lett. 2008 May 24. [Epub ahead of print] No abstract available.

31. Kunnumakkara AB, Anand P, Aggarwal BB. Curcumin inhibits proliferation, invasion, angiogenesis and metastasis of different cancers through interaction with multiple cell signaling proteins. Cancer Lett. 2008 May 12. [Epub ahead of print]

32. Anand P, Sundaram C, Jhurani S, Kunnumakkara AB, Aggarwal BB. Curcumin and cancer: An "old-age" disease with an "age-old" solution. Cancer Lett. 2008 Aug 18;267(1):133-64.
33. Harikumar KB, Aggarwal BB. Resveratrol: a multitargeted agent for age-associated chronic diseases. Cell Cycle. 2008 Apr;7(8):1020-35. Epub 2008 Feb 15.
34. Goel A, Jhurani S, Aggarwal BB. Multi-targeted therapy by curcumin: how spicy is it? Mol Nutr Food Res. 2008 Apr 2. [Epub ahead of print]
35. Goel A, Kunnumakkara AB, Aggarwal BB. Curcumin as "Curecumin": From kitchen to clinic. Biochem Pharmacol. 2007 Aug 19; [Epub ahead of print]
36. Aggarwal B.B, C. Sundaram, N. Malani and H. Ichikawa, "Curcumin: The Indian solid gold", in The Molecular Targets and Therapeutic Uses of Curcumin in Health and Disease (ed by B.B. Aggarwal, Y-J. Surh, S. Shishodia), Springer Publishing Company, New York., 2007, p. 1-76.
37. Aggarwal BB, Shishodia S, Sandur SK, Pandey MK, Sethi G. Inflammation and cancer: how hot is the link? Biochem Pharmacol. 2006 Nov 30;72(11):1605-21.
38. Aggarwal BB. Nuclear factor-kappa B: a transcription factor for all seasons. Expert Opin Ther Targets. 2007 Feb;11(2):109-10.
39. Jagetia GC, Aggarwal BB. "Spicing up" of the immune system by curcumin. J Clin Immunol. 2007 Jan;27(1):19-35.
40. Aggarwal BB, Sethi G, Ahn KS, Sandur SK, Pandey MK, Kunnumakkara AB, Sung B, Ichikawa H. Targeting signal-transducer-and-activator-of-transcription-3 for prevention and therapy of cancer: modern target but ancient solution. Ann N Y Acad Sci. 2006 Dec;1091:151-69.
41. Garodia P, Ichikawa H, Malani N, Sethi G, Aggarwal BB. From ancient medicine to modern medicine: ayurvedic concepts of health and their role in inflammation and cancer. J Soc Integr Oncol. 2007 Winter;5(1):25-37.
42. Ahn KS, Aggarwal BB. Transcription factor NF-kappaB: a sensor for smoke and stress signals. Ann N Y Acad Sci. 2005 Nov;1056:218-33.
43. Aggarwal BB, Shishodia S. Molecular targets of dietary agents for prevention and therapy of cancer. Biochem Pharmacol. 2006 ;71(10):1397-421.
44. Aggarwal BB, Kumar A, Bharti AC. Anticancer potential of curcumin: preclinical and clinical studies. Anticancer Res. 2003 Jan-Feb;23(1A):363-98.
45. Aggarwal BB. Signalling pathways of the TNF superfamily: a double-edged sword. Nat Rev Immunol. 2003 Sep;3(9):745-56.
46. Aggarwal BB. Nuclear factor-kappaB: the enemy within. Cancer Cell. 2004 Sep;6(3):203-8.
47. Dorai T, Aggarwal BB. Role of chemopreventive agents in cancer therapy. Cancer Lett. 2004 Nov 25;215(2):129-40.
48. Aggarwal BB, Takada Y, Oommen OV. From chemoprevention to chemotherapy: common targets and common goals. Expert Opin Investig Drugs. 2004 Oct;13(10):1327-38.
49. Aggarwal BB, Shishodia S. Suppression of the nuclear factor-kappaB activation pathway by spice-derived phytochemicals: reasoning for seasoning. Ann N Y Acad Sci. 2004 ;1030:434-41.
50. Aggarwal BB, Bhardwaj A, Aggarwal RS, Seeram NP, Shishodia S, Takada Y. Role of resveratrol in prevention and therapy of cancer: preclinical and clinical studies. Anticancer Res. 2004 Sep-Oct;24(5A):2783-840.
51. Ahn KS, Aggarwal BB. Transcription Factor NF-{kappa}B: A Sensor for Smoke and Stress Signals. Ann N Y Acad Sci. 2005 Nov;1056:218-33.
52. Shishodia S, Sethi G, Aggarwal BB Curcumin: getting back to the roots. Ann N Y Acad Sci. 2005 Nov;1056:206-17.
53. Garg AK, Buchholz TA, Aggarwal BB. Chemosensitization and radiosensitization of tumors by plant polyphenols. Antioxid Redox Signal. 2005 Nov-Dec;7(11-12):1630-47. Review.
54. Aggarwal BB, Shishodia S, Takada Y, Jackson-Bernitsas D, Ahn KS, Sethi G, Ichikawa H. TNF blockade: an inflammatory issue. Ernst Schering Res Found Workshop. 2006;(56):161-86.
55. Aggarwal BB, Ichikawa H, Garodia P, Weerasinghe P, Sethi G, Bhatt ID, Pandey MK, Shishodia S, Nair MG. From traditional Ayurvedic medicine to modern medicine: identification of therapeutic targets for suppression of inflammation and cancer. Expert Opin Ther Targets. 2006 Feb;10 (1):87-118.

56. Aggarwal BB, Shishodia S, Sandur SK, Pandey MK, Sethi G. Inflammation and cancer: How hot is the link/ Biochem Pharmacol. 2006 Nov 30;72(11):1605-21
57. Aggarwal, B.B. and Shishodia S., Surh Y-J (eds.) *The Molecular Targets and Therapeutic Uses of Curcumin in Health and Disease* Springer Publishers, New York, 2007
58. Aggarwal, B.B. and Shishodia S., (eds.) *Resveratrol in Health and Disease*. Taylor and Francis Books, Inc., Boston, pp. 1-679, 2006

S-14

MOLECULAR MECHANISMS OF FLAVONOID QUERCETIN IN ENHANCING APOPTOSIS IN CHRONIC LYMPHOCYTIC LEUKEMIA

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Quercetin is the most abundant flavonoid present in the diet and its disease preventing properties have been largely investigated. Among these, quercetin is able to modulate several hallmarks of cancer, including resistance to apoptosis. Previous studies from our group demonstrated the capacity of quercetin to sensitize several leukemia cell lines and B-cells isolated from patients affected by chronic lymphocytic leukemia (CLL) to death ligand agonists (e.g., anti-CD95 and rTRAIL). Moreover, in association with canonical and innovative chemotherapeutic drugs (fludarabine, ABT-737 and BH3-mimetics), quercetin synergistically enhances the drug response against CLL. This effect is mediated by changes in the expression and activity of anti-apoptotic proteins belonging to the Bcl-2 family. Among these, Mcl-1 has been associated to apoptotic resistance in CLL. Aim of this presentation is to review the apoptotic-enhancing activity of quercetin in vitro (leukemic cell lines) and ex vivo (B-CLL cells) models, depending upon the down-regulation of Mcl-1. Using a relatively new cell line, HG3 derived from primary B-cells immortalized with Epstein-Barr virus, we will show how the association between BH3-mimetics and quercetin synergistically induces apoptosis through the inhibition of PI3K/Akt signaling pathway. We also identified the protein kinase CK2 as the direct and primary target of quercetin, since CK2 activity is inhibited by the flavonoid within one minute from the treatment. In the case of ABT-737, a BH3-mimetic compound which binds and inactivates Bcl-2, Bcl-XL, BCL-W members, but weakly interacts with Mcl-1, we demonstrated that the combined treatment of ABT-737 and quercetin represents a promising new therapeutic strategy against B-CLL. In fact, since quercetin contributes to remove the resistance due to Mcl-1 over-expression, it allows ABT-737 to exhibit its therapeutic efficacy against pro-survival Bcl-2 factors. Considering the rapid uptake of quercetin and its low toxicity against normal peripheral blood cells, we are designing a clinical study on CLL patients aimed to demonstrate the potential use of the molecule in the adjuvant chemotherapy against CLL.

S-15

Development Of Novel Targeted Therapies For Solid Tumors

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S-16

MER RECEPTOR TYROSINE KINASE AND MACROPHAGE PHAGOCYTOSIS OF APOPTOTIC CELLS

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One of the hallmark changes associated with apoptosis is the exposure of anionic phospholipids such as phosphatidylserine (PtdSer) and phosphatidylethanolamine on the outer leaflet of the plasma membrane. Phagocytic cells express a variety of receptors that confer the capacity for recognition of this membrane alteration, including BAI-1 and Tim4. In addition, phagocytes express receptors that interact with PtdSer-binding molecules that act as a bridge between the phagocyte and the apoptotic cell. Mer (gene name Mertk) is a receptor tyrosine kinase expressed by a variety of different cell types, including macrophages derived from most tissues. Mer serves as a receptor for Protein S and Gas6, PtdSer binding proteins that rapidly opsonise apoptotic cells. Mer can mediate tethering of Protein S-opsonised apoptotic cells and activation of the intrinsic kinase activity of Mer signals apoptotic cell internalisation. In addition, TLR-dependent pro-inflammatory cytokine production by macrophages is influenced by Mer-dependent signalling. We have examined Mer expression during human monocyte differentiation to macrophages *in vitro* and explore the relationship between monocyte expression of Mer and the capacity to bind the Mer ligand, Protein S. Together, our new data reveal a novel role for Mer in the control of monocyte function.

S-17
Unequal Cell Death in Differentiating T helper Cells

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T lymphocytes play a pivotal role in the immune response as a key regulatory and effector cells. Upon antigenic stimulation, naive CD4 T cells can active, proliferate and differentiate into unique signature cytokine expressing effector T helper (Th) cells for instance Th1, Th2 and Th17. Recently discovered Th17 cells are known to play a critical role in various inflammatory pathologies including Multiple sclerosis (MS), Rheumatoid arthritis (RA) and cancer. Th1 cells are involved in cell-mediated immune response and are responsible for the clearance of intracellular pathogens. Th2 cells are involved in humoral immune responses, allergies and responds to some parasites. They secrete IL-4, IL-13, IL-10 and IL-5. Th17 cells are linked to several autoimmune diseases and mediate immune response to bacterial and fungal infections. They secrete IL-17, IL-21 and IL-22 cytokines.
Apoptosis is a process that involves sequential activation of a series of proteins such as caspases after an appropriate stimulation. Apoptosis can be carried out by external (receptor-ligand interaction) or internal (mitochondria) mediated pathways. Fas, FasL, DR5 and TRAIL are apoptotic proteins whereas FLIP and Bcl-xL are anti-apoptotic proteins, which involve in apoptotic pathways. It is known in the literature that Activation Induced Cell Death (AICD) is important in eliminating activated T cells. The Fas/FasL pathway is very important in T cell death. TCR signaling can lead to the deletion of activated peripheral T cells through apoptosis. The survival or death of effector T cells is affected on IFNγ, IL-4 and IL-2.
T helper cells show different susceptibility to apoptosis. For instance, Th17 cells are less sensitive than Th1 cells to Fas mediated apoptosis because they have a higher expression of FLIP. Th17 are more sensitive to Fas mediated apoptosis than Th2 cells due to their higher expression of FasL. Re-stimulated Th17 cells undergo AICD through a Fas/FasL mediated pathway that is unaffected by IFNγ. Human Th17 cells are phenotypically resembles differentiated memory T cells but they are distinct from central memory, exhausted and senescent T cells. Human Th17 cells are long lived cells. Overall data showed that there is an unequal cell death in differentiating T helper subsets. The further investigation of cell survival and death signal networks in T helper cells will help us better understand the molecular mechanisms of various pathologies such as autoimmunity, cancer and infection diseases. This work was supported by a grant from the Scientific and Technological Research Council of Turkey (TUBITAK). Project grant number is 110T412 and awarded to Dr. Ayten Nalbant.
Key words: T cells, T helpers, Th1, Th2, Th17, AICD and apoptosis

S-18

Gabriel LOPEZ-BERESTEIN

S-19

APOPTOSIS AS A POWERFUL MEANS OF IMMUNE MODULATION FOR THE TREATMENT OF TYPE 1 DIABETES

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Type 1 diabetes (T1D) is a debilitating autoimmune disease affecting a significant portion of population worldwide. Insulin treatment as standard of care is often ineffective in preventing recurrent hyperglycemic episodes with long-term undesired adverse effects. Transplantation of pancreatic islets as a source of beta cells producing insulin has proven effective in improving metabolic control/quality of life and preventing severe hypoglycemia in patients with T1D. However, islet transplantation suffer from immune rejection irrespective of chronic use of immunosuppression and its complications. Both autoimmunity and islet graft rejection are initiated and perpetuated by an imbalance between pathogenic T effector (Teff) and protective T regulatory (Treg) cells. Teff cells upregulate Fas receptor on their surface following activation and become sensitive to Fas/FasL-mediated apoptosis. Therefore, Fas-mediated apoptosis has a great potential to specifically purge out auto and allo-reactive pathogenic Teff cells for the prevention and treatment of T1D. We have generated a novel form of FasL protein with improved apoptotic activity and demonstrated its utility for the induction of tolerance to auto and allogeneic antigens for the treatment of T1D in various rodent models. Tolerance was initiated by a regulatory circuit involving apoptosis of Teff cells, phagocytosis, secretion of TGFβ, and generation/expansion of T regulatory cells. T regulatory cells were not only important in tolerance induction but also maintenance. This concept is presently being pursued for clinical development.

S-20

ABNORMAL BRAIN GANGLIOSIDE ACCUMULATION TRIGGERS APOPTOSIS IN EARLY ONSET TAY - SACHS DISEASE MOUSE MODEL

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Tay-Sachs disease is a severe lysosomal storage disorder caused by mutations in the HEXA gene coding for α subunit of lysosomal β-Hexosaminidase A enzyme, which converts GM2 to GM3 ganglioside. Unexpectedly, the $HexA^{-/-}$ mice have a normal lifespan and show no obvious neurological impairments until at least 1 year of age, owing to the ability of these mice to catabolize stored GM2 ganglioside via sialidase(s) removing sialic acid into glycolipid GA2 which further processed by β-Hexosaminidase B, thereby bypassing the HexA defect. To elucidate whether sialidase Neu3 can contribute to GM2 ganglioside degradation, we generated mice model with combined deficiencies of β-Hexosaminidase A and Sialidase Neu3. $HexA^{-/-}Neu3^{-/-}$ mice are healthy at birth but died at 1.5-4.5 months of age. Thin layer chromatography and mass spectrometry analysis of brain from $HexA^{-/-}Neu3^{-/-}$ mice showed abnormally accumulated GM2 ganglioside level. Slow movement, ataxia and tremor are among neurological abnormalities. In the current study, we delineate whether there is apoptosis in Tay-Sachs mice model's brain. In order to profile the expression of 84 key genes related to apoptosis in the cerebellum and cortex from 4.5 months old mice, we used RT2 Profiler PCR Array system specific for apoptosis. We found that mRNA levels of pro-apoptotic genes such as TNF and caspase 4 increased 5.8X and 2.1X respectively in $HexA^{-/-}Neu3^{-/-}$. On the other hand, mRNA levels of anti-apoptotic genes such as Bag1 and Bcl2 (1.6 X), Cd40lg (1.8X) and Naip (7.1X) decreased in cerebellum of the same mice as compared to $HexA^{-/-}$ mice revealing the apoptosis in early-onset Tay-Sachs mice model. We suggest that once a critical threshold of GM2 ganglioside storage is reached in the cerebellum, a signaling cascade is triggered which activates neuronal death.

➢ ORAL PRESENTATIONS

OP-1

ANDROGEN RESPONSE IS REQUIRED FOR DNA DAMAGE RESPONSE IN OXIDATIVE STRESS CONDITIONS

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Aim: As it has been shown that inflammatory microenvironment leads to the proteosomal degradation of androgen receptor (AR), prostatic inflammation was associated with development of carcinoma in previous studies. In addition, androgen signaling also contributes to distinct cellular mechanisms including antioxidant response regulation. To investigate the role of androgens in oxidative stress conditions in prostate cells oxidative DNA damage and subsequent damage response was examined. Material and
Method: We used a model of inflammatory microenvironment to mimic the chronic inflammation and/or oxidative stress conditions by treating prostate cells with inflammatory conditioned media (CM) with known cytokine concentrations or H2O2. Then, the cellular responses on the ROS generation and DNA damage response was investigated.

Results: We demonstrated that the treatment of the prostate cells with oxidative stress inducers (menadione and H2O2) resulted with increased formation of reactive oxygen species (ROS) and oxidative DNA damage upon AR depletion. Consistently, while DNA damage (H2AX and AP sites) was increased after AR silencing, DNA damage recognition was disrupted that was shown by decreased ATM phosphorylations in these cells suggesting that AR signaling is required for DNA damage response in prostate cancer cell lines. Furthermore, androgen-independent LNCaP 104r2 cells were found to be more tolerant to oxidative stress with higher ROS levels and activated SIRT1 expression resulting reduced oxidative DNA damage.

Conclusions: Thus, loss of AR function results in increased DNA damage upon abrogated DNA damage response mechanism. In addition, androgens has a significant role in controlling oxidative stress state and further damage response cascade of the cell. Therefore, though androgen ablation is known as the main therapeutic approach for prostate cancer cure, loss of androgen signaling seems to induce further oxidative stress resistance and DNA damage generation by promoting tumorigenic alterations in prostate cells.

Keywords: Androgen receptor, oxidative stress, DNA damage, γH2AX, prostate cancer

OP-2

NEUROPROTECTIVE ROLE OF CURCUMIN ON CNS DAMAGE IN OFFSPRING OF PREGNANT RATS EXPOSED TO AROCLOR 1254

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In this study, it is aimed to identify the damages to the central nervous system in offspring of rats exposed to Aroclor 1254, toxic Polychlorinated biphenyls (PCBs), during critical peri-od of pregnancy and, invivo and invitro, to reveal out neuroprotective effect of curcumin, a powerful antioxidant, on the the damages. In the study, 3 groups were constituted and 2 fe-males and 1 male Wistar rats were mated in each groups. Newborn pups (n = 10) were ran-domly selected from each grups. The first (control) group was allocated without any pro-cessing to pups. The pregnant rats of second group were exposed to Aroclor 1254 with 1 mg/kg/BW dose during 7th to 21st days of gestation. The pregnant rats of third group were exposed to dose of Aroclor 1254 within same days and dose. The rat pups in last group were also orally treated with curcumin dissolved in 50% DMSO with 200 mg/kg/BW dose during the first postnatal three days. The damages were compared histopathologically in the last two groups. 8-OHdG, 4HNE the MBP expressions and DNA in situ fragmentation positivities (by TUNEL method) were immunohistochemically evaluated and decided how curcumin to de-creased at those 8OHdG and 4HNE activities and increased at MBP activity. Rat F98 glioma line were applied with Aroclor 1254 and curcumin separately and together. MTT, LDH and cytotoxicity test were performed and evaluated statistically. Especially 8OHdG and TUNEL reactions gave similar results in prosenchephalon and mesencephalon. Invitro studies show that curcumin reduce on cytotoxicity and DNA damage in cells due to oxidative stress by Aroclor 1254 on glioma cells. In conclusion, curcumin has role in reducing oxidative stress and returning effects of DNA damage processed by Aroclor 1254. It is thought on neuroprotective effect of curcumin that its higher doses may be more effective over the entire CNS.

Keywords: offspring rat, Aroclor 1254, curcumin, CNS, invivo and invitro

OP-3

PROTECTIVE EFFECTS OF DIFFERENT ANTIOXIDANTS AGAINST THE TOXICITY OF 3,5-DIMETHYLAMINOPHENOL IN HUMAN BLADDER CELL LINE

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Monocyclic aromatic amines (MAAs) are widespread environmental contaminants with multiple sources. Exposure to MAAs mainly cause toxic effects on bladder and increase in bladder cancer incidence. Their main oxidative metabolites are their o- or p-phenol derivatives. As a leading example, the phenolic derivative of 3,5-dimethylaniline, namely 3,5-dimethylaminophenol (3,5-DMAP), is readily oxidized to the quinone imine following administration and mammalian cells treated with this compound exhibit elevated levels of reactive oxygen species (ROS) for several days following exposure. The main aim of this study was to investigate whether this compound caused an oxidant/antioxidant imbalance in human bladder cells (UROtsa cells). Furhermore, we investigated whether this compound causes DNA damage and increases in the caspases (caspase 3 and 8), which are main enzymes that play essential role in apoptosis. Protective effects of organic (selenomethionine) and inorganic (sodium selenite) selenocompounds, ascorbic acid and N-acetylcysteine (NAC) against its toxicity was also assessed. We observed that at inhibitory concentration (IC50) of 3,5-DMAP was approximately 50 µM. Besides, 3,5-DMAP caused a dose dependent increase of ROS. In both cytoplasm and nucleus, 3,5-DMAP caused alterations in the antioxidant enzyme activities and decreases cellular redox ratio. Lipid peroxidation and as a consequence protein oxidation were elevated in particularly cytoplasm. Single strand DNA breaks were also induced by 3,5-DMAP; however no changes were observed in double strand DNA breaks. Moreover, apoptosis was also triggered by 3,5-DMAP exposure as evidenced by increases in caspase 3 and caspase 8 activities. Selenocompounds, ascorbic acid and NAC were found to be both partially protective against the cellular toxicity of 3,5-DMAP.

Keywords: 3,5-dimethylaminophenol, alkylaniline, reactive oxygen species, cyctotoxicity, apoptosis

OP-4

REDUCTION OF LIVER CIRRHOSIS BY TGF-BETA TYPE 1 RECEPTOR KINASE INHIBITOR, LY-364947

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Object: Cirrhosis is a common and consequence of chronic liver disease that characterized by the diffuse fibrosis, scar tissue and regenerative nodules. Transforming growth factor (TGF) beta type 1 is one of the major profibrogenic mediators which takes part in the development of liver cirrhosis. The balance between cell proliferation and apoptosis is very important in the formation of liver cirrhosis. In this study, we aimed to determine the apoptotic and antioxidant mechanisms in liver cirrhosis and the effect of the TGF-β type I receptor kinase inhibitor, LY-364947 on liver cirrhosis.

Material and Method: 6 groups were included as Control, Cirrhosis,Cirrhosis + DMSO, Cirrhosis + LY-364947(100µg/kg/week),Cirrhosis + LY- 364947(300µg/kg/week), after Cirrhosis + LY-364947(100µg/kg/week). Experimental liver cirrhosis was developed by N-Nitrosodiethylamine. Control, Cirrhosis, Cirrhosis + DMSO, Cirrhosis + LY- 364947(100µg/kg/week), and Cirrhosis + LY- 364947 (300µg/kg/week) groups were killed when cirrhosis was established by evaluation of histopathological investigations in rats which were chosen randomly from the Cirrhosis group. After cirrhosis was established LY-364947 was administered for 4 weeks to rats Cirrhosis+ LY- 364947 (100µg/kg/week) group. Apoptosis was evaluated by measurement of caspase -3,-8 and -9. Antioxidant capacity and lipid peroxidation were evaluated by measuring the levels of reduced glutathione (GSH) and malondialdehyde (MDA), respectively. The evaluations were carried out by colorimetric methods according to the assay instructions. Histopathological investigations were also carried out.

Results: Caspase -3,-8 and -9 enzyme activities and also increased lipid peroxidation were significantly suppressed by LY- 364947(300µg/kg/week). Histopathological findings indicated that cirrhosis development was reduced by LY- 364947(300µg/kg/week) whereas administration of LY- 364947(100µg/kg/week) after cirrhosis was failed to point out the same affect. GSH depletion was observed in all groups compared with control.

Conclusion: LY- 364947 may be considered as a promising antifibrotic agent in liver cirrhosis by suppressing apoptosis and lipid peroxidation and reducing liver cirrhosis.

Keywords: liver cirrhosis, LY- 364947,TGF-β type I, caspase, apoptosis

OP-5

THE ABILITY OF THYMOQUINONE TO INDUCE CASPASE-INDEPENDENT CELL DEATH IN DIFFUSE LARGE B CELL LYMPHOMA IS MEDIATED BY INTRACELLULAR CALCIUM RELEASE

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Introduction: Disruption of the apoptotic pathways in diffuse large B cell lymphoma (DLBCL) remains a challenge for standard treatment and results in refractoriness and poor prognosis. Thymoquinone (TQ), an active compound from a medicinal plant Nigella sativa, has been reported to kills cancerous cells through different cell death pathways. Nevertheless, the molecular mechanisms underpinning the interplay between different cell death modalities are still unclear. Herein, we explored the in vitro anticancer activity of TQ against DLBCL and trend to understand the mechanisms of its anticancer activity.

Results: TQ greatly inhibited the proliferation and induced cell death of DLBCL cell lines and primary refractory DLBCL cells with a minimal effect on normal lymphocytes. Molecular investigations revealed that TQ activates the UPR pathway in highly responsive cell lines, identified through sXBP-1, GRP78, CHOP and HSPA1A up-regulation. In parallel, TQ activated the mitochondrial pathway of apoptosis revealed by caspase-3 and -9 activation consecutive to cytochrome c release. However the mitochondrial events don't played a critical role in highly responsive cells, in fact, caspase inhibition by z-VAD-fmk failed to rescue them from TQ-induced cell death, while this was partially abrogated in the lowly responsive one. In contrast, cytosolic calcium chelation by BAPTA-AM strongly repressed TQ cell death effect in highly responsive cell lines. Indeed [Ca2+]c measurement showed that TQ leads to an acute [Ca2+]c rise in highly responsive cells with a moderate rise in the less responsive one. The observed [Ca2+]c rise was derived mainly from the ER calcium stores and mediated by IP3 receptors, in fact their inhibition by 2-APB strongly prevented TQ-induced cell death.

Conclusion: This work demonstrates requirement [Ca2+]c rise in the ability of TQ to induce non-apoptotic cell death, which may offer an effective strategy for antilymphoma therapy in DLBCL intrinsically resistant or refractory to chemotherapy induced apoptotic cell death.

Keywords: TQ: Thymoquinone ER: endoplasmic reticulumUPR: unfolded protein response[Ca2+]c : cytosolic calcium sXBP1 : spliced XBP1

OP-6

MOLECULAR SIGNIFICANCE OF AUTOPHAGY IN GAUCHER DISEASE

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Autophagy is a lysosomal-dependent catabolic pathway contributing to cellular homeostasis by sequestering cytosolic macromolecules in double or multimembrane vesicles and deliver them to lysosomes for degradation. Gaucher disease is the most frequent lysosomal storage disorder (LSD) caused by deficiency of acid-β glucosidase and is characterized by the accumulation of glucosylceramide or other gycolipids in visceral organs or central nervous system. Although the relevance of autophagy is shown in different LSDs, the underlying molecular mechanism in Gaucher disease is poorly understood. Here, we investigated molecular significance of autophagic pathway in fibroblasts cells obtained from Gaucher patients homozygous for L296V mutation, as well as for the most common mutations, N370S, L444P, and D409H. We observed significant attenuation in the expression of key autophagy-related genes (BECN1, ATG5 and LC3) and accumulation of their proteins in mutant cells. We found that decrease ability of autophagosomes to fuse with lysosomes is associated with elevated lysosomal pH and reduced lysosomal enzyme activity. Analysis of proteasomal degradation machinery showed decreased proteolytic activity of proteasome, which consequently leads to increased susceptibility to cell death. Our data indicate that both autophagic pathway and ubiquitin-proteasome system are affected by mulfunctional lysosomes and may underlie the mechanism of clinical severity of Gaucher patients. (This project is supported by TUBITAK-3501-National Young Researchers Carreer Development Program, Project No: 112T130).

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Keywords: Autophagy, gaucher disease, mutant fibroblasts

OP-7

PROTEASOME INHIBITORS TRIGGER NEUROPATHY BY DESTABILIZING CYTOSKELETON

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The proteasomal system controls different cellular processes via turnover of many crucial cellular proteins. Therefore, proteasomal system is the target of treatment strategies for several diseases, including cancer. Since the approval of clinical usage of the first proteasome inhibitor, bortezomib, by FDA, proteasome inhibitors have been used in the clinic for the treatment of different cancers. As many other chemotherapeutic agents, peripheral neuropathy is one of the most abundant side effects of proteasome inhibitors and main reason for treatment limitations. Aim of the present work was tracing the reasons of peripheral neuropathy triggered by proteasome inhibitor bortezomib. With this purpose in mind, we also compared bortezomib with a second generation proteasome inhibitor carfilzomib, which is known to be less toxic for neuron cells. As a beginning step, we checked the inhibitory effects of drugs on proteasome activity. Bortezomib was 5-fold more effective on inhibiting 20S proteasome after 3 h treatment, but then inhibitory effects of drugs were similar. Then we performed proteomic analysis with nanoLC-MS/MS, and our data showed significant changes in cytoskeleton members, especially in actin filament polymerization, vimentin and nestin expression, and also in HSP expression. We performed confocal imaging which also showed significant change in actin filament polymerization after 3 h drug treatment, and microtubule stabilization after 24 h treatment. Western blotting analysis showed that, while ARP2 expression increased about 2-fold after 3 h treatment in both groups, coronin 1C expression increased only in bortezomib group (1.2 fold), and transgelin expression change was greater in carfilzomib treated group after 3 h treatment. Coimmunoprecipitation of HSP70-β-actin (both directions) showed that β-actin monomers interacted with HSP70 chaperons after proteasome inhibitor treatment, and this interaction was higher in bortezomib treated group. We can conclude that, the less toxic effect of carfilzomib may be related to actin polymerization stability and potential.

Keywords: bortezomib, carfilzomib, neuropathy

OP-8

LONG NON-CODING RNAS: POTENTIAL REGULATORY PLAYERS OF APOPTOSIS

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Apoptosis is a type of Programmed Cell Death (PCD) which is essential for cellular homeostasis and proper development. Diseases like autoimmune diseases and cancer are associated with aberrant apoptosis. Despite the well-known role of certain proteins and microRNAs in apoptosis, the potential regulatory role of long non-coding RNAs (lncRNAs) is still unclear. In this study, we used two cancer therapeutics drugs, cisplatin and doxorubicin, and two ligands, Fas mAb and TNFalpha, to identify pathway-drug specific and/or global lncRNAs that are differentially expressed in apoptotic HeLa cells. Induction of apoptosis was measured by Flow Cytometry and was further verified by Fluorescence Microscopy and western blotting. Three replicates of total RNAs were deep-sequenced using the Illumina platform. Treatment with cisplatin, doxorubicin, anti-Fas and TNF-alpha led to the differential expression of 1644, 506, 584 and 807, respectively. Total number of differentially expressed lncRNAs that are common for all agents was ~250 (2-fold or higher, $P < 0.01$). Interestingly, a fraction of dysregulated lncRNAs appear to be antisense/sense to or located in the close proximity to protein-coding genes or miRNAs known to be key regulators of apoptosis. Four candidates were selected for validation by qPCR. GapmeR-mediated silencing of the candidates showed a relationship between apoptosis and candidates. These results indicate that many lncRNAs are differentially expressed upon induction of cell death via treatment with cisplatin, doxorubicin, anti-Fas and TNF-alpha in HeLa cells under our experimental design. Mechanistic and further functional characterization of candidate lncRNAs may give an important insight into apoptosis at the molecular level.

Keywords: apoptosis, long non-coding RNA, deep-sequencing

OP-9

HYPOXIA INDUCED APOPTOSIS IN MOUSE NEUROBLASTOMA CELL LINE

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Object: Neuroblastoma (NB) is the most common extracranial pediatric solid tumor. Hypoxia is a well-known feature of solid tumors and is a key prognostic factor for tumor progression and poor clinical outcome. Apoptosis is a progress of programmed cell death that occurs in response to distinct signals such as hypoxia, excessive oncogene activation or chemotherapeutic agents. In this study we aimed to investigate the hypoxic effect on apoptosis in neuroblastoma cell line.

Methods: The mouse neuroblastoma cell line (NA2B) was cultured in DMEM F-12 supplemented with 10% fetal calf serum and 1% L-glutamine. Cells were cultured in 24 well plate and divided into two groups. For hypoxic condition group; cells were exposed to 3% O2, 92% N2, 5% CO2 gas mixture to create 3% hypoxic condition for 36 hours in hypoxia chamber. For control group; cells were incubated under normal culture conditions in humidified atmosphere at 37°C in 5%CO2. After 36 hours incubation cells fixed in 4% paraformaldehyde and were stained with indirect immunoperoxidase technique in order to determine distributions of cytochrome-c, bcl-2, Bax and caspase-3. DNA fragmentation was detected by terminal deoxynucleotidyltransferase-biotin nick end-labelling (TUNEL) method. Results evaluated with the One-Way ANOVA statistical test. Results: In hypoxic group, very strong; caspase-3, strong; Bax and cytochrome-c and mild to moderate; Bcl-2 immunoreactivities were detected. Immunoreactivities of caspase-3, cytochrome-c, Bax and Bcl-2 were less in control group. TUNEL positive stained cells were more in hypoxic group when compare to control group.

Conclusion: Hypoxia is an important environmental factor that regulates cell differentiation, apoptosis and survival. In our study we demonstrated that hypoxia can induce apoptosis via the caspase activation accompanying cytochrome c release with the increase of proapoptotic molecule Bax. High malignancy potential of NB can be correlated with resistance to apoptosis and hypoxia can be used to trigger the apoptosis of the NB.

Keywords: Apoptosis, hypoxia, neuroblastoma

OP-10

PROTEASOMAL SYSTEM AND HEAT SHOCK PROTEINS IN CANCER CELL DEATH MECHANISMS

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Proteasomal degradation is crucial to prevent the accumulation of cellular damage. The removal of the damage is a required process for healthy organisms to keep the integrity while in cancer cells this situation may induce drug resistance. Regarding chemotherapy, degradation mechanisms such as proteasomal system and autophagy have been focused recently and proteasomal inhibition in cancer cells have been shown to induce autophagy. This induced pathway may prevent the cancer cells from death or can cause autophagic cell death. There are many preclinic studies to improve the results and on the other hand heat shock proteins are accepted to be protective which may bring new approach. The object of the studies performed in our laboratory was to confirm the role of proteasomal system and heat shock response in cancer cell death. In this direction, several cancer cell lines have been tested from different aspects of proteasomal activity. MCF7 and MDA-MB-231 breast cancer cells were treated with proteasome inhibitors bortezomib and carfilzomib in different concentrations. HCT116 colon cancer cell line and HT22 hippocampal tumor cell line have been used to confirm the role of heat shock response related to proteasomal degradation. Results showed that following the treatment of breast cancer cells with bortezomib and carfilzomib, proteasome inhibition induced the autophagic protein degradation and apoptotic cell death. In this model, autophagic protein degradation has been found to be a resistance against apoptotic cell death. In HCT116 cell line, heat treatment induced HSP70 protein expression which caused increased proteasomal degradation and autophagic protein degradation and decreased apoptosis. Additionally, HT22 cells showed the same results in direction with the other cell lines. In these cells, Nrf2 pathway has been found to be involved in the activation of proteasome activation and heat shock response. We can conclude that the activation of proteasome activity and regulation of the proteasomal system by heat shock proteins should be targeted in detail to highlight the chemotherapy resistance.

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Keywords: proteasome, heat shock response, cancer, chemotherapy, resistance

OP-11

THE ROLE OF HYPOXIA ON APOPTOTIC MARKERS IN PRIMARY AND METASTATIC CANCER CELL LINES

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Aim: Cancer microenvironment is characterized by significantly lower oxygen concentration. Hypoxic condition is a common feature of solid tumor, and is a direct stress that triggers apoptosis in many human cell types and promote cell survival, motility and tumor angiogenesis that is essential to tumor. In this study, we aimed to investigate the effect of hypoxia on apoptotic markers in primary and metastatic cancer cell lines.

Materials and Methods: Human primary colon (Colo-320) and breast (Mcf-7), human metastatic colon (Colo-741) and breast (M4A4) cancer cell line were used. They were cultured in RPMI-1640 media supplemented with 10% FBS, 1% L-glutamine, 1% penicillin and streptomycin until 80% confluency. All type of cells divided into two groups; control group cultured with 5% CO2 and 37°C, hypoxia group were cultured in hypoxia chamber which have a gas mixture of 5% CO2, 3% O2 and 92% N2 to provide hypoxic condition for 36 h. After fixation with 4% paraformaldehyde, apoptotic cells were determined by TUNEL assay, distribution of Caspase-3, Bax, Bcl-2 and Cytochrome-c were analyzed using indirect immunohistochemistry method. Immunostaning results were evaluated as H-SCORE in comparison with the One-Way ANOVA statistical test.

Results: The number of TUNEL positive cells in Colo320 under hypoxic condition was higher than other hypoxic and other groups. The strong immunoreactivity of Caspase-3 was detected especially was detected hypoxic conditions of Colo320 cells. The ratio of Bcl-2/Bax was higher in primary cancer cells lines (Colo320 and Mcf-7). The moderate/strong immunoreactivity of Cytochrome-c was detected in all groups.

Conclusion: Hypoxia is important in colon cancer occurrence and progression. Hypoxia condition promotes apoptosis and inhibits proliferation in various cancer cells in vitro. Our results showed that hypoxia triggered apoptotic pathway especially in primary colon cancer cell line however both colon and breast metastatic cancer cell lines were rescued from hypoxia induced apoptosis.

Keywords: hypoxia, primary, metastatic, breast, colon, cancer, apoptosis

OP-12

HL-60 CELL LINE GENERATED IN VITRO ENVIRONMENT ON ANALYSIS OF THE BIOCHEMICAL EFFECTS OF SORAFENIB AND LITHIUM CHLORIDE

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Acute leukemia is characterisized with decreasing mature cells and aggregation of leucocyte precursors. In this study we aimed to investigate the effects of Sorafenib and Lithium Chloride (LiCl) on HL-60 human Acute Promyelocytic Leukemia (APL) cell line. The HL-60 APL cell cultures are subjected to Sorafenib and LiCl separately and in combination for 72 hours during experiments. Tumor cells are counted under Cell Count Hemocytometer and the apoptose ratios are investigated with flow cytometry method. The changes in the cells in cell cultures are evaulated by transmission electron microscopy. To determine the possible the mechanism of action of the drugs, the caspase 3 actvities, phosphorylated GSK-3β, phosphorylated AKT, total AKT, phosphorylated p38, total p38, phosphorylated ERK, total ERK, phosphorylated IκBα, total IκBα , phosphorylated c-jun, total c-jun, phosphorylated STAT3 ve total STAT3 levels are investigated with ELISA method. Separately and in combination Sorafenib and LiCl decreased the number of cells and increased the apoptotic ratio in HL-60 APL cell line in a statistically significant manner when compared with the control group.

Keywords: HL-60, Sorafenib, Lithium Chloride, Apoptosis, Leukemia

OP-13

REGULATION OF MIRNA ON COLON CANCER STEM CELLS

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Colorectal cancer is one of the most commonly diagnosed and lethal cancers worldwide. It is a multistep process that requires the accumulation of genetic/epigenetic aberrations. There are several issues concerning colorectal carcinogenesis that remain unanswered, such as the cell of origin and the type of cells that propagate the tumor after its initiation. Cancer stem cells (CSCs) have similar properties with normal stem cells but, they grow rapidly and differentiate to tumor cells. CSCs therefore seem to have an important role in cancer recurrence. The biological behavior of cancer, including carcinogenesis and functional heterogeneity, can be explained by the CSC hypothesis. The regulation of CSCs at the molecular level is not well-understood. MicroRNAs (miRNAs) are a class of endogenous non-coding RNAs that play an important role in the regulation of several cellular, physiological, and developmental processes. Stem cell-specific miRNAs play important roles in tumor initiation and development. Regulation of miRNA expression is controlling with several proteins.

In this study, the human primary colon carcinoma cell line, Colo 320, RPMI-1640 cultured in 10% FCS, 1% L-glutamine and 1% penicilin-streptomycine containing culture medium. Colon carcinoma stem cells characterized by CD133 surface protein were isolated from primer (Colo 320) colon carcinoma cell line by magnetic-activated cell sorting (MACS) technique. Magnetically labeled CD133+ and unlabeled CD133− cells were separated magnetically. CD133+ and CD133− cells were passaged after reaching 80% monolayer confluency. They were then fixed with 4% paraformaldyde and distribution of anti-cyclin D1, anti-c-myc, anti-beta-catenin, anti-dicer, anti-drosha, anti-eIF2α and anti- eIF2C were investigated using indirect immunoperoxidase staining. Results show that the efficiency of the separations is 30%, and the separation is more successful by MiniMACS column. Strong immunreactivity of cyclin D1 was observed in primer colon carcinoma CD133+ and CD133- cells. Dicer, drosha, cyclin D1, c-myc, beta-catenin immunoreactivities were less in CD133+ cells. However, higher immunoreactivities of eIF2α and eIF2C were detected in CD133+ cells. As a result CD133+ cells have higher expression of eIF2α and eIF2C therefore miRNA expressions may control with these molecules in CSCs. In addition, both CSCs and unlabeled Colo 320 cells have similar cell cycle controlling. Because of differences of regulation of miRNA expression between CD133+ and CD133- cells, treatment protocol should be different in tumors, which have cancer stem cells.

Keywords: Cancer stem cell, colon, cell cycle, miRNA

OP-14

THE USE OF KERATINOCYTES AND PRP IN EXPERIMENTAL BURN WOUND MODEL OF DIABETIC RAT

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Object: Diabetes mellitus (DM) is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. DM affects all three phases of wound healing (inflammation, proliferation and remodeling), leading to delayed closure and poor esthetic and functional scarring. A keratinocyte is the predominant cell type in the epidermis. The primary function of keratinocytes is the formation of a barrier against environmental damage. Besides, platelet rich plasma represent a key source of cytokines and growth factors extensively used for tissue regeneration; wound healing and tissue repair.In this study we aimed to investigate the effects of keratinocytes which was differentiated from rat adipose stem cells and PRP in experimental burn wound model of diabetic rat (1,2,3).

Material and Method: Male Sprague–Dawley rats were injected intraperitoneally with 60 mg / kg streptozocin and after 4 weeks DM status was defined as blood glucose levels higher than 280 mg/dl. Rat ADSCs were isolated from retroperitoneal adipose tissue and washed with 5% penicillin-streptomycin containing phosphate buffered saline (PBS). Briefly, they were minced into small pieces, treated with 0.075% collagenase with α-MEM (includes 15% FCS, 1% L-glutamin, 1% penicillin-streptomycin) and incubated at 37 0C, 5% CO2 for 30 minutes. The cells were cultured until 80% confluency and then were differentiated to keratinocyte using 0.5 nM bone morphogenetic protein-4 on matrigel coated culture plate. Wound healing rat model was obtained with 100 °C heated 7 mm diameter copper that applied to the back of rats for 45 seconds. They were then separated as untreated, keratinocyte, PRP, and keratinocyte+PRP applied groups. After transferring the keratinocytes, tissue samples were collected on day 3rd, 7th, 10th and 14th. They were fixed with 10% formalin solution. Routine paraffin embedding processes were done. Sections were stained both H-E and indirect-immunohistochemical protocols used for analysing anti-EGF, anti-FGF2, anti-VEGF, anti-MCP1, anti-collagen1, anti-cytokeratin8, anti-cytokeratin14, anti-TGFβ-1, anti-PDGF immunoreactivity.

Results: Cells were positively stained with early and late Keratinocyte differentiation marker cytokeratin-8 and cytokeratin-14, respectively. Cytokeratin-8 immunoreactivity was found more positive than cytokeratin-14. Histochemical analyses showed that, ADSC derived keratinocyte accelerate the epithelialization and improve wound healing, but keratinocyte + PRP treatment were presented greatest improvement. In addition, molecular markers for progression of wound healing were more detectable in this group according to others.

Conclusion: Although applying ADSC derived keratinocytes trigger the healing, best development seen when keratinocytes used with PRP. This study is a basic attempt to understand the physiology of keratinocytes and PRP before using them for the further studies.

Keywords: wound healing, stem cells, keratinocyte, diabetes mellitus

OP-15

PROTECTIVE EFFECTS OF ADIPOSE-DERIVED MESENCHYMAL STEM CELLS ON OVARIAN ISCHEMIA-REPERFUSION INJURY IN RATS

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Object: We aimed to investigate the immunohistological and biochemical effects of Adipose Derived Stem Cells (ADSC) on ovarian ischemia-reperfusion (I/R) model in rats.

Material and Method: Twenty-five female, virgin adult Wistar albino rats were divided into four groups as Group 1 (Sham), Group 2 (ADSC), Group 3 (Bilateral torsion+detorsion), Group 4 (Bilateral torsion+detorsion+ADSC). Laparotomies were applied to all groups, adnexal detorsion was applied to Groups 3 and 4 for 6 h period. After 6-h period of torsion/detorsion procedures, bilateral ovaries were exposed and 1x105 cell/mL. ADSC were seperately administered with a microinjector in a volume of 20 µl PBS media in Group 4. After 6 hour period over laparatomy, ADSC were administered to bilateral ovaries in Group 2. Laparotomies were performed in all animals after a 7-day recovery period and bilateral ovaries were removed and blood was taken from inferior vena cava. For immunohistological analysis, TUNEL, iNOS, Caspase-3 and Brdu staining were determined in experimental groups. Advanced protein oxidation protein products (AOPP) and total sulfhydryl (TSH) levels and superoxide dismutase (SOD) activity were measured in serum and ovarian tissue.

Results: Serum AOPP levels as protein oxidation marker were significantly increased in Group 3 when compared to Group 1 and Group 2 ($p<0.05$), whereas tissue and serum AOPP levels in Group 4 were significantly decreased compared to Group 3 ($p<0.05$). Tissue SOD activity in Group 3 and 4 were significantly decreased compared to sham groups ($p<0.05$). iNOS was more strengthened via torsion and detorsion in Group 3, but diminished after ADSC implementation in Group 4. ADSC treatment after torsion-detorsion have led to a decrease in apoptotic cell count and Caspase-3 immunoreactivity. These changes were all consistent with iNOS immunoreactivity and TUNEL staining.

Conclusion: We concluded that ADSC might have protective effects on ovarian I/R model.

Keywords: Key Words: Adipose Derived Stem Cell, Ischemia, Ovarian, Rat, Torsion

OP-16

INVESTIGATION OF ANTICARCINOGENIC EFFECT OF POLYPHENOL COMBINATIONS AS A DRUG ON MCF-7 AND MCF-10A CELL LINE

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Object: Recently, it has been showed that flavonoids have anticarcinogenic features. In our previous researches, we detected the cell viability and cytotoxic effect of more than 30 flavonoids and phenolic acids in breast cancer. In present study, we aimed that investigating the interactions and synergistic effects of some flavonoids and phenolic acids upon MCF-7 breast cancer and MCF-10A normal epithelial cell lines.

Material and Methods: For this purpose, we designed artificial drug cocktails based on these IC50 values of them, which contains these substances such as several flavonoid cocktails. Then, we performed the cell proliferation assay by using WST-1 reagent, analysis of apoptosis by using Annexin V-PI assay and cell cycle assay by using Muse Cell Analyzer. After determining the most effective doses of drug cocktail, we performed the whole genome microarray analysis and real-time PCR experiment carried out to validate the microarray data.

Results: In consequence of microarray data, %75 of flavonoid cocktail is the most effective dose to MCF-7 and MCF-10A cell lines. In MCF-7 breast cancer cell line, this dose lead to more than 60 cell division related genes in crucial points like cell cycle, histones, centomere and kinetochore, got down regulated. In addition, more than 30 DNA repair related genes were down regulated and various breast cancer associated genes were affected. In contrast, there is no such a similar effect upon MCF-10A breast epithelial cell line. It has been showed that a designed flavonoid cocktail prevents the cell division of breast cells in many pathways.

Conclusion: In this manner, we consider that flavonoids can be use as an anticancer drug in further researches.

Keywords: Flavonoids, phenolic acid, breast cancer, whole genom expression, cell proliferation

OP-17

EFFECT OF TEA POLYPHENOLS ON PHASE I AND PHASE II ENZYME ACTIVITIES AND APOPTOTIC CELL DEATH MECHANISM IN BREAST CANCER CELLS

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Object: In this study the anti-proliferative effect of green and black tea polyphenols on breast cancer cell lines were investigated. The effects of polyphenol extracts on phase I and phase II enzymes were also evaluated. Finally, the effects of these polyphenols on apoptotic cell death mechanisms were investigated.

Materials and Methods: Polyphenols were extracted from tea samples and obtained in powder form with spray-drying. Extracts were then applied to breast cancer cell lines (MCF-7 and MDA MB-231) and their effects on cell viability and apoptotic mechanisms were investigated by flowcytometry. Additionally quinone reductase (QR), glutathione transferase (GST) activities were determined.

Results: In MCF-7 cells, tea polyphenols increased apoptosis (% 17.7) and necrosis (% 36.5) values significantly ($p<0.05$) when compared to control group (%4.67 and % 4.97). This effect was not observed in MDA MB-231 cells. Tea polyphenols increased green tea group quinone reductase activity (26.5 U/ml) in MCF-7 cells compared to the control group (17.2 U/ml). For MDA MB-231 cells, almost no change in activity was observed. Quinone reductase and glutathione transferase activites were increased significantly ($p< 0.001$) in MCF-7 cells when compared to the control group. Glutathione transferase activity was increased only in black tea group in MCF-7 cells, whereas it was decreased in MDA MB-231cells after both tea polyphenols application.

Conclusion: According to our results, polyphenols appear to effect different pathways depending on the molecular properties of breast cancer cell lines. MCF-7 cells were more sensitive to the effects of polyphenols than MDA MB-231 cells. Apoptotic cell death was induced in MCF-7 cells, whereas in MDA MB-231 cells other death pathways were activated.

Keywords: Breastcancer, Polyphenols, Cell death, Cell cycle, Enzyme activities.

OP-18

THE APOPTOTIC EFFECT OF PISTACIA VERA NUT SKIN EXTRACT ON HUMAN GASTRIC CANCER CELL LINE HGC-27

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Objective: Gastric cancer (GC) is a major world-wide health problem. It is the third leading cause of cancer-related death. The median overall survival is less than one year for advanced GC patients; thus, there is an urgent need to develop novel therapy for GC. Bioactive natural products are a good source for development of novel cancer preventive and therapeutic drugs. In this study, we investigated the apoptotic activity of water extract of the pistachio (Pistacia vera L.) nut skin on the human GC cell line HGC-27.

Material and Method: HGC-27 cells were cultured in DMEM medium supplemented with 10% fetal bovine serum. When cells reached 70% of confluence, they were treated with water extract or saline during 24 h. Apoptotic cells were quantified by Annexin-V/7AAD-positive staining, using an Annexin-V-FITC/7AAD kit from Beckman Coulter. For the cell cycle assay, BD Cycletest™ Plus DNA Reagent kit (Biosciences, Germany) was used. Apoptosis and cell cycle analysis were performed by using flow cytometer (NAVIOS Beckman Coulter, USA). For gene expression study, mRNA was isolated from cells by using miRNeasy Mini Kit (Qiagen GmbH, Germany). Then cDNA was produced with the Ipsogen RT Kit (Qiagen GmbH). qRT-PCR was performed by Rotor Gene 6000 (Qiagen GmbH, Hilden, Germany).

Results: Apoptosis of the HGC-27 cells was stimulated with water extract (8.6%) when compared to saline (0.6%). Proliferation was found to be stimulated in cell cycle assay (water extract: G0/G1 57.0%, G2 0.0%, and S 43.0%; saline: G0/G1 45.1%, G2 0.4%, and S 54.5%). NFKB (2.5±0.4 fold), p27 (0.8±0.2 fold), and p53 (4.1±2.3 fold) gene expressions were significantly modified with water extract treatment.

Conclusion: Our results showed that water extract of the Pistacia vera nut skin exhibited its anti-tumor activity against HGC-27 cells through promoting apoptosis, inducing proliferation, and elevated p53 gene expression. This study was supported by a Gaziantep University project (TF.13.10).

Keywords: Apoptosis, cell cycle, flow cytometer, gastric cancer, gene expression

OP-19

RESVERATROL CAN EFFECT CELL FATE DIFFERENTLY RELATED WITH P53 MUTATION AND SIRT1 ACTIVITY LEVEL OF HCT116 COLON CARCINOMA CELLS

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Introduction: Resveratrol is a natural polyphenol synthesized by more than 70 plant species. Anti-carcinogenic effects of Resveratrol have been proved in a variety of cancer cells. However recent studies emphasized that Resveratrol has an interaction with Sirtuin1 (SIRT1), a member of histone deacetylase enzymes that having a role on critical genes and deacetylation of p53 in carcinoma. Considering the interaction of Resveratrol with SIRT1 activity, the influence of Resveratrol on gene expressions and its relation with p53 mutation is gaining great importance.

Aim: To investigate the effects of Resveratrol in SIRT1 inhibited and uninhibited conditions on cell viability, apoptotic cell death levels and fold changes of some proliferative or anti-proliferative gene expressions, which may have important effects on colon carcinoma progression, in p53(+/+) and p53(-/-) HCT116 colon carcinoma cells.

Material and Method: IC50 doses of resveratrol were determined by WST-1 assay (Roche©). The cell death ratios in treatments of Resveratrol and Sirtinol (a SIRT1 inhbitor) were determined by Annexin-V-FITC/PI assay (BD Pharmingen©) for flow cytometry (BD-FACS Canto II™). The activity of SIRT1 was evaluated by fluorometric activity assay (Enzo Life Sciences©). The changes of some the proliferative (CCND1, FRA1, PPARD, EGFR, BIRC5, PCNA, MCL1, STAT3, FOS, JUN) and anti-proliferative (P27, ATF4, TRAIL, PUMA, GADD45A, RB1, FASLG, TNF, SOCS3, STAT1) gene expressions were evaluated in all groups as fold changes by using Bio-Rad CFX Connect™ Real Time PCR Detection System. All data were statistically analysed by Student's t test using Graphpad Prism 5.04 software.

Results: IC50 level of Resveratrol was found as 54 µM in HCT116p53(+/+) and 31 µM in HCT116p53(-/-) cell lines for 48 hour. According to viability data, we decided to use 60 µM Resveratrol concentration for both of the cell lines. SIRT1 inhibition dosage and timing of Sirtinol were found as 80 µM and 24 hour respectively. Resveratrol treatment (60 µM) causes 27.6% apoptotic cell death in SIRT1 uninhibited condition but it causes 33.4% apoptotic cell death in HCT116p53(+/+) cell line with SIRT1 inhibited condition. The same treatment conditions cause 17.9% and 18.2% apoptotic cell death for HCT116p53(-/-) cell line respectively. Gene expression results revealed that except for Survivin and FOS genes, in SIRT1 inhibited condition Resveratrol causes significant ($p<0.05$) increase of the proliferative genes in HCT116p53(+/+) cell line. However all proliferative gene expressions were found higher significantly ($p<0.05$) in HCT116p53(-/-) cell line for the same treatments. On the other hand, SIRT1 inhibited condition causes significant ($p<0.05$) increase of anti-proliferative genes except for RB1 and SOCS3 in HCT116p53(-/-) cell line. The same condition makes also significant rise ($p<0.05$) of anti-proliferative genes in HCT116p53(+/+) cell line except for RB1 and GADD45A.

Conclusion: This research has revealed that Resveratrol (60µM) causes decrease in cell viability and increase in apoptotic cell death in HCT116p53(+/+) and HCT116p53(-/-) cell lines. In addition, our research proved for the first time that the effects of Resveratrol could change according to p53 mutation and SIRT1 activity level of HCT116 colon carcinoma cells. We proposed that Resveratrol might show proliferative and/or apoptotic effects related with p53 mutation and SIRT1 activity of colon cancer cells. Consequently, we predicted that unconscious usage of Resveratrol in colon cancer patient might cause adverse effects. This study was supported with Scientific Research Projects Coordination Unit of Dokuz Eylul University with 2013.KB.SAG.055 project number.

Keywords: Resveratrol, SIRT1, p53, Colon Cancer, Signal Transduction Pathways

OP-20

ANTICANCER ACTIVITIES OF DCM-MEOH EXTRACTS OF SALVIA FRIGIDA ON LUNG, BREAST AND PROSTATE CANCERS

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Object: The use of medicinal plants and their derivatives in the treatment of various disease is rapidly increasing. Salvia frigida is a medicinal plant that belongs to salvia genus. Although it has been used for a long time due its medical activities, its anticancer activities is largely unknown and remains elusive. In this study, our aim was to evaluate the anticancer effects of Salvia frigida in lung, breast, and prostate cancer cell lines.

Material and Method: MTT was used to determine the cell viability. Apoptosis induction and cell cycle phases of cells were evaluated by flow-cytometric approach. Also, effects of the extracts on DNA fragmentation were evaluated. Cellular antioxidant activities of extracts were tested. Important phytochemicals were determined by using a HPLC and GCMS. Lastly, expression of genes involved in the apoptosis, DNA repair and anti-oxidant system were determined by a high-throughput approach. Findings: Active doses of DCM and MeOH extracts was not induced apoptosis in A549 cells. Also, DCM extract was shown to be induced apoptosis in DU-145 cells, yet MeOH showed poor apoptotic effect. Additionally, MeOH was increased the number of apoptotic cells with 19.2 % ratio. In cell cycle analysis, both extract was not effective in A549 cells. In DU-145 cells, while aneuploidy S cells were reduced, aneuploidy G2 cells increased after DCM extract treatment. While DCM extract had no effect on cell cycle of MCF7 cells, MeOH induced significant changes. Also, both extracts showed strong antioxidant activities in all cells. Phenolic acids of catechin, syringic acid, epicatechin, routine and benzoic acid were determined by HPLC. In GCMS analysis, 28 phytochemicals in DCM and 20 phytochemicals. Lastly, significant changes were determined in gene expression analysis after extract treatments.

Conclusion: Our findings suggest that Salvia frigida can be novel natural anticancer agent.

Keywords: Anticancer, Apoptosis, Cancer, Medicinal Plants, Salvia frigida

OP-21

THE EFFECT OF PROPOLIS AND CAFFEIC ASID PHENYL ESTER (CAPE) IN PANCREATIC CANCER

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Introduction: Pancreatic cancer (PaCa) is one of the most lethal human cancers with a 5-year survival rate of 3–5 % (approximately 6 months survive). The most important features of PaCa is its early invasion and metastasis, resistance to chemotherapy and radiotherapy and aggressive tumor progression. Propolis and its compounds could be potentially useful as chemotherapeutic or chemopreventive anticancer drugs. Bioactive components from the propolis have been extensively explored to possess anticancer activity. Caffeic acid phenethyl ester (CAPE), a naturally occurring compound isolated from the extract of propolis with well-known antioxidant activity, has been reported as an inhibitor of certain enzyme activities such as xanthine oxidase and cyclooxygenase as well as transcriptional factor NF-κB activation.

Material & Method:
Cell Culture: PANC-1 cells were cultured in DMEM containing 10 % FBS, 2 mM L-glutamine, %1 penicilin/streptomycin.
WST1: Sensitivity of the Panc1 cell line to increased doses of propolis and CAPE was determined by culturing 5×10^4 cells/ml for 24 and 48 h, incubated with WST1.
Clonogenic Assay: The effectiveness of propolis and CAPE on the survival and proliferation of Panc1 cells was tested by clonogenic assay.
Apoptotic Assessment: The apoptotic activity of propolis and CAPE on Panc1 cells was tested by Annexin V and TUNEL assays.

Result: We investigated the effects of propolis and CAPE in pancretic cancer cells and found that they both inhibit pancreatic cancer cell proliferation and induce apoptosis, but propolis is found to be more efficient in comparison to CAPE.

OP-22

ANTI-CANCER EFFICACY OF DEGUELIN AGAINST LUNG CANCER CELLS WITH AND WITHOUT DOCETAXEL

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OBJECTIVES: 58,7% of every new 100.000 cases that were diagnosed to cancer in between the years 2008 and 2012 are lung cancers and bronchial carcinomas. Lung cancers are divided in two main groups as Small Cell Lung Cancers(SCLC) and Non-Small Cell Lung Cancers(NSCLC). Despite the recent improvements of the treatments, the response and remission rates observed on the patients are relatively nominal.Dosetaksel(DTX) is a chemotherapeutic that has an anti-tumor activity against various solid tumors. The growing resistance against DTX still continues to be the biggest obstacle for the treatment success of NSCLC patients. Deguelin is a natural plant derivative rotenoid and has an encouraging activity against a lot of human cancers. The comparison of the treatment activity of the separate and combined usage of Deguelin,which is a potential chemotherapeutic agent, and Dosetaxel,which is used in standard treatment,is aimed in this study.

MATERIAL-METHOD: The IC50 doses of dosetataxel and deguelin on the A549 and H1299 NSCLCcell lines were determined via the cell vitality tests in our study. The active concentrations determined were applied to NSCLC cell lines as deugelin,dosetaxel and their combinations. The impacts of the medicine are studied by applying flow cytometric analyzes(apoptosis,cell cycle),glutathione and reducted glutathione,colony formation,migration and angiogenesis analyzes on the treated cells and measuring the Oxidative Stress Index(OSI).Statistical analyse program,Rstudio(v.0.98.501) and the R-script language were used to examine the differences between the agents. The states in which the p-value was lower than 0.05 were accepted as statistically meaningful.

RESULTS: We found that Deguelin has pro-apoptotic, anti-migratory and cytotoxic potential on lung cancer cells. Deguelin amplified DTX-related anti-cancer efficacy(increased apoptotic cell content and cytotoxicity, reduced migration). Also,Deguelin pre-treatment sensitized the cells DTX-treatment(reduced IC50 values).These effects were remarkable in p53-mutant cells.

CONCLUSION: Deguelin, solely, has anti-cancer potential on NSCLCcells.Both Deguelin pre-treatment and combinantion with standart chemotherapeutics result in enhanced anti-cancer efficacy.

OP-23

SYNERGISTIC EFFECTS OF ACHILLEA BIEBERSTEINII AND 5-FU AGAINST COLORECTAL CANCER CELLS

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Object: Colorectal cancer (CRC) is a major reason of cancer-related death, approximately 1.2 million new cases are reported each year in the worldwide and more than half die from the disease. 5-Fluorouracil (5-FU) is the backbone in the clinical treatment of advanced CRC. However, beside the antitumor effect most toxicities attributed to the drug and observed severe side effects. Therefore, novel treatments are still needed. 5-FU-based chemotherapeutic regimens are established as a fundamental standart treatment for metastatic colorectal cancer [1]. Antimicrobial, antioxidant, antiinflammatory, spasmolytic, antidiabetic, antiulcer, antitumor, choleretic and hepatoprotective activity, and cytotoxic effects of different Achillea species have been previously reported [2]. A. biebersteinii Afan. (Asteraceae) is a perennial herb, villose, stems erect, simple or branched from the base; 30–60 cm high; leaves up to 10 cm; flowering period, April-May [3]. In this study, antiproliferative and apoptotic effects of A. biebersteinii and it's combination with 5-FU were analyzed with various methods.

Material and Method: HT-29 colorectal adenocarcinoma cell line were obtained from ATCC. Cell Proliferation Kit I (MTT) and Cell Death Detection Elisa Kit were purchased from Roche Diagnostics, Germany. Other chemicals and reagents were obtained from Sigma and Merck. Cell viability was determined by MTT assay. HT-29 cells were treated with the different concentrations of A. biebersteinii, 5-FU and A. biebersteinii+5-FU. Cell Death Detection Elisa Kit was used according to the manufacturer's protocol for detect the apoptotic effect. pTEN, AKT, MAPK, mTOR, VEGF Receptor 2, p53 and β-actin gene expression levels were measured by RT-PCR. Western blot analyze were used to determine pTEN, AKT, MAPK, mTOR, VEGF Receptor 2, p53 and β-actin protein levels.

Results: Results are provided as the mean of independent experiments, each assay were performed in triplicate.

Conclusion: In vitro cytotoxic and apoptotic effect of A. biebersteinii+5-FU showed that this combination can be a candidate for colorectal cancer treatment.

References: [1] MEHMET KADİR ERDOĞAN, HAKAN AŞKIN. INTERNATİONAL SYMPOSİUM ON APPM, 2-3 APRİL 2015, PP 35. [2] TABANCA, N., DEMİRCİ, B., GURBUEZ, I., DEMİRCİ, F., BECNEL, J.J., WEDGE, D.E., & BASER, K H. (2011). NPC, (6), 5, 701-706. [3] AKKOL, E. K., KOCA, U., PESİN, I., & YİLMAZER, D. (2011). EVİDENCE-BASED COMPLEMENTARY AND ALTERNATİVE MEDİCİNE, 38.

Keywords: Achillea, apoptosis, 5-FU, mTOR

OP-24

ANTI-PROLIFERATIVE AND ANTI-INVASIVE EFFECTS OF FERULIC ACID IN TT MEDULLARY THYROID CANCER CELLS INTERACTING WITH URG4/URGCP

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OBJECT: Ferulic acid (4-hydroxy-3-methoxycinnamic acid; FA), a common dietary plant phenolic compound, is abundant in fruits and vegetables. The aim of present study is to investigate the effects of FA on cell cycle, apoptosis, invasion, migration, and colony formation in the TT medullary thyroid cancer cell line.

MATERIAL AND METHOD: TT human thyroid cancer cell line treated with 50 µM - 1mM FA by solving in medium for 24, 48 and 72 h considering a time- and dose-dependent manner. The effect of FA on cell viability was determined by using CellTiter-Glo Cell Viability Assay. Expression profiles of certain cell cycle and apoptosis genes such as URG4/ URGCP, CCND1, CDK4, CDK6, p53, PARP, PUMA, NOXA, BAX, BCL2, BID, CASP3, CASP9, MMP2, MMP9 and TIMP1 were performed on real-time PCR according to the SYBR Green protocol. Effects of FA in TT cells on invasion, colony formation and cell migration were determined by matrigel chamber, wound-healing and colony formation assay, respectively. Statistical analysis were performed with RT2Profiles PCR Array Data Analysis which is assessed statistically using the Student's t test.

RESULTS: IC50 dose of FA in the TT cells was detected as 150 µM. It was determined that FA caused a decrease in the expression of URG4/URGCP, CCND1, CDK4, CDK6, BCL2, MMP2 and MMP9, a significant increase in the expression of p53, PARP, PUMA, NOXA, BAX, BID, CASP3, CASP9 and TIMP1 genes in TT cell line. It was also found that FA in TT cells suppressed invasion, migration, and colony formation by using matrigel invasion chamber, wound healing and colony formation assay, respectively.

CONCLUSION: FA indicates anti-carcinogenesis activity by affecting cell cycle arrest, apoptosis, invasion, migration, and colony formation on TT cells. It is necessary to conduct further studies to discover therapeutic effect and the molecular mechanism of FA on thyroid cancer.

Keywords: Ferulic acid, Thyroid cancer, URG4/URGCP, apoptotic and cell cycle genes

OP-25

RESVERATOL INDUCES APOPTOSIS THROUGH OXIDATIVE STRESS IN BIPHASIC MALIGNANT PLEURAL MESOTHELIOMA CELLS

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AIM: Malignant pleural mesothelioma (MPM) is an asbestos-related tumor arising from the mesothelial surface of the pleural cavity with a poor prognosis. Biphasic malignant pleural mesothelioma (BMPM) is the one of the most resistant subtype of MPM to radiation and chemotherapy. Resveratrol (RSV) is a promising natural compound in the treatment of cancer. Its potent anti-cancer activity on BMPM cells are shown in previous studies. However the underlying molecular mechanisms of this effect is unclear. Therefore in this study we investigated the mechanism of the anti-proliferative effect of RSV in BMPM cells

MATERIAL AND METHOD: Human BMPM cells (MSTO-211H) were treated with RSV at concentrations from 5 µM to 150 µM for different exposure times (24h, 48h, 72 h) and then cell viability were measured. To evaluate apoptosis, the cells were treated with RSV for 24 hours and then annexin V-FITC-propidium iodide (PI) double staining is performed. ROS production in the cells is measured by flow cytometry and protein expressions of anti-oxidant enzymes are measured. Furthermore, since NFKB pathway is a key in prevention of apoptosis, we examined the protein expression of it.

RESULTS: RSV treatment reduced the cell viability in a dose and time dependent manner. Apoptosis was significantly increased in RSV-treated cells. Intracellular ROS levels were not changed with treatment but RSV increased SOD2 protein expression although it didn't change GPX and SOD1 protein expressions.

CONCLUSION: Our results revealed that RSV exhibited significant anti-proliferative effect on BMPM cells in a dose and time dependent manner by inducing apoptosis and this induction of apoptosis in the cells may have occurred via increased ROS production. And this oxidative stress may be counteracted with increased SOD2 level. These data suggest that RSV may be an important substance to investigate clinically for the treatment of BMPM which is a very resistant subtype of MPM to therapy.

Keywords: Resveratrol, Biphasic Malignant Pleural Mesothelioma, Apoptosis

OP-26

EVALUATION OF ANTIPROLIFERATIVE ACTIVITY AND ANTIAPOPTOTIC EFFECT OF THYMBRA SINTENISII SUBSP. ISAURICA EXTRACTS IN HUMAN CANCER CELL LINES

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Object: Cancer is a pathological state that is a genetic and developmental process occurring due to the excessive proliferation of the cells and the loss of their apoptosis functions. Side effects and drug resistance cause problems in the usage of synthetic drugs. Therefore, researchers try to elucidate new drug candidate compounds from bioactive natural products to lessen-prevent side effects and resistance. Traditional medicine from plants leads a pathway for this purpose. Thymbras also display antioxidant activities, anti-tumour and antimicrobial activities. Several plant species have been used for pharmaceutical purposes and yet many plants need to be studied in molecular detail. Thymbras have rich flora and attracted researchers for elucidating their composition. In the present study, we aimed to investigate antiapoptotic effect, anticancer, antioxidant activity of Thymbra sintenisii subsp. isaurica extracts.

Material and Method: Antiproliferative Activity The breast cancer cell line (MCF-7), human osteosarcoma cell line (MG-63), mouse fibroblast cell line (L929) were treated with the different concentrations (5, 10, 20, 40, 80, 160 µg/mL) of Thymbra sintenisii subsp. isaurica extracts in 24 and 48 hours and the analysis of cytotoxicity was tested with XTT. The IC50 values were calculated. Antioxidant Activities Antioxidant activities of Thymbra sintenisii subsp. İsaurica extracts was investigated by means of free radical scavenging activity (DPPH assay) and total phenolic compounds. Annexin V Analysis Antiapoptotic Effect of Thymbra sintenisii subsp. İsaurica extracts was evaluated in terms of apoptosis using flow cytometry.

Results: As a result of the study, it has been found that rich in polyphenolic compounds, high radical scavenging activity and anticancer activity of Thymbra sintenisii subsp. İsaurica extracts.

Conclusion: Thymbra sintenisii subsp. İsaurica extracts showed potent anticancer activity against cancer cell lines.

Keywords: Thymbra sintenisii subsp. İsaurica, Apoptosis, Cancer, Polyphenol

OP-27

BACTERIAL HEAT SHOCK PROTEIN GROEL TRIGGERS HUMAN PRIMARY T CELL APOPTOSIS

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Objectives: Modulation of apoptosis could be a critical factor in defining the outcome of an infection by bacterial virulence factors. *Aggregatibacter actinomycetemcomitans*' Hsp is a 64-kDa GroEL-protein which has been shown to influence the host and immune system cells but apoptotic capacity of GroEL protein on T cells is not known yet. The purpose of the present study was to investigate the ability of endogenous GroEL protein of *Aggregatibacter actinomycetemcomitans* to induce human T cell apoptosis.
Methods: Endogenously expressed GroEL protein was purified from *A. actinomycetemcomitans* (ATCC 29522) by ATP affinity chromatography and electroeluted from SDS-PAGE. Purified GroEL protein was confirmed by western blot and LC-ESI-MS. LPS concentration in purified GroEL protein was analyzed by LAL Chromogenic Endpoint assay kit. Detoxi-Gel Endotoxin Removing Gel was used to remove LPS from purified samples. Purified GroEL protein was used as antigen at different doses. PBMCs from healthy donors were cultured with GroEL from 0-96 hours. Apoptosis related changes in T cells were measured by flow cytometry and western blot.
Results: The data showed that phosphatidylserine exposure as an early apoptotic event was dose dependent in GroEL treated T cells. The kinetics of plasma membrane changes of T cells were also time dependent. GroEL treated T cells were positive for active caspase-3 in a dose dependent manner. Additionally, the rate of GroEL induced apoptosis was suppressed by the addition of general caspase inhibitor Z-VAD-FMK. Furthermore, cleaved caspase-8 bands (40/36kDa and 23kDa) were identified in GroEL responding cells.
Conclusions: Overall data presented in this study demonstrated that endogenous heat shock protein GroEL of *A. actinomycetemcomitans* mediates T cell apoptosis suggesting a role for Hsp's to modulate T cell immune response.

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Keywords: T cells, Apoptosis, GroEL, Bacterial heat shock protein, Hsp60, *Aggregatibacter actinomycetemcomitans*

OP-28

CAN ADV36 INDUCE 3T3-L1 PREADIPOCYTES INTO MATURE ADIPOCYTES?

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Aim: 3T3-L1 adipocytes originally derived from Swiss mouse embryo tissue have been fundamental in metabolic disease research for 30 years. The 3T3-L1 system has been pivotal in advancing the understanding of basic cellular mechanisms associated with diabetes, obesity and related disorders. In this study, it was aimed to investigate the differentiation of preadipocytes infected with Adenovirus 36 using morphological, histochemical and immunohistochemical methods.

Method: Following the removal of 3T3-L1 preadipocyte cell lines from stock, cells were incubated in a 3T3-L1 preadipocyte liquid medium (ZenBio, Inc, USA) for ten days. The medium was composed of DMEM, HEPES, Bovine calf serum together with a selection of antibiotics (i.e., Penicillin, Streptomycin) and an antifungal (i.e., Amphotericin B). The preadipocyte culture medium was replaced every 2 days with fresh medium during the 10 day incubation period. The subsequent formation of ~80% of confluent cells allowed the passage of cells into two groups using 6-well plates. Following two days of incubation, one of the passaged cell lines were infected with Adv36 (ATCC VR-1610) while the other remained uninfected and served as the control cell line. Cells were collected from both the infected and control cell lines at 2,5,7,9,14 and 21 days and used in phase contrast microscopy, Oil Red staining and leptin immunoreactivity analyses. Result Phase contrast microscopy analysis revealed an increase in the number of fat vacuoles amongst the Adv36 infected preadipocytes when compared with the uninfected cell line. Furthermore, the differentiation of the infected preadipocytes into mature adipocytes markedly increased particularly after the 7th day of infection. This observation was confirmed with the microscopy studies, together with Oil red staining and leptin distribution detection methods.

Conclusion: Adv36 infection can trigger 3T3-L1 preadipocytes into mature adipocytes in addition to increasing the number of lipid vacuoles.

Keywords: Adenovirus 36, 3T3-L1 cell, preadipocyte, mature adipocyte

OP-29

NUCLEOFECTION EFFECT ON EARLY APOPTOTIC CHANGES IN HUMAN NAIVE CD4 T CELLS

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Objective: Naive CD4 T cells play a major role in mediating immune response. These cells are difficult to transfect with viral based methods because of some drawbacks. The aim of this study is to investigate the effect of nucleofection, a non- viral transfection method, on Naive T cell apoptosis.

Methods: Blood form healthy donors is obtained with the permission of Dokuz Eylül Faculty of Medicine noninvasive ethics committee. Human peripheral blood mononuclear cells (PBMC) are isolated with the ficoll density gradient centrifugation Naive CD4 T cell are isolated by utilizing Variomax. After the purity of the cells are measured, the AMAXA 4D Nucleofection system is used on these cells for the introduction of GFP. The cells are then cultured in full IMDM for 24 and 48h. Then, the cells were analyzed with both CD4, CD25, CD69 markers for cell activation and Annexin V and 7AAD markers for cell death by using Flow Cytometry.

Result: The rate of CD4 positive GFP positive cells is found 0.3% after 24h incubation and 35% after 48h incubation. The cells are also analyzed with activation markers such as CD25, CD69. Both the rate of CD25 positive GFP positive cells and CD69 positive GFP positive cells are found 0% after 48h incubation . The viability of the nucleofected cells are searched by Annexin V and 7AAD. The rate of Annexin V positive GFP positive cells is found ~25% and the rate of 7AAD positive GFP positive cells is found ~8% after 48h incubation.

Conclusion: In conlusion, Nucleofection is an easy and rapid method that gives short electrical pulses to cell membrane and make holes in the membrane through which nucleic acids can pass. Due to the changes in phospholipid structure, the continuity of cell function can effect and cells can undergo apoptosis.

OP-30

INHIBITION OF CERAMIDASES IS A NEW POTENTIAL TARGET FOR LIVER CANCER THERAPY

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Object: Human liver hepatocellular carcinoma (HCC) is one of the most common malignancies in the world with an estimated half a million deaths annually and its incidence is on the rise in the USA, Europe and Asia. HCC is highly resistance to chemotherapy. Ceranib-2 is a ceramidase inhibitor that has shown significant antitumor activity in a variety of tumor cells. We intended to investigate whether ceranib-2 inhibits/induces cell proliferation and apoptosis the of hepatoma cancer cell lines HepG2 and SK1. To evaluate if ceranib-2 has an activity against liver cancer and with an aim to identify the altered cellular factors upon ceranib-2 treatment. Human HepG2 and SK-1 cancer cell lines was used as a model and cell death approach was utilized to elucidate the molecular mechanisms underlying ceranib-2's antitumor activity.

Material and Method: HepG2 and SK1 cell lines were purchased from ATCC. HepG2 cells were maintained in Dulbecco's modified Eagle's medium and SK1 in cells EMEM medium at 370C and 5% CO2 in atmosphere. Cells growth inhibition was measured by MTT method and apoptosis was detected by flow cytometry. Ultrastructural changes in HepG2 and SK1 cells were investigated under transmission electron microscope and morphological changes under confocal microscope.

Results: Ceranib-2 remarkably inhibited the proliferation of HepG2 and SK1 cancer cells and induced apoptosis in ceranib-2 treated groups. According to our transmission electron microscopy results ceranib-2 altered the ultrastructure in both of SK-1 and HepG2 cells. In ceranib-2 treated and acridine orange and phalloidin stained cells the morphology of SK-1 and HepG2 cells was altered indicated apoptotic changes. Flow cytometric analysis underlies the apoptotic action of ceranib-2 both in SK-1 and HepG2 cells.

Conclusion: On the basis or our findings we suggest this agent for further research for cancer therapy.

Keywords: Ceranib-2, liver cancer, apoptosis.

OP-31

THE EFFECTS OF BOR COMPOUNDS ON INFLAMMATORY GENES IN BREAST CANCER

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Objective: Breast cancer is the most common women's cancer in the world. Bor compounds have anti-cancer properties. The aim of this study was to evaluate the effects of bor compounds such as boric acid (BA) and disodium pentaborate (DSP) in breast cancer cells via apoptosis and inflammatory gene expressions.

Material and Methods: Triple negative (estrogen, progesterone and ERB2) and positive breast cancer cell lines, MDA-MB-231 and HTB-20 cells, treated with BA(0-200uM) and DSP(0-7.5uM). Cell proliferation and apoptosis determined with using WST-1 and Flow cytometric Annexin-V/PI measurements. Inflammation related 84 gene expressions were evaluated with RT-PCR array. Kruskal-Wallis and Mann-Withney U test and also t- test were used for statically analysis with using SPSS 15.0 program. ≥10 fold and more changes were accepted as a significant gene expression level.

Results: Both of BA (50uM) and DSP (5uM) decreased the cell viability by dose dependent manner at 24 hours' incubation. Apoptotic cell death was also induced mainly in HTB-20 triple positive breast cancer cells. BA treated MDA-MB-231 cells showed that increased CCL15, CCR1, CSF2, CXCL11, IL-15 while decreased CCL22, IL-7, OSM, TNSF10 gene expressions. DSP increased the expressions of CSF1, CXCL3, IL10RA, IL16, IL27, LTB, MIF, TNFRS11B but decreased the expressions of IL5 in same cells. In HTB-20 cells, BA reduced CCL1, CCL4, CCL5, CCR1, CD40LG, CXCL2, CXCL6, CXCR1, FASLG, IL27, IL5, NAMPT, OSM, TNFS13B gene expressions. AIMP1, CCL13, CCL8, CXCL12, CXCL5, CXCR1, FASLG, IL10RA, IL13, IL1R1, IL7, TNSF10, TNSF4 gene expressions increased with DSP in that cells.

Conclusion: BA and DSP affected the expressions of inflammation related genes in breast cancer cells in the opposite directions. This study showed that BA and DSP have different effects on specially inflammation related genes by directed estrogen, progesterone receptors and HER2 in breast cancer.

Keywords: Boric acid, Disodium pentaborate, breast cancer, inflammatory genes, apoptosis

OP-32

EVALUATION OF THE APOPTOTIC EFFECTS OF WORTMANNIN AND THALIDOMIDE ON BREAST CANCER CELL LINES

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Object: The aim of this study was to investigate the effects of PI3K inhibitor Wortmannin and angiogenesis inhibitor Thalidomide on intrinsic and extrinsic apoptotic pathways on breast cancer cell lines which have low (67NR) and high (4T1) metastatic potential using TUNEL and indirect immunohistochemical techniques.

Material and Method: 67NR and 4T1 breast cancer lines were cultured in DMEM-F12, medium containing 5% FBS, 1% NEA, 1% L-glutamine and 1% penicillin/streptomycin. IC50 values were determined as 2,5 µM for Wortmannin and 25 µM for Thalidomide by using MTT assay. Apoptotic cells were detected via TUNEL method and TUNEL index were calculated. Anti-Caspase-3, anti-FasL, anti-Apaf-1, anti-Cytochrome-c and anti-Bcl-2 primary antibodies were performed immunohistochemically after 24h and 48h drug administration. The mean values of the staining intensities (mild, moderate, strong and very strong) and percentage of positively stained cells were calculated using H-Score.

Results: It was observed that TUNEL index had a statistically significant increase in the drug administered group when compared to the control groups ($p<0.05$). Immunoreactivities of Caspase 3, Fas Ligand, Apaf-1 and Cytochrome C were seen as mild, mild/moderate, strong and moderate/strong in 67NR control group, respectively. While immunoreactivity of Caspase 3 was seen as moderate in 4T1 control group, immunoreactivities of Fas Ligand, Apaf-1 and Cytochrome C were observed as moderate/strong in this group. Statistically significant increased immunoreactivity scores were determined in the drug administered groups when compared to the control groups ($p<0.05$). The immunoreactivity of Bcl-2 was seen as strong in all groups and there was no statistically significant difference for Bcl-2 immunoreactivity ($p>0.05$).

Conclusion: It was concluded that Wortmannin and Thalidomide had an important role on both intrinsic and extrinsic apoptotic signalling pathways on 67NR and 4T1 breast cancer cell lines. As future expectation, these drugs might be used therapeutically to control cancer development via apoptosis in addition to classical treatments on cancer treatment.

Keywords: Breast Cancer Cell Line, Wortmannin, Thalidomide, apoptosis.

➢ POSTER PRESENTATIONS

PP-1

A SCAFFOLD PROTEIN IS AN INTERACTION PARTNER OF ATG5 AND A NOVEL REGULATOR OF AUTOPHAGY

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Autophagy is a biological mechanism allowing recycling of long-lived proteins, abnormal protein aggregates and damaged organelles under cellular stress conditions. Following sequestration in double or multimembrane autophagic vesicles, the cargo is delivered to lysosomes for degradation. ATG5 is a key component of an E3-like ATG12- ATG5-ATG16 protein complex that catalyzes conjugation of the MAP1LC3 protein to lipids, thus controlling autophagic vesicle formation and expansion. Accumulating data indicate that ATG5 is a convergence point for autophagy regulation. Here, we describe a scaffold protein, as a novel ATG5 interactor and an autophagy protein. Using several independent techniques, such as Yeast Two Hybrid Screen, immunoprecipitation assays, immunoflourescence analysis, gel filtration tests and SILAC based proteomic analysis, we showed that the scaffold protein interacted with ATG5, and both proteins co-localized in stress-responsive dot-like structures in the cytosol. Importantly, classical autophagy inducers (starvation or mTOR blockage) stimulated the interaction between ATG5 and the scaffold protein. Moreover, we identified the critical aminoacid regions of the scaffold protein for the interaction using site directed mutagenesis as well as molecular dynamics simulations. Knockdown of the scaffold protein or prevention of its binding to ATG5 using mutagenesis blocked autophagy activation. Therefore, the scaffold protein is a new ATG5-interacting protein and an important and novel component of the autophagy pathways.

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Keywords: Autophagy, lysosome, RACK1, ATG5, ATG12-5-16, signaling, mTOR, p70S6K, proteinproteininteraction

PP-2

LACCASE- GAMA CD ENCAPSULATED PCL NANOFIBERS FOR BETTER BIOCATALYTIC ACTIVITY

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The enzymes, as bio-catalysts, have opened a door to use eco-friendly, green and sustainable processes in the field of synthetic chemistry and thereby getting results by natural methods in the production have been possible. Since the enzymes came out of the reactions that they catalyzed unaffected and are so expensive compounds, in order to enable their reuses and avoid undesirable mixtures with the post-production products, the processes called immobilization should be carried out. Our study examines the effects of cyclodextrin use on enzymatic activity following enzyme immobilization into nanofibers. Analysis have been conducted on enzyme stability, enzyme activity and reaction performing potentials of enzymes. Electrospinning is a common and versatile method in nanofiber spinning from the polymer solutions, polymer mixtures or blends that can produce fibers from nanometer level to micron level. Nanofibers produced by electrospinning are formed by the creation of polymer droplets which is followed by the whipping, stretching and thinning of visco-elastic liquid under high electrical voltage application while after electrical field surpasses the surface energy of the droplet. Cyclodextrins (CDs), which offer functional solutions by creating the complex structures (inclusion complex), have unique properties, i.e. have nontoxic nature, improve solubility, and reduce undesired odor and some others. Well known and broadly used laccase enzyme which belongs to phenol oxidase group was chosen as a reference material for this study. Analysis that have been carried out have not exactly confirmed the laccase-γCD inclusion complex formations, but it has been observed that different structural formations have been created by complex formations of a different kind of interaction. Then, both FTIR data and the images of complex samples obtained have confirmed this condition. Subsequently, It has been gained insight on whether the structures produced by electrospinning creates nanofiber or not by SEM analysis. Uniform fiber formations were observed and it was realized that fiber diameters have been further thinned in cases of encapsulation. Nanofiber systems and cyclodextrin use on enzyme activity were analyzed and it has been indicated that the both nanofibers and cyclodextrin use, seperately, has been positively affected on the enzyme activity and increased the stability. Moreover it was shown that the immobilization of the enzymes treated in physical mixture of γCD and laccase and the creation of inclusion complex following by introduction of those materials into PCL polymer nanofibers has occurred a significant increase in the values of enzyme activation.

Keywords: Laccase, long stability.

PP-3

DECELLULARIZATION OF LIVER AND HISTOLOGICAL EXAMINATION OF OBTAINED MATRIX

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Abstract In tissue engineering, the importance of the natural or synthetic scaffold in a conversion and transplantation of three-dimensional structure of the produced cells is large (1). The formation of a functional vascular network resulting in new tissue is required for a successful tissue repair (2). Therefore using of decellularizated tissue as carrier support has advantages both in terms of comprising natural matrix and vascular supplying. In our study, decellularization methods of liver tissue for forming artificial liver and histologic examination of obtained matrix. The liver tissues were fixed in 10% neutral formalin and then embedded in paraffin blocks. Sections (2-4 µm thickness) were obtained using a sliding microtome from the prepared paraffin blocks. These sections were stained by Hematoxylin-Eosin (H-E). Later, the liver tissue sections were analyzed in the photomicroscope (Fig 1,2,3). Histological evaluation with H-E staining revealed less nuclei or cytoplasmic staining in decellularize groups (Fig 2,3) compared to normal rat liver (Fig 1). We showed that it was especially observed noticeable reduction hepatosit after the 5-hour desellülarization. In this study, it shown that the matrix with vascular network which is very important for tissue-engineering can be obtained in the liver, with an easy way, by desellülarization of tissue. In this way, produced hepatocytes can be easily clinging on a natural matrix and proliferating (3, 4).

Figures:

Fig 1. Control-liver, Normal liver histology (H-E, x10).

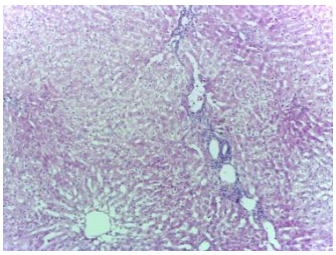


Fig 2. 2-hour decellularization-liver, moderate parenchymal staining (H-E, x10).

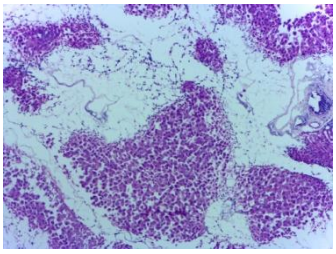
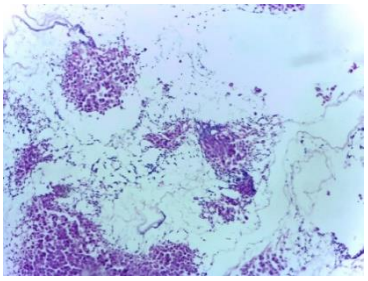


Fig 3. 5-hour decellularization-liver, less parenchymal staining (H-E, x10).



Keywords: Keywords: Decellularizated liver, tissue engineering.

PP-4

ASSOCIATION TRANSIENT RECEPTOR POTENTIAL MELASTATIN 2 GENE POLYMORPHISMS WITH PRETERM BIRTH

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Objective: Preterm or premature birth is defined as delivery of an infant before 37 completed weeks of gestation. Preterm birth is the leading cause of neonatal death and infant mortality, often as a result of respiratory distress syndrome due to immature lung development. Transient receptor potential (TRP) channels are non-selective channels permeable to monovalent and divalent cations. TRP melastatin (TRPM) 2 channel can be activated by micromolar levels of H2O2 and agents that produce reactive oxygen/nitrogen species, providing a direct link to inflammation, oxidative stress, and cell death. The aim of this study was to investigate a possible association between TRPM2 gene polymorphisms and preterm birth in a Turkish population.

Material and Method: A total of 90 women in preterm labor and 94 women in term labor with similar age and sex were enrolled to this study. Genomic DNA from the participants was analyzed by a BioMark 96.96 dynamic array system (Fluidigm, South San Francisco, CA, USA). For calculation of the significance of differences in genotype and allele frequencies, the chi-square test or Fisher's exact test was used.

Results: We observed that the CC genotype (15.3% vs. 1.2%, p=0.0034) and C allele frequencies (28.8% vs. 19.0%, p=0.048) of TRPM2 rs1612472 polymorphism were high in preterm birth group when compared to controls. There were significant changes in the genotype (TT, 60.9%; TC, 25.3%; CC, 13.8%) and allele (T, 73.6%; C, 26.4%) frequencies for the TRPM2 rs933151 polymorphism in preterm birth when compared to the controls (TT, 56.4%; TC, 11.7%; CC, 31.9%, p=0.0038; T, 62.2%; A, 37.8%, p=0.0285). However, no association was found with the TRPM2 rs1618355 polymorphism.

Conclusion: Our results are the first to demonstrate that TRPM2 gene polymorphisms may modify individual susceptibility to preterm birth in the Turkish population

Keywords: polymorphism, preterm birth, TRPM2

PP-5

ASSOCIATION TRANSIENT RECEPTOR POTENTIAL MELASTATIN 2 GENE POLYMORPHISM WITH PREECLAMPSIA

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Objective: Preeclampsia is characterized by maternal hypertension, proteinuria, oedema and, in 30% of cases, by intrauterine growth retardation. The precise factors involved in the pathogenesis of preeclampsia remain unclear and it is considered as a multisystem disorder. The oxidative stress, resulting from deficient remodelling of spiral arteries, is an important consequence of preeclampsia. The Ca2+ homeostasis is perturbed in preeclamptic placentas, most likely caused by a high oxidative stress level and lack of ATP. Transient receptor potential (TRP) channel is a nonvoltage-gated Ca2+-permeable cation channel superfamily activated by a variety of physicochemical stimuli. TRP melastatin (TRPM) 2 channel has been found to respond to oxidative stress by increased channel activity. The aim of this study was to investigate a possible association between TRPM2 gene polymorphisms and preeclampsia in a Turkish population.

Material and Method: A total of 94 patients with preeclampsia and 94 healthy controls with similar age and sex were enrolled to this study. Genomic DNA from the participants was analyzed by a BioMark 96.96 dynamic array system (Fluidigm, South San Francisco, CA, USA). For calculation of the significance of differences in genotype and allele frequencies, the chi-square test or Fisher's exact test was used.

Results: There were marked changes in the genotype (TT, 67.0%; TC, 26.6%; CC, 6.4%) and allele (T, 80.3%; C, 19.7%) frequencies for the TRPM2 gene rs933151 polymorphism in patients when compared to the controls (TT, 56.4%; TC, 11.7%; CC, 31.9%, $p<0.0001$; T, 62.2%; A, 37.8%, $p=0.0002$). However, no associations were found with the TRPM2 rs1612472 and rs1618355 polymorphisms.

Conclusion: To the best of our knowledge, these results are the first to demonstrate the contribution TRPM2 gene variants in preeclampsia. Our data showed that TRPM2 gene rs933151 polymorphism may modify individual susceptibility to preeclampsia in the Turkish population.

Keywords: polymorphism, preeclampsia, TRPM2

PP-6

CHARACTERIZATION AND CYTOTOXICITY OF BORON NITRIDE (BN) NANOPARTICLES: EMPHASIS ON TOXICOGENOMICS

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Boron nitride (BN) nanoparticles were synthesized chemically and characterized by using X-ray crystallography (XRD), transmission electron microscope (TEM), scanning electron microscope (SEM) and energy-dispersive X-ray spectroscopy (EDX) techniques. BN nanoparticles applied on the human lung alveolar epithelial cell line (HPAEpiC). To evaluate cytotoxicity of BN nanoparticles MTT, LDH and NR assays were carried out after 72 hours incubation. According to MTT results IC20 value for W2B was determined to isolate total RNA from cultures and investigate in microarray analysis. Finally, microarray data were investigated with The Database for Annotation, Visualization and Integrated Discovery (DAVID) analysis and functional categories for these genes revealed to understand effects of BN on biological pathways. The main aim of this article is to reveal characteristics, living cell interactions and any cytotoxic effect of BN.

In conclusion, there were so many publications about toxic effects of boron including compounds and the majority of these researches claim adverse outcome of molecules. Main aim of this project is to find out characteristics and cytotoxicity of boron nitrite (BN) nanoparticles, and also overcome the general judgement about notoriety of boron molecules. In accordance with all cell viability tests, MTT, LDH and NR analysis give similar results, and all of them confirm that appropriate amount of BN doesn't have lethal response on the HPAEpiC cells. Microarray results put forth anticancer/apoptotic, anti-metastatic and development regulatory effects of BN. Also, positive impact of the molecule on diabetes and stroke pathophysiology can be inferenced from gene analysis.

"This research was supported by National Boron Research Institute (BOREN) (Grant number: Ç0391)."

Keywords: Boron Nitride (BN) Nanoparticles, In vitro, Gene expression, Alveolar epithelial cells, Microarray

PP-7

CYTOTOXIC ACTIVITIES AND CELLULAR MECHANISM EXERTED BY WALTERINESSIA MORGANI COBRA VENOM AGAINST NEURONAL CANCER CELLS

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Snake venom is a complex mixture of many substances, including toxins, enzymes, growth factors, activators, and inhibitors, with a wide spectrum of biological activities and designed to affect vital processes, such as the function of nerves and muscles, the action of the heart, the circulation of the blood, and the permeability of membranes. Cancer and neurodegeneration are often thought of as disease mechanisms at opposite ends of a spectrum; one due to enhanced resistance to cell death and the other due to premature cell death. Many of the genes associated with either cancer and/or neurodegeneration play a central role in cell cycle control, DNA repair, and kinase signaling. Cancer therapy is one of the main areas for the use of protein peptides and enzymes originating from animals of different species. Cytotoxic effects of snake venom have potential to kill tumor cells. In this study, we investigated functional and activity of cobra crude venom of W. morgani on neuronal cancer cells. For this purpose, we determined cytotoxic effects of W. morgani crude venom against U87MG, SHSY5Y, KELLY and SK-NAS cells by MTT assay. Crude venom showed high cytotoxic effect on nerve cells with IC50 values varying between 0,15 - 5,2 µg/ml according to the viability percent. This results indicated that W. morgani cobra venom have potential for further studies to determine mechanisms of actions

In conclusion, W. morgani venom contain a large number of pharmacologically highly active substances through a specific mode of action each with the potential of becoming a potent drug. The formulations or combinations of this venom for targeted drug delivery could be used to for cancer and neurodegenerative diseases treatments. This study is ongoing to determine mechanistic effects of W. morgani venom on CXCR4 expression and Na+/K+-ATPase activity by flow cytometer.

Keywords: Walterinnessia morgani, Snake Venom, Neurodegeneration, Neurotoxin

PP-8

EXPRESSION PROFILING AND PATHWAY ANALYSIS OF IRON OXIDE (FE2O3) NANOPARTICLES TOXICITY ON HUMAN LUNG ALVEOLAR EPITHELIAL CELL LINE (HPAEPIC) USING MICROARRAY ANALYSIS

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[3]| Atatürk University

The main aim of this work to find out toxic effects of Fe2O3 on gene expression patten and pathway relationships of human lung alveolar epithelial cells (HPAEpiC).

Chemically synthetized Fe2O3 was characterized via using X-ray crystallography (XRD) and transmission electron microscope (TEM) techniques. Cell viability and cytotoxicity were determined by 3-(4,5-dimethyl-thiazol-2-yl) 2,5-diphenyltetrazolium bromide (MTT), neutral red (NR) and lactate dehydrogenase (LDH) release test. Whole genome microarray expression analysis was performed to be able to find out the effects of Fe2O3 on gene expression in HPAEpiC cell cultures. For further analyses, these genes were functionally classified by using DAVID (The Database for Annotation, Visualization and Integrated Discovery) with gene ontology (GO) analysis.

According to cytotoxicity assays LC20 value for Fe2O3 is 20.451 mg/L and this value is enough to call Fe2O3 nanopartcle high toxicity molecule. DAVID annotation analysis indicated that Fe2O3 mediated toxicity directly or indirectly affects regulation of cell proliferation, response to hormone stimulus, estrogen stimulus, cytokine activity and blood circulation by activating diverse genes.

"This research was supported by National Boron Research Institute (BOREN) (Grant number: Ç0391)."

Keywords: Iron oxide (Fe2O3) nanoparticles, Microarray Analysis, Toxicogenomics, Human Lung Alveolar Epithelial Cells (HPAEpiC)

PP-9

CYTOTOXIC AND ANTI-ADHESIVE PROPERTIES OF POLYLACTIC-CO-GLYCOLIC ACID COATED POLYPROPYLENE MESHES

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Object: Abdominal adhesions are frequently observed complications such as trauma, infection, pain and function disorder after intraperitoneal and pelvic operations. These complications are major problems for patients during healing process after such operations.Therefore anti-adhesive membranes are employed during operations in order to prevent or diminish dangerous complications. Synthetic meshes were introduced and used more than forty years. Polypropylene (PP) mesh could be useful as implantable mesh structures in surgery due to its high durability and elasticity. On the other hand, it can lead post-surgical adhesion after implantation. Bilayer meshes are used in clinical applications to prevent adhesion.

Material and Method: In this study, bilayer meshes composed of electrospun (polylactic-co-glycolic acid (plga) and plga-chitosan composite) coating on the PP mesh were developed. After incubating human peripheral mononuclear blood cells with meshes for 72 hours; annexin V – propidium iodide test for evaluating cytotoxic potential of meshes was used. For measuring anti-adhesive properties of meshes, human fibroblasts were seeded on mesh coated tissue culture plate wells and after 72 hours of incubation, LDH test was performed. Tests were performed as triplicates.

Results: After 72 hours of incubation, decreased rates of necrotic cells for both bilayer meshes were observed compared to PP mesh, whereas apoptotic cell rates were higher for bilayer meshes. Compared to non coated tissue culture wells, decreased rates of adhered fibroblasts were observed for all meshes.

Conclusion: PP mesh is a nano-sized product has high surface area porosity and it is an excellent replacement for a natural extracellular matrix. This design would prevent post-surgical adhesion in addition to increasing tissue-material integrity.

Keywords: Polypropylene, Polylactic-co-glycolic Acid, Adhesion, Cytotoxicity

PP-10

CASPASE-8 AND -9 MEDIATED APOPTOSIS IN MUCOSAL DISEASE (MD) FATAL VARIANT OF BOVINE VIRAL DIARRHEA (BVD)

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Object: Apoptosis have been defined as programmable cell death in many literatures. Generally apoptosis can be induced by two major regulatory pathways. One of them is extrinsic apoptotic pathway by a receptor/ligand interaction mechanism which involves the recruitment of regulatory caspase-8 to death receptor complex. Intrinsic pathway is associated with caspase-9 and the further cleavage of effector caspases. Particularly, cancer and many diseases evade cell death mechanism and caspases. Bovine Viral Diarrhea Virus (BVDV) is a unique agent for health of ruminants and pigs that causes high economic losses in both livestock and dairy industry. BVDV has two biotypes, one of them is non-cytopathogenic (ncp) blocks to process apoptosis mechanism and no apparently induces cytopathogenic effects on cell culture. Thus, ncp biotype causes persistent infection (PI) and infecting animals spreads to healthy animals throughout their lives. Cytopathogenic (cp) biotype led to acute infection or superinfection in persistent animals named as Mucosal Disease (MD), is highly fatal variant of BVDV. It is aimed to examine effects of both biotypes belonging to MD, have been detected by Polymerase Chain Reaction (PCR) in blood sample of infected animal, on cell culture.

Material and Method: Molecular test and virus isolation were examined from blood sample of an infected calf. To detect the expressions of caspase-8 and 9 by 4',6-diamino-2-phenylindole (DAPI) staining in MDBK cell culture. Cell culture fixation process are performed at 0., 12., 24., 48., 72. hr after virus inoculation to detect expressions of caspase-8 and -9 in different time period. Results: In microscopic assessment, caspase-8 and caspase-9 were stained from 12. hr and their expressions gradually increased in parallel to time. Furthermore, these findings showed parallelism with molecular results.

Conclusion: In conclusion, obtained results indicated that both caspase -8 and caspase-9 pathways could be activated by MD. It is tempting to speculate that caspases activities may be similar in both MD and acute BVD infections.

Keywords: Keywords: Caspase-8, Caspase-9, Apoptosis, BVDV/MD, DAPI

PP-11

EFFECTS OF ARGAN OIL ON MICRONUCLEUS AND MEGAKARYOCYTIC EMPERIPOLESIS IN RATS EXPOSED TO ACRYLAMIDE

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Object: Our study aimed to determine the effects of argan oil (AO) on the frequency of megakaryocytic emperipolesis (ME) and micronucleus (MN) against acrylamide (AA)-induced toxicity in rats.

Material and Method: Twenty healthy rats were obtained from the Experimental Animal Center of University of Ondokuz Mayıs (Samsun, Turkey). The study was approved by the Medical Research Ethics Committee of that university (HADYEK 2014/24). Animals in control group were orally gavaged with a constant volume of 1 ml/kg bw of 0.9% NaCl solution. While AA was administered intraperitoneally in a dose of 50 mg/kg/day, AO was administered by oral gavage in a dose of 6 mg/kg/day every other day for 30 days. Animals were also treated with mixture of AA (50 mg/kg/day) and AO (6 mg/kg/day) every other day for 30 days until they were euthanized. Bone marrow samples were analyzed for the frequencies of ME an MN.

Results: AA significantly increased the formation of ME and MN and decreased the ratio of polychromatic erythrocytes (PCEs) in bone marrow. No significant differences were observed in the animals received the AO compared to the control group. Co-treatment with AA+AO significantly ameliorated the MN, ME and PCEs values in bone marrow.

Conclusion: These findings suggest that AO may play a beneficial role in reducing the cell damage induced by AA.

Keywords: Acrylamide, argan oil, micronucleus, megakaryocytic emperipolesis, bone marrow.

PP-12

ND6 IN MITOCHONDRIAL DNA MIGHT HAVE ROLE ON BETA-CELL DEATH TARGETING BY MIR-29A

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Diabetes Mellitus is a common metabolic disorder in the World. This disorder characterized by high blood glucose level and related with leak insulin production by the pancreatic beta cells. Also the beta-cell death is important for T2DM. Moreover, genetic players seem to be involved in the development of T2DM such as MiRNAs. MiRNAs are post-transcriptional regulators of protein expression and they bind to complementary sequences in the 3'UTRs of their target mRNA either performing transcript degradation or translational inhibition. It is accepted that they play a role in the defect of β- cells to secrete enough insulin so trigger type 2 diabetes mellitus (T2DM). This project focuses on miR-29a that has been shown to be involved in glucose-induced beta-cell dysfunction, previously. Recently two publications have shown that microRNAs are present in the mitochondria, suggesting that microRNAs can regulate mitochondrial gene-expression. So in this study the target sequence was searched for miR29-a for mitochondrial gene ND6. ND6 is a vital gene for mitochondrial function and if mitochondrial function is damaged in beta-cells, beta cell dysfunction and death are observed. As method, the potential targets of miR-29a in ND6 sequence was identified and the oligos were designed. Then these oligos were transferred into a vector which has luc2 gene. These vectors were copied in E.coli cells then transfected to HEK cells for luciferase assay. The primary aim of the luciferase assay was to determine, whether miR-29a interacts with the predicted target site in ND6. Results of the luciferase assay's data and t-test showed that there was not a high quality complementation in ND6 between miR-29a. But for say a certain answer this experiment must repeat with another candidate target sites. For the future the existing connection to T2DM between miR-29a and ND6 because of mitochondrial functions could be motivating.

Keywords: miR-29a, microRNA, mtDNA, diabetes mellitus, Beta-cell death.

PP-13

EFFECT OF OZONE TREATMENT ON RADIATION COLITIS IN RATS

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Object: Radiation colitis occurring after radiation treatment of cancer and a major side effects that reduce the patient's quality of life. The formation of radiation colitis is thought to be caused by oxidative mechanism. We aimed to investigate the efficiency of intraperitoneal ozone application of radiation-induced colitis in rats.

Material and method: 42 Wistar albino rats were divided into 5 groups including Sham, Radiation, Ozone, Radiation+Oxygen and Radiation+Ozone. Sham group had no application. Medical ozone was administered to Ozone group intraperitoneally 1 mg/kg/day dose the third day to the seventh day. 25 Gray gamma ray exposure was applied to the abdominal-pelvic region of Radiation, Radiation+Oxygen and Radiation+Ozone groups. All groups made no application for two days. Exposure of radiation from day 3th to day 7th at 1 mg/kg/day dose of oxygen was administrated to Radiation+Oxygen group and was administrated ozone to Radiation+Ozone group. There has been no application until the end of the experiment the Radiation group. TUNEL staining was performed to determine apoptosis occurring in the intestinal tissue. The difference between groups was evaluated semi-quantitatively. Percentage of positive cells to all the cells in the image area was evaluated under a light microscope; -(None), +1(little), +2(medium), +3(severe) as scored.

Results: The immunohistochemical evaluation; apoptotic cells was not observed in the groups without radiation. Apoptotic cells score were severe(+3) in Radiation and Radiation+Oxygen groups, little(+1) in Radiation+Ozone group. Percentage of apoptotic cells reduction in the Radiation+Ozone group according to the radiation and Radiation+Oxygen groups was statistically significant ($p<0,05$).

Conclusion: We demonstrated that medical ozone application reduced severity of radiation-induced colitis in the rat intestinal epithelial. We further demonstrated that medical ozone possessed both anti-apoptotic and anti-inflammatory properties after radiation injury. These findings suggested the potential role of medical ozone against intestinal epitelial injury during radiotherapy.

Keywords: Keywords: Apoptosis, Ozone, Radiation Colitis, Radiotherapy

PP-14

EFFECTS OF CURCUMIN AND QUERCETIN ON REACTIVE OXYGEN SPECIES AND APOPTOTIC PROCESS IN CHRONIC MYELOID LEUKEMIA (K562) CELLS

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Aim: We have used quercetin and curcumin, two natural polyphenols, to induce apoptosis of chronic myeloid leukemia cells. Curcumin has limited clinical use because of its low bioavailability (< 2%). Therefore, we have tried synergistic combination of curcumin with quercetin to increase its apoptotic effects.

Methods: Cell proliferation was analyzed by WST-1 method and IC50 values were determined. Synergistic effects of the two polyphenols were analyzed by the CalcuSyn combination analysis program. Annexin-V staining was used for detection of apoptosis, DCFDA was used for detection of reactive oxygen species and JC-1 dye was used for detection of mitochondrial membrane potential. All of the above analyses were performed by flow cytometry. Fluorometric assay was used to measure intracellular glutathione. Chromatin condensation was shown using Hoechst 33342 dye by florescence microscope. Protein expressions associated with apoptotic mechanisms were analyzed using the Western blot method.

Results: The rate of apoptosis was enhanced by the synergistic combination of quercetin and curcumin. Using the combination doses the potency of quercetin and curcumin was decreased 1.6 to 7.3 fold, respectively. Quercetin and curcumin caused concentration-dependent decrease in cell proliferation and induced apoptosis (% 92.48) in CML cells after 48 hours. Combination of quercetin with curcumin reduced both cell viability and intracellular GSH (% 17.2). However, increased ROS and mitochondrial membrane potential as well as apoptosis rate was noted. The analysis of variance (ANOVA) was used for the comparison of all experiments.

Conclusion: We suggest combined use of the two polyphenols, quercetin and curcumin to increase their bioavalibility. Our results indicate that quercetin and curcumin exhibit a high level of strong synergism in combination, with enhanced bioactivity thereby reducing the required effective dose for each agent

Keywords: Chronic myeloid leukemia (CML), quercetin, curcumin, apoptosis, reactive oxygen species (ROS).

PP-15

CAN TAURINE PREVENT IFOSFAMIDE NEUROTOXICITY?

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Ifosfamide is often used in the treatment of childhood cancer. Ifosfamide have central nervous system toxicity such as changes in consciousness, cerebral infarction due to seizures, paralysis, neuropathy, leukoencephalopathy and ototoxicity. In this study, we aimed to histological analyses of Taurine because of antioxidant properties on the effect of ifosfamide neurotoxicity and nephrotoxicity. Wistar-Albino rats were divided into 4 groups; Group 1 was given intraperitoneal injection of 50 mg / kg ifosfamide, Group 2 was given intraperitoneal injection of 50 mg / kg ifosfamide + 1g / kg oral taurine (7 days), Group 3 was given intraperitoneal saline (control group), Group 4 was given oral 1 g / kg taurine (7 days). All animals were sacrificed after 8 days of studies and brain tissue were fixed in 10% formalin solution. After paraffin embedding procedure, sections were stained either TUNEL for apoptotic cell detection, or indirect immunoprexidase staining for distributions of Bax, cytochrome-C, caspase-3, caspase-8, i-nos, e-nos and n-nos. After histochemical analyses, in ifosfamide given group, chromatin condensation of neurons and neuroglia cells, edema around the neuroglia cells and demyelination of oligodendrocyte cells were observed. In Ifosfamide and Taurine treated group, chromatin condensation of neurons and neuroglia cells were rare. The number of TUNEL positive cells was higher in ifosfamide given group when compared with other. Increased caspase-3 and i-nos immunoreactivity were also detected in ifosfamide given group. While bax immunoreactivity was negative in neurons in ifosfamide given group, only weak Bax immunoreactiivty was observed in neuroglia cells. Cytochrome C, caspase-8, e-nos and n-nos immunoreactivities were negative in all groups. In conclusion; ifosfamide induced apoptotic pathways expressiong of Bax, caspase-3 and i-nos. Taurine may protects ifosfamide neurotoxicity inhibiting cellular damage.

Keywords: apoptose, caspase-3, NOS

PP-16

THE EFFECTS OF ZIZIPHUS JUJUBA ON SKIN CANCER CELLS

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Objects: Reactive oxidant species may result in oxidative stress in the cellular and extracellular environment and have been implicated in the etiology and progression of many diseases mainly chronic. Cutaneous melanoma is one of the most serious skin cancers. It is caused by neural crest-derived melanocytes -pigmented cells normally presented normally in the epidermis and, sometimes, in the dermis. Ziziphus jujuba Mill. (ZJ) distribute in the tropical and subtropical regions of Asia and have been employed as essential oriental folk medicine for thousands of years. This study was carried out to investigate the anticancer and antioxidant effects of ZJ on melanoma cells.

Materials Methods: Cell survival was quantified by colorimetric MTT assay with time and dose response. Melanoma cells were treated with 100 µmol Ziziphus jujuba essential oil for three hours. The morphology of cells were monitored and pictured. Then, the cell homogenates were taken after treatment period. Glutathione (GSH), Total oxidant capacity and total antioxidant capacity (TOC, TAC) and nitric oxide levels were estimated using highly specific spectrophotometric methods.

Results: Ziziphus jujuba inhibited growth and proliferation, and increased total antioxidant capacity. MTT assays indicated that Ziziphus jujuba significantly decreased cell viability. The results demonstrated that Ziziphus jujuba prevented decrease in antioxidant levels and nitric oxide levels in melanoma cells. Also GSH levels were improved after ZJ treatment.

Conclusion: Ziziphus jujuba demonstrated potent antiproliferative and antioxidative effects in melanoma cells. The study provides a scientific and ethnopharmacological rationale for the therapeutic use of ZJ fruit

Keywords: Keywords: Ziziphus jujuba, Melanoma, Oxidative stress

PP-17

IN VITRO EFFECTS OF OLEUROPEIN ON MELONOMA CELLS

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Objects: Oxidative stress is considered to be involved in the pathophysiology of all cancers. Melanoma is the main cause of death in patients with skin cancer. It is caused by neural crest-derived melanocytes -pigmented cells normally presented normally in the epidermis and, sometimes, in the dermis. Oleuropein is the main phenolic compound of olive tree and is responsible for the characteristic bitterness of olive fruits. Oleuropein is a heterosidic ester of elenolic acid and hydroxytyrosol and possesses beneficial effects on human health. The aim of this study was to determine in vitro effects of oleuropein on melanoma cells.

Materials Methods: Viability of the cells was quantified by MTT assay in a time and dose response manner. Melanoma cells were treated with 100 µmol for three hours. The morphology of cells was monitored. Then, the cell homogenates were taken after treatment period. Glutathione (GSH), Total oxidant capacity and total antioxidant capacity (TOC, TAC) and nitric oxide levels were identified using specific colorimetric methods.

Results: Oleuropein treatment increased cell viability in melanoma cell line in a dose-dependent manner. The cells treated with oleuropein with optimum dose and time. Oleuropein inhibits the activation of nitric oxide and prevented decreased levels of GSH. Also total oxidant capacity decreased significantly after treatment.

Conclusion: Oleuropein decreased cell death and triggerred antioxidant system positively. These findings suggested that oleuropein has potent anticancer and antioxidant properties on melanoma cells. However, further in vivo studies are required to determine the exact potential of this agent.

Keywords: Keywords: Oleuropein, Melanoma, Oxidative stress

PP-18

PROTECTIVE EFFECTS OF PLANTAGO HOLOSTEUM SCOP. EXTRACT ON HYDROGEN PEROXIDE-INDUCED DAMAGE IN L929 FIBROBLAST IN RELATION TO THE ANTIOXIDANT ACTIVITY

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The genus Plantago (Plantaginaceae) is represented by 21 species in Turkey. Several effects are described for the genus Plantago such as antitumoral, anti-inflammatory, antifungal, antibacterial, analgesic, antispasmodic, antiviral and hepatoprotective. Plantago species are known not only as a food plant, but also an old medicinal plant that has been used externally to treatment of wound, abscess and acnes, internally to treatment of diabetes, urinary infections and cancer as a decoction, common cold and viral infections as infusion in Anatolia. Earlier investigations performed on Plantago species resulted in the isolation of mainly iridoid glucosides, phenylethanoid and flavonoid glycosides, caffeic acid derivatives, polysaccharides and lipids. Wound healing consists of different stages of inflammation, proliferation and remodeling stage. In this study; antioxidant, proliferative and protective effect of extract were investigated against H2O2 damaged L929 murine fibroblasts to understand wound healing properties of the extract. As a result of our study, water extract of P. holosteum showed radical scavenging activity against DPPH, NO, SO and ABTS radicals comparable to that of known antioxidants BHA and ascorbic acid and water extract of P. holosteum did not increase the proliferation of the fibroblast in the concentration range of 10-200 □g/mL. In the case of protective effect of the extract against H2O2 injury, cytotoxicity of hydrogen peroxide was found dose-dependent in L929 fibroblasts. While pre-incubation with the extract protects fibroblasts against oxidative damage of H2O2, incubation with the extract after H2O2 application did not repair the oxidative injury. This result correlates with the antioxidant potential of the extract. Our study on P. holosteum will continue to investigate other parameters of wound healing in different cell lines.

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Keywords: Plantago holosteum, hydrogen peroxide, wound healing, antiinflammatory

PP-19

THE NEUROTOXIC, CYTOTOXIC, APOPTOTIC AND ANTIPROLIFERATIVE ACTIVITIES OF THE EXTRACTS OF SOME MARINE ALGAE ON NA2B CELL LINE

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Marine algae are natural compounds and their cytotoxic, antiproliferative and apoptotic effect has been shown for many cancer types. Anticancer effect of these algae may also important nervous system tumours. Therefore, these effects of the algae extracts on mouse neuroblastoma cell line (NA2B) were investigated in culture. Extracts from Petalonia fascia, Jania longifurca and Halimeda tuna were harvested in the Aegean Sea shores of Turkey. 15000 cell/ml/well were treated by algae extracts at 1 to 0,00007 µg/ml dilution rates for NA2B cell survival and proliferation by using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT). Cytotoxic effect of algae extracts on the NA2B cells were also investigated for oxidative stress with Nitric Oxide Synthase (NOS) immunocytochemistry and apoptosis with terminal deoxynucleotidyl transferase dUTP nick end labeling (TUNEL). Moderate toxic effect was examined by neurite inhibition with neurotoxicity screening test (NST) at the IC50 dilutions of extracts. It was observed by MTT results that J. longifurca extracts were more toxic compared to P. fascia and H. tuna extracts. There was clear increase of endothelial and inducible NOS immunostaining for oxidative stress and TUNEL for apoptosis after extracts application. There was inhibition of neurite outgrowth with a statistical meaning due to moderate toxic effect of algae. This study suggested that the compounds of algae could be useful for the anticancer treatment of nervous system tumour. Meanwhile, their spread on the marine environment may dangerous for aquatic animals and also for human health because of the food chain.

Keywords: Cytotoxicity, Marine algae, NA2B cell line, Neurite inhibition, Neurotoxicity screening test, TUNEL

PP-20

SERUM HYPOXIA INDUCIBLE FACTOR-1ALPHA LEVELS IN PATIENTS WITH POLYCYSTIC OVARY SYNDROME

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The Polycystic Ovary Syndrome (PCOS), unknown etyology, is major healthy problem in women worldwide. Resently some studys have shown that hypoxia inducible factor (HIF)-1alpha, the oxygen-sensitive transcriptional activator, plays a key role in the development of mammalian ovarian follicular development and ovulation. In this study, we aimed to investigate to the levels of HIF-1 alpha in individual with PCOS. The study was carried out on 48 women consisted of 28 patients with PCOS and 20 healthy ones as control. In serums obtained from blood samples taken from patient and control groups, the HIF-1 alpha concentrations were measured by ELISA kit, a specific enzyme-linked immunosorbent assay. There was no different betwen the groups in mean of age distrubution, $p>0.05$. It was found that serum HIF-1 alpha concentrations were higher in patient with PCOS than in control group (50.92± 34.73, 41.16± 40.98, respectively) but there was no statistically significant difference, $p>0.05$. Because of there is no statistically significant difference between two groups in our study, we suggest that are studied with more than number of samples.

PP-21

THE EFFECTS OF NMDA RECEPTOR SUBUNITS AND CHOLESTEROL ON AMYLOID BETA TOXICITY IN SHSY-5Y CELLS

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Objective: There is an increased sensitivity to N Methyl D aspartate (NMDA) receptor and irreversible neuronal cell death in Alzheimer's Disease (AD). Increase of intracellular calcium levels, which occurs after NMDA receptor induction, can lead to neuronal cell death. It has been suggested that amyloid beta (AB) peptides at toxic concentrations may increase pregnenolone sulfate (PS) level in the presence of cholesterol. It's known that micro molar level of PS has an agonistic effect on NMDA receptor, which has a heterogeneous structure. For the effect of PS on NMDA receptor, a combination of receptor subunits (NR2A, NR2B and NR1) is required. However, the role of NMDA subunits and the effect of PS on AB toxicity in the presence of high cholesterol levels has not to be revealed yet. The aim of this study is to determine the effects of NMDA receptor subtypes on AB toxicity and to examine possible connection with the change of PS level.

Material and Method: SH-SY5Ycells were treated with 10 µM AB 1-42 after the peptides were incubated at 37ºC for three days and cholesterol (1.2-9 mM). To examine the effects of NMDA receptor inhibitors, the cells were also treated with NVP-AAM077 (NR2A inhibitor, 12nm), ifenprodil (NR2B inhibitor, 0.5 µM) and MK801/memantin (total inhibitor, 5µM) for 24, 48 and 72 h. Changes in cellular cholesterol and PS levels were determined simultaneously in a dose-and time-dependent manner. The cell viability was also evaluated.

Results and Conclusion: The treatment together with cholesterol (9mM) and AB peptides decreased cell viability nearly 50% of control and significantly increased PS levels ($p<0.05$). The treatment of ifenprodil and memantin significantly protected SHSY-5Y cells from the toxicity induced by the treatment together with cholesterol and AB peptide ($p<0.05$). NMDA receptor NR2B subunit may play a key role in neuronal cell death induced by AB peptides. The changes in PS levels and its NMDA receptor activity may affect neuronal cell survival.

Keywords: NMDA receptor, amyloid beta toxicity, neuronal cell death, cholesterol, pregnenolone sulfate, NR2 receptor subunit

PP-22

NUCLEIC ACID DAMAGE IN WOMEN WITH POLYCISTIC OVARY SYNDROME: 8-HIDROKSI 2'-DEOKSI GUANOZIN LEVELS

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Polycystic ovary syndrome (PCOS) is the most common gynecological endocrine disorder in reproductive-aged women. The etiology of PCOS is not fully understood. Free radicals causes damage or death to the cells. Hydroxyl radical (OH-) reacts with DNA purine bases and genetates purine radicals. Oxidative DNA damage uses as a measure of the damage indicator and usually measures as 8-Hydroxy-2'-deoxyguanosine (8-OHDG) nucleosides. In this study, we aimed to investigate for the levels of 8-OHDG in patients with PCOS. The levels of 8-OHDG, FSH, LH were assessed in 103 patients with PCOS and 47 healty control group. Isolation of DNA from all blood samples was performed used by manifacture kit. 8-OHDG levels were studied with high performance liquid chromatography (HPLC-UV and -EC). Because variables showed abnormal distribution, Manny Whitney U test, nonparametric test, was applied for staistical analysis in the present study. As statistical, $p< 0.05$ values were considered significant. The levels of FSH and LH in PCOS group were significantly higher than control group, $p<0.05$. There was significantly different in 8-OHDG levels between PCOS and control groups (1.96± 0.83, 0.45± 0.15 respectively), $p<0.001$. According to the results, our study was shown that 8-OHDG levels were increased in patient with PCOS. We suggest that levels of 8-OHDG may be evalueted as an indicator of oxidative stress in PCOS

Keywords: 8-OHDG, PCOS

PP-23

THE EFFECTS OF LIQUIDAMBAR ORIENTALIS OIL ON HUMAN GLIOSBLASTOMA CELLS IN H2O2 INDUCED OXIDATIVE STRESS

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Objects: Glioblastomas, the most malignant form, are characterized by increased proliferation and invasion into the surrounding normal brain tissue. The essential oil of Liquidambar orientalis var. orientalis, an endemic tree species in Turkey, has medicinal and cosmetic properties; and its antioxidant properties were investigated in H2O2 induced oxidative stress in human glioblastoma cells. Materials Methods: Cell survival was quantified by colorimetric MTT assay with time and dose response. U87 human gliblastoma cells were pretreated with H2O2 100 µM after 30 minutes, 100 nM Liquidambar orientalis essential oil was added to the cells for three hours. The morphology of cells were monitored and pictured. Then, the cell homogenates were taken after treatment period. Glutathione (GSH), Total oxidant capacity (TOC) and total antioxidant capacity (TAC) and nitric oxide levels were estimated using highly specific spectrophotometric methods. Results: We demonstrated that the treatment of glioma cells with Liquidambar orientalis antagonized H2O2 induced oxidative stress. In U87 cells exposed to H2O2, GSH was significantly depleted. The results suggest that Liquidambar orientalis regulated nitric oxide and TOC levels which was increased by H2O2 induction. TAC was increased after treatment of oil. Conclusion: Liquidambar orientalis triggers antioxidant system in human glioma cells. The present findings may have useful implications for the potential use of these oil in the medical field as well as in the food industry

PP-24

EVALUATION OF VITAMIN D, VITAMIN B12, IRON LEVELS IN A SMALL GROUP OF MEDICAL FACULTY STUDENTS

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Aim: Humans get vitamin D from exposure to sunlight anf from diet. In young people vitamin D is important for calcium absorption and bone growth. Low levels of vitamin D is found to be related to some diseases. Cancer, skeletal diseases, diabetes mellitus; multiple sclerosis and also it is about menthal development. We wondered what is vitamin D levels in our medical faculty students, how often do they benefit from daylight. And we also evaluated their some other biochemical parameters.

Methods: Initially 60 medical faculty students joined to work. They were divided according to living with family, once again divided girls and boys.

Results: In male students living with their family, Vitamin D levels higher than others. B12, ıron, hematologic parameters found similar in all groups. Vitamin D cut off value is known as 20 ng/ml. When divided to only cutt off, in high vitamin D group (34,48 ng/ml) , it is found that B12(372,4 pg/ml), ferritin(56,95 ng/ml) and iron(88,70 µg/dl) average levels were higher than low vitamin D groups (12,50 ng/ml) respectively 349,71 pg/ml, 49.85 ng/ml, 79,5 µg/dl. İn high vitamin D group the duration of exposure to direct sunlight is 2,7 hour when compared others 2.22 hours in day. we also measured Superoxid dismutase, glutatyon peroxidase, totalantioxidant (TAS) and totaloxidant (TOS)capacity.

Conclusion: It is important to be aware of benefit of vitamin for healthy life and so everyone should know their own biochemical state.

Keywords: Keywords: Liquidambar orientalis var. orientalis, U87-MG, Oxidative stress

PP-25

ANTICANCER EFFECT OF SOME SILVER(I)-N-HETEROCYLIC CARBENE COMPLEXES ON DIFFERENT CANCER CELL LINES

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Metal-based drugs have many effective in the treatment of a variety of diseases especially for cancer. So, the metallo-organic diagnostic agents' attention has been increase in recently. Although metal-containing drugs which such as cisplatin, fluorouracil have known as effective therapeutics, Ag are one of the metallo-organic compounds that see interest in last five years. This work was to inform of anticancer activity of some Ag (I)-N-Heterocyclic carbene (NHC) complexes against different cancer cells.

Liu and coworker identified in their work that some Ag(I)-NHC complexes anticancer effects against breast cancers (MCF-7 and MDA-MB-231) and colon carcinoma (HT-29) cells. While, the 4-hydroxyl substituted compound exhibited antiproliferative activities with IC50 9.2-16.2 µM, the fluoro and methoxy-substituted compounds were anticancer activities in the three cancer cell line with IC50 values under 10 µM (1).

Explained that Ag-(I)-NHC complexes induced HL60 colon cancer cell death independent of the caspase cascade via the mitochondrial AIF pathway (2).

According to a study published in 2003; the researchers identified that a Ag (I)-NHC complex showed significant antiproliferative activity with inhibited thioredoxin reductase. The inhibition of this selenoenzyme identified with alteration of the cellular redox environment (3).

In a work; determined that the apoptosis mechanism of new Ag (I)-NHC complexes on lung cancer cell (A549). In the result identified that while Ag complexes having the IC50 level 7.1±0.78, cisplatin was IC50 value of 6.59±0.63. Ag complexes inhibited the growth of A549 cancer cells by inducing G2/M phase cell cycle arrest and apoptosis. Ag complexes induced apoptosis with the levels of intracellular ROS. Also these complexes disrupted the mitochondrial membrane potential. Further; induced the caspase-3 activation and led to the translocation of apoptosis inducing factor and endonuclease G to the nucleus (4).

Reported that 4-Alkylated Ag-NHC complex exhibited cytotoxic effects in leukemia cells with IC50 values of 27 μM (5).

PP-26

CYTOTOXIC EFFECTS OF NON-HEMOLYTIC SKIN-PAROTOID GLAND SECRETIONS OF THE BUFONID TOADS FROM TURKEY

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Object: Skin-parotoid secretions of amphibians contain a large number of biologically active compounds which are thought to play several roles, either in the regulation of the physiological functions of the skin or in defense mechanisms against predators or microorganisms. Especially, the biodiversity of biochemical compounds in the auricular and skin glands of toads makes them unique sources for new therapeutic agents.

Material and Method: Common Toad-Bufo bufo, Caucasian Toad-Bufo verrucosissimus and Variable Green Toad-Bufotes variabilis were collected in field, then skin secretions obtained by stimulator, while parotoid gland secretions obtained by manual compressing. Each individual was rinsed with ultra-pure water into the tubes, then snap-frozen by liquid nitrogen and then lyophilized. Protein content was determined by BCA assay kit. Cytotoxic effects was determined against HeLa, A549, Caco-2, MPanc-96, PC-3, MDA-MB-231 cancer cells and HEK-293 as a non-cancerous cell line by MTT assay. Parthenolide was used as a positive cytotoxic control agent. Percentages of surviving cells and IC50 values in each cells were calculated after incubation with secretions using GraphPadPrism 5. Hemolytic activity of secretions was also determined on rabbit red-blood cells.

Results: Protein concentrations of B. bufo, B. verrucosissimus and B. variabilis secretions were calculated as 3100 µg/ml, 3300 µg/ml, 3480 µg/ml, respectively. Crude skin-parotoid gland secretions of all taxa were showed strong cytotoxic effect on all cancer and non-cancerous cells with an IC50 values varying between <0.1-6.02 µg/ml. No hemolytic activities at concentrations between 0.5-50 µg/ml were observed.

Conclusion: Further investigations need to focus on to purify the active components from these skin-parotoid secretions and determine the possible mode of action of secretion-induced cytotoxicity to obtain a better understanding of their potential use as anticancer agents.

Keywords: Toad, skin-parotoid gland secretion, cytotoxicity, hemolytic activity.

PP-27

MICRORNA376 FAMILY AND CANCER

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Object: Autophagy, is one of the most well known catabolic processes whose activation can degrade accumulated proteins as well as damaged organelles for maintaining cellular homeostasis. Beside this, autophagy was found to be associated with several abnormalities including cancer and metabolic diseases. In addition, miRNAs have been implicated in several fundamental biological processes including development, differentiation, apoptosis and stem cell maintenance. Moreover, evidence also suggests that miRNAs play a role in cellular transformation and carcinogenesis. Thus, understanding the regulation of autophagic mechanisms through miRNAs might have tremendous importance in the field of cancer. Material and Method Overexpression of MIR376B in MCF-7 cells has been utilized and several mono clone cells picked and cultured under selection condition. For further analysis, mono clones were evaluated by their autophagic capacity via LC3 shift, p62 accumulation and miR-376b target protein status. After the characterization of clones, several growth analyses were performed either short or long term assays in vitro. Clonogenic potential of MIR376B stably overexpressing and control cells were analyzed by their ability to create colonies by colony formation assay. On the other hand, Gamma-H2AX foci analysis and ROS measurement by DCFDA was carried out to identify the DNA damage and oxidative stress, respectively.

Result: As a consequence of autophagy deregulation, accumulation of p62 was observed in miR-376b stable cells. Intriguingly, intracellular ROS level was also increased and accumulation of ROS localized around the mitochondria. In addition to susceptibility of oxidative stress, loss of autophagy makes cells more prone to DNA damage. Although in short term assays, growth attenuation of miR-376b stable cells was observed; in colony formation assay, those cells formed more and bigger colonies.

Conclusion: We identified for the first time that MIR376B as a key miRNA which might has a role in tumorigenesis in breast cancer.

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Keywords: Macroautophagy, mammalian autophagy regulation, microRNA, MIR376B, cancer

PP-28

THE CYTOTOXIC EFFECTS OF ETOPOSIDE, PYCNOGENOL® AND THEIR COMBINATION ON A549 CELL LINE

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Object: Chemotherapy is the most common method used in treating cancers and directly targets at eliminating cancer cells using chemicals with cytotoxic effect. Because the target of chemotherapy is tumor cell, the agent is used in high doses during application and this creates toxic responses causing side effects on normal cells. Hence chemotherapy is combined with some cancer treatments to increase the therapeutic efficacy by reducing side effects. Etoposide belongs to the class of drugs known as podophyllotoxin derivatives. A topoisomerase II inhibitor within etaposide promotes apoptosis in cancer cells and it has been used for the treatment of lung cancer. Pycnogenol® is a kind of flavonoid and it can be used as the dietary supplement. It is rich of many phytochemicals of medical value, is extracted from the French maritime pine bark Pinus pinaster. In this study the cytotoxic effects of Etoposide and Pycnogenol combination on A549 cells was researched.

Material and Method: A549 cells were cultured in RPMI 1640 medium containing 10% fetal bovine serum, 10 µg/ml gentamicin and 5% sodium pyruvate. The cells were incubated in 5% CO2 at 37 °C. The cells were treated with various concentrations (500-12,5 µg/mL) of Etoposide, Pycnogenol and their combination. Cell viability was measured after 24 h treatments. Cytotoxic effects of drugs were investigated using MTT cell proliferation kit.

Results: Etoposide alone caused 34.20%, 27.87% and 24.28% cell death at 500, 400 and 300 µg/mL concentrations, respectively. IC50 values for Pycnogenol and Pycnogenol+Etoposide are 485 and 195 µg/mL, respectively.

Conclusion: At the end of 24 h Pycnogenol and Etoposide combinations at different doses exerted synergistic effect on A549 cell death

Keywords: Etoposide, Pycnogenol®, Cytotoxicity, Combine therapy

PP-29

THE APOPTOTIC EFFECT OF BONGARDIA CHRYSOGONUM ON HUMAN GASTRIC CANCER CELL LINE HGC-27

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Objective: Despite the great progress in the treatment of gastric cancer (GC), it is still the third leading cause of cancer death worldwide. GC still carries a poor prognosis. Therefore, it is imperative to identify novel, low toxic and effective anticancer drugs. In this study, we investigated the apoptotic activity of hexane extract of the tuber skin of Bongardia chrysogonum on the human GC cell line HGC-27.

Material and Method: HGC-27 cells were cultured in DMEM medium supplemented with 10% fetal bovine serum. When cells reached 70% of confluence, they were treated with hexane extract or DMSO during 24 h. Apoptotic cells were quantified by Annexin-V/7AAD-positive staining, using an Annexin-V-FITC/7AAD kit from Beckman Coulter. Positive cells were analyzed on a flow cytometer (NAVIOS Beckman Coulter, USA).For the cell cycle assay, BD Cycletest™ Plus DNA Reagent kit (Biosciences, Germany) was used. Cell cycle analysis was performed by using flow cytometer. For gene expression study, mRNA was isolated from cells by using miRNeasy Mini Kit (Qiagen GmbH, Germany). Then cDNA was produced with the Ipsogen RT Kit (Qiagen GmbH). qRT-PCR was performed by Rotor Gene 6000 (Qiagen GmbH, Hilden, Germany). Western blot analysis was performed for protein expression.

Results: Apoptosis of the HGC-27 cells was stimulated with hexane extract of the tuber skin (2.8%) when compared to DMSO (0.5%). No marked changes was observed in cell cycle assay (hexane extract: G0/G1 58.9%, G2 0.1%, and S 41.0%; DMSO: G0/G1 60.0%, G2 0.3%, and S 39.7%). We found that NFKB, p27, and p53 gene and protein expressions were not significantly modified with hexane extract treatment ($p>0.05$).

Conclusion: Our results showed that hexane extract of the tuber skin of Bongardia chrysogonum exhibited its anti-tumor activity against HGC-27 cells through promoting apoptosis.

This study was supported by a Gaziantep University project (TF.13.10).

Keywords: Apoptosis, cell cycle, flow cytometer, gastric cancer, gene expression

PP-30

ANALYSIS OF CELL DEATH IN HCT-116 UNDER HYPOXIC CONDITION

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Object: Colorectal cancer is a common malignant neoplasm prevalent in both developed and developing countries. This disease ranks second among causes of cancer-related deaths worldwide, comprising 10%–15% of all forms of cancer. Hypoxia is a common feature of the microenvironment of solid tumors. Hypoxia-regulated genes are known to be involved in multiple biological processes, such as proliferation, angiogenesis, metabolism, immortalization, migration, carcinogenesis and apoptosis expression. Apoptosis play an important role in the involvement of hypoxia. In this study, we aim to identify the analyses of cell death in hypoxia condition on human colon carcinoma cell line.

Material and Method: Human colon carcinoma cell line (HCT-116) was purchased from ATCC. The cells were cultured in RPMI-1640 medium supplemented with 10% fetal bovine serum, 2 mM L-glutamine and 1% penicilin-streptomycine 37◦C in a 5% CO2 humidified atmosphere. The cells were passaged after reaching 80% monolayer confluency and separated in two groups. Group 1 cells were cultured in normal condition, Group 2 cells were cultured in hypoxia chamber which have a gas mixture of 5% CO2, 3% O2 and 92 % N2 for 36 h. They were then fixed with 4% paraformaldyde and cell death analyses were performed with TUNEL. The distribution of anti-Bax, anti-Bcl2, anti-caspase 3 and anti-cytochrome-c were investigated using indirect immunoperoxidase staining. Results TUNEL positive cells were detected in both group 1 and group 2. In group 1, the distribution of caspase 3, Bcl2 and cytochrome-c were observed as a weak staining, they were more detectable in group 2. In addition, positive immunoreactivity of Bax was detected only group 2.

Conclusion: The sensitivity of HCT-116 cells is associated with the upregulation of Bcl-2, Bax, caspase-3 and cytochrome-c activities in hypoxic condition. The hypoxia may trigger the expression of anti-apoptotic protein bcl- 2 to prevent the cell death, and also, intrinsic apoptotic pathway was induced to controlling of cell death

Keywords: Colon carcinoma, hypoxia, apoptosis

PP-31

INVESTIGATION OF THE MDM2 GENE POLYMORPHISM IN PATIENTS WITH ACUTE MYELOID LEUKEMIA

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Aim : Leukemia is malign disease which originated from bone marrow lymphatic and hematopoietic stem cells. Acute myeloid leukemia is a class of leukemia which is shown that both of phenotypic and genotypic heterogeneity. This disease is descripted over all 100 cytogenetic aberattion and vary genes mutation. MDM 2 (Murinedoubleminute 2) gen is a proto-oncogen and MDM2 gen polymorphisms which studiedsome different cancer types and this is associated with lots of cancertypes.

Method: This study is research of the relationship between occuring MDM2 gen 354 A/G and (MboII) -410 T\G (SNP 309) single nucleotid polymorphisms and AML. MDM2 gen polymorphisms one of them is 354 A\G region which Adenine nucleotide convert to guanine nucleotide and the other one is -410 T/G region which Timin nucleotide convert to guanine nucleotide are determined in this study.

Result: For that reason 80 AML patients who have diagnosted AML and 20 healthy people are used in this study. DNA is isolated from AML patients and control group's blood. Both of these region are worked by RT-PCR. Data analyses of 354 d A/G polymorphism is determine that all of the people who are inclueded in this study have AA genotype.

Conclusion: Compared of MDM2 gen 354 A/G polymorphisms between patient and control groups have not found meaningful by istatistically ($p<0,005$). -410 T\G region polymorphism result is found that the 19 wild type (TT) of 80 AML patients, 25 heterozygote genotype (TG) AND 36 patients are mutant type (GG) genotype. At the end of the statistical analysis of MDM2 gen -410 T\G polymorphisms has found statistically significant between two groups ($p<0,05$).

Keywords: MDM2, Acute Myeloid Leukemia, RT-PCR

PP-32

ACTIVATION OF ENDOPLASMIC RETICULUM STRESS MIGHT NOT BE SUFFICIENT TO INDUCE APOPTOSIS IN METASTATIC BREAST CANCER: POSSIBLE ROLE CYTOPLASMIC EXPRESSION OF CHOP.

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Object: Endoplasmic reticulum (ER) is the principal organelle responsible for multiple cellular functions including protein folding, maturation and the maintenance of cellular homeostasis (1). ER stress is part of the unfolded protein response (UPR) which is involved in cell death(2). Markers of ER stres include GRP78, a protein kinase, CHOP and p-PERK which under normal conditions activates apoptotic pathway and may have anti-tumoral effects(2-3). Therefore, the goal of the present study is to determine the expression patterns of ER stress proteins in metastatic breast cancer.

Material and Method: We previously isolated liver (4TLM) and heart (4THM) metastatic as well as non-metastatic 67NR cells formed by 4THM murine breast carcinoma using an orthotopic model (4). 4TLM, 4THM (100.000 cells/mouse) and 67NR (1.000.000 cells /mouse) cells were inoculated into the right upper mammary pad of 8-10 weeks old female Balb-c mice. Necropsies were performed 25-27 days after injection. p-PERK, GRP78, CHOP, Bcl-2 immunoreactivities were examined in primary tumors by using immunohistochemistry. Immunoreaction intensity was quantified using image J software.

Results: We observed the signs of ER stress such that p-PERK, GRP78 expressions were significantly increased in highly metastatic 4TLM primary tumor tissues compared to non-metastatic tumors. Differently ER stress protein CHOP which supposed to be nuclear under stress conditions were localized in cytoplasm(5). It is known that cytoplasmic CHOP in the presence of Bcl-2 expression prevents apoptotic cell death due to ER stress. Interestingly CHOP and Bcl-2 immunoreactivity was higher in non-metastatic tumor groups.

Conclusion: These results demonstrated that ER stress increased in more aggressive metastatic tumors (4TLM) and expression of cytoplasmic CHOP might counteract these apoptotic signals contributing to aggressiveness of 4TLM tumors.

Keywords: ER stress, breast cancer, CHOP, Bcl2, immunohistochemistry

PP-33

CHARACTERIZATION OF THE EFFECT OF SEVERAL ETODOLAC DERIVATIVES ON CANCER CELL LINES

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Object Etodolac is a nonsteroidal anti-inflammatory drug with analgesic and antipyretic properties. It has inhibitory effects on cyclooxygenase-2 (COX-2) activation and similar to other COX-2 inhibitors, it shows anti-tumorigenic effects. We studied anti-proliferative effects of hydrazone and triazole derivatives of etodolac (SGK 206 and SGK 242, respectively) synthesized by Dr. Kucukguzel [1] on two breast cancer cell lines, MCF-7 and MDA-MB-231.

Material and Methods Using the MTT colorimetric method, derivatives of etodolac were evaluated in vitro against the breast cancer cell lines, for cell viability and growth inhibition at different doses following 24, 48, 72 and 96 hours incubations. Apoptosis was also evaluated in both cell lines after treatment in various concentrations using the Tali image-based cytometer. In addition, PGE2 concentrations were measured in the conditioned media using PGE2 ELISA kit (Enzo).

Results: We first checked Cox-inhibiting activity of the drugs with PGE2 assay. All drugs inhibited PGE2 release, but derivatives are more efficient in low concentrations. MTT results indicated that etodolac doesn't have toxicity on the cells, even after long incubation periods in 100 µM concentration. Our derivatives, on the other hand, had substantial effect in much lower doses (10 µM). Especially SGK-206 showed a regular dose-response curve, with an IC50 value of 32 µM against the MCF-7 and 38 µM against the MDA-MB-231. Corresponding values for SGK-242 were 12 µM and 19 µM, respectively. Higher concentrations showed toxic effects in both drugs (>25 µM). Apoptosis also increased in both cell lines in a dose-dependent manner.

Conclusion: Our results show that derivatives of etodolac were more effective both in PGE2 inhibition and cytotoxicity. Similarly apoptotic effect of etodolac was very low compared to the other compounds. We are going to extend our studies on the search of low-dose effects (<25 µM) of these drugs in different cancer cell lines.

Keywords: etadolac, breast cancer, apoptosis, cell cytotoxicity.

PP-34

ANTI-CYTOTOXIC AND ANTI-GENOTOXIC EFFECTS OF CONJUGATED LINOLEIC ACID AND WHEY PROTEIN IN ACROLEIN-TREATED RATS

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Object: Our study aimed to determine the effects of conjugated linoleic acid (CLA) or whey protein (WP) on the frequency of micronucleus (MN) and the ratio of polychromatic erythrocytes (PCEs) against acrolein (AC)-induced toxicity in rats.

Material and Method: Thirty-six healthy rats were obtained from the Experimental Animal Center of University of Ondokuz Mayıs in Samsun, Turkey. The study was approved by the Medical Research Ethics Committee (HADYEK 2014/24) of the university. Animals were orally gavaged with CLA (200 mg/kg/day), WP (200 mg/kg/day), AC (5 mg/kg/day), CLA+AC (200+5 mg/kg/day) and WP+AC (200+5 mg/kg/day) six days per week for 30 days.

Results: It was found that AC significantly increased the formation of MN in bone marrow. It also significantly decreased the ratio of PCEs. No significant differences in MN and PCEs values were observed in the animals received the CLA or WP alone compared to the control group. Co-treatment with CLA+AC or WP+AC significantly increased the ratio of PCEs. Although the co-treatment with CLA+AC or WP+AC decreased the formation of MN, the decreases were not found significant when compared to control group.

Conclusion: Our results indicate that CLA and WH may play a beneficial role in reducing the cytotoxicity and genotoxicity induced by AC in rats.

Keywords: Acrolein; Whey protein; Conjugated linoleic acid; Cytotoxicity; Genotoxicity

PP-35

PISTACIA EURYCARPA INHIBITS CELL PROLIFERATION AND INDUCES APOPTOSIS IN COLORECTAL CANCER BY MODULATING APOPTOTIC PATHWAY GENES

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Object: Colorectal cancer (CRC) is the third most common cancer. Its global incidence and mortality have been on the rise. Recent strategy of therapies has involved the use of non-steroid anti-inflammatory drugs and cyclooxygenase-selective inhibitors [1]. The genus Pistacia consists of small trees of the cashew nut family Anacardiaceae and is native to tropical and subtropical Asia where its members have long been cultivated for a variety of uses. The trunk of Pistacia species produces a characteristic exudate called mastic gum. The mastic gum and oil are medicinally used against rabies, snake bites, baldness, scabies, as well as in prescriptions for stomach, intestine, bladder, and liver inflammations, oral and dental diseases [2]. In this study, growth-inhibiting and pro-apoptotic effects of hexane, chloroform and methanol extracts of Pistacia eurycarpa in HT- 29 colorectal cancer cell line were investigated.

Material and Method: Aerial parts of Pistacia eurycarpa were collected in Bingöl province. Hexane, chloroform and methanol extraction were done by Soxhlet extractor. Dose and time dependent cytotoxic and apoptotic effects of Pistacia eurycarpa were evaluated by MTT Cell Proliferation Kit and Cell Death Detection Elisa Kit, respectively (Roche Diagnostics, Germany). Manufecturer's protocol was followed for analyses. Combination of 5-FU and Pistacia eurycarpa were also applied to HT-29 colorectal cell line for detecting the synergism. pTEN, AKT, MAPK, mTOR, VEGF Receptor 2, p53 and β-actin gene expression levels were measured by RT-PCR. Western blot analyze were used to determine pTEN, AKT, MAPK, mTOR, VEGF Receptor 2, p53 and β-actin protein levels.

Results: According to Cell viability rates, gene and protein expression levels results, there is a synergism between Pistacia eurycarpa and 5-FU.

Conclusion: In conclusion, Pistacia eurycarpa extracts represents a potential source for anti-proliferative and apoptotic agents in combating CRC.

References: [1] KWOK, A. H. Y., WANG, Y., & HO, W. S. (2016). CYTOTOXİC AND PRO-OXİDATİVE EFFECTS OF IMPERATA CYLİNDRİCA AERİAL PART ETHYL ACETATE EXTRACT İN COLORECTAL CANCER İN VİTRO. PHYTOMEDİCİNE. [2] DEMİRCİ, F., BASER, K.H.C., CALİS, I., & GOKHAN, E. (2001). CHEMİSTRY OF NAT COMP, 37(4), 332-335.

Keywords: Colorectal cancer, Pistacia, extract, MTT, p53

PP-36

ANTI-CANCER ACITIVITY OF A HYDRAZONE DERIVATIVE OF ETODOLAC IN K562 LEUKEMIA CELL LINES

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Object: Etodolac is a nonsteroidal anti-inflammatory drug that selectively inhibits Cox-2 enzyme. Cox-2 inhibitors have been reported to have anti-tumorigenic effects on cancer cells through inhibition of proliferation and induction of apoptosis. Hydrazones constitute an important class of compounds for new drug development. Studies implies that hydrazide-hydrazone derivatives of anti-cancer drugs could be more effective and less toxic.

Here, we studied anti-proliferative effects of a hydrazone derivative of etodolac (SGK 205) on leukemia cell line. Here we report its effects on myelogenous leukemia cell line K562.

Material and Methods: Anti-proliferative effects of the drug was determined using MTT colorimetric method, at different doses following 24 and 48 hour incubations. Apoptosis was evaluated by two different methods, where cells were either stained with Annexin-V/PI and analyzed using the Tali image-based cytometer or with JC-1 probe, which was used to detect mitochondrial membrane potential changes.

Results: There was a dose-dependent decrease in proliferation detected by MTT assay and IC50 values were 21.3 and 17.7 µM at the end of 24 and 48 hour incubations, respectively. Apoptosis assays also displayed a dose-dependent increase until 40 µM of the drug. After this concentration drug was highly toxic for the cells. Etodolac didn't show neither anti-proliferative nor apoptotic effects at indicated doses and incubation times.

Conclusion: Several etodolac hydrazone derivatives were synthesized by Dr.Küçükgüzel et al. and cytotoxic effect of compound SGK-205 was tested at a concentration of 10 µM against the full panel of 60 human cancer cell lines by the National Cancer Institute (NCI). These preliminary results displayed a strong effect on leukemia cell lines.

Our further characterization of the compound, this study showed that SGK-205 had indeed anti-proliferative and apoptotic effects on K562 cells in a dose-dependent manner, but concentrations higher than 40 µM results in toxicity.

PP-37

MYC INHIBITION MAY BE A PROMISING THERAPEUTIC STRATEGY FOR A SIGNIFICANT FRACTION OF SCLC

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Object: Small cell lung cancer (SCLC) is the most aggressive type of lung cancer with high mortality. One of the MYC family genes, MYC, MYCL1 or MYCN, is amplified and overexpressed in ~20% of SCLC; therefore, we have selected these MYC family genes as strong candidates of targets for therapy for SCLC, and are investigating the biological significance of MYC family gene amplification for SCLC cells by either down-regulation of MYC family genes or suppression of their activities with inducible shRNAs and siRNAs.

Material and Method: SCLC cell lines were used in this study. To check the effect of Myc expression on cellular proliferation we designed constitutive Myc expression vector by using pCDH-CMV vector as a backbone. After constitutive vector construction, we planned to construct a vector containing an inducible system for the expression of c-Myc by transfering MYC cDNA cloned into pCDH vector into pTRIPZ vector. In the same way, a constitutive system for knockdown of c-Myc by short hairpin expression is not suitable for the evaluation the effect of c-Myc expression by expression profiling. Therefore, we planned to construct an inducible system for knockdown of MYC using pTRIPZ. The effect of Myc expression on cellular proliferation and gene expression profiling were evaluated in Myc overexpressed and knocdown SCLC cells.

Results: Constitutive and inducible expression of Myc induced cellular proliferation in Myc non-amplified SCLC cells. Importantly, c-Myc downregulation markedly reduced the enhanced proliferation of SCLC cells with ectopic expression of c-myc. And in MYC overexpressed cells we evaluated inhibitory effects of shRNAs on cellular proliferation. Due to changes in Myc expression, we evaluated the changes of some proliferation and apoptosis markers by western blot and qRT-PCR.

Conclusion: The results indicate that SCLC cells are addicted to Myc proteins for their growth and, therefore, are highly sensitive to MYC inhibition.

Keywords: SCLC, MYC, shRNA, apoptosis, proliferation

PP-38

EFFECTS OF BOROTUNGSTATE AGENT ON CELL SURVIVAL AND MORPHOLOGY IN NON SMALL LUNG CANCER CELL LINE

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Object: Despite of clinical treatments, lung carcinoma is one of the most common causes of death by way of cancer. Recent studies demonstrated that polyoxometalates (POMs) exhibit potent antitumor activity. Borotungstate (K16[Ni(H2O)6]2[BW12O40]4.48H2O) is a POM. It's a compound containing boron atom which was previously synthesized in our lab. Since the function of borotungstate has not been defined yet, we decided to test the possible proliferative and morphological effect on human. non small cell lung (H460) carcinoma.

Material and Method: The cells were inoculated at a concentration of 1x104 cells/well. Then they were treated with 10, 25, 50, 75 and 100µM borotungstate for 24 or 48 hr. At the end of incubation periods, cell survival was measured with MTT assay. In this study we used 10, 25, 50, 75 and 100 µM doses of carboplatin as positive control for 24 and 48hr. Furthermore, the cells were treated with 10 to 100 µM borotungstate doses for 24 hr and morphological changes were detected under an inverted light microscope.

Results: Borotungstate decreased the survival of H460 cells in a dose and time dependent manner. The half maximal inhibitory concentration values of borotungstate were determined as 51 µM for 24 hr and 31 µM for 48 hr respectively. Borotungstate treatment of 25 µM decreased cell counts 50, 75, and 100 µM doses changed the cell morphology and declined the cell counts.

Conclusion: In this study a new synthetic borotungstate agent, examined for the first time in a human non small lung cancer cells and obtained some preliminary results about the cytotoxic and morphological effects of this agent

Keywords: Borotungstate, MTT, cell morphology, lung carcinoma

PP-39

APOPTOTIC EFFECT OF FERULIC ACID ON DU145 PROSTATE CANCER CELLS

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Object: It is known that phenolic compounds are important phtochemicals to have the strong activities. However, information about their anticancer activity including cell cytotoxicity, antiproliferation and apoptotic effects on cancer cells is limited. Hence, in present study, we aim to determine of anticancer activity of ferulic acid (FA), which is synthesized naturally in plants, on prostate cancer cells DU145.

Material and method: MTT test was used to determine the antiproliferative and cytotoxic effects of FA on DU145 cells. 0.2, 1, 5, 25 µg/ml doses of FA were used for application. After the application of doses of FA, apoptosis were determined by using a FACS with staining Annexine V and Propodium iodide (PI). Then, apoptotic genes; caspase 9, caspase3 and Bax, and antiapoptotic genes; Bcl-2 and Bcl-Xl expression levels were determined by Real Time PCR.

Results: According to our results, cytotoxic effect on DU145 cell was only observed in 25 µg/ml dose of FA. However, any of the application doses showed an apoptotic effect on DU145 cells. They exhibited a necrotic effect rather than the apoptotic effect. Although alterations in expression levels of caspase 9, caspase3, Bax, Bcl-2 and Bcl-Xl genes were determined, this alterations was statistically insignificant.

Conclusion: Consequently, we can say that FA have not a apoptotic effect on DU145 prostate cancer cell lines.

Keywords: Ferulic acid, antiproliferation, apoptosis, Caspas, Bcl-2,

PP-40

CAN ARZANOL, A POTENT MPGES-1 INHIBITOR, USE AS A TARGET FOR PRIMARY OR METASTATIC BREAST CANCER CELLS?

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Arzanol is a prenylated heterodimeric phloroglucinyl α-pyrone chemically known as 3-(3-acetyl-2,4,6-trihydroxy-5-(3-methylbut-2-en-1-yl)benzyl)-6-ethyl-4-hydroxy-5-methyl-2H-pyran-2-one. Prostaglandin E2 (PGE2) is a bioactive lipid that can elicit a wide range of biological effects associated with inflammation and cancer. In our study, the effects of Arzanol on primary and metastatic breast cancer cell lines via caspase-3 expressions were investigated. Human primary (MCF-7) and metastatic (M4A4) breast adenocarcinoma cell line were cultured in RMPI-1640 containing 10% FCS, 1% L-glutamine and 1% penicillin-streptomycin until 80% of confluence. Cells were separated in two groups; control and Arzanol treated groups. Control group of MCF-7 and M4A4 cells were cultured with normal culture medium. In Arzanol treated group of MCF-7 and M4A4 were cultured with different dosage of Arzanol (0.5 µg/ml, 10 µg/ml, 20 µg/ml and 40 µg/ml) for 96 hours. Cell cultures were collected for LDH assay. After fixation of cells with 4% paraformaldeyde, distribution of caspase-3 was detected with indirect immunoperoxidase techniques. After LDH analyses, Arzanol was more cytotoxic in MCF-7 cells when compare with M4A4 cells. In addition, toxicity was higher in 40 µg/ml dosage of Arzanol. Moderate or strong caspase-3 immunoreactivity was detected all dosage of Arzanol in M4A4 cells. In control group of MCF-7 and M4A4, caspase-3 immunoreactivity was negative. Arzanol was induce caspase-3 secretion in M4A4 cells, metastatic breast adenocarcinoma cells, than MCF-7, primary adenocarcinoma cell line. In addition, Arzanol was cytotoxic for MCF-7 cells, therefore weak caspase-3 immunoreactivity was observed.

Keywords: Breast carcinoma, Arnazol, caspase-3

PP-41

THE EFFECTED GENES AND PATHWAYS BY USNIC ACID INSIDE THE CELL

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Object: Usnic acid (UA) is a dibenzofuran derivative metabolite found in lichen. It has anticancer properties in many cancers such as colon, lung, breast cancer and leukemia. But the mechanisms of anticancer effect is still unknown. Singh at all showed that UA stopped the growth of A549 lung cancer cells and made them to undergo apoptosis. In our study, we tried to find out which molecules were effected during apopitosis by using UA on A549 cells. So we could explain the intracellular mechanisms of UA; thought to be an ideal antitumoral agent, on molecular basis for the first time in the literature.

Materials and Methods: A549 cells were obtained from ATCC. Toxic dose of the UA is determined using the "MTT cell proliferation assay" method. The cultured cells were used for RNA isolation and than cDNA synthesis. The gene expression analyses were done for Procaspase 3, Bcl-xL and BAX genes by RT-PCR.

Results: According to MTT results, the dose of UA that we used in our study (50-100 µM) was determined to be non-toxic. In our study, Procaspase 3 gene expression was found as 0,003 in the control group and it was found same as 0.003 in the 50 µM concentration group and 100 µM concentration group. The gene expression levels of Bcl-XL were found as 0.19 in control group, 0.55 in the 50 µM concentration group and 0.2 in 100 µM concentration group. In control group BAX gene expression was found as 0.98 in the control group while it was found 0.64 in the 50 µM concentration group and 1.57 in the100 µM concentration group.

Conclusion: According to the results, the doses of UA that we used in our study, has no toxic effect on A549 cells. According to the gene expression analysis, no change was showed in gene expression of Procaspase 3 between three groups. Bcl-XL gene expression increased in the 50 µM concentration group. The gene expression levels were found similar in the 100 µM concentration group and control group. BAX gene expression decreased in the 50 µM concentration group compared to the control group and increased in the100 µM concentration group. Consequently, while UA was effective on the genes of the final pathway of apoptosis (Bcl-XL and BAX genes), was not effective on Procaspase 3 gene known as involved in the mitochondrial pathway of apoptosis. Our findings indicate that UA is effective through apoptotic pathways within the cell.

Keywords: Usnik asit, akciğer kanseri, apopitoz, gen ekspresyonu

PP-42

THERAPEUTIC APPROACHES FOR PANCREATIC CANCER STEM CELLS

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Pancreatic cancer (PaCa) is cur¬rently the fourth most frequent cause of can¬cer-related deaths in the US.Pancreatic Ductal Adenocarcinoma (PDAC), the major histological subtype comprising 90% of all pancreatic cancers, displays local invasion and metastasis during early stages of the diseasevia developing intrinsic resistance to most terapeutics, contributing to itsnotoriously poor prognosis (~6 monhts of median survival with 1-5% 5-year survival rates). PDAC is a highly complex malignancy due to many molecular alterations, including Mutated KRAS (~90% of the cases), TP53, TGF-β, hedgehog and Wnt signaling pathways. Given that cancer stem cells (CSCs)have a crucial role not only in tumor initiation and progression but also recurrence of the disease, they are proposed to be excellent targets for effective novel therapeutic approaches. Here, we presented the recent therapeutic strategies targeting pancreas CSCs such as chemotherapeutic drugs, miRNAs, immunotherapy and particularly natural compounds

PP-43

THE EFFECTS OF ALPHA LIPOIC ACID AS A POWERFUL ANTIOXIDANT IN BUSULFAN TREATMENT FOR CHRONIC MYELOID LEUKEMIA

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Purpose: Chronic myeloid leukemia (CML)is a myeloproliferative disease causing leukocytosis in pheripheric blood which is characterized by uncontrolled clonal proliferation and decreased apoptosis of myeloid stem cells. Busulfan has been used for CML as a conventional chemotherapy. Alpha lipoic asid (ALA); is the unique antioxidant that is soluble both in lipid and water. ALA is named as a global antioxidant because of its role in preventing of radical metabolite production, clearing of free radicals, repairing of damage cells, inhibition of chain reactions which produce secondary radicals and increase endogenous antioxidant capacity. Busulfan, has several side effects such as testis damage and ALA is a candidate protective agent for this side effect. The aim of this study is to search the effect of ALA, both alone and in combination with busulfan on K562 CML cell line, in order to evaluate whether ALA would interact the antitumoral effect of busulfan.

Method: CCL-243 (K562) human cells were cultured at 37°C 5% CO2 with RPMI. Then ALA alone (0.05-1 miliM) and Busulfan (20-200 microM) were applied. After that optimized dosage for ALA and busulfan is determined they were applied as a combination in these concentrations (0,5 mM ALA ve 40 µM Busulfan). After 24 hours, cell viability, necrosis and apoptosis was measured by using Annexin-V-PI and differences between groups were analyzed by Mann-Whithney-U test with SPSS 15.0 program.

Results: Busulfan and ALA decreased viability of cells in a dose dependent manner ($p<0.05$). Combination of ALA and Busulfan did not change the viability of cells compared with busulfan. Conclusion: In this study; single ALA application to tumor cells, did not proliferative CML cells. Besides it showed cytotoxic effect. When it was combined with busulfan, it did not affect the cytotoxic effect of busulfan. Our next step will be evaluating side effect protective capacity of ALA against busulfan testis damage in animal models

Keywords: Chronic myeloid leukemia, Busulfan, Alpha lipoic acid

PP-44

DIFFERENTIAL EXPRESSIONS OF SIRTUIN FAMILY OF GENES IN BREAST CANCER

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Object: Mammalian sirtuins have been shown to perform distinct cellular functions and play crucial roles in a variety of cellular processes including regulation of cell death. Also, deregulated expression of these genes was reported to be involved in development of various malignancies including breast cancer. An increasing number of evidence indicates that sirtuins have both tumor promoter and tumor suppressor functions. However, the roles of sirtuins have not been well-reported in breast cancer. Accordingly, in the present work, we aimed to reveal the role of Sirtuin family of genes in breast cancer.

Material and Method: In the present study, quantitative expression levels of sirtuins (SIRT1-7) in breast cancer patients and breast cancer cell lines (MCF-7 and SKBR3) and control cell line (CRL-4010) were assessed by using a high-throughput real-time PCR method.

Results: As a result, sirtuins was found to be differentially expressed in breast cancer tissues and cancer cell lines. Particularly, expressions of SIRT1 and SIRT4 were found to be significantly down-regulated in breast cancer tissues and SKBR3 breast cancer cells. In contrast, SIRT2, SIRT3, and SIRT5 genes were shown to be up-regulated in our study. Although SIRT6 and SIRT7 were also up-regulated in breast cancer tissues, these expression changes were statistically insignificant. Additionally, SIRT2, SIRT3, SIRT5, SIRT6 and SIRT7 were found to be differentially expressed in breast cancer cell lines. Yet, these changes were not well-correlated with tissue expression levels.

Conclusion: In conclusion, sirtuins family of genes shows differential expressions in breast cancer tissues and cells and SIRT1 and SIRT4 seems to play key tumor suppressor roles in breast cancer development.

Keywords: Breast Cancer; Cancer Cells; Expression; Sirtuins; SIRT; SIRT1

PP-45

PAPER BASED MOLECULAR DIAGNOSIS OF HUMAN PAPILLOMAVIRUS AT THE POC

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Object: Cervical cancer is a major problem for the developing world and low-resource settings where standard screening techniques are not accessible. The current detection methods require complex and expensive equipment which are not accessible in low-resource settings. Early detection is critical for cervical cancer, as it is one of the few cancers that can be successfully treated when caught early. We propose to create a point-of-care (POC) paper microfluidic chip that provides isolation, non-enzymatic amplification and detection of human papillomavirus (HPV), the etiological agent of cervical cancer.

Material and Method: First, cervical cells lysed, releasing HPV DNA that was extracted via paper matrices. The chip does not require enzymatic amplification; electrochemical detection was used instead of the traditional polymerase chain reaction (PCR) that requires silver nanoparticles and magnetic microbeads. Lastly, the Ag modified DNA were detected with a hallow channel included paper device, generating electrochemical readout. The entire process has been completed in less than 30 minutes, allowing doctors to diagnose, advice and potentially treat patients in the same visit.

Result and Conclusion: In Conclusion, I have reported a new paper extraction support material, a novel paper based electrochemical detection device which includes modification of DNA for POC applications of HPV16 and 18. I also applied the method to the Patient samples which are provided from Beth Israel Hospital. It can extract at least 10 copies per µL DNA from patient samples approximately in 10 minutes and can detect low label concentrations as low as 530 FM in only 4.6 min. This device is inexpensive, compatible to simple and rapid fabrication techniques, user friendly, sensitive, quantitative and robust. Asymptomatic patients positive for HPV 16 and 18 could be screened more closely, thus allocating precious resources to those most at risk, a beneficial use in both low resources settings and in the worldwide

Keywords: Human Papillomavirus (HPV), Paperfluidic, Analytical Detection

PP-46

CYTOTOXICITY OF ALOE VERA LEAF LECTIN (ALOCTIN) COMBINED WITH IMATINIB ON AGS HUMAN GASTRIC TUMOR CELL LINE AND SAOS-2 HUMAN OSTEOSARCOMA CELL LINES

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Object: The biological effects of A. vera are due to various chemical compounds including anthraquinones, glycoproteins, polysaccharides, vitamins and enzymes. It has been reported that the lectin of A. vera plays role in many biological activities of A. vera. Furthermore, the anticancer potential of Aloe is also available on many publications. Although, several in vitro, in vivo assays, and clinical studies were undertaken on A. vera, the mechanisms which accompanies the pharmacological effects of the plant have not been clarified. The present study was designed to evaluate possible chemopreventive effects of the recently purified Aloctin and its combination with imatinib mesylate (IM) in vitro.

Material and Method: The gel portion of leaves of A. vera (L.) Burm. f. was separated from the leaf skin and discarded, a single lectin (named Aloctin) was isolated from leaf skin by ammonium sulphate precipitation, affinity chromatography. The cell proliferation was detected by MTT assay. AGS human gastric tumor cell line and Saos-2 human osteosarcoma cell line were used to evaluate the effects of IM (25µM), Aloctin (0.5µg/ml) and their combination on cell proliferation. The concentrations of Aloctin (0,3 µg/ml – 3 µg/ml) and IM (0,1µM – 100 µM) at which concentration causing 50% inhibition of the cell proliferation (IC50) were calculated.

Results: The results indicated significant cytotoxic activity of Aloctin alone with an IC50 of less than 0.3 µg/ml, and of IM alone with an IC50 of 65 µM and 108 µM for AGS and Saos-2 cells respectively, while the combination group induced highest decrease in cell proliferation.

Conclusions: In this study, we showed that the combination of Aloctin with IM was able to enhance cytotoxic effect in vitro. Our results suggest that the Aloctin could be considered as a chemotherapeutic agent which may help to reveal important findings for the future use of Aloctin in cancer research by the drug combination.

Keywords: Aloe vera lectin (Aloctin), Imatinib mesylate, Cytotoxicity, AGS human gastric tumor cell lines, Saos-2 human osteosarcoma cell lines.

PP-47

IN VITRO EVALUATION OF HPV STATUS AND ITS RELATION WITH TREATMENT STRATEGIES IN HEAD AND NECK CANCERS

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Purpose: To compare the treatment responses of primary cell culture HPV positive and HPV negative head and neck cancers to radiotheraphy and chemotheraphy and to determine the optimal treatment modality depending on the HPV status of the tumor.

Method: Tumor resection samples were obtained from 34 patients with head and neck cancer. Single cell suspensions were acquired from tumor samples and cultivated in RPMI 1640 medium supplemented with 10% FBS, %1 penicillin/streptomycin, 1% L-glutamine and 1% fungisomel. 8 subgroups were planned for each patient's primary cell culture as control, cisplatin, cetuximab, combination of cisplatin +cetuximab and their 15 Gy radiotherapy combinations. Cell proliferation (BrdU), DNA damage (H2AX) and apoptosis (PARP) levels were analyzed after 24 hours by flow cytometry. Expression of HPV 16 and 18 was also detected by Real time PCR. Data was statistically analyzed considering clinical outcomes of patients.

Results: There were 24 male and 10 female patients with a mean age of 58.44 (23-80). Tumor localisation was oral cavity in 10 patients, oropharynx in 4 patients, larynx in 14 patients and hypopharynx in six patients. HPV 16 was positive in 5 cases (14.7%). In vitro evaluation of cytotoxic, apoptotic and DNA damage among HPV16 positive and negative cases did not show statistically significant difference in cisplatin, cetuximab, combination of cisplatin and cetuximab and their 15 Gy radiotherapy combinations (p:>0.05) . There was statistically significant difference for cell proliferetion index, apoptosis and DNA damaged cell percentages among HPV 16 positive and negative cases in 15 Gy radiotherapy alone group. Radiotherapy alone caused more cell death, apoptosis and DNA damage in tumor cells of HPV16 negative group.

Conclusion: In this in vitro study, chemotherapy and radiotherapy sensitivity of primary cancer cell culture of head and neck cancers were analysed and compared among HPV positive and negative cases by proliferation (BrdU), DNA damage (H2AX) and apoptosis (PARP) methods. Our results indicate that cisplatin and cetuximab did not have an additional effect in both groups and radiotherapy was found to be more effective in HPV negative cases.

Keywords: Head and neck cancer, HPV, treatment strategies

PP-48

EFFECTS OF TESTOSTERONE PROPIONATE ON BONE MARROW DERIVED MESENCHYMAL STEM CELLS' PROLIFERATION AND VIABILITY

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Object: Mesenchymal stem cells (MSCs) can be found in various tissues such as adipose tissue, bone marrow and umblical cord; they have CD73, CD90 and CD105 surface markers whereas they lack hematopoietic markers; they are able to attach plastic surfaces; they can differentiate and have self renewal capacity. There are various studies, indicating effects of testosterone on different cell types (both healthy and cancer cells). But currently effects of testosterone on mesenchymal stem cells are unknown. In this study, we aimed to show effects of testosterone on human bone marrow derived mesenchymal stem cells after 24 hours of incubation.

Material and Methods: We expanded commercially available MSCs in vitro and added a short-acting form of testosterone; testosterone propionate (TP) at specific concentrations. We used Annexin V – Propidium Iodide viability and apoptosis test for detecting viability and CFSE staining for detecting proliferation. Measurements were performed on flow cytometry.

Results: This study shows that testosterone promotes proliferation in a dose dependent manner. Whereas 10^{-8} M TP showed highest proliferation promotion and highest increase at viability. Doses over this concentration were found to be toxic for MSCs.

Conclusion: Our study is the first to show effects of testosterone on MSCs' viability and proliferation. Testosterone as a culture media supplement may help to shorten the culture period for MSCs. Our findings may also help to future treatment studies of diseases which are thought to be linked with mesenchymal stem cells.

Keywords: Mesenchymal Stem Cells, Testosterone Propionate, Cell Culture, Proliferation, Cell Death

PP-49

THE EFFECT OF BONE MARROW MESENCHYMAL STEM CELL APPLICATION ON CASPASE 3 ENZYME IN RATS WITH CRUSHED MUSCLE DAMAGE.

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Aim: In this study, the effect of mesenchymal stem cell application on Caspase 3 Enzyme in rats with crushed muscle damage was researched.

Material and Methods: In this study, the right soleus muscles of 56 female Wistar Albino rats were used. The animals were randomly divided as two experimental groups (a Muscle Damage Group (MD) and a Muscle Damage + Mesenchymal Stem Cell Group (MD+MSC)) and a control group (C). The experimental groups were divided into the sub-groups of the 7th, 14th and 21st day (each sub-group n=8). The group from which tissue samples were obtained without generating muscle damage was used as the control group (n=8). Tissue samples were taken from each experimental group on the 7th, 14th and 21st day and their Caspase-3 activities were measured. The tissue samples were examined by means of immunohistochemical dyeing.

Results: When the Caspase 3 activities values of the groups were compared with the control group, the increase in the Caspase-3 activities in the MD+MSC on the 14th day and 21st days were found statistically significant. There was a statistically significant increase in the Caspase-3 activities of the MD on the 21st day when compared to MD+MSC on the 21st day. In all the MD+MSC sub-groups, the existence of MSCs with green fluorescent protein (GFP) and that they transformed into muscle cells were observed.

Conclusions: In addition, the fact that the Caspase-3 activity increased in the MSC group on the 14th and 21 st days can be interpreted as the fact that the stem cells replaced the crushed cells and transformed into muscle cells; and that the crushed cells turned into apoptosis.

Keywords: Stem Cell, Caspase-3 , Crushed Muscle Damage

PP-50

EFFECT OF TGF-B1 OVEREXPRESSION ON BIOLOGICAL CHARACTERISTICS OF HUMAN DENTAL PULP DERIVED MESENCHYMAL STEM CELLS

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Object: In our study were investigated how the effected the surface markers, multilineage differentiation, viability, apoptosis, cell cyle, DNA damage and senesence of TGF-β1 gene therapy on human dental pulp derived mesenchymal stromal cells (hDPSC).

Material and Methods: hDPSCs were isolated from human teeth and cultured with %20FBS in α-MEM. The plasmid TGF- β1 (Sino Biological Inc, China) was amplified in Escherichia coli host strain DH5α and purified by plasmid isolation with the Endo-Free Maxiprep Plasmid Isolation Kit. TGF- β1 gene transfer was performed into hDPSCs by electroporation method after the plasmid was prepared. For transfection efficiency were performed western blot and flow cytometry analyses and GFP transfection. MSC markers, multilineage differentiation, cell proliferation, apoptosis, cell cycle, DNA damage and cellular senescence were performed to compared transfected and non-transfected cells. Statistical analyses were performed using GraphPad Prism version 6.00 for windows (GraphPad Software, San Diego California USA).

Results: Flow cytometry analysis detected strong expression of TGF-β1 in pCMV-TGF-β1-transfected hDPSCs. TGF-β1 transfection efficiency was measured as 95% (Fig 1D). Also, western blot analysis shown that TGF-β1 protein levels increased at third and sixth days in pCMV-TGF-β1-transfected hDPSCs. The continuous TGF-β1 overexpression in hDPSCs did not influence the immunophenotype and surface marker expression of MSCs. Our results showed that increased osteogenic and chondrogenic differentiation of TGF-β1 but reduced to adipogenic differentiation. Overexpression of TGF-β1 was increased proliferation and decreased total apoptosis in hDPSCs ($p<0,05$). TGFB1 transfection was increased the number of cells at 'S'phase ($p<0,05$). There was no significant difference at DNA damage. Cellular senescence decreased in TGFB1 transfected group ($p<0,05$).

Conclusion: These results reflect that TGF-β1 has major impact on MSC differentiation. TGF-β1 transfection has no effect on cell surface markers but differentiation. TGF-β1 transfection has positive effect on proliferation, cell cycle and prevents cellular senescence and apoptosis.

Keywords: Dental pulp derived Mesenchymal Stem Cells, TGF-β1 Transfection, Apoptosis, DNA Damage,Cellular Senescence

PP-51

MONOSODIUM GLUTAMATE HAS NO CYTOTOXIC EFFECT ON MOUSE MESENCHYMAL STEM CELLS

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Aim/Backround: Monosodium glutamate as a food additive is a substance added to food to preserve flavor or enhance its taste and appearance. Monosodium glutamate is the sodium salt of glutamic acid, one of the most abundant naturally-occurring non-essential amino acids. U.S. Food and Drug Administration reported that monosodium glutamate is safe for use in food. However, the excessive use of monosodium glutamate causes dizziness, nausea and vomiting. Due to reports, it causes obesity. We aimed to find out the cytotoxiciy of monosodium glutamate and the role on the selected genes, which have role on obesity.

Methods: In our study, we used mice which are known to be the most sensitive in terms of toxicity. We were studied the cytotoxic effects of monosodium glutamate on mouse mesenchymal stem cells at monosodium glutamate doses below. Also we analyzed leptin-lep and ghrelin / obestatin in prepropeptide-GHRL gene expressions in order to find out the role of monosodium glutamate in obesity.

Result: Monosodium glutamate below the toxic dose does not have a cytotoxic effect on mouse mesenchymal stem cells. Also no expression change observed in genes which are known to be associated with obesity.

Conclusion: Our results support that monosodium glutamate has no toxic effect on stem cells in uses in certain doses.

Keywords: Monosodium glutamate, mouse mesenchymal stem cells, leptin gene, ghrelin/obestatin prepropeptide gene.

PP-52

ALTERED EXPRESSION OF APOPTOSIS-RELATED GENES IN CANCER STEM CELLS IN A PROSTATE CANCER MODEL

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Objective: In contrast to low-tumorigenic bulk tumor cells (non-CSCs), cancer stem cells (CSCs) are a subset of tumor cells with the potential to self-renew and differentiate into different cancer subtypes. CSCs are thought to be responsible for tumor recurrence, distant metastasis, angiogenesis, and drug/radiation resistance. Understanding the properties and characteristics of CSCs is key to future study on cancer research, such as the isolation and identification of CSCs, cancer diagnosis, and cancer therapy. The present study focuses on the differences of expression levels of apoptosis-related genes between CSCs and non-CSCs, using prostate CSCs as a model system.

Material and Method: Cluster of differentiation (CD) 133+high/CD44+high prostate CSCs were isolated from the DU145 human prostate cancer cell line by flow cytometry cell sorting. Total RNA from both CSCs and non-CSCs was isolated and extracted followed by cDNA synthesis. The qRT-PCR array was used to measure the expression levels of apoptosis-related genes.

Results: Significant up-or down-regulation in gene expression was detected in 18 genes in the prostate CSCs compared with non-CSCs. The studied genes could be classified as Caspase, BCL-2, IAP, CARD, TNFR families and genes involved in P53 signaling, NF-Kb signaling and FAS signaling pathways. By comparing the gene expression levels between the two groups, more significant differences were mainly detected in the FAS, BIRC3, BCL2L11, FADD, TRAF5, HSP90B, HMGB1 and FAM96A genes. In addition, it was found that several anti-apoptotic genes including BIRC3 and FAS were greatly up-regulated in CSCs, revealing an association with the high survival rate of these cells. Conclusion: There are some differences in the expression profile of apoptosis related genes in CSCs compared to non-CSCs. These findings may provide key information for more effective therapeutic approaches.

Keywords: Cancer stem cell; Apoptosis; Gene expression; Prostate cancer

PP-53

THE POTENTIAL EFFECTS OF PROPOLIS ON APOPTOSIS AND CELL CYCLE PATHWAYS IN HER2-POSITIVE BREAST CANCER CELLS

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Object: In this study, we aim to investigate the effects of propolis on ER/PR-, HER2/neu+ human breast cancer cell line SKBR3 with intend to clarify the efficiency of propolis in HER2+ breast cancers in overview of whole genome expression for the first time.

Material And Method: Eight different samples were collected from different regions, including China, Argentina and Turkey. Propolis samples were dissolved in ethanol and water solutions. The SKBR3 (HER2 + breast cancer cell) and MCF10A (normal breast epithelial cell) were incubated for 24, 48 and 72 hours after the addition of different concentrations of propolis extracts. Cell growth and cytotoxicity levels were measured by WST-1 assay. Apoptotic cell death rates were determined by flow cytometry using Annexin V-FITC. Microarray analysis was performed to examine the gene expression profiles by using Gene-Spring software.

Results: Propolis extract from Turkey, 50 µg/ml and 48th hour determined as the most effective extract, dose and time conditions which were selected according to decrease in proliferation and increase in apoptotic effects of SKBR3 cancer cells. At this point for the whole genome, we determined more restricted cutoff ($p < 0.01$), totally 248 genes were significantly regulated, among which 91 were up-regulated and 157 down-regulated in SKBR3 cells at the 48th hour. We detected that altered expression levels of 7 genes on cell cycle pathway and 3 genes on apoptosis pathway had a statistically significant difference. Furthermore, no such alteration was detected in MCF10A cell line under the same conditions.

Conclusion: We exhibited that HER2-positive breast cancer cells are reduced proliferation changing expression levels of cell cycle markers and increased apoptosis with altered expression levels of apoptotic and antiapoptotic markers by İstanbul-Kartal propolis. These data provide a potential mechanistic basis for one of the oldest natural agents used in medicine and cancer treatment.

Keywords: Propolis, HER2-Positive Breast Cancer, Apoptosis, Cell Cycle, Microarray

PP-54

FERULIC ACID DECREASES CELL VIABILITY, INVASION, MIGRATION AND COLONY FORMATION IN MIA PACA-2 HUMAN PANCREATIC CANCER CELLS IN VITRO

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Objects: Novel and combinatorial treatment methods are becoming sought after entities in cancer treatment and these treatments are even more valuable for pancreatic cancer. The scientists are always on the lookout for new chemicals to help them in their fight against cancer. In this study, we examine the effects of ferulic acid (FA), a phenolic compound, found in various nutrients, on gene expression, viability, colony formation and migration/invasion in the cultured MIA PaCa-2 human pancreatic cancer cell.

Material and Method: The effect of FA on cell viability was determined by CellTiter-Glo® Luminescent Cell Viability Assay. Expression profiles of certain cell cycle and apoptosis genes such as CCND1, CDK4, CDK6, RB, p21, p16, p53, caspase-3, caspase-9, caspase-8, caspase-10, Bcl-2, BCL-XL, BID, DR4, DR5, FADD,TRADD,PARP, APAF, Bax, Akt, PTEN, PUMA, NOXA, MMP2,MMP9, TIMP1 and TIMP2 were determined by real-time PCR. Furthermore, effects of FA in MIA PaCa-2 cells on invasion, colony formation and cell migration were determined by matrigel chamber, wound-healing test and colony formation assay, respectively.

Results: IC50 dose in MIA PaCa-2 cells was detected as 500 µM/ml at the 72nd hour. Additionally, effects of FA on colony formation and invasion were also investigated. It was observed that FA caused a significant decrease in the expression of CCND1, CDK4/6, Bcl2 and caspase 8 and 10 in the MIA PaCa-2 cells while causing an increase in the expression of p53, Bax, PTEN caspase-3 and 9. FA was observed to decrease colony formation, invasion and migration of MIA PaCa-2 pancreatic cancer cells.

Conclusion: FA is thought to behave as an anti-cancer agent by affecting cell viability, cell cycle, apoptotic, invasion and colony formation behaviour of MIA PaCa-2 cells. Therefore, FA is placed as a strong candidate for further studies aimed at finding a better, more effective treatment approach for pancreatic cancer

Keywords: Ferulic acid, Pancreatic cancer, Apoptotic genes, Cell cycle genes

PP-55

ANALYSING THE CYTOTOXIC EFFECTS OF CERANIB-2 ON HEPATOCELLULAR CARCINOMA CELLS

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Object: Ceramide induces cell death in cancer cells on the contrary sphingosine-1-phosphate leads to cell proliferation. Ceramide/sphingosine-1-phosphate ratio is critical for the viability of the cells and this level is regulated by ceramidase enzymes. Ceranib-2, an anticancer agent is an inhibitor of human ceramidases. Inhibiting of ceramidases cause imbalance in ceramide/sphingosine-1-phosphate ratio. Herein, ceranib-2 was investigated for its cytotoxic effects in human hepatocarcinoma cells Hep3B2.

Material and Method: Stock solution of ceranib-2 was prepared in dimethyl sulphoxide (DMSO; Sigma Aldrich, UK) and further dilutions were made with fresh culture medium in the range of 5-110µM. MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide) colorimetric assay was used to find the cell viability for 24 hours. IC50 value was determined according to the ELISA reader (ELx808) results.

Results: The viability of Hep3B2 cells reduced in dose-dependent manner. In our results it is shown that ceranib-2 exhibited high antiproliferative effects on Hep3B2 cells being cytotoxic in low concentrations.

Conclusion: On the basis of our findings ceranib-2 is found to be a good inhibitör of cell proliferation of Hep3B2 cells and we suggest this agent for further investigation for drug designing for cancer therapy.

Keywords: Human hepatocarcinoma, Liver cancer, Cytotoxicity

PP-56

CYTOTOXIC EFFECT OF MORUS RUBRA EXTRACT ON HUMAN COLON CANCER CELLS THROUGH INCREASED ENDOPLASMIC RETICULUM STRESS AND SUPPRESSED TELOMERASE

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ABSTRACT Object: Cancer cells have some characteristic features, such as evasion of apoptosis, dysregulation of cell cycle control, self-sufficiency in growth signalling, and telomerase activation. Telomerase therefore has an important role in unlimited proliferation of cancer cells, so inhibition of telomerase activity is a potentially highly selective target for cancer treatment. In recent years, induction of endoplasmic reticulum (ER) stress-mediated apoptosis by CCAAT/enhancer-binding protein homologous protein (CHOP) in cancer cells is represented as an alternative approach for cancer therapy. Morus rubra, known as "red mulberry", belongs to family of Moraceae and genus of Morus. It is rich in polyphenols and its biological functions are attributed to these compounds. Many studies have evaluated the cytotoxic effects of different Morus species, but there is no study about cytotoxic effect of M. rubra on colon cancer cells. The aim of this study was to investigate the cytotoxic effect of the M. rubra extract on colon (WiDr) cancer cells in terms of ER stress and telomerase expression for the first time.

Material and Method: The cytotoxic activity of extract was determined by MTT assay. The effect of M. rubra extract on telomerase and CHOP expression was demonstrated by RT-PCR technique. Results: Morus rubra extract exhibited selective cytotoxicity on colon cancer cells compared to normal fibroblast cells. M. rubra extract significantly repressed the telomerase expression at all treatment time (24, 48, and 72 h) in a dose dependent manner in colon cancer cells. Moreover, treatment with M. rubra extract significantly induced CHOP expression at 72 h in these cells.

Conclusion: Our results demonstrated that M. rubra extract induces cytotoxicity in colon cancer cells through, at least in part, increasing ER stress-related signalling and repressing telomerase expression.

PP-57

MORUS RUBRA EXTRACT INDUCES APOPTOSIS AND INHIBITS PROLIFERATION IN HUMAN PROSTATE CANCER CELLS

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ABSTRACT Object: Cancer is a heterogeneous disease, two of whose characteristic features are uncontrollable cell proliferation and insufficient apoptosis. Morus rubra, known as "red mulberry" belongs to family of Moraceae and genus of Morus. In folk medicines, the fruits of Morus species are used to treat many diseases and their biological effects are attributed to their phenolic contents. Many studies have evaluated the cytotoxic effects of different Morus species, but there is no study about cytotoxic effect of M. rubra. In this study, we aimed to evaluate phenolic content, phenolic composition, and cytotoxic effect of M. rubra extract.

Material and Method: Total polyphenol content, phenolic composition, and cytotoxic effect of M. rubra extract were determined using Folin-Ciocalteu method, HPLC, and MTT assay, respectively. Then, mechanisms of cytotoxic effect of M. rubra extract on human prostate (PC-3) cancer cells were examined in regard to cell cycle, and apoptosis using flow cytometric methods.

Results: Total polyphenol content of acidified dimethyl sulfoxide extract of M. rubra was found as 11.9±0.1 mg GAE per to g sample. Ascorbic acid, gallic acid, and epigallocatechin gallate were detected as major antioxidant compounds in the extract. M. rubra extract exhibited selective cytotoxicity against prostate cancer cells compared to normal fibroblast cells. We determined that M. rubra extract increased cell cycle arrest at G1 phase and exhibited apoptotic features in prostate cancer cells.

Conclusion: Our results showed that M. rubra extract has apoptotic and antiproliferative effect in prostate cancer cells. Further studies are needed to better understand the molecular mechanisms underlying this effect of M. rubra extract

Keywords: Apoptosis, Cell Cycle, Cytotoxicity, Morus rubra, Prostate Neoplasms

PP-58

THE EFFECTS OF RESVERATROL GAIN IMPORTANCE ACCORDING TO P53 MUTATION IN HCT116 COLON CANCER CELL LINES

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Cancer is a disease that includes heterogenic and complex molecular changes. Anti-carcinogenic effects of Resveratrol, a natural polyphenol, have been proved in a variety of cancer cells. Considering the effects of Resveratrol, the influence of the signal transduction pathways in the presence or absence of p53 of colon cancer cells is gaining importance. Our aim was to investigate the effects of Resveratrol in the presence or absence of p53 on cell viability, apoptotic cell death ratio and fold changes of proliferative or anti-proliferative gene expressions, which may have important effects on colon cancer, in HCT116 colon carcinoma cells. IC50 doses of Resveratrol were determined by WST-1 assay. The apoptotic cell death ratios in treatments of Resveratrol were determined by Annexin-V-FITC/PI assay for flow cytomety. The changes of CCND1, FRA1, PPARD, EGFR, BIRC5, PCNA, MCL1, STAT3, FOS, JUN, P27, ATF4, TRAIL, PUMA, GADD45A, RB1, FASLG, TNF, SOCS3, STAT1 gene expressions were evaluated by Real Time PCR. All data were statistically analysed by Student's t test. Our research has revealed that Resveratrol (60µM) causes decrease in cell viability and increase in apoptotic cell death in HCT116p53(+/+) and HCT116p53(-/-) cells significantly ($p<0.05$). The fold changes of the gene expressions have shown that Resveratrol has significant ($p<0.05$) and different effects on the expressions of the genes related with the existence of p53 in HCT116 cell lines. Hence we proposed that Resveratrol might show proliferative or apoptotic effects related with p53 mutation of colon cancer cells and we predicted that unconscious consumption of Resveratrol in colon cancer patient might cause adverse effects. This study was supported with Scientific Research Projects Coordination Unit of Dokuz Eylul University with 2013.KB.SAG.055 project number

Keywords: Resveratrol, p53, Colon Cancer, Signal Transduction Pathways

PP-59

THE EFFECTS OF MIRTAZAPINE ON DIABETIC NEPHROPATHY

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Diabetes mellitus is a serious health problem and is associated with severe acute and chronic complications that influence quality of life. Diabetic nephropathy has became the most common cause of end-stage renal disease. Hyperglycemia has been shown to induce in vitro apoptosis of several cells and apoptosis in endothelial cells in diabetic vascular complications. Mirtazapine is a noradrenergic and specific serotonergic antidepressant. It is also known that mirtazapine is shown to inhibit the production of enzymatic/ non-enzymatic oxidant parameters. We aim that how mirtazapine effect on diabetic nephropathy. A total of 21 male Sprague Dawley rats, 8 weeks old and weighting 180-200g, were purchased from Eskisehir Osmangazi University Medical and Surgical Experimental Research Center (Eskisehir, Turkey). 14 rats were randomly selected and injected once intraperitoneally with streptozotocin (55mg/kg). After 72h, blood-glucose levels were measured by glucometer and levels higher than 300mg/dl were accepted for diabetes. After 4 weeks, 7 out of the 14 rats was administered 20mg/kg for 14 days by intragastric gavage. Control group was injected with an equal volume of citrate buffer. In control group healthy cortex layer, negative Caspase 1, high NLRP3 and negative TUNEL reactions were seen. In diabet group Class II b diabetic nephropathy, diffuse expansion of mesangium, tubular atrophy, high expression of Caspase-1 and NLRP3 in glomerular endothelial and tubules and TUNEL positive reaction in glomerul and tubular nucleus were seen. In Mirtazapine group healthy glomerulus, low expression of Caspase 1, mild NLRP3 expression of NLRP3 and TUNEL positive reaction in glomerulus and tubular nucleus (Figure 1). Parallel reactions were seen in western blot too (Figure 2). Our results show that diabetes mellitus induced apoptosis but not by NLRP3 inflamation pathway and mirtazapine administration to the diabetic nephropathy
may be worth considering as a new candidate for preventing cell death.

Keywords: Diabetic nephropathy, mirtazapine, NLRP3

PP-60

TUBULAR CELL APOPTOSIS IN PATHOGENESIS OF TYPE 1 CARDIORENAL SYNDROME AND EFFECT OF ERYTHROPOIETIN

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Objective: Condition of renal failure occurring in heart failure is termed cardiorenal syndrome. This is attributed to hemodynamic derangement, with reduced renal perfusion and increased venous pressure. Erythropoietin (EPO) is a cytokine, with significant homology to mediators of growth and inflammation. We aimed to research early changes in renal tissue after myocardial infarction and effect of EPO on this tissue.

Material and Methods: Coronary artery ligation was performed to induce myocardial infarction in Wistar rats. Rats (n=36) were divided into five groups; Group 1: Sham-operated control rats. Group 2: Rats receiving a single intraperitoneal injection of saline immediately after ligation and sacrificed 6 hours after surgery; Group 3: Rats receiving a single intraperitoneal injection EPO (5000 U kg−1) (Eprex 4000 IU/0.4 mL) immediately after ligation and sacrificed 6 hours after surgery; Group 4: Rats receiving a single intraperitoneal injection EPO (10000 U kg−1) immediately after ligation and sacrificed 6 hours after surgery; Group 5: Rats without treatment sacrificed 1 hour after ligation. We investigated changes with light microscopy, immunohistochemistry (caspase 3).

Results: We observed renal morphology was regular in Group 1 and Group 5. However in Group 2, we observed tubular infiltration (p=0,007), tubular necrosis (p=0,000), tubular dilatation (p=0,015), luminal congestion (p=0,020) with loss of brush border (p=0,000) and spillage of epithelial cells into tubule lümen (p=0,000). Significantly less tubular damage was observed in EPO-treated groups (Group 3 and Group 4).

Caspase 3 immunostaining was observed at 6 hours in renal cortex (p=0,002) (Group 2), at 1 hour in renal medulla (p=0,016) (Group 5). Caspase-3 immunostaining decreased in the medulla of EPO-treated groups (p=0,016) (Group 3 and Group 4).

Conclusion: Cardiac reasons that can cause acute kidney injury influences particularly the medulla in a very short time. EPO treatment promotes a protective effect on the cardio-renal axis, which might be attributed to its anti-apoptotic properties.

Keywords: Erythropoietin, caspase 3, cardiorenal syndrome

PP-61

A NOVEL REGULATOR OF MITOPHAGY IS A PUTATIVE AUTOPHAGY-UPS COORDINATOR PROTEIN

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Autophagy and ubiquitin proteasome systems are the major degredation sytems in mammalian cells for the recycling of cellular contents ranging from soluble proteins to intracellular organelles. Previous studies demonstrated these two independent degredation pathways have compansatory effet on eachother when one of them is inhibited in order to prevent cells from toxicity of the protein aggregates the other pathway is activated. Additionally, recent findings suggest that mitophagy and UPS work coordinatively in order to eliminate dysfunctional mitochondria. However no direct connection between autophagy and UPS has been identified untill now. Due to the role of mitochondrial function in aging and age-related diseases such as Parkinson's disease, Alzheimer's disease and Huntington's disease, understanding the nature of mitochondrial dynamics has great importance. The purpose of the study to introduce for the first time direct link between autophagy and UPS in the regulation of mitochondrial homeostasis. Protein-protein interactions evaluated through colocalization studies, immunoprecipitation and gel filtration technique. Total cell lysates were analysed for in vitro determination of the mitochondrial protein degredation. Subcellular fractionation experiments performed for the determination of the stress-induced recruitment of cytoplasmic proteins to mitochondria. In this study, we show that proteasome shows direct physical interaction with autophagic machinery under mitochondrial stress conditions. The interaction between autophagic machinery and proteasome subunit is important for retranslocation of cytoplasmic proteins to mitochondria.Taken together , our study introduced a regulatory subunit of proteasome as a novel regulator of mitophagy and an interactor of ATG5, and described as a direct coordinator of mitochondrial clearance and UPS. These findings might have implications for proteinopathies involving mitophagy defects, including Parkinson's Disease.

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Keywords: Autophagy, Proteasome, UPS, MItochondrIa, MItochondrIal DynamIcs

PP-62

AUTOPHAGY TARGETS AN IBMPFD-RELATED VCP/P97 MUTANT PROTEIN

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Here, we studIed the role of an IBMPFD-related mutant of VCP/P97 (P137L mutant) In autophagy. In contrast wIth wIld-type VCP/P97 or commonly studIed mutants (R155 or R191), the P137 mutant was aggregate-prone. We showed that, unlIke other mutants, the P137 mutant proteIn stImulated both autophagosome and autolysosome formatIon. Moreover, P137 mutant proteIn Itself was a substrate of autophagy. StarvatIon- and mTOR InhIbItIon-Induced autophagy led to the degradatIon of the P137 mutant, whIle preservIng wIld-type and functIonal VCP/P97.Our results IndIcate that cellular mechanIsms leadIng to IBMPFD dIsease may be varIous, and underlIne the Importance of studyIng dIfferent dIsease-associated mutatIons to better understand human pathologIes and tailor mutatIon-specIfIc treatment strategIes.

Keywords: Autophagy, VCP, IBMPFD, autophagosome maturatIon, autophagy target, ubIquItIn-proteasome system, mItophagy

PP-63

OXADIAZOLE-BASED PYRAZOLINE DERIVATIVES AS NEW ANTITUMOR AGENTS

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In recent years, the development of resistance to anticancer drugs has emerged as a major obstacle in the fight against cancer and therefore a considerable amount of research has been carried out on the discovery of new selective anticancer agents with enhanced activity and limited toxicity [1,2]. In an effort to develop potent anticancer agents, some oxadiazole-based pyrazoline derivatives were synthesized via the reaction of 1-(chloroacetyl)-3-(2-furyl)-5-aryl-2-pyrazolines with 5-substituted-1,3,4-oxadiazole-2(3H)-thiones. MTT assay was carried out to determine cytotoxic effects of the compounds on MCF-7 human breast adenocarcinoma, A549 human lung adenocarcinoma and NIH/3T3 mouse embryonic fibroblast cell lines. 1-[(5-Benzhydryl-1,3,4-oxadiazol-2-yl)thioacetyl]-3-(2-furyl)-5-phenyl-4,5-dihydro-1H-pyrazole was found to be the most effective agent in this series against MCF-7 cell line with an IC50 value of 125 µg/mL when compared with cisplatin (IC50= 35.31 µg/mL) and low toxicity to NIH/3T3 cell line (IC50 > 500 µg/mL). On the other hand, this compound showed low anticancer activity against A549 cell line (IC50= 339 µg/mL) when compared with cisplatin (IC50= 42.57 µg/mL).

Keywords: Oxadiazole, Pyrazoline, Cancer, Drug Design

PP-64

DNA SYNTHESIS INHIBITORY EFFECTS OF NEW THIAZOLE DERIVATIVES

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Cancer is one of the leading causes of death throughout the world and therefore extensive efforts have been devoted to the discovery of new potent and selective anticancer agents. The clinical efficacy of tiazofurin and its analogues, bleomycins and dasatinib pointed out the importance of thiazole scaffold in the field of cancer treatment.1 In the current work, new thiazolyl hydrazone derivatives were synthesized and evaluated for their cytotoxic effects on A549 human lung adenocarcinoma, C6 rat glioma and NIH/3T3 mouse embryonic fibroblast cell lines using MTT assay. Furthermore, Cell Proliferation ELISA, BrdU (colorimetric) kit was used to determine DNA synthesis inhibitory effects of the most effective compounds. 2-[2-(4-(1H-1,2,4-triazol-1-yl)benzylidene)hydrazinyl]-4-(4-nitrophenyl)thiazole was the most promising agent in this series due to its significant inhibitory effect on C6 cell line with an IC50 value of 13.00±1.00 µg/mL when compared with cisplatin (IC50=12.67±3.06 µg/mL) and low cytotoxicity against NIH/3T3 cell line (IC50= 733.33±256.58 µg/mL). DNA synthesis inhibition percent of this compound was 62.2% at its IC50 value (13.00±1.00 µg/mL), whereas the inhibition percent of cisplatin was 53.95% at IC50 value (12.67±3.06 µg/mL). On the other hand, this compound did not show significant cytotoxic activity against A549 cell line (IC50= 453.33±80.83 µg/mL).

Keywords: Thiazole, Cancer, DNA Synthesis

PP-65

SYNTHESIS AND EVALUATION OF DNA INHIBITORY EFFECTS OF NOVEL THIOSEMICARBAZONE DERIVATIVES

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Cancer is a very common condition which affects people all over the world. Lots of diseases or alterations can cause cancer so it is not a single disease. A change occurs in the body then cells start to grow and multiply uncontrollably1. Thiosemicarbazones possess a wide range of biological effects including antineoplastic, antibacterial, antifungal and antiviral effects and they give rise to a great variety of coordination modes depending on their chemical structure2. Triapine, a thiosemicarbazone derivative, is a promising anticancer agent inhibiting ribonucleotide reductase enzyme, which is important for cell proliferation3. In the current work, 4-substituted benzaldehyde N-(1,3-benzodioxol-5-yl)thiosemicarbazones were synthesized and evaluated for their cytotoxic effects on C6 rat glioma and NIH/3T3 mouse embryonic fibroblast (healthy) cell lines using MTT assay. Cell Proliferation ELISA, BrdU (colorimetric) kit was also used to determine DNA synthesis inhibitory effects of the most effective compounds. Biphenyl substituted compound can be identified as the most promising anticancer agent against C6 cell line (IC50= 32.67±6.43 µg/mL) compared to cisplatin (IC50= 14.33±2.31 µg/mL). Furthermore, this compound showed no significant cytotoxicity against NIH/3T3 cell line (IC50= 143.33±32.14). On the other hand, DNA synthesis inhibition percent of the compound was found as 54.99 %, whereas the inhibition percent of cisplatin was found as 53.95 %.

Keywords: Thiosemicarbazone, Cancer, DNA Synthesis Inhibitory Effect

PP-66

INVESTIGATION OF CARMOFUR INDUCED ULTRASTRUCTURAL CHANGES ON HUMAN LUNG ADENOCARCINOMA CELLS

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Object: The incidence of cancer has been experiencing arise in recent years. Lung cancer is one of type common cases in the world. Lung cancer is one of type common cases in the World. In Turkey, lung cancer incidance in mans is at about %62 and %5 in womans. The frequent case in cancer therapy is developing a resistance to anti-cancer agent. Thus, there is a need for new anti-cancer agents. Carmofur, a primidine analogue, has been used in colorectal cancer therapy. In the present study we investigated the cell the effects of carmofur on the A549 cells ultrastructure.

Material and Method: For ultrastructural evaluations a stock solution of carmofur was prepared in dimethyl sulfoxide (DMSO) and the IC50 concentration detected in our previous study was applied for 24 hours to the A549 cells and fixed in glutaraldehyde overnight at +4 °C. Following fixation, A549 cells were post-fixed in osmium tetroxide, dehydrated in ethanol then embedded in EPON 812 epoxy resin. Sapmles were sectioned and stained in uranyl acetate and lead citrate then observed under a transmission electron microscope (TEM).

Results: In our results we demonstrated that carmofur was highly cytotoxic in low doses in A549 cells and caused damages in the cell ultrastructure namely ondulations and blebbings on the cell membrane, condensation and fragmentations of the nuclei, shrinkage of the cells that indicate apoptosis that is the prefered cell death in cancer cell lines.

Conclusion: From our results it can be concluded that carmofur has a big potential in killing the A549 cells. After further investigations carmofur may be claimed as an useful agent for anti-cancer drug designing.

Keywords: Carmofur, lung adenocarcinoma, cytotoxicity, TEM

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