**Introduction:** The ethnopharmacological use of Cymbopogon spp. dates back from ancient times. Traditionally used in tropical and semi-tropical countries for the repellent properties of their essential oil, the consumption of Cymbopogon spp. infusions is growing all over the world. This is not only due to the unique aroma, widely appreciated by the consumers, but also because of the antimicrobial, anti-inflammatory and sedative properties.

**Methods:** The chemical characterization of infusions and ethanol:water (50:50, v/v) extracts from Cymbopogon citratus and Cymbopogon schoenanthus was achieved by HPLC-DAD. The anti-inflammatory potential of the extracts was assessed by cell and cell-free assays.

**Results:** HPLC-DAD analysis allowed the identification of several caffeic acid derivatives and flavonoids in the infusions and in the ethanol:water extracts of both species. The different extracts displayed scavenging activity against superoxide anion and nitric oxide (NO) radicals, and capacity to significantly reduce NO production by LPS-stimulated macrophages (RAW 264.7 cell line). In addition, the extracts were able to prevent hyaluronic acid degradation via inhibition of hyaluronidase, an enzyme recognized to participate in a number of physiological and pathological processes, including inflammation. No toxicity was observed on human gastric adenocarcinoma and hepatocyte carcinoma cell lines, at a maximum concentration of 2.0 mg/mL.

**Conclusion:** This study provided scientific evidence on the ethnopharmacological use of Cymbopogon species on inflammatory conditions, encouraging infusion consumption and future incorporation of Cymbopogon spp. extracts into nutraceuticals.

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**PS153**

**HuR prevents c-fos mRNA degradation by proteasome-associated ribonuclease in vitro**

E. Zhigalova 1,*, A. Mittenberg 2

1 Skolkovo Institute of Science and Technology, Moscow, Russian Federation
2 Institute of Cytology Russian Academy of Science, St. Petersburg, Russian Federation

E-mail address: katty_z@inbox.ru

(E. Zhigalova).

**Aim:** To estimate HuR protective activity against proteasome-associated ribonuclease for c-myc and c-fos mRNAs.

**Introduction:** Proteasome-associated proteins are attractive targets for multiple myeloma treatment. One of them is HuR protein known to selectively bind ARE-containing mRNAs and protect them from degradation. HuR is supposed to play a role in cancerogenesis since its expression is elevated in many cancer types and it stabilizes a lot of mRNAs encoding proteins involved in oncogenesis. Previously, it was shown that proteasome in addition to its main function – protein degradation – may act as a selective RNase. Moreover, HuR and proteasome have common targets – c-myc and c-fos protooncogene mRNAs.

**Methods:** HuR-GST fusion protein has been cloned, expressed and purified by affinity chromatography. Fragments of c-myc and c-fos were cloned and mRNAs have been transcribed in vitro. Proteasomes have been isolated from K562 cell line (human proerytroleukemia) and Im-9 cells (human multiple myeloma). mRNAs were treated by proteasomes in presence and absence of HuR. The estimation of mRNA cleavage was held by gel-electrophoresis.

**Results:** GST-HuR has specifically bound ARE-containing fragments of c-myc and c-fos mRNAs. Proteasomes extracted from Im-9 and K562 cells cleaved target mRNAs in absence of HuR. It was shown that HuR prevents degradation of c-fos mRNA by proteasomal endoribonuclease, whereas c-myc mRNA was cleaved in the...
same conditions. GST protein didn’t bind with target mRNAs and didn’t affect proteasome cleavage activity.

**Conclusion:** HuR protects c-fos mRNA from proteasome ribonuclease cleavage in vitro, but can’t prevent c-myc mRNA degradation. HuR and proteasome compete with each other for manifestation of their opposite activities. Thus, a new mechanism of regulation of proto-oncogenes expression was observed. However, the functional role of this process in vivo should be evaluated in further studies.1-4

**References**


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**PS156**

**Metformin interferes with glucose cellular uptake by both estrogen and progesterone receptor-positive (MCF-7) and triple-negative (MDA-MB-231) breast cancer cell lines**

I. Amaral 1, C. Silva, A. Correia-Branco, F. Martel

*i3S – FMUP, Portugal E-mail address: inesamaral@ua.pt* (I. Amaral).

**Aim:** Transport experiments with 3H-DG, culture growth and proliferation rate assays were performed. This work aimed to investigate the possible interference of metformin with glucose uptake by MCF-7 and MDA-MB-231 human breast adenocarcinoma cell lines as a mechanism contributing to its anticarcinogenic effect.

**Introduction:** Breast cancer, the most common cancer among women, remains one of the leading causes of mortality among women worldwide.1 Metformin has been widely used as a treatment for type 2 diabetes for over 40 years.2 The first report of a reduced risk of developing cancer for diabetic patients treated with metformin was published in 2005.3 Several mechanisms of action of metformin appear to be implicated in this effect.4,5

**Methods:** Transport experiments with 3H-DG, culture growth and proliferation rate assays were performed.

**Results:** Acute (26 min) exposure of MCF-7 cells to metformin significantly inhibited uptake of 3H-deoxy-D-glucose (3H-DG) (maximal inhibition found with metformin 0.5 mM: 27 ± 2% reduction). Chronically (24 h), metformin induced a concentration-dependent increase in 3H-DG uptake (maximal increase observed with metformin 1 mM: 81 ± 15% increase). Acute (26 min) exposure of MDA-MB-231 cells to metformin slightly inhibited uptake of 3H-DG (maximal inhibition found with metformin 1 mM: 10 ± 3% reduction). Chronic (24 h) exposure to metformin significantly increased 3H-DG uptake by MDA-MB-231 cells (maximal increase observed with metformin 1 mM: 30 ± 8% increase).

Chronic (24 h) exposure of both cell lines to metformin (1 mM) decreased culture growth/cell mass; in contrast, it increased cell proliferation rates. Combination of metformin (1 mM) with the facilitative glucose transporter (GLUT) inhibitor kaempferol (30 μM) did not result in a more marked effect on culture growth and cell proliferation rates.

**Conclusion:** Summarizing, chronic exposure of MCF-7 and MDA-MB-231 cells to metformin induces a marked increase in glucose uptake, associated with an anticarcinogenic effect of the drug. We suggest that the increase in glucose uptake is a compensatory mechanism to cellular energy depletion induced by metformin.

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**PS162**

**Endocannabinoids induce placental trophoblast reticulum stress**

S.C.F. Pereira 6-*, M. Almada 1,2, B.M. Fonseca 1,2, L. Midão 1,2, J. Maia 1,2, N.A. Teixeira 1,2, G. Correia-da-Silva 1,2

1 Laboratório de Bioquímica, Faculdade de Farmácía Universidade do Porto, Porto, Portugal
2 UCIBIO-REQUIMTE, Porto, Portugal
3 Faculdade de Ciências e Instituto de Ciências Biomédicas da Universidade do Porto, Porto, Portugal
4 Departamento de Química, Universidade de Aveiro, Aveiro, Portugal

**E-mail address:** saracatarinapereira@gmail.com (S.C.F. Pereira).

**Aim:** We aim to investigate in cytotrophoblasts whether these effects on cell viability loss are due to endoplasmic reticulum (ER) stress mediated apoptosis.

**Introduction:** Placental development relies on a balance between proliferation, differentiation and apoptosis of trophoblasts, a process tightly regulated by growth factors, cytokines and hormones. Endocannabinoids (eCB), such as 2-arachidonoylglycerol (2-AG) and anandamide (AEA) may play a role in these processes. We previously demonstrated that both eCB induced trophoblast cell death.1,2 Here we investigated in cytotrophoblasts whether these effects on cell viability loss are due to endoplasmic reticulum (ER) stress mediated apoptosis.

ER stress is caused by the accumulation of unfolded proteins leading to an unfolded protein response (UPR) triggered by transmembrane ER signaling proteins including: pancreatic ER kinase (PKR)-like ER kinase (PERK), inositol-requiring enzyme 1 (Ire1) and Activating transcription factor 6 (ATF6). The dissociation of Grp78 (BiP) from these sensors triggers a series of mechanisms that can restore homeostasis or lead to apoptosis. Placental stress has been implicated in the pathophysiology of pregnancy complications, including growth restriction and pre-eclampsia.

**Methods:** BeWo cells (ATCC, USA), an accepted model of cytotrophoblast stem cells were treated with AEA or 2-AG (10 micromolar) for 24 h. Through quantitative real time polymerase chain reaction (qPCR), we evaluated mRNA levels of ER stress markers: CHOP, Grp78, ATF4 and spliced mXBP1. Protein expression of CHOP was evaluated by western-blol.

**Results:** After 24 h of treatment with both eCB, we found an increase in mRNA levels of ER stress markers: CHOP, Grp78, ATF4 and spliced mXBP1. Protein expression of CHOP also increased in both cases.