JNK INHIBITOR SP600125 DECREASED LIPID PEROXIDATION IN ISOLATED HEPATOCYTES

Adeyemo Blessing Oluwatosin
Scientific supervisor: ass. prof. Krasilnikova O.A.
National University of Pharmacy, Kharkiv, Ukraine
meceqween@gmail.com

Introduction. JNK is a protein kinase, which belongs to the MAP-kinase family, is activated in response to numerous intra- and extracellular stimuli and takes part in cell division and differentiation, apoptosis, cancerogenesis. The JNK1 kinases play a central role in obesity-driven insulin resistance by direct phosphorylation of IRS leading to reduced of the PI3K signaling pathway in response to insulin. The use of JNK inhibitors is a promising direction in the treatment of insulin resistance. Therefore, the study of the biological activity of JNK inhibitors is an important and urgent problem.

Aim. The purpose of this study was to study the antioxidant activity of the JNK inhibitor SP600125in vitro in isolated hepatocytes.

Materials and methods. The studies were conducted on female rats weighing 190±15 g, kept under standard conditions in the vivarium NUPh. Hepatocytes were isolated from intact rats by Seglen method and incubated in Eagle medium during 3 hours at 37^{o}C in the presence of 10 μmol JNK activator acetaminophen (APAP). In some cases, 10 minutes prior to the adding of APAP hepatocytes were incubated with the JNK SP600125 inhibitor (10 μmol). Lipid peroxidation intencity was evaluated by TBARS and conjugated diens (CD) levels. The data obtained were processed statistically.

Results and discussion. It was shown that adding JNK inhibitor SP600125 to the hepatocytes incubation medium did not change TBARS and CD content. The results obtained indicate that this compound does not possess antioxidant activity. Incubation of hepatocytes with APAP increased the CD level in 1.7 times and TBARS level in 2.1 times. Cells preincubation with SP600125 decreased TBARS and CD accumulation. APAP are partly metabolized by cytochrome P450 into highly reactive intermediate metabolite N-acetyl-p-benzoquinone imine, which stimulates H_2O_2 production and JNK phosphorylation (pJNK) and activation. pJNK translocates to mitochondria and causes its dysfunction and ROS release. These ROS continue to phosphorylate and activate JNK. This leads to the pathological loop formation. Cells preincubation with SP600125 decreased TBARS and CD accumulation.

Conclusions. The use of SP600125 decreased lipid peroxidation by affecting JNK. The data obtained indicate also that the APAP toxicity is mediated by JNK activation.

HYPOGLYCEMIC ACTION OF APPLE POLYPHENOL EXTRACT

Alaa Mahmoud Omar Abdel Naby
Scientific supervisor: Ph.D., ass. prof. Kravchenko G.B.
National University of Pharmacy, Kharkiv, Ukraine
annabk2014@gmail.com

Introduction. Diabetes mellitus type 2 (DM2) is the most abundant endocrine disease in the world and one of the biggest challenge for the health care system. There is a plenty information that medicinal plants normalize glucose level and improve secondary disorders of metabolism caused by DM2, in particular, provide prevention of liver and kidney diseases and cardiovascular disorders.

Aim. The aim of the recent experiment was to study hypoglycemic activity of apple polyphenol extract (PE) under dexamethasone induced DM2.

Materials and methods. The white male inbred rats 18 month earth old were randomized into 4 groups: 1 – intact animals (IA); 2 – control pathology (DM); 3 – animals with DM that *per os* were administered PE (obtained at NPhU Pharmacognosy department) 1 hour before glucose loading in dose 50 mg/kg bw (DM+PE); 4 – diabetic animals that were administered herbal tea “Arfazetin” in dose 24 mg/kg bw 1 hour before glucose loading (DM+Ar). The DM2 was developed under subcutaneous dexamethasone injections in dose 0.125 mg/kg bw during 13 days. The glucose homeostasis was

evaluated by oral glucose tolerance test (OGTT). On the 14th day the fasting blood glucose level (FBG) was determined in all groups of animals. Then animals were intragastrically loaded glucose solution in dose 3 mg/kg bw. Glucose concentration was determined in blood samples from gingival vein using glucometer "One Touch Select" (LifeScan, USA) in 15, 30, 60, 120 min after loading.

Results and discussion. Glucose loading caused significant increase in blood glucose level in IA and DM groups compared with FBG, put particular maximum was reached after 1 h (in 2.2 and 2.5 times respectively). Glucose concentration in animals, which were administered PE and "Arfazetin", also was highest at 1 h, but less than in DM animals by 23.4% and 21.1% respectively. However, it was staying significantly lower compared with DM group under all control measurements. By the end of the second hour glucose level decreased and practically reached the FBG in all groups except for DM. The revealed hypoglycemic action may be mediated by quercetin – the main component of PE, which, presumably, improves cell insulin sensitivity by adiponectin synthesis stimulation.

Conclusions. Summarizing up the obtained results we can make a conclusion that PE administration caused hypoglycemic effect under DM2 in rats comparable with registered in Ukraine herbal decoction "Arfazetin".

APPLE POLYPHENOLS IMPROVE GLUTATHIONE METABOLISM IN THE RAT LIVER UNDER EXPERIMENTAL INSULIN RESISTANCE

Amhal Nour Eddine
Scientific supervisor: ass.prof. Krasilnikova O.A.
National University of Pharmacy, Kharkiv, Ukraine
noureddineamhal@gmail.com

Introduction. Glutathione is a tripeptide composed of glutamic acid, cysteine and glycine. The reduced glutathione (GSH) is the main component of the body's antioxidant system. Glutathione protective properties manifestation is associated with its oxidation. Glutathione reduction in cells occurs with the participation of glutathione reductase. Insulin resistance and its complications development is accompanied by hyperglycemia, oxidative stress and lipid peroxidation. Oxidative stress and activation of lipid peroxidation lead to the cell antioxidant system depletion. The use of plant polyphenols, which exhibit antioxidant properties is a promising direction in the treatment of insulin resistance.

Aim. The aim of the work was to investigate the apple polyphenol extract effect on the GSH level and glutathione reductase activity in the liver of rats under experimental insulin resistance.

Materials and methods. The experimental study was conducted on 180±20 g rats from vivarium NUPh. Animals were kept on a high-fructose diet during 5 weeks to induce experimental IR. Apple polyphenol extract (APE) was administered during two weeks in dose 9 mg polyphenols/kg. The animals were decapitated under chloralose-urethane anesthesia. The liver was perfused with cold physiological solution and homogenized. In the liver, GSH level and glutathione reductase activity were determined with Ellman's reagent. The protein level was determined by the Lowry method. The data obtained were processed statistically.

Results and discussion. It was shown that glutathione reductase activity was reduced from 1.64±0.10 (intact) to 1.12±0.04 nmol NADPH/min/mg protein (group IR) in rat liver. GSH level was also reduced from 8.03±0.25 (intact) to 4.96±0.11 4.96±0.11 μg/mg protein (group IR). GSH level is decrease due to an inhibition of the glutathione reductase activity and the enhancement of free radical oxidation under these conditions. APE administration increased glutathione reductase activity up to 1,37±0,04 nmol NADPH/min/mg protein and the GSH level up to 6,85±0,19 μg/mg protein times the liver of rats under experimental insulin resistance.

Conclusions. Thus, apple polyphenols improve antioxidant system activity in rat liver under experimental insulin resistance. Apple fruit polyphenols applying is promising in the treatment of insulin resistance, diabetes and its complications.