**PP098—NICORANDIL INDUCED ENDOTHELIMUM-INDEPENDENT RELAXATION OF ARTERIAL BYPASS GRAFT**

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**Introduction:** Spasm of the human internal mammary artery (HIMA) is a rare but life-threatening complication after coronary artery bypass grafting (CABG). The reversal of this vasospasm is often challenging, and the most effective therapy is not well defined. The present study was aimed to investigate vasorelaxant effect of nicorandil, K+ channel opener, on the HIMA and to define the role of different K+ channel subtypes in nicorandil action on this blood vessel.

**Patients (or Materials) and Methods:** Discarded segments of HIMA were collected from patients undergoing CABG and studied in organ baths. HIMA rings were precontracted with phenylephrine (10 µM) followed by cumulatively adding increasing doses of nicorandil. The endothelium was removed mechanically.

**Results:** Nicorandil (0.001 µM–300 µM) induced a concentration-dependent relaxation of HIMA rings precontracted by phenylephrine. Glibenclamide (10 µM), a highly selective blocker of ATP-sensitive K+ (KATP) channels, partially inhibited relaxation of HIMA induced by nicorandil. Tetraethylammonium (TEA, 1 mM), a nonselective blocker of Ca2+-activated K+ (KCa) channels, as well as iberiotoxin (100 nM), a most selective blocker of large-conductance KCa (BKCa) channels, partly antagonized relaxation of HIMA. A nonselective blocker of voltage-gated K+ (Kv) channels, 4-aminopyridine (4-AP, 0.5 mM), as well as margatoxin (10 nM), a potent inhibitor of Kv1.3 channels, did not significantly modify the nicorandil-induced relaxation of HIMA.

**Conclusion:** The results from our study demonstrate that, in HIMA, nicorandil has a potent vasorelaxant effect which is endothelium-independent. It seems that mechanism of this relaxation includes KATP and 4-AP-sensitive K+ channels located in the smooth muscle of HIMA.

**Disclosure of Interest:** None declared.

**PP100—POTENTIAL OF NATTOKINASE AS AN ANTITHROMBOTIC & FIBRINOlytic AGENT**

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**Introduction:** Thrombosis is 1 of the major causes of death worldwide. Atherothrombotic diseases such as myocardial or cerebral infarction are serious consequences of the thrombus formed in blood vessels. Nattokinase is new fibrinolytic enzyme with a molecular weight of 27,728 Da that cleaves directly cross-linked fibrin in vitro. In this study, we investigated the effect of nattokinase supplementation on thrombus formation using different animal models.

**Patients (or Materials) and Methods:** To study the fibrinolytic activity, in vitro clot dissolution assay was done in which clot was formed in helix and then kept in contact with different concentrations of nattokinase and streptokinase (standard drug) and its clot dissolution property was studied. Further it was studied for its antithrombotic activity in venous thrombosis. Rats were pretreated orally for 7 days with Nattokinase (100 and 200 mg/kg). One hour after the last dose, stasis was developed in the inferior vena cava in rats, and its activity was intensified with the help of ferric chloride. It was also studied for its antithrombotic activity in arterial thrombosis using arteriovenous shunt-induced thrombosis in rats. In this rats were pretreated orally for 7 days with Nattokinase (100 and 200 mg/kg) and on last day 1 hour after last dose thrombosis was induced by arterio-venous shunt. In both the above studies, % inhibition of thrombus formation was calculated. Because hemorrhage is an important side effect of antithrombotic and thrombolytic therapies, the drug was studied for its effect on hemorrhage using rat tail transaction method. Here, effect of 7 days pretreatment of 2 doses of nattokinase (100 and 200 mg/kg) on hemorrhage was studied.

**Results:** In vitro clot dissolution assay, there was significant and dose-dependent clot dissolution with different concentration of nattokinase. Maximum activity, 94.43 % inhibition, was seen in 3000ug/ml concentration which was comparable with 1000 IU of streptokinase. In venous thrombosis model, it showed significant and dose-dependent inhibition of thrombus formation as indicated by decrease in weight of thrombus. Antithrombotic potential of nattokinase at the highest dose (200 mg/kg) showed 45.431 % inhibition with respect to saline control group. In arterio-venous shunt, it showed no significant inhibition of thrombus formation on thrombogenic surface. In the rat tail transaction model, both the doses of nattokinase (100 and 200 mg/kg) has significantly increased the bleeding time with not much effect on blood loss. Also, the platelet MDA was significantly decreased in both groups treated with 100 and 200 mg/kg of nattokinase.

**Conclusion:** The results of various in vitro and in vivo models studied indicated that nattokinase has a good potential as an antithrombotic and fibrinolytic agent.

**Disclosure of Interest:** None declared.

**PP101—REACTIVITY MODIFICATIONS IN HEART RESISTANCE VESSELS TO ANGIOTENSIN II IN ISOLATED PERFUSED HEART OF HYPERTENSIVE RATS**

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**Introduction:** High blood pressure (HBP) is a disease with high morbidity and mortality worldwide and is considered 1 of the main etiologic factors of multiple cardiac pathologies like cardiac hypertrophy, myocardial infarction, among others. One of the regulatory mechanisms of blood pressure is the renin angiotensin system (RAS), so much so that the first-line drugs in the treatment of hypertension are directed to some of the components of the RAS. However, these treatments frequently lead to the development of changes in morphology and physiology characterized by vascular complications. It is therefore necessary to consider adjuvant therapies to improve the outlook and quality of life of patients with HBP. A therapy that has been shown clinically to improve the condition of patients with HBP is hyperbaric oxygenation (HBO), which consists in subjecting a subject at a pressure >2 atmospheres absolute with 100% oxygen. This therapy has been shown to improve vascular smooth muscle relaxation, although the mechanism of action has not yet been specified.
**Clinical Therapeutics**

Patients (or Materials) and Methods: Male Wistar rats were used (340 ± 20 g) at standard conditions. Hypertension was induced by a previously described surgical method (PAGE), which was obtained with measurements HAS stable (plethysmographic method) from the seventh postoperative day, the eighth day began therapy OHB, for 5 days per week for 4 weeks. At the end of treatment, the rats were sacrificed getting heart; this was mounted on a system of isolated organ (Langendorff system), and it was stimulating to logarithmic doses of angiotensin II (Ang II) to measure variations in vascular reactivity associated with the RAS in coronary resistance arteries. The results are analyzed with 2 way ANOVA and post hoc Von Ferroni.

Results: The diastolic pressure measurement was obtained a pressure of 150 ± 15 mm Hg, consistent with the definition of hypertension. When analyzing perfusion pressure resistance in coronary arteries, the HBP group without treatment showed an increase in vascular reactivity compared with the healthy group. Whereas the HBP with HBO group no presented changes in vascular reactivity to any dose of Ang II, compared with healthy control group.

Conclusion: HBO therapy reduces vascular reactivity in coronary resistance arteries under Ang II stimulation in a model of hypertension of renal origin. This suggests that HBO could be used with hypertensive patients to reduce the development of heart disease and in another way it could help to improve the functionality of the heart and reduce the use of drugs in the HBP treatment.

Disclosure of Interest: None declared.

**PP102—MOLECULAR DOCKING RESEARCH & IN-VITRO ANALYSIS OF NOVEL NATURAL AND SYNTHETIC PTP 1B INHIBITORS AS POTENTIAL THERAPEUTIC TARGET FOR DIABETES MELLITUS**

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Introduction: Augmented perversiveness of type 2 diabetes mellitus and obesity has amplified the medical necessitate for new agents to treat these disease states. Both type 2 diabetes and obesity are connected to the resistance to the hormones insulin and leptin. Protein tyrosine phosphatase 1B (PTP1B) has been shown to function as a negative regulator of insulin signaling as well as leptin signal transduction. This research exertion shows the molecular docking analysis of novel synthetically prepared compounds and new-fangled isolated natural PTP 1B inhibitors as novel target for type 2 diabetes.

Patients (or Materials) and Methods: Molegro Virtual Docker (MVD) has been used to dock novel natural PTP1B inhibitors with the Discovery Studio 3.0 visualizer.

Results: Molecular docking of novel natural PTP1B inhibitors showed some compounds with excellent dock score with greater binding affinity exhibiting pi-pi interactions

Conclusion: The plausible mechanism of action of various natural PTP1B inhibitors has been explained supported by in vitro experimentation data. Traditional drugs with unknown mechanism of action were experimented for the PTP1B inhibitory activity and therefore are attention-grabbing biologically lead compounds.

Disclosure of Interest: None declared.

**PP103—EFFECTS OF CLOFIBRATE ON OUABAIN-INDUCED ARRHYTHMIA IN ISOLATED RAT ATRIA**

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Introduction: Cardiac arrhythmia is 1 of the critical health conditions. Clofibrate is a peroxisome proliferated-activator receptor-α (PPAR-α) agonist that is widely used for reducing triglycerides in hyperlipidemic patients. Because this drug has showed several beneficial effects in relief of some cardiovascular diseases, the aim of the present study was to evaluate antiarrhythmic effects of clofibrate on ouabain-induced arrhythmia in rats.

Patients (or Materials) and Methods: Twenty male rats weighing 220 to 230 g were divided into 2 equal groups randomly. Group 1 (treatment) received clofibrate (300 mg/kg) solved in olive oil (1 mL/kg) once daily for 14 days intraperitoneally. Group 2 (vehicle control) only received olive oil (1 mL/kg) once daily for 14 days intraperitoneally. After induction of anesthesia, heart was rapidly removed and the atria were immersed in a tissue bath containing modified Krebs solution and attached to isometric force transducer of PowerLab machine. Time of onset of arrhythmia and asystole, atrial beating rate, and contractile force were recorded and analyzed statistically with paired and Student’s t test in treatment and control groups.

Results: Clofibrate significantly postponed time of onset of arrhythmia (23.57 [4.69] minutes) rather than control group (2.04 [0.27] minutes; P = 0.024). A significant increase in the onset time of asystole in treatment group (66.19 [12.33] minutes) was observed, while this time for control group was 22.77 (7.17) (P = 0.004). Incubation of ouabain increased the atrial beating rate in control group significantly (P = 0.022), while it does not show such effect in treatment group (P = 0.845). Incubation of ouabain had no effects on contractile force in control and treatment groups (P = 0.063 and P = 0.539, respectively).

Conclusion: Based on the findings, clofibrate may possess potential to reduce some kinds of cardiac arrhythmias. Further studies especially on in vivo arrhythmial models can more elucidate these beneficial effects of this drug.

Disclosure of Interest: None declared.

**PP105—THE ROLE OF POLY (ADP-RIbose) POLYMERASE (PARP) PATHWAY ON ENDOTHELIN-1 (ET-1)-INDUCED ENDOTHELIAL DYSFUNCTION IN RAT THORACIC AORTA**

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Introduction: The aim of this study was to investigate whether activation of the nuclear enzyme poly(ADP-ribose) polymerase (PARP) contributes to the development of endothelin-1 (ET-1)-induced endothelial dysfunction.

Patients (or Materials) and Methods: To evaluate vascular reactivity, isometric tension studies were performed in response to vasodilator agents, acetylcholine (ACh) and sodium nitroprusside (SNP), and constrictor agents, potassium chloride (KCl) and phenylephrine (Phe), in rat thoracic aorta rings incubated with or without ET-1(10-3 M, 18 hours). To investigate mechanisms of ET-1 action, additional sets of experiments involving rings incubation with ET-1 alone or with addition of polyelectrolyte glycol-superoxide dinsmute (PEG-SOD), a cell permeable superoxide radical scavenger, 41 U/mL plus apocynin (a NADPH oxidase inhibitor, 300 µM), and PJ34 (an inhibitor of poly(ADP-ribose) polymerase, 3 x 10-6 M) for 18 hours, and both relaxant and constrictor responses were evaluated. Moreover, PARP-1 and poly(ADP-ribose) (PAR, an end-product of PARP activity) expressions were evaluated by Western blot and immunohistochemistry.

Results: The results of this study demonstrated that incubation of thoracic artery rings with ET-1 (10-3 M, 18 hours) resulted in significant inhibition of response to ACh (an endothelium-dependent vasodilator) while SNP (an endothelium-independent vasodilator)-