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Oral presentations

BASIC MEDICAL SCIENCE AND PHARMACY

Determination of the total phenolic content and the reducing activity *in vitro* of bee pollen in impact of different storage conditions and duration

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Background. Bee pollen, containing proteins, vitamins, and phenolic compounds, is essential for bees, and has the nutraceutical potential for humans. Bee pollen may be used as fresh and dried products. In order to preserve it biological activity, preparation and storage conditions of bee pollen are of great importance, as they can lose their biological activity over time.

Aim. The aim of this study was to evaluate the influence of storage conditions and duration of bee pollen on the total phenolic content and reducing activity *in vitro* and to evaluate the botanical origin of bee pollen.

Methods. Bee pollen were collected in May from apiary, located in Pasvalio district, Talkoniai 55.9598° N, 24.3422° E. One part of pollen was dried, another part was fresh frozen in a freezer at –20°C and at –80°C. Botanical origin was detected by melissopalynological analysis. The total phenolic content was evaluated using Folin–Ciocâlteu method. The reducing activity was determined by FRAP spectrophotometric assay.

Statistical analysis: All the experiments were carried out in triplicate. Means and standards deviations were calculated with the SPSS 20.0 software (Chicago, USA). The significant differences were calculated by one-way ANOVA, followed by Tukey post hoc comparison test. Differences were considered to be significant at $p<0.05$.

Results. After melissopalynological determination, pollen from the *Salix* spp.*, Taraxacum officinale* L., *Aesculus hippocastanum* L., *Quercus* spp. and *Brassica napus* L. was detected.

Fresh bee pollen contained 24.52±0.66 mg GAE/g DW amount of total phenolic content and reducing activity was 84.28±3.17 μmol TE/g DW.

The values of total phenolic content were in a range of 22.06±0.55 mg GAE/g DW to 24.40±0.30 mg GAE/g DW (after 3 months) and of 19.09±0.68 mg GAE/g DW to 21.62±1.24 mg GAE/g DW (after 6 months). The highest levels of total phenolic content were detected in bee pollen samples, stored in a freezer at –80°C.

The reducing activity of bee pollen was in a range of 75.91±5.58 μmol TE/g DW to 80.04±1.29 μmol TE/g DW (after 3 months) and of 62.33±2.71 μmol TE/g DW to 76.04±3.22 μmol TE/g DW (after 6 months). The lowest reducing activity was determined in dried bee pollen samples after 6 months storage (62.33±2.71 μmol TE/g DW).

Conclusion. No significant difference of the total phenolic compounds between frozen bee pollen samples in 0 to 6-month period was detected. Whereas the lowest reducing activity *in vitro* was determined in dried bee pollen samples after storage.

Total phenolic compounds of *Potentilla erecta (L.) Raeusch.* in above-ground parts during vegetation period

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Background. *Potentilla erecta* (L.) Raeusch. (Rosaceae) is most common in the Northern hemisphere (especially in Scandinavia and Russia), in cold or temperate climates, arctic and Alpine zones [1,2]. The plant material is used for treating inflammation, wounds, certain types of cancer, infections caused by bacteria, fungi, viruses, diarrhoea, diabetes mellitus, and other conditions [2]. Phytochemical studies related *P. erecta* has led to the isolation and identification of tannins, ellagitannins, agrimoniin as the most active chemicals with biological effects [3]. High content of phenolics compounds, condensed and hydrolysable tannins serves as the basis for its beneficial effect [1].

Aim. The aim of the current study was to determine the amount of phenolic compounds of wild silverwort *(P. erecta*) grown in Lithuania.

Methods. Aboveground parts of *P. erecta* collected from VU Šiauliai Academy Botanical Garden during the plant's vegetation period of June–September. Extracts were prepared using 0.2 g of dry matter and 20 mL of 70% acetone. The total phenolic compounds were determined according to Folin–Ciocalteu method using a spectrophotometer.

Results. In different vegetation periods the total phenolic content was in a range of 50.88±2.77 GAE mg/g to 92.42±0.12 GAE mg/g. The results have shown that the greatest amount of phenolic content was determined in August (92.42±0.12 GAE mg/g) during massive flowering period and the lowest was September (50.88±2.77) GAE mg/g at the end of vegetation.

Conclusion. The total amount of phenolic compounds varies in different vegetation periods, so it is important to determine in which vegetation period the plant accumulates the highest amount of phenolic compounds, as this can ensure the best quality of the raw material and therapeutic potential. The greatest amount of phenolic compounds was determined in the plant material collected during flowering.

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Precision-guided interventions: the promise of aptamer targeting β1-integrin

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Background. Triple-negative breast cancer (TNBC) accounts for ~20% of all invasive breast cancer cases. TNBC tumours are negative for expression of human epidermal growth factor receptor 2 (HER-2), progesterone receptor (PR), and estrogen receptor 2 (ER), rendering TNBC resistant to endocrine therapy. Combination of chemotherapy followed by surgery is used as a treatment strategy for early TNBC, while chemotherapy is used to treat advanced and metastatic TNBC. Treatments with immune-checkpoint inhibitors (ICIs) targeting programmed cell death protein 1 (PD-1) and programmed death-ligand 1 (PD-L1) have augmented the therapeutic choices for patients with PD-L1+ TNBC in recent years. However, present treatments do not yet provide optimal therapy options.

Aptamers are short (20–100 nt), single-stranded DNA or RNA oligonucleotides that bind to their target molecules due to a specific three-dimensional structure. Their affinity and specificity are comparable to antibodies; however, aptamers are smaller (6–30 kDa versus 150–180 kDa for antibodies) and can be chemically synthesised, resulting in minimal to no batch-to-batch variability and straightforward scale-up.

Aim. The aim of the study was to identify the protein target of GreenB1 aptamer and to determine its trafficking pathway within the cell.

Methods. The target protein on MDA-MB-231 cells for the GreenB1 aptamer was identified using the proximity labelling method. Biotin labelled GreenB1 was conjugated to streptavidin-conjugated horseradish peroxidase (HRP). HRP, in the presence of hydrogen peroxide, creates a highly reactive tyramide species that labels nearby proteins. Biotinylated proteins were pulled down using streptavidin-coated magnetic beads and eluted using excess biotin and further analysed using mass spectrometry. Imaging flow cytometry was used to determine GreenB1 aptamer co-localization with anti-β1-integrin antibody and LysoTracker.

Results. β1-integrin binding aptamer GreenB1 which selectively binds MDA-MB-231 cells *in vitro*, is quickly internalized in cells but does not affect the amount of β1-integrin available for binding on the cell surface.

Conclusions. GreenB1 translational applications are of great interest in the future and might lead to innovative targeted protein breakdown or therapeutic approaches.

Acknowledgements. This study was supported by UL fundamental research grant "Research of biomarkers and natural substances for acute and chronic diseases' diagnostics and personalized treatment".

Co-culture approaches with organoids of the human exocrine pancreas and pancreatic progenitors

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Background. The public need for therapies to treat pancreas-related conditions like diabetes, exocrine pancreatic insufficiency and cancer is increasing. Protocols to establish human pancreatic organoids (hPOs) from primary tissue and pluripotent stem cells (PSCs) have recently developed. hPOs offer new ways to model pancreatic disorders and study the aspects of development and regeneration of this organ. Organoid cultures can be further complicated by integrating other cell types. Such co-culture approaches can help to dissect the function of the cells in the progenitor niche or tumour stroma. The role of the peripheral nervous system (PNS) in such contexts has often been overlooked; nevertheless, mounting evidence supports its active involvement.

Aim. The aim of our work is to establish hPOs from healthy and tumor tissue and from PSCs, co-culture them with neural crest-derived glia and mesenchymal cells to study the interaction of exocrine pancreas with stromal components in various culture conditions.

Methods. hPOs were established from donor tissue material and PSCs. Primary and commercial human Schwann cells (SC) and mesenchymal cells were used for co-culture with pancreatic organoids and in PSC differentiation protocols. hPOs and co-culture constructs were characterized using immunofluorescence in gel sections and using whole-mount approaches and targeted DNA sequencing.

Results. We describe successful derivation of normal and tumour hPOs, as well as primary human SC cultures. hPOs express various markers of the exocrine pancreas including CK19, SOX9, PDX1 and CDH1. SCs and mesenchymal cells are identified in co-cultures by vimentin expression. Both cell types survive in various co-culture setups and support hPO growth even in depleted medium. SCs tend to form network-like structures around pancreatic organoids. DNA sequencing data of primary hPO cultures suggest the importance of such analysis to validate the tumour identity of established cell lines. Glial cells in co-culture with PSC-derived pancreatic progenitors (PPs) show marked tropism towards PPs under the acinar differentiation conditions. Additionally, we discuss the experiments of hPO inclusion in microfluidic devices.

Conclusion. hPO and pancreatic progenitor co-culture with SCs and mesenchymal cells offers opportunity to study the role of the stromal components in pancreatic deficiencies and development.

Acknowledgements. Authors declare no conflict of interest. The presented research is funded by Postdoctoral research project nr. 1.1.1.2/VIAA/4/20/623, by the UL fundamental research grant "Research of biomarkers and natural substances for acute and chronic diseases' diagnostics and personalized treatment" and VPP-EM-FOTONIKA-2022/1-0001.

Environmentally friendly approach based on mechanochemical synthesis for the development of ferric ferrocyanide with improved adsorption capacity of radionuclides in nuclear incidents

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Background. The problem is of outstanding importance since the 24th of February 2022 due to the raised nuclear attack hazards in Europe due to the invasion of Russian Federation in Ukraine and its aggressive politics. This fact has affected all the European Union countries to revaluate their nuclear defence strategies especially attributed to individual protection measures.

Aim. The aim of this study is to develop sorbent composites for binding radioactive and non-radioactive isotopes, based on "green" synthesis using non-toxic, easily accessible raw materials. Main attention was focused on Cs-137 because it can be easily incorporated in the living organisms due to chemical similarity to potassium.

Methods. The synthesis was modified using firstly agate mortar at room temperature followed by optimisation of the time and the synthesis scheme using a planetary ball mill. The optimal settings were determined experimentally. The obtained material was tested with an X-ray diffraction spectroscopy technique and compared to database to confirm that obtained powder is pure ferric ferrocyanide. Obtained results were confirmed using Fourier–transform infrared spectroscopy method. Mechanically synthesized ferric ferrocyanide was tested for Cs sorption at different pH (water, artificial gastric and artificial intestinal solutions) using inductively coupled plasma mass spectrometry method. Determination of particle size and Zeta potentional measurements also was done.

Results. Within optimisation studies of several parameters (time, pH, concentration and grinding method), a ferric ferrocyanide mechanochemically obtained pharmaceutical substance was tested for radionuclide adsorption based on stable salts of cesium, strontium, rubidium, and cobalt, and showed sufficient adsorption in different media compared to the commercial antidote Radiogardase®, known as the solid form of Prussian blue, effective for cesium and thallium. It is the only one antidote accepted by the European Medicines Agency and the United States of America Food and Drug Administration for Cs binding. The synthesized sorbent had higher adsorption of Cs in acidic conditions, compared to the Radiogardase®.

Conclusion. The developed scheme based on "green chemistry" and mechanochemical synthesis allows to provide controlled development of ferric ferrocyanide antidotes with regulated chemico-physical properties for the application in nuclear incidents. It seems that in the case of radioactive fallout ferric ferrocyanide sorption is sufficient to reduce the quantity of active pharmaceutical substance preparation, reducing possible side effects (stomach discomfort and constipation).

Acknowledgements. This work was possible thanks to the selfless sharing of knowledge by professors and researchers of the UL, and it is funded by UL Foundation and "Mikrotīkls".

Analysis of commercial effervescent tablets containing magnesium and vitamin B6 using X-ray microtomography

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Background. The density analysis of the tested tablets can be performed using non-invasive scanning of the inner microstructure with X-ray microtomography. X-rays are absorbed by the given object in proportion to its density, which is reflected in the microtomographic image by the gray level, the linear relationship between the density of the tested object and the brightness of the image is observed. "Bright" pixels represent areas of high density, while "dark" pixels represent areas of low density. The present study aimed to compare the density, thus the homogeneity of the effervescent tablets with magnesium and vitamin B6, expired and unexpired.

Methods. We analyzed expired (n=20; expiration date April 2021) and unexpired (n=20; expiration date March 2024) effervescent tablets containing magnesium and vitamin B6 (Zdrovit; Natur Produkt Pharma sp. z o. o., Ostrów Mazowiecka, Poland). Randomly selected unexpired tablet was additionally dried at a vacuum dryer at 50°C for 24 hours. X-ray microtomographic scanning was performed using the Phoenix v|tome|x apparatus (GE Sensing & Inspection Technologies GmbH, Germany). The tablets were scanned at an energy of 180 kV. The image was recorded with a resolution of 2024×2024 pixels with a total number of 2000 scans for each tablet. The Micro-CT HA Phantom D32 calibration phantom was scanned simultaneously with the tested tablets to establish the grayscale level of reference density. Data were analyzed with the use of Statistica 13.0 software.

Results. We observed a complete correlation between the brightness of all the tested tablets (from microtomographic scans) and the density was 1.00. The significant difference in the density between the three analyzed types of effervescent tablets ($p<0.001$). In the post-hoc analysis, significantly higher density was demonstrated in unexpired effervescent tablets than in expired (1.268±0.010 vs. 1.261±0.009 g/cm^3, respectively; $p<0.001$) as well as in unexpired tablets *vs* dried (1.268±0.010 vs. 1.238±0.022 g/cm^3, respectively; $p<0.001$). In addition, the difference in mean density between expired and dried tablets was also significant ($p<0.001$). Therefore, the best homogeneity of ingredients distribution was reported for the unexpired tablets, while the worst, for dried tablets.

Conclusion. The density of the expired effervescent tablets and tablets dried in the vacuum drier was significantly lower than in unexpired effervescent tablets with magnesium and vitamin B6.

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Assessment of national trends in the use of reimbursed oral lipid lowering medications for primary and secondary cardiovascular disease prevention in Latvia from 2012 to 2021

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Background. Lipid-lowering medications (LLMs) are the mainstay of primary and secondary prevention of atherosclerotic cardiovascular diseases (CVD).

Aim. To characterize the nation-wide state-reimbursed LLMs used for either primary or secondary CVD prevention in Latvia.

Methods. Using data from the National Health Service database on all state-reimbursed prescriptions in Latvia from 2012 to 2021, we retrospectively and longitudinally assessed numbers of dispensed oral LLMs. The total amount of units dispensed annually was calculated using the number of tablets or capsules in each package for each dose. Dispensed LLMs were further analyzed based on the international classification of diseases (ICD) codes indicated in prescription to ascertain if they were intended for primary or secondary prevention. Doses of 40 to 80 mg of atorvastatin and 20 to 40 mg of rosuvastatin were considered as high-intensity statins. Version 22 of IBM SPSS Statistics was used to analyze the data.

Results. From 2012 to 2021, total consumption of all statins tripled from 6,970 million units to 19,852 million for primary prevention and doubled from 12.747 million units to 23.594 million for secondary prevention. Ezetimibe became reimbursed for primary CVD prevention in 2020, and number of dispensed units increased from 5,852 to 16,408 in 2021. Ezetimibe dispensed for secondary CVD prevention rose from 184,744 units in 2012 to 4.669 million units in 2021. The consumption of high-intensity statins for the primary prevention of CVD was 17.0% of all statins in primary prevention in 2012, which doubled to 34.4% in 2021. Consumption of high-intensity statins for secondary CVD prevention increased from 39.0% in 2012 to 54.3% in 2021. In 2012, the average dose in primary prevention was 18.0±10.7 mg for atorvastatin and 16.4±7.2 mg for rosuvastatin, while in 2021 it was 22.3±11.9 mg and 18.2±8.0 mg, respectively ($p<0.001$ for both).

In secondary prevention, from year 2012 to 2021 the average dose of atorvastatin and rosuvastatin increased from 28.4+17.4 mg to 32.4±20.3 mg and from 18.0±7.9 mg to 19.4±8.7 mg, respectively ($p<0.001$ for both).

Between 2012 and 2021, for primary prevention the proportion of atorvastatin 80 mg increased from 6.0% in 2012 to 11.4% in 2021, and rosuvastatin 40 mg increased from 19.4% to 30.8%.

Conclusions. Over a 10-year period, the use of statins and ezetimibe for both primary and secondary CVD prevention has improved substantially as seen by more units dispensed, increase of mean statin doses and more frequent use of high-intensity and maximal doses.

Selected drug effects on lipid accumulation and release of IL-6 and IL-8 from HepG2 cells

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Background. Non-alcoholic fatty liver disease (NAFLD) is the most common chronic liver disease worldwide. Free fatty acids stimulated lipogenesis and release of pro- and inflammation cytokines are the main causes of NAFLD. There is currently no approved pharmacotherapy for NAFLD and there is no complete picture of how the medicines act in the lipid-loaded liver. Non-steroidal anti-inflammatory drugs (NSAIDs) and occasionally statins have been associated with hepatic side effects; however, the frequency of these side effects is uncertain. Their hepatotoxic evidence is mostly demonstrated as aminotransferase elevation. However, the drug effects on lipid accumulation and pro-inflammatory cytokine production in the fatty liver cells are less investigated.

Aim. To elucidate the role of selected drugs on lipid and pro-inflammatory cytokine IL-6 and IL-8 production in HepG2 cell fatty liver model.

Methods. Steatosis in HepG2 cells was caused by addition of 0.5 mM oleic acid and palmitoleic acid (FA) mix 2:1 for 24 h. Effects of silymarin, diclofenac, metformin, atorvastatin and celecoxib on HepG2 cell viability were tested using CCK-8 kit (Sigma-Aldrich). Then, all tested compounds were added to the cells simultaneously with FA. Lipid accumulation was measured using Nile Red dye and observed under a fluorescence microscope. The IL-8 and IL-6 levels in the cultured medium were assayed using ELISA kits according to the manufacturer's eBioscience instructions. Absorbance or fluorescence in all experiments were recorded by Infinite 200 PRO plate reader and i-control software (Tecan Trading AG, Switzerland). Statistical analyses were performed by one-way ANOVA followed by Dunnett's Multiple Comparison test. The experimental results were statistically analysed by GraphPad Prism 7 software (San Diego, CA, USA).

Results. Treatment of HepG2 cells with a FA induced 7-8 fold lipid accumulation, whereas all five compounds decreased lipid accumulation. FA caused 8-9 fold IL-6 and 2.5 fold increase of the IL-8 release. All compounds inhibited excessive production of the IL-6 and IL-8. Celecoxib the most effectively inhibited IL-6 release showing effect already at concentration 12.5 μM. Silymarin, celecoxib and atorvastatin reduced IL-8 level at 25 and 50 μM, diclofenac and metformin at 100 μM.

Conclusion. Tested here four drugs and silymarin showed anti-inflammatory and anti-lipogenesis effects in short-term fatty liver model *in vitro*.

Acknowledgements. The authors declare the absence of conflict of interest. The study was supported by UL Foundation grant No 2469 and funded by '*Mikrotīkls*' *Ltd*.

Collagen fibre changes in the ascending aorta for patients with an altered aortic valve

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Background. The aorta is a flexible type of artery that provides systemic circulation. It can be structurally changed if congenital abnormalities of bicuspid aortic valves (BAVs), altered tricuspid aortic valves (TAVs), age-related changes or hypertension are present. Deposits of inflammatory and fat cells, as well as morphological alternations of collagen fibres in the aortic wall layers (*tunica intima, tunica media, tunica adventitia*) can cause dissection, aneurysms, or stenosis.

Aim. To evaluate collagen fibre changes in the ascending aorta of both patients with altered aortic valves (AVs) and controls.

Methods. We used surgical material from 28 patients with altered AVs and autopsies from 13 individuals defined as controls. Tissue samples were stained using Sirius Red and Masson's trichrome methods. Light microscopy was used to estimate the relative thickness, fragmentation, and organization of collagen bundles. The statistical analysis was performed using SPSS v27. The Kruskal-Wallis test was performed to detect statistically significant differences between groups. Correlations between the parameters were determined using Spearman's rho test.

Results. The organization of collagen fibres in the *tunica intima* differed statistically significantly ($H=11.15$, $p=0.004$) when three groups were compared. More organized collagen fibres were observed in controls (mode=1) compared to patients with TAVs (mode=2) and BAVs (mode=2). Also, the relative density of collagen fibres in the *tunica adventitia* differed statistically significantly in all groups ($H= 28.07$, $p<0.001$). The relative density of collagen fibres in the *tunica adventitia* in the BAVs group (mode=2) was less dense than in controls (mode=3) or TAVs group (mode=3). A significant correlation was found in *tunica intima* between the fragmentation and organization of collagen bundles (controls, $p<0.001$, BAVs group, $p=0.009$, TAVs group, $p<0.001$). A significant correlation was found between the relative density and fragmentation of bundles in *tunica media* (BAVs group, $p=0.027$, TAVs group, $p=0.02$). In addition, *tunica adventitia* showed that there was a correlation between the relative density of collagen bundles and fat cell deposits (controls, $p=0.01$, BAVs group, $p=0.002$).

Conclusion. Controls showed a more organized *tunica intima* layer than the other groups, which may indicate that patients with altered BAVs and TAVs had disrupted hemodynamics that directly affected the *tunica intima*. The BAVs group had less dense *tunica adventitia*, which could be due to fat cell deposits, which were more common in this group. Correlations indicate that more fragmented collagen fibres tend to be more disorganized, which can happen because of their rupture.

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Effects of nanosecond pulsed electric field on LAG-3 expression in melanoma cells

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Background. LAG-3 (Lymphocyte activation gene 3) protein is a checkpoint receptor that interacts with LSEC-tin, Galectin-3 and FGL1. This interaction leads to reduced production of IL-2 and IFN-γ. LAG-3 is widely expressed in different tumor types and modulates the tumor microenvironment through immunosuppressive effects. Differential expression in various tumor types influences patient prognosis, which is often associated with coexpression with immune checkpoint inhibitors, such as TIM-3, PD-1 and CTLA-4.

Aim. Here, we discuss expression profiles in different melanoma types after treatment of the cells with nanosecond pulsed electric field (nsPEF). Besides, we described the effects of the field on melanoma cells' invasion and proliferation potential.

Methods. C32 and A375 cells were exposed to 4-16kV/cm, 200ns, 100p, 10Hz nsPEF. The long-term viability of the cells was analyzed by mitochondrial activity assay and Presto Blue assay. The permeabilization of the melanoma cells in response to PEF treatments was analyzed by YOPRO-1 uptake studies. The morphological changes of tumor cells were analyzed by holotomographic and confocal microscopy.

Results. LAG-3 inhibitors suppress T-cell proliferation and activation by disallowing the interaction between LAG-3 to MHC-II. The process enhances the anti-tumor immune response. The study proves, that nsPEF may increase the expression of membrane-associated proteins, including LAG-3 and PD-1.

Conclusion. Our study sheds light on a novel method in which we may modulate cancer cells to overexpress cancer-specific molecules thus sensitizing tumors to the targeted anticancer therapy.

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Nanosecond pulsed electric field induces exocytosis from pancreatic cancer cells

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Background. Nanosecond pulsed electric field (nsPEF) find its application in oncology by inducing cancer death and enhancing the efficacy of chemotherapy. No studies have considered the potential of nsPEF in releasing of cancer-related molecules in microvesicles yet.

Aim. The aim of the study was to derive the parameters of nsPEF, which would lead to the release of microvesicles. Besides, the study aimed to characterize the content of the vesicles and their role in cancer resistance to the therapy.

Methods. For the study, primary pancreatic cancer cells were used. After treatment with nsPEF, cancer cells were observed with holotomographic microscope. Further, the effects of microvesicles release on the resistance of the cells to paclitaxel were assessed. Next, the content of multidrug resistance proteins was characterized with confocal microscopy. In the end, 3D cell culture models of pancreatic cancer were formed and the effects of nsPEF treatment on spheroid growth were analyzed.

Results. By treatment of the cells with 100 pulses of 200 ns, 10 kHz and 8 kV/cm, we may induce the release of microvesicles from pancreatic cancer cells. The vesicles contain multidrug resistance proteins and after the treatment, cancer cells become more vulnerable to paclitaxel. Moreover, nsPEF reduces growth of the tumor and enhances its adhesive properties.

Conclusion. nsPEF may be used to sensitize cancer cells to chemotherapy via releasing of multidrug resistance proteins in microvesicles.

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Selection of malaria lactate dehydrogenase specific aptamers for point-of-care diagnostic device development

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Background. Malaria is a life-threatening mosquito-borne infectious disease common in subtropical and tropical areas caused by *Plasmodium* genus parasites. The WHO Global Malaria Programme aims for a 90% reduction in the global malaria burden by 2030. To achieve that, effective diagnostic testing should be implemented. Currently, microscopic examination of the blood smear is the golden standard diagnostic method for malaria (CDC). Rapid diagnostic tests based on antibody-antigen reactions are an alternative, however, they have limited storage stability. The aptamer is a single-stranded oligonucleotide that forms a three-dimensional structure enabling binding to the target molecule like the antibody. The advantage of the aptamer use in the diagnostic device is its increased stability and affordable costs for synthesis. To enhance point-of-care diagnostics for malaria, the development of the aptamer-based paper microfluidic diagnostic device is a potential strategy.

Aim. The aim of the study was to select *P. falciparum* lactate dehydrogenase (Pf-LDH)-specific aptamers for the development of a point-of-care diagnostic device.

Methods. Aptamers were selected using protein-SELEX (systematic evolution of ligands by exponential enrichment) method. In brief, randomized ssDNA aptamer library (Integrated DNA Technologies) was incubated with recombinant His-Tag Pf-LDH (SPAN Diagnostics) covered NTA magnetic beads (Dynabeads™). Bound aptamers were eluted and amplified by PCR to increase the Pf-LDH specific aptamer pool. After four SELEX cycles, the aptamer pool was sequenced using the next generation sequencing method (Illumina MiSeq). Selected lead sequences were assessed for their ability to bind to Pf-LDH by ELONA method. In brief, Ni-NTA HisSorb™ plate was coated with Pf-LDH, blocked and then incubated with biotinylated aptamers. Next, poly-HRP streptavidin reagent (Thermo Scientific™) was added, following by TMB substrate incubation. The optical density was measured at 450 nm by Tecan Infinite M200 plate reader. Results were calculated using GraphPad Prism software.

Results. Enrichment of Pf-LDH binding aptamers within the initial oligonucleotide library was observed by detecting an increasing number of retrieved aptamers after each subsequent protein SELEX cycle. Based on enrichment analysis of HTS data, five aptamer sequences were identified as possible Pf-LDH binders.

Conclusion. Pf-LDH aptamers identified in this study offer an alternative to previously identified aptamers binding to malaria-related proteins.

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Antimicrobial resistance patterns among pathogens isolated from blood and synovial fluid samples

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Background. Evaluation of the bacterial etiology and antimicrobial resistance profiles of pathogens causing bloodstream and joint infections is important in assessing the best treatment modality. Delayed appropriate antimicrobial treatment is associated with increased morbidity and mortality in patients with both septic and joint infections.

Aim. To determine the diversity of pathogens causing bloodstream and synovial fluid infections, to evaluate antimicrobial resistance patterns, and to compare results for 2020 and 2022.

Methods. Microorganisms were isolated from clinical blood and synovial fluid samples by *BBL*™ *Crystal*™ *system*, antibiotic susceptibility test was performed by *Bauer-Kirby*™ disc diffusion test and E™-test. Data were obtained from the laboratory information system and isolated bacterial strains were typed according to antibiograms. Data processed using MS Excel 2019.

Results. In 2020 from total of 101 isolates gram-positive bacteria (GPB) accounted for 85.15% (n=86) and gram-negative bacteria (GNB) accounted for 14.85% (n=15) cases. In 2022 from total 126 isolates gram positive were 73.80% (n=93) and 26.20% (n=33) were gram-negative. The most frequently isolated bacteria in 2020 were *Staphylococcus epidermidis* (n=36), *Staphylococcus aureus* (n=29) and *Streptococcus agalactiae* (n=13) while in 2022 most common was *Staphylococcus aureus* (n=52) following *Staphylococcus epidermidis* (n=22) and *Escherichia coli* (n=9).

Methicillin resistant *S. aureus* were 3.45% (n=1) in 2020, but 13.46% (n=7) in 2022. Multi-drug resistant (MDR) *S. epidermidis* isolates were 25% (n=9) and 59.09% (n=13) in 2022. There were no MDR *Streptococcus agalactiae* and *Enterococcus faecalis* in either year.

GPB in 2020 were mainly resistant to erythromycin 15.12% (n=13), GNB in 2020 were mostly resistant to ampicillin and amoxiclavum 60% (n=9). GPB in 2022 were mainly resistant to ciprofloxacin 25.81% (n=24), GNB in 2022 were mostly resistant to cefazolin 48.48% (n=16)

Conclusion.

1. In 2020 gram-positive bacteria exhibited high resistance to erythromycin 15.12% (n=13) while in 2022 GPB were mainly resistant to ciprofloxacin 25.81% (n=24)
2. In 2020 gram-negative bacteria exhibited high resistance to ampicillin and amoxiclavum 60% (n=9) while in 2022 GNB were mainly resistant to cefazolin 48.48% (n=16)
3. In 2020 of all gram-negative isolates, 33.33% (n=5) showed multi drug resistance, while in 2022, 45.45% (n=15) were multidrug resistant
4. In 2020 of all gram-positive bacteria, 16.28% (n=14) showed multi drug resistance, while in 2022, 24.73% (n=23) were multidrug resistant
5. The results show that local monitoring of antimicrobial resistance is an important tool for improving empirical treatment and clinical decision making.

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Vitamin D, total IgE, and high-sensitivity C-reactive protein in patients with multiple sclerosis in the Latvian population

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Background. Multiple sclerosis (MS) is a chronic inflammatory disease of the central nervous system. Vitamin D deficiency predisposes to gastrointestinal infections by altering the gut microbiota, can increase immunity to food proteins, and promote the development of food allergies. Immunoglobulin E (IgE) plays a critical role in the allergic inflammatory process. It is assumed that in MS patients under conditions of vitamin D deficiency, there is an increase in inflammatory markers such as total IgE and high-sensitivity C-reactive protein. However, their prognostic role and relationship with MS progression have remained unnoticed in the literature.

The aim of the study was to estimate serum vitamin D, total IgE, and high-sensitivity C-reactive protein levels in patients with relapsing and progressive forms of MS.

Methods. The examined cohort included 134 patients diagnosed with MS, stratified by the course of the disease, therapy (yes/no), and sex. The investigated parameters were determined in blood serum by means of enzyme-linked immunosorbent assay (ELISA). Statistical analysis was performed with SPSS.25 Statistical Package.

Results. The average vitamin D level was 23.22 ng/mL and did not differ in groups stratified by sex, and disease courses. It was below normal in 39.55% of MS patients and was statistically different (p=2.31E-02) among patients stratified by therapy (yes/no; 22.7 vs. 19.00 ng/mL, respectively). Mean IgE levels were found to be significantly higher in men (29.32 IU/mL) compared to women (14.32 IU/mL) and go beyond the norm in 48.94% of patients with MS. Median CRP levels did not differ in MS patients' cohort stratified by sex and by therapy (yes/no); it was found to be statistically significantly different (p=3.33E-02) among relapsing-remitting and secondary progressive MS groups (0.52 vs. 1.02 mg/ml).

Conclusions. In the examined MS cohort, the following statistically significant increases in the studied parameters were revealed: the average level of vitamin D among patients increased statistically significantly during therapy; the level of IgE was found to be predominant in the patient's group of men compared to women. Serum CRP was found to be elevated in a group of patients with secondary progressive MS and may be useful as a diagnostic tool to detect and monitor inflammation in these patients.

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Vitamin D receptor gene polymorphisms are associated with multiple sclerosis in a case/control study in a Latvian population

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Background. Vitamin D is a lipid-soluble vitamin and hormone. It has a critical role in the pathogenesis of immunity-related diseases including multiple sclerosis (MS) and acts by binding to special vitamin D receptors (VDRs). Genetic variants of the *VDR* gene have been studied as potential factors that affect *VDR* activity. Among the known *VDR* polymorphisms, the most common that influence *VDR* expression within the immune system are Bmsl (rs1544410), ApaI (rs7975232), TaqI (rs731236) and FokI (rs10735810) (Kamel et al. 2014). Our previous bioinformatic analysis demonstrated the potential of their use as molecular markers of MS. In this study, we confirm our results by genotyping in a case/control study in the Latvian population.

Aim. To analyze *VDR* gene genetic variations for a possible association with multiple sclerosis in a case/control study in the Latvian population.

Methods. The case/control study included 296 patients diagnosed with MS and 253 healthy individuals. *VDR* (Bmsl (rs1544410), ApaI (rs7975232), TaqI (rs731236) and FokI (rs2228570)) were genotyped by restriction enzyme site polymorphism. Statistical analysis was performed using the statistical package SPSS.25.

Results. As a result of the study, no statistically significant association between MS and polymorphisms FokI (rs2228570), ApaI (rs7975232), and TaqI (rs731236) was found. However, in the case of Bmsl (rs1544410), a statistically significant risk effect was found for rare allele A ($p<0.02$, OR=1.33, 95% CI [1.04–1.69]) and homozygote AA with rare allele included ($p<0.01$, OR=1.93, 95% CI [1.18–3.16]. The analysis of four-locus (rs2228570-rs1544410-rs7975232-rs731236) haplotypes revealed statistically significant associations related to MS: two protective haplotypes C-G-G-T (the most common in the study cohort), and T-G-T-T ($p<0.001$, OR=0.28, 95% CI [0.14–0.55]); three risk haplotypes: T-G-G-T ($p<0.001$), C-G-T-T and C-A-G-T ($p<0.00001$).

Conclusion. As a result of the case/control study, potential genetic markers associated with multiple sclerosis in the Latvian disease cohort were identified. The interaction of these markers with vitamin D levels in different MS progression groups in Latvians is planned in future studies.

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Genetic variations in the vitamin D binding protein *GC* gene are associated with multiple sclerosis in the Latvian population

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Background. Vitamin D is a nutrient and hormone that plays an important role in the pathogenesis of different autoimmunity-related diseases. Numerous studies have shown that there is a correlation between serum vitamin D levels and the risk of developing multiple sclerosis (MS). Vitamin D acts by binding to a specific vitamin D binding protein (VDBP) which is involved in vitamin D transport and storage. The genetic variations of the VDBP *(GC)* gene have been studied as a potential risk factor for vitamin D deficiency. Our previous meta- and bioinformatic analysis of *GC* gene SNPs rs7041 and rs4588 illustrated the potential of their use as molecular markers of MS. These results need further investigation by genotyping in a case/control study.

Aim. To identify the potential association of *GC* gene genetic variations with multiple sclerosis in the Latvian disease cohort.

Methods. The examined MS cohort included 296 patients, and the control group included 253 healthy individuals. The *GC* (rs7041 and rs4588) were genotyped by the restriction enzyme site polymorphism method. Statistical analysis was performed with SPSS.25 Statistical Package.

Results. Statistical analysis revealed a significant association with MS for both *CG* gene loci studied for common alleles and for homozygotes involving common alleles (rs4588, $p<0.001$ and rs7041, $p<0.01$, respectively). The homozygote genotypes with rare alleles included: AA (rs4588) and TT (rs7041) have been identified as clinical protective factors in the MS cohort ($p<0.001$, OR=0.39, CI 95% [0.24–0.64]) and ($p<0.01$, OR=0.40, CI 95% [0.21–0.76]), respectively. Two-locus risk genotype GG-CC (including common alleles) ($p<0.01$, OR=1.86, 95% CI [1.25–2.76], multiplicative model) and G-C risk haplotype with included common alleles, confirm the found associations.

Conclusions. We present evidence that the *GC* (rs7041 and rs4588) may contribute to the risk of multiple sclerosis in the Latvian disease cohort.

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Heterogeneity of Pierre Robin sequence

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Background. Pierre Robin Sequence (PRS) is a rare genetic disorder that affects the development of craniofacial structures, including the jaw and tongue. The exact prevalence of PRS is not well known, but it is estimated to affect 1 in 8,500 to 1 in 22,000 live births. The aetiology of PRS is not fully understood, but it is considered a complex disorder that results from a combination of genetic mutations and environmental factors that affect the development of craniofacial structures. Studies have shown that mutations in genes such as *SOX9*, *TBX1*, and *DGCR2* can lead to the development of PRS. The *SOX9* gene, which is important for the development of the skeletal system, has been identified as a potential genetic cause of PRS. PRS can also be associated with other syndromes such as Stickler syndrome, velocardiofacial syndrome, Treacher Collins syndrome and CHARGE syndrome. The syndromic forms of PRS are caused by mutations in genes that are involved in the development of craniofacial structures and other organs.

Aim. The aim of the current study was investigating the genetic basis of PRS.

Methods. A total of 3 patients' cases were analysed by whole genome sequencing.

Results. Gene list for PRS case studies was created, which included *SOX9*, *BMPR1B*, *PVRL1*, *COL11A2*, *GAD67*, *COL11A1*, *COL9A1*, *COL2A1*, *TBX1*, *DGCR2* genes.

Two candidate variants were identified in the study.

One was a genetic variant located on chromosome 22 in the *DGCR2* gene. The specific variant is a missense change c.1180C>T or p.Arg394Cys. The *DGCR2* gene encodes a protein called DiGeorge syndrome critical region 2 protein, which plays a role in the processing of microRNAs, small non-coding RNAs that regulate gene expression. The second was a genetic variant located on chromosome 22 in the *TBX1* gene. The specific variant is a missense change c.112T>G or p.Tyr38Asp. The *TBX1* gene plays a role in the development of the heart and other organs during embryonic development. Variations in this gene have been linked to a condition called DiGeorge syndrome or velocardiofacial syndrome which is a complex disorder that affects multiple body systems, including the heart, immune system, and face.

Conclusion. To the best of our knowledge this is the first attempt to identify genetic risk factors in the development of PRS by whole genome sequencing. Limitations are small sample size, nevertheless the interesting variants have been identified, which validate further studies.

PAEDIATRICS

Occurrence of birth defects by region, 2011–2020

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Background. About 3%–4% of babies are born with some type of birth defect. Congenital anomalies (CA) can be diagnosed before birth through ultrasound, amniocentesis, or other methods. In other cases a birth defect is diagnosed after birth through physical examination or a blood test that screens for several disorders in newborns. The presence of CA during delivery can significantly affect infant mortality and morbidity.

Aim. An analysis of occurrence of birth defects in the Latvia regions in 2011–2020.

Methods. A retrospective epidemiological analysis of congenital anomalies from the database of the Register of Patients with Particular Diseases regarding Patients with congenital anomalies. Data from 2011–2020 about live birth with CA (ICD-10; Q00–Q99) were used (n=6635). Place of residence was categorized by region: Riga, Pieriga, Vidzeme, Kurzeme, Zemgale, Latgale.

Results. A total 3.3% (2011–2020) of live birth had anomalies, 3.8% (n=250) of them died, malformations of the circulatory system represent an important part (50.4%; n=126) of them. From all live newborns 23.5% (n=1503) are in age group 0–4 years and 76.5% (n=4882) 5–14 years in 2022. During the study time period highest proportion was observed in Riga 33.3% (n=2124) and Pieriga region 21.0% (n=1340), in other regions around 10% from all birth defects (Kurzeme and Latgale 9.8%, Vidzeme 9.9%, Zemgale 11.7%). Congenital malformations of the circulatory system (Q20–Q28) present 20.4% (n=1305) of all registered CA and are themselves the most frequent birth defect group in births 65.6 per 10000 live births then (Q60–Q64) urinary system 16.7% (n=1067) of all CA and 53.6/10000, (Q65–Q79) musculoskeletal system 14.3% (n=915) and 46.0/10000, (Q50–Q56) genital organs – 12.2% and 39.3/10000. From all CA cases 8.9% (n=568) abnormal findings were indicated that detected prenatally. Higher proportion of prenatally detected cases was observed in Riga ($p<0.001$) – 36.6% (n=208) and Pieriga region ($p<0.01$) 13.9% (n=79) as in other regions. Malformations of the circulatory system most frequently indicated prenatally – 24.5% (n=139), urinary system – 16.7% (n=95) and musculoskeletal system – 17.4% (n=99).

Conclusion. Differences in CA frequencies between regions were found. Higher proportion of CA at births were observed in Riga and Pieriga region as well as prenatally detected. There is not enough information of prenatally detected CA.

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The composition of infant faecal microbiota and association with allergy and introduction of complementary food

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Background. Previous studies on infant gut microbiota suggests that microbiota contributes to the manifestations of allergic diseases and have demonstrated that certain environmental exposures, for example, diet affects the composition of gut microbiota.

Aim. To uncover the composition of the infant faecal microbiota and its relevance to the allergy and diet.

Methods. Study was performed at primary healthcare centres. The parents of children filled out a questionnaire and brought the child's faecal samples: one in 0–6 months of age, another 6–12 months. The 16S rRNS gene sequencing was performed to identify the bacterial taxonomic units. Statistics: t tests, Mann-Whitney test.

Results. In total, 23 children were included (52%, 12/23 boys). Mean age in group 0–6 months was 4 months (CI: 3.4–4.7), in group 6–12 months – 11.0 (CI: 10.9–12.0). The most abundant bacterial families were as follows: *Bifidobacteriaceae* (37%), *Lachnospiraceae* (11%), *Enterobacteriaceae* (10%) in infants till 6 months of age, in infants 6–12 months – *Bifidobacteriaceae* (44%), *Lachnospiraceae* (31%), *Ruminococcaceae* (7%); while most abundant genera were: *Bifidobacterium* (37%), *Escherichia-Shigella* (9%), *Veillonella* (5%), *Blautia* (2%) in 0–6 months; *Bifidobacterium* (47%), *Blautia* (8%), *Veillonella* (3%) in 6–12 months.

There was higher relative abundance of family *Lachnospiraceae* at the age of 6–12 months in infants with allergic manifestations in first half-year of life compared with infants without allergy (0.02 (CI 0.02 to 0.05) *vs* 0.38 (CI 0.08 to 0.68); p=0.02). Similarly, higher relative abundance of *Lachnospiraceae* was at the age of 6–12 months for infants with allergic manifestations in second half-year (0.06 (CI 0.01 to 0.13) *vs* 0.36 (CI 0.22 to 0.49); p=0.0007).

Most infants in our research group started to eat fish, eggs, gluten-containing foods, and soy at the age of 7–12 months (68%, 15/22). Infants who received all these potentially allergic products at the age of 7–12 months had less abundance of genus *Escherichia-Shigella* and *Ruminococcus gnavus group* compared to infants who did not (0.0006 (CI 0.0007 to 0.002) *vs* 0.005 (CI 0.0007 to 0.01); p=0.02 and 0.002 (CI 0.002 to 0.006) *vs* 0.02 (CI 0.005 to 0.04); p<0.001).

Conclusion. Most of infants had normal faecal microbiota with dominance of *Bifidobacterium*. Furthermore, it tends to change during the time. The introduction of complementary foods is an important dietary event during infancy that has an impact on the developing gut microbiome. Further research is needed to understand the whole ecosystem of the infant gut microbiome and to describe bacterial responses to dietary change.

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Albumin and diuretic therapy indications and effectiveness treating paediatric patients with nephrotic syndrome

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Background. Nephrotic syndrome (NS) manifests as massive proteinuria (⩾3 g/l or ⩾3 g in daily urine), oedema, hypoproteinaemia, hypoalbuminaemia, and hyperlipidaemia. The main treatment of NS is corticosteroids. Diuretics are prescribed for patients with significant swelling. If the patient has significant hypoalbuminaemia (<20 g/l), oliguria and severe oedema albumin infusion therapy is prescribed together with furosemide.

Aim. The aim of this study was to evaluate albumin and diuretic therapy indications and effectiveness treating paediatric patients with nephrotic syndrome.

Methods. A retrospective analysis was performed based on the medical histories of the paediatric patients treated in the Department of Children Diseases, Lithuanian University of Health Sciences Kaunas Clinics, in the period of 2017–2021. A total of 71 cases of nephrotic syndrome were identified. The patients who were treated with albumin and/or furosemide infusion were evaluated. Pearson χ^2 criterion and Mann–Whitney test were used in statistical analysis.

Results. Albumin infusion was administered to 66.2% of children. Albumin infusions were more often given to patients with elevated haematocrit level (p=0.033). Multiple albumin infusions were given according to the following indications: 1) hypoalbuminaemia (<20 g/l); 2) oliguria; 3) persistent oedema; 4) increased weight. A single dose of albumin was effective in 25.5% of patients, as there were no criteria for re-dosing. 66.7% of patients treated with furosemide were additionally prescribed more equal 3 times albumin infusions (p=0.002). Patients with higher daily proteinuria were more often prescribed furosemide (p=0.001) and more often required multiple doses of albumin (p=0.014). The subjects were divided into two groups by prescribed treatment: 1) steroids only, steroids and furosemide, steroids and albumin (n=32) and 2) steroids together with albumin and furosemide combination (n=39). In the first group, diuresis at hospital discharge was normal in 75% of patients and in the second group, in 94.9% of patients (p=0.017). Remission of nephrotic syndrome (based on negative urine dipstick readings or trace of protein (<1+) on three consecutive days) was equally achieved in both the first and second groups.

Conclusion. Administration of combined albumin and furosemide is equally effective as treatment with separate medications. Medicaments and their combinations are prescribed strictly according to the indications.

Acknowledgements. The authors declare the absence of conflict of interest.

Follow-up outcomes in patients with hypoxic-ischemic encephalopathy who were treated with therapeutic hypothermia

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Background. Neonates with moderate and severe asphyxia at birth are at risk for neurodevelopmental impairment such as speech disorders, movement disorders, epilepsy, hyperactivity, attention deficit, autism and cerebral palsy.

Aim. To analyze patients' outcomes in an 18-month period who have been diagnosed with hypoxic ischemic encephalopathy (HIE) and treated with therapeutic hypothermia.

Methods. We identified neonates who have been diagnosed with HIE and treated with therapeutic hypothermia in Children's Clinical University Hospital in 2017–2020. Patients were followed-up and analyzed every 3 months using anthropometric data and specific scales such as Munich Functional Developmental diagnosis (MFDD), Hammersmith infant neurological examination (HINE) and Modified checklist for Autism in toddlers, revised (M-CHAT-R). The data were statistically analyzed in IBM SPSS statistics v.29.0.

Results. Overall 46 patients with gestational age from 37 to 40 weeks and 2 days were included in this study: 43.48% (n=20) girls and 56.52% (n=26) boys with mean birth weight of 3455 grams [2015; 4342] in girls and 3691 g [2650; 5470] in boys.

In this patient cohort there were 52.2% (n=24) patients with moderate HIE and 47.8% (n=22) patients with severe HIE. In this patient cohort we did not find significant correlation between gender and stage of HIE (p=0.796).

According to MFDD at 18 months of age 71.7% (n=33) patients had normal psychomotor development and 28.3% (n=13) had abnormal development, among them 10.9% (n=5) had speech disorders, 4.3% (n=2) had attention deficit hyperactivity disorder and 13% (n=6) had psychomotor delay.

There was a negative correlation between developmental disorders and patient's psychomotor development evaluation according to MFDD at the age of 18 months ($p<0.001$; $R=-0.679$).

There was a negative correlation between patient neurological evaluation according to Hammersmith infant neurological examination and development of any psychomotor or speech delay ($R=-0.164$, $p=0.277$).

Conclusion. In patients treated with therapeutic hypothermia we did not find any positive statistically significant correlation between severity of HIE and applied scores of physical, psychomotor, neurological development at the age of 18 months. To identify risk factors for developmental delay in this group of patients' further investigations are needed.

Validation of the VDR gene methylation profile and its genetic background on the association with childhood bronchial asthma in the Latvian population

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Background. Bronchial asthma (BA) is a chronic inflammatory, genetically complex, heterogeneous disease. It has been proposed that epigenetic mechanisms could be involved in BA pathogenesis. DNA methylation of the genes related to vitamin D genetic background has not been described in BA yet.

Aim. To study the level of DNA methylation profiles and genetic variations at the *VDR* gene in patients with childhood BA.

Methods. The case/control group included 115 children diagnosed with asthma and 37 healthy individuals.

1. The *VDR* gene SNPs rs1544410, rs731236, rs7975232, and rs2228570 were genotyped by restriction enzyme site polymorphism on BA main and sex-specific association.
2. Determination of the DNA methylation status in the *VDR* promoter region was performed by real-time PCR procedure, using OneStep qMethyl™ Kit (Zymo Research). DNA methylation extent was calculated using threshold cycle values and the following equation: $100*2(-\Delta C_T)$.
3. Statistical analysis was performed with SPSS.25 Statistical Package.

Results. DNA methylation profile was analyzed in 115 patients and 37 controls. Mean methylation profiles statistically differ by gender in the BA cohort (p=4.36E-02) and were found to be higher in the female disease group (39.2, 95% CI=31.13–47.26), than in the male (30.67, 95% CI=24.82–36.52).

In the control group, rs1544410 and rs731236 rare alleles homozygous were associated with the highest methylation level (p=1.28E-02, η=0.49, and p=1.59E-02, η=0.46, respectively). In turn, the genotype GG (rs7975232) with rare alleles included was found in association with the lowest methylation profile (p=2.59E-02, η=0.41).

The rare alleles genotypes related to rs1544410 and rs731236, and the common alleles genotype TT (rs7975232) were found to be associated with the lowest methylation profile in the disease cohort (p=1.46E-04, η=0.68, p=8.02E-03, η=0.46 and p=7.43E-03, η=0.41, respectively), and thus, can be considered as BA risk factors.

Conclusion. Our study demonstrated differences in *VDR* gene methylation profiles in BA patients stratified by gender. Changes in the level of DNA methylation at promoter regions of the *VDR* gene were found to be associated with its genetic background.

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GASTROENTEROLOGY AND GASTROINTESTINAL ONCOLOGY

Effectiveness of antibiotic prophylaxis and adherence of doctors to it before percutaneous transhepatic cholangiography: one centre retrospective research

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Background. Percutaneous transhepatic cholangiography (PTC) is associated with increased morbidity and mortality rates. Infectious complication rates, that have been previously reported, are between 24 and 40.6%. Quality Improvement Guidelines for PTC, Biliary Drainage, and Percutaneous Cholecystostomy published by the Society of Interventional Radiology (SIR) in the Journal of Vascular and Interventional Radiology recommend administration of antibiotic prophylaxis before PTC. In 2022, in order to reduce complication rates at Pauls Stradiņš Clinical University Hospital, new local guidelines for antibiotic prophylaxis before endoscopic and minimally invasive procedures have been recently elaborated.

Aim. The aim of this study is to evaluate effectiveness of antibiotic prophylaxis in reducing complication rates and adherence of doctors to the guidelines at Pauls Stradiņš Clinical University Hospital.

Methods. A retrospective analysis of 63 oncological patients who had a PTC procedure at Pauls Stradiņš Clinical University Hospital from January 2019 to September 2022. Data was collected about indication of PTC, number of PTC procedures, type of stent inserted, antibiotics given prior to PTC, blood test and culture results, antibiotic sensitivity, and post-PTC complications.

Results. Data of 63 patients with malignancy who underwent PTC were included in the current study; information for 55 patients (median age 67.5 years, 40% were women) was complete, while 8 patients were excluded due to lack of data. Patients were divided into the following groups: group I with antibiotic prophylaxis (21 patients), group II – without (34 patients). Altogether 25 out of 55 (45.5%) patients developed PTC-related complications, 8 (38%) from group I and 17 (50%) from group II. In group I, 23.5% patients developed cholangitis, 9.5% pancreatitis, and 8.8% sepsis, in group II – 42.9% cholangitis, 14.7% pancreatitis, and 19% sepsis. Although in the first group only in 45% cases antibiotic prophylaxis were administered prior to the PTC procedures, that allowed to lower complication rates of cholangitis by 19.4%, of pancreatitis by 5.2% and of sepsis by 10.2%. One patient from group II developed a non-infectious complication – PTC caused bleeding. Risk factors for infectious complications are drain obstruction and repeated interventions.

Conclusion. In 2022, at Pauls Stradiņš Clinical University Hospital implementation of the local guidelines for antibiotic prophylaxis before endoscopic and minimally invasive procedures significantly reduced complication rates after PTC compared to data from 2019–2021. Adherence of doctors to the guidelines must be improved.

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Leading factors in the development of colorectal cancer in Latvia and their distribution by region

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Background. Colorectal cancer is one of the most common types of cancer in Latvia. It is known that the main factors in the development of colorectal cancer are age (>65 years), sedentary lifestyle, adenomatous polyposis, male sex, active and passive smoking, oncological diseases for 1st degree relatives, red meat consumption, alcohol consumption, diabetes mellitus, cholecystectomy, colon polyps and passive smoking.

Aim. The purpose of the work is to determine which risk factors for the development of colorectal cancer dominate in Latvia and to check if there is a difference depending on the region. The study may help determine which risk factors have a greater influence on the development of colorectal cancer in Latvia.

Methods. The study is retrospective. The total number of respondents was 168 patients. In this study data were obtained on modifiable and non-modifiable risk factors and the region of residence using a medical database. Two risk factors – adenomatous polyposis and consumption of red meat – were excluded from the study, due to the lack of data.

Results. Based on the data obtained, it can be concluded that all regions of Latvia have similar dominant risk factors for the development of colorectal cancer, no significant differences between the regions of Latvia and risk factors were found. The main risk factor for colorectal cancer in Latvia was a sedentary lifestyle which was found in 81% (n=136) of all respondents. In second place among the risk factors for colorectal cancer was age, 66% (n=111) of all respondents were over 65 years old. In third place was male gender 50% (n=84) and active smoking 50% (n=84) of all respondents. Less common colorectal cancer risk factors in Latvia were cancer in first-degree relatives 42% (n=71), alcohol consumption 17% (n=29), diabetes mellitus 13% (n=22), passive smoking 7% (n=11), cholecystectomy 6% (n=10) and colon polyps 4% (n=6) of all respondents.

Conclusions. Dominant risk factors for colorectal cancer in Latvia are sedentary lifestyle, age >65 years, male sex and active smoking, while the least common risk factors are oncological diseases for 1st degree relatives, alcohol consumption, diabetes mellitus, cholecystectomy, colon polyps and passive smoking. No significant differences between the regions of Latvia and risk factors were found. It should be noted that to clarify the results, it is necessary to involve a larger number of patients.

Acknowledgements. The authors declare absence of conflict of interest.

Machine learning based breath sensor signal analysis for colorectal cancer detection

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Background. Although colorectal cancer (CRC) screening is included in organized programs of many countries worldwide, there is still a place for better screening tools. Sensor-based exhaled breath analysis has shown significant promise for early cancer detection in scientific and clinical practice because of its high accuracy, low cost, non-invasiveness, and ease of operation. With advances in technology, the breath analysis approach could hold the key to the detection of CRC cancer.

Aim. The aim of the current study was to build machine learning models to classify the sensor responses into cancer and non-cancer groups.

Methods. Altogether 131 study subjects were enrolled in the study: 43 colorectal cancer patients and 88 control group patients. Study subjects' breaths were measured with a hybrid-sensor breath analyzer. The sensor readings for each patient were pre-processed by normalizing against the room air to reduce the impact of environmental air, then smoothing using a median filter and extracting features like minimum, maximum, average and saturation values (at the end of the measurement). The processed data set was used to build machine learning models using C4.5, Random Forests and Neural Network algorithms to classify the sensor responses into cancer and non-cancer groups.

Results. Of 43 cancer patients, 62.7% were males and 37.2% were females; of 88 control patients, 29.5% were males and 70.4% were females. The best classification results were acquired using cost-sensitive Random Forest algorithm with doubled cost for cancer misclassification to compensate for class disbalance. The resulting model showed an overall accuracy of 77.1%, sensitivity of 67.4%, and specificity of 81.8%.

Conclusion. Although the specificity is high, the sensitivity should still be improved before use in any larger tests and pilot screenings. We are hoping to solve this by increasing the data set size (especially the number of cancer patient breath analyses) that would allow us to build more accurate and more sensitive models.

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Multivariate analysis of dietary and lifestyle factors associated with serologic gastric mucosa atrophy

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Background. Gastric cancer prevention includes the eradication of *H. pylori*, reduction of salt consumption, and smoking cessation. Gastric mucosal atrophy (GMA) is known as a status of increased risk of developing gastric cancer, but little is known about the role of dietary factors in the development of GMA. Serum pepsinogen is a good and reliable marker for the indirect assessment of GMA.

Aim. The study aimed to identify different dietary and lifestyle factors associated with decreased serological GMA rates indicative of gastric precancerous lesions using multivariate analysis.

Methods. Participants (40–64 years) of the "Multicenter randomized study of H. pylori eradication and pepsinogen testing for prevention of gastric cancer mortality" (GISTAR study) from two recruitment centers in Jēkabpils and Kuldīga, Latvia, were invited to fill a short dietary questionnaire based on memory recall. For food items included in the questionnaire, the frequency of consumption was graded from "never", "once a week" and up to "multiple times a day". Additionally, by study personnel, the data on sex, age, smoking, and alcohol use were collected and blood samples were taken to analyze serum pepsinogen (Eiken Pg I, II, and Eiken Pg I/II ratio) and assess serological gastric mucosa status. Statistical analysis was done using IBM SPSS22 with binary logistic regression.

Results. A total of 907 adults (mean age 53.72 (SD 6.66), 69% of females) filled out the diet questionnaire and had data on serological pepsinogen measurements. Sixty of them (8.5%) had serologically detected GMA. In multivariate analysis, eating garlic, onions, and leeks was positively associated with GMA ($p=0.01$). Paradoxically, smoking was negatively associated with GMA ($p=0.03$). Consumption of alcohol, buckwheat, red meat, and meat products, as well as typical Nordic fruits like apples, pears, and plums, did not show a statistically significant association in this study.

Conclusion. Eating garlic, onions, and leeks was associated with serologically detected GMA. The observed association between smoking and lower rates of serological GMA could be explained by the lower specificity of pepsinogen tests in smoking individuals. Additional studies of the impact of dietary factors on gastric atrophy would be necessary to elucidate the role of the diet.

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SURGERY

Laparotomy closure technique impact on wound healing

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Background. The problem is of outstanding importance since there are still no strong evidence of best technique to close emergency laparotomy incisions to prevent early complications such as wound dehiscence and infection that may lead to higher mortality rates, reoperations, readmissions, and higher costs.

Aim. The aim of the current study was to collect single centre data from Riga East Clinical University hospital general surgery department of both elective and emergency laparotomies to compare used abdominal closure techniques to find the best abdominal wound closure technique which could prevent such complications as wound dehiscence and infection.

Methods. Altogether 223 patients who underwent surgery with laparotomy incision were enrolled in the study dating from January 2021 to September 2022. 155 were patients with emergency laparotomy, but 68 patients had elective laparotomy. Data about laparotomy closure technique for example used suture, suturing technique, length between sutures, skin closure technique and other data that might influence wound healing were collected. After surgery patients were observed for complications such as wound dehiscence and wound infection.

Results. All together 6.3% (n=14) of patients had wound healing complications after laparotomy incision. 4% (n=9) of patients were diagnosed with wound dehiscence. Although more wound dehiscence was found in group with interrupted suture (3 from 33 cases (9.1%), comparing continuous suture 6 from 190 cases (3.2%)), the difference was not statistically significant (p=0.132). But there was statistically significant difference comparing suture distance impact on wound infection. In group with wound infection suture count on 1 cm wound length was 0.75 (IQR 0.48–0.75) comparing control group where suture count on 1 cm wound length was 1.20 (IQR 1.00–1.67) (p=0.04) therefor indicating more benefits from "small bites" technique comparing "large bites" technique.

Conclusion. Among all data collected there are significant variation in laparotomy incision closure technique between patients who suffered from wound dehiscence or wound infection. Therefore, there needs to be detailed investigation in possible other causes, not only suturing technique that may impact on wound healing.

Acknowledgements. The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Intraoperative balloon dilatation of the *papilla Vateri* as a treatment method in patients with choleidoholtiasis

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Background. Choledocholithiasis is a common complication of gall stones disease. Two-stage therapy is the standard treatment for choledocholithiasis with calculous cholecystitis. The first stage is to make endoscopic retrograde cholangiopancreatography with papillotomy and stone evacuation from common bile duct (CBD) and second stage is laparoscopic cholecystectomy (ERCP/LS). An innovative, one-stage method - laparoscopic cholecystectomy with intraoperative balloon dilatation of the papilla Vateri and anterograde evacuation of gallstones to duodenum (IBDPV/LS) evaluated in our hospital.

Aim. The aim of this study is to compare one-stage IBDPV/LS with two-stage ERCP/LS.

Methods. A retrospective, comparative study in Riga East Clinical University Hospital Gailezers was done from January 2021 to December 2022. All patients is divided in to the two groups. The first group underwent one-stage treatment as an approbation treatment method for patients with choledocholithiasis and calculous cholecystitis. The second patient group underwent two-stage therapy. Hospitalization time, lipase level after procedure, gallstones diameter, number and type of postoperative complications were analysed.

Results. A total 64 patients with choledocholithiasis and calculous cholecystitis were performed from 2021 to 2022. Of which 18 patients underwent IBDPV/LS and 48 patients used ERCP/LS treatment. The mean diameter of gallstones in the CBD was 6 mm in first group, while in the second group mean was 7 mm. The median hospitalization duration for patients with one-stage treatment was 8 days (IQR 7–9 days), and for two-stage therapy it was 14 days (IQR 10–20.5 days) ($p<0.01$). Two patients developed mild acute postoperative pancreatitis in IBDPV/LS group but in the ERCP/LS group, moderate postoperative acute pancreatitis developed in 4 patients after ERCP. The median lipase level in the IBDPV/LS group was 38.2 U/L (IQR 30.5–75.15 U/L), but in the ERCP/LS group, it was 57 U/L (IQR 25.05–172.5 U/L).

Conclusion. Laparoscopic cholecystectomy with intraoperative balloon dilatation of the papilla Vateri and anterograde evacuation of gallstones is associated with a shorter hospitalization time, lower complication rate compared with a two-stage method.

Single-stage ERCP and laparoscopic cholecystectomy versus preoperative ERCP followed by laparoscopic cholecystectomy in the management of cholecysto-choledocholithiasis

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Background. Choledocholithiasis is the presence of biliary stones within the common bile duct in patients with gallstone disease. It is estimated that up to 15% of patients undergoing cholecystectomy have choledocholithiasis. Treatment options include laparoscopic and endoscopic approach. These methods can be done gradually (two-step) or in combination (one-step), therefore there are controversies regarding the best management option.

Aim. To evaluate and compare efficiency and safety of the one-step to the two-step approaches for managing choledocholithiasis.

Methods. A retrospective study was done in Jelgava City Hospital, Latvia. Totally 206 patients with proved choledocholithiasis by magnetic resonance cholangio-pancreatography or computed tomography were included in the study between January 1, 2016, and November 30, 2022. Main outcomes included hospital stay, length of anaesthesia, length of procedures as well as postoperative and postERCP morbidity according to Clavien–Dindo classification were evaluated. The statistical analysis was done by SPSS Statistics 21.0 software.

Results. A total of 206 patients were included in the study. One-step approach was done in 122 patients, two-step approach in 84 patients. The median length of hospital stay (10 days, IQR 6–14) in the two step group was statistically significantly longer than that in the one-step group (4 days, IQR 2–6) ($p<0.001$). The median anaesthesia length (150 min, IQR 125–175) in the one-step group was statistically significantly shorter than it was in the two-step group (205 min, IQR 160–250) ($p<0.001$). The median duration of the operation and procedure in the one-step group was 90 min (IQR 55–125), which was statistically significantly shorter than it was in the two-step group (120 min, IQR 85–155) ($p<0.001$). Frequency and severity of complications were significantly more common in the two-step than one-step group (7.1% versus 0.8%, $p<0.002$). One patient suffered duodenal perforation (two-step approach group) and 3 patients suffered mild acute post-ERCP pancreatitis (1 in one-step group and 2 in two-step group).

Conclusion. One-step approach for treating cholecysto-choledocholithiasis in this study proved to be a treatment modality that was associated with lower complication rate, as well as shorter hospital stay and shorter anaesthesia and procedure times, compared to the two-step approach group.

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Change in spinopelvic parameters after single-level anterior lateral interbody fusion for degenerative spinal deformity

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Background. Anterior lateral interbody fusion (ALIF) is a treatment option for degenerative lumbar disc disease, which causes loss of lumbar lordosis and spinopelvic parameters, leading to chronic back pain, stiffness, and decreased quality of life. Restoration of lumbar spinopelvic parameters has been proven as an important determinant of outcomes in spine surgery.

Aim. To evaluate the changes in the spinopelvic and regional lumbar parameters after a single-level (L5–S1) anterior lateral interbody fusion.

Methods. Patients who underwent ALIF surgery from 2016 to 2022 were included in a single-centre retrospective cohort study. Patients with a history of multi-level or combined ALIF and posterior correction were excluded. The spinopelvic parameters that were evaluated in radiographs before and after surgery included pelvic incidence (PI), pelvic tilt (PT), sacral slope (SS), lumbar lordosis (LL), segmental lumbar lordosis (LLseg), regional lumbar lordosis (LLreg), and disk height changes at the L3–L4 and L2–3 level.

Results. Twenty-two patients were included with mean pelvic incidence of 53° (SD, 14.4°). After surgery, regional lordosis increased from mean 42.5° (SD, 10.2°) to mean 44.5° (SD, 8.6°), sacral slope increased from mean 34° (SD, 9.2°) to mean 37.1° (SD, 10.2°) (p=NS), pelvic tilt decreased from mean 20° (SD, 7.6°) to mean 15.3° (SD, 6.7°) (p=NS), and overall lumbar lordosis decreased from mean 52.4° (SD, 12.9°) to mean 49.1° (SD, 11.4°). Segmental lumbar lordosis was found to be significantly increased from mean 27.86° (SD, 9.3°) to 34.5° (SD, 7.5°) (p=0.013).

Conclusions. Our findings suggest that one-level ALIF prosthesis significantly increases segmental lordosis and improves sagittal spinopelvic parameters. Results indicate insignificantly decreased overall lumbar lordosis following the surgery, likely due to the compensatory mechanisms in upper lumbar spine, which need to be assessed in further research.

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CARDIOVASCULAR AND REGENERATIVE MEDICINE

The difference of oxidative stress readings in patients diagnosed with chronic heart failure

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Background. With respect to structural and functional cardiac disorders, heart failure (HF) is divided into HF with reduced ejection fraction (HFrEF) and HF with preserved ejection fraction (HFpEF). HFrEF is known to be the outcome of myocardial ischemia and infarction. Notably, HFpEF is associated with dysregulated metabolism and chronic hypertension, contributing to oxidative stress and myocardial dysfunction. Oxidative stress (an imbalance between the increased formation of reactive oxygen species (ROS) and the elimination or neutralization of ROS by an antioxidant system) plays an important role in the development of chronic HF and correlates with left ventricle dysfunction and hypertrophy in the failing heart. So, oxidative and antioxidative stress biomarker levels should be higher in HFpEF.

Aim. The aim was to compare oxidative stress (nitrotyrosine, dityrosine, protein carbonyl, malondialdehyde, oxidized HDL) and antioxidative biomarker (total plasma antioxidant capacity, catalase (CAT)) levels in the blood between HFrEF and HFpEF patient groups.

Methods. A total of 60 CHF patients were enrolled in the study and divided into two groups: HFpEF of <40% (n=27) and HFrEF of ⩾40% (n=33). Nitrotyrosine, dityrosine, protein carbonyl, malondialdehyde, oxidized high density lipoprotein cholesterol (HDL) and antioxidative readings' (total plasma antioxidant capacity, catalase (CAT)) concentrations were measured in the blood.

Results. No statistically significant differences between the groups in the aforementioned variables were observed in the current study. Further, we divided the entire study sample into two groups based on the median values of malondialdehyde, protein carbonyl and oxidized HDL levels. No statistically significant differences of serum oxidative/antioxidative stress markers and left ventricular ejection fraction values were found between the groups of different malondialdehyde concentration (⩽114.29 vs. >114.29 μg/L), different protein carbonyl (⩽259.95 vs. >259.95 U/mL) and oxidized HDL levels (⩽3.06 vs. >3.06 pg/L). There was no correlation between the readings too.

Conclusion. Oxidative stress and antioxidative biomarker levels in the blood between HFrEF and HFpEF patient groups do not differ.

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A new D-shape endograft for patients with abdominal aortic aneurysms

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Background. The new D-shape endograft is a balloon-expandable device that consists of 2 bilateral aortic endografts and 2 iliac endografts, and its research takes place in Pauls Stradiņš Clinical University Hospital (PSCUH) since 2014. The purpose of the D-shape endograft is to exclude aneurysm from the circulation and to reduce intra-aneurysmal pressure for patients with infrarenal AAA. Specific complications observed during endovascular aneurysm repair (EVAR) are endograft migration and endoleaks. Multi-center prospective, controlled clinical trial (2011–2015) early results have shown that Altura Endograft System is a safe method for treating patients (N=90) with AAA, and is associated with favorable midterm results (Krievins et al., 2018). The results of a five-year period (N=100) in a single-center study (PSCUH) reveals the potential of D-shape endograft and underlines the importance of further D-prosthesis research (Krievins et al., 2019).

Aim. To prove the efficacy and safety of D-shape endograft treatment for patients with abdominal aortic aneurysms.

Methods. This study covers all patients with implanted *Altura* device in Latvia since 2014 (N=175). Prospective study with retrospective data analysis took place in PSCUH Scientific Institute. Follow-up CTA and clinical examination were done in first month and every year after the reconstruction. Analysis of CT scans were done by radiologist.

Results. A total of 175 patients (age 72.4±8.9 years) with AAA (diameter 57.4±11.4 mm) were treated. Endografts were successfully implanted in all patients (100%). There have not been any peri-operative deaths within the first 30 days. Mean fluoroscopy time was 22±11 min, mean contrast 156.7±71.3 mL, mean blood loss 63.8 mL.

Cumulative major adverse event rate (MAE: death, stroke, paraplegia, MI, respiratory failure, bowel ischemia and blood loss ⩾1000 ml) was 0% in the 1st year, 2.4% in the 2nd year (2/88) with no additional events up to 5 years. Clinical success (freedom from type I/III endoleak or re-intervention) was 95.4% at 30 days (8/175), 94.4% at one year (6/112), 86.7% at 2 years (11/88), 88.3% at 3 years (7/60), 92.5% at 4 years (3/40), and 91.7% at 5 years (2/24). Twenty-nine (16.6%) type II endoleaks were observed. During median follow-up of 24 months (1–60 months), there were no aneurysm ruptures or AAA-related deaths. Secondary procedures were performed in 28 patients: 8 for type Ia endoleak, 2 for type Ib endoleak, 5 for iliac stenosis, and 13 for type II endoleak.

Conclusion. The new D-shape endograft is an effective and safe method for treating AAA.

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Effect of lipid extract of sea buckthorn seeds on patients with residual cardiovascular disease risk

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Background. Coronary heart disease (CHD) remains one of the leading causes of death worldwide. Despite the availability of effective preventive therapies, CHD events still occur due to the residual risk. Supplementation with plant-derived extracts could play a role in risk reduction.

Aim. To determine whether sea buckthorn seed lipid extract (SBS-LE) containing polyunsaturated, monounsaturated fatty acids and fat-soluble vitamins could reduce the residual risk for CHD patients.

Methods. 35 of 100 CHD patients with optimal treatment were included in this pilot trial at the Latvian Centre of Cardiology. At the baseline (V_0) body mass index, blood pressure, and heart rate were measured, blood samples were taken for lipid profile (total cholesterol (TC), triglycerides (TG), low-density lipoproteins (LDL), high-density lipoproteins (HDL)), glucose, C-reactive protein (CRP) and uric acid. Two SBS-LE soft gelatine capsules (2000 mg) were prescribed per day for three months in addition to standard medical therapy with statins and ezetimibe. SBS-LE was produced by supercritical fluid extraction with CO_2. After one month (V_1), patients were questioned about possible side effects and adverse events. After three months (V_3), patients were observed for adverse events, change of medication and blood samples were taken for the same analysis as at the baseline. The efficacy of the SBS-LE was evaluated as changes in measured parameters.

Results. Of the first 35 patients, 17 (49%) were men, and the mean age was 69.28±10.48 years. Thirty-three (94.29%) patients were adherent to the SBS-LE supplement. There were no side effects, and the tolerability was good. One patient (2.86%) was hospitalised with worsening heart failure. In 2 patients (5.71%) with hypertension, antihypertensive medication was changed. In 4 patients (11.43%), the dose of statins was increased.

At 3 months, the systolic blood pressure was reduced by 3.15±7.86 mmHg from 140 [IQR=125; 140] mmHg at V_0 to 135 [IQR=125; 135] mm Hg at V_3 (p=0.048). There was also a trend to a lower blood glucose level 5.63 [IQR=5.11; 6.35] mmol/l at V_3 vs. 5.95 [IQR=5.47; 6.55] mmol/l at V_0 vs (p=0.106). There were no significant differences in other parameters.

Conclusion. Preliminary results of our study show a reduction in systolic blood pressure. SBS-LE seems a promising additional preventive strategy to reduce CHD residual risk. Enrolment in this study continues.

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True and complex bifurcation stenting – treatment techniques and long-term outcomes

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Background. PCI for bifurcation disease is known to be technically challenging and has historically been associated with lower procedural success rates and worse clinical outcomes than non-bifurcation lesions.

Aim. The aim of this study was to evaluate intrahospital and long-term outcomes of patients who underwent percutaneous coronary intervention (PCI) for true bifurcation lesions involving main vessel and side branch with diameter more or equal 2.5 mm and how often systematic double-stenting technique was used in complex coronary bifurcations.

Methods. A retrospective analysis of the ongoing Coronary Bifurcation Treatment registry in Latvia Centre of Cardiology (PCI performed from 01.01.2017. to 31.12.2022.) and follow-up after 1 year. Study population was divided into two groups: provisional single stenting (1 stent) and systematic double stenting (2 stent). Analysed were complex coronary bifurcations according to DEFENITION criteria. A total of 651 patients were included in this study (1 stent – 528 patients, 2 stent – 123 patients).

Results. Complications were perforation (1 stent 0% vs. 2 stent 0.8% (n=1), p=0.189), SB occlusion (1 stent 2.8% (n=15) vs. 2 stent 0.8% (n=1), p=0.330), no reflow phenomenon (1 stent 0.2% (n=1) vs. 2 stent 0%, p=0.629), cardiogenic shock (1 stent 0% vs. 2 stent 0.8% (n=1), p=0.189), periprocedural myocardial infarction (MI) (1 stent 4.0% (n=21) vs. 2 stent 6.5% (n=8), p=0.223) and intrahospital stent thrombosis (1 stent 0% vs. 2 stent 0.8% (n=1), p=0.189). One-year follow-up till now was possible in 486 patients (1 stent – 404 patients, 2 stent – 82 patients). There were cases of death (1 stent 2.4% (n=10) vs. 2 stent 3.5% (n=3), p=0.472), hospitalization because of MI (1 stent 0.7% (n=3) vs. 2 stent 0%, p=0.435), hospitalization because of stroke (1 stent 0.3% (n=1) vs. 2 stent 1.2%, (n=1), p=0.308), target lesion revascularization (1 stent 1.6% (n=6) vs. 2 stent 1.2% (n=1), p=0.840) and target vessel revascularization (1 stent 3.4% (n=13) vs. 2 stent 3.8% (n=3), p=0.747). There were no cases of definite late stent thrombosis. 64 cases of complex coronary bifurcations were identified (1 stent 7.0% (n=37) vs. 2 stent 22.0% (n=27), p<0.001)

Conclusion. Intrahospital and long-term complication rate in the treatment of true coronary bifurcation lesions was low. There were no cases of definite late stent thrombosis. In complex coronary bifurcations preferred was systematic double-stenting technique.

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Cardiovascular events in patients with familial hypercholesterolemia before and after inclusion in the Latvian Registry of Familial Hypercholesterolemia

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Background. Familial hypercholesterolemia (FH) is the most common genetic disease which causes premature coronary artery disease (CAD) due to accumulation of low-density lipoprotein (LDL). There has been no previous follow-up study of FH patients in Latvia.

Aim. To evaluate the incidence of cardiovascular events in patients with suspected familial hypercholesterolemia before and after inclusion in the Latvian Registry of Familial Hypercholesterolemia (LRFH).

Methods. This retrospective study involved patients included in the LRFH from February 2015 to November 2022. Inclusion criteria: previously consented for communication, probands with definitive/probable FH according to Dutch Lipid Clinic Network score and their first-degree relatives with LDL-cholesterol >95%. Patients were interviewed by phone using a pre-formed questionnaire in November and December 2022. Results were processed using IBM SPSS 29 statistics.

Results. By November 2022, a total of 513 patients were diagnosed with clinical FH, and 302 patients were eligible for this study. Only 252 patients (83.4%) were reachable by phone, and 18 of them refused the interview. Among the interviewed 234 patients, the majority were women (62%, n=145), mean current age was 44±11.2 years in men and 57±11.9 years in women. The main results of cardiovascular outcomes before and after inclusion in LRFH are summarized in Table. Premature CAD (men <55 years, woman <60 years) was diagnosed in 58 (24.7%) and 31 (13.2%) patients, respectively (n=89, 38.0% in total). Out of 123 CAD (52.5% in total) cases, 82 patients (35.0%) had asymptomatic CAD, 37 patients (15.8%) before inclusion and 45 (19.2%) during follow-up.

	Before inclusion in LRFH	During follow-up	Total
CAD	72 (30.7%)	51 (21.8%)	123 (52.5%)
MI	14 (6.0%)	1 (0.4%)	15 (6.4%)
PCI	25 (10.6%)	28 (11.9%)	53 (22.5%)
CABG	2 (0.8%)	4 (1.7%)	6 (2.5%)

Conclusion. Although CAD was present or newly diagnosed in half of interviewed FH patients and about one fourth of them had undergone revascularization, there were very few major cardiovascular events at the follow-up. These data imply a generally good prognosis of FH patients managed in Latvia. However, health status was not known in a substantial number (16%) of patients, which necessitates a more systematic approach to acquire high-quality data, preferably with administrative data integration.

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Relationship between segments with prominent myocardial noncompaction of the left ventricle and conduction disturbances

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Background. Usually referred to as an isolated condition, left ventricular noncompaction is a structural defect of the left ventricle that can occasionally be linked to other structural cardiac diseases. It is yet to be determined whether a distinct noncompaction in particular segments of the left ventricle correspond to specific abnormalities in ECGs.

Aim. The aim of this study was to assess conduction disturbances in patients with noncompaction cardiomyopathy at Pauls Stradiņš Clinical University Hospital.

Methods. This retrospective single-center study included adult patients who had cardiac MRI performed at Pauls Stradiņš Clinical University Hospital in the timespan from January 1, 2021, to October 1, 2022. In order to assess the compacted and noncompacted layers of the myocardium, syngo.CT Cardiac Function software was used. Mathematical data processing was performed using descriptive statistics.

Results. Out of 811 cardiac MRIs that were performed in the respective period of time, noncompaction cardiomyopathy was detected in 5.5% (n=45) of the cases. There were 49% (n=22) of women and 51% (n=23) of men. In 71% (n=32) of the cases, ECGs were available for assessment, 19% (n=6) of which presented with atrial fibrillation, 34% (n=11) with ventricular extrasystoles, 22% (n=7) with left bundle branch block, 3% (n=1) showed sinoatrial node block, and 3% (n=1) nonspecific ST-T segment changes. In 19% (n=6) of the ECGs there were no abnormal findings. It was concluded that most of the conduction disturbances presented in patients who had the highest noncompacted versus compacted myocardial layer ratio in segments 6, 12, and 13.

Conclusion. In our study, most of the conduction disturbances were observed in patients who displayed the highest noncompacted versus compacted layer ratio in anterior and lateral portions of the left ventricle.

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Arrhythmias in discharged patients after acute coronary syndrome with ST segment elevations

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Background. Studies have shown that patients after myocardial infarction may develop some form of arrhythmia.

Aim. The aim of the current study was to assess the patients' risk factors, incidence, and types of arrhythmias in discharged patients after acute coronary syndrome.

Methods. Altogether 29 patients were enrolled in the study after revascularization of the culprit vessel. After discharge an ECG-monitoring device was attached to the chest. The device recorded patients' ECG for 7 days. Patients' echocardiographic, laboratory, angiographic data were collected and correlation with arrhythmias were sought.

Results. Of the 29 study patients 83% were male and 17% were female. The mean age was 57.34 years. A total of 69% of patients reported current smoking. Pre-existing coronary artery disease was known in 41% of patients, arterial hypertension in 76% of patients, atrial fibrillation in 10% and 23% of patients already had the diagnosis of chronic heart failure. History of percutaneous coronary intervention was present in 24% of patients, and 20% of patients had previous myocardial infarction. On study population mean total cholesterol was 5.84 mmol/L, LDL 3.7 mmol/L and triglycerides 1.23 mmol/L. Mean CK-MB upon admission was 68.55 ng/ml, while troponin-I was 18 555 ng/L. Before the hospitalization 17% of patients were taking aspirin and 10% statins.

Non-sustained monomorphic ventricular tachycardia episodes were documented in 24% of patients. There was one patient who developed atrial flutter with a total of 13 episodes. The most common rhythm disturbance was ventricular premature beats with average 113 premature beats per day. A total of 17 patients developed sinus bradycardia episodes. The longest bradycardia episode during the day was 63 min, while during the night, 80 min.

Patients with lower ejection fraction had higher heart rate during the night ($r=-0.37$; $p=0.048$). Higher maximum CK-MB correlated with higher average heart rate during the day ($r=0.391$; $p=0.036$) and lower during the night ($r=-0.457$; $p=0.013$). There was a positive correlation between BNP and average ventricular ectopic beats per hour ($r=0.366$; $p=0.051$). Higher total cholesterol values had a positive correlation with the amount of atrial run episodes ($r=0.373$; $p=0.047$).

Conclusion. The study population were high cardiovascular risk patients. The most common rhythm disorder in the patient population was ventricular premature beats. Non-sustained ventricular tachycardia was not a frequent finding in the patient population. Patients with heart failure markers and higher cholesterol were at a greater risk of developing arrhythmias.

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Efficacy of pulmonary vein isolation to treat atrial fibrillation in Pauls Stradiņš Clinical University Hospital

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Background. Atrial fibrillation (AF) is the most common sustained cardiac arrhythmia in adults AF. Pulmonary vein isolation (PVI) with catheter ablation is a well-established treatment for the prevention of AF recurrences. Estimation of the efficacy of AF catheter ablation is important to improve outcome of those procedures.

Aim. Evaluate efficacy of the PVI by catheter ablation to treat AF in Pauls Stradiņš Clinical University Hospital.

Methods. We included AF patients to whom PVI catheter ablation was done in Pauls Stradinš Clinical University Hospital Latvian Cardiology Centre.

PVI was reached using radiofrequency or cryoablation catheter ablation. Preoperative, intraoperative and postoperative data were collected. Patients were followed up for 3, 6, and 12 months. AF recurrences were estimated by Holter monitoring and clinical AF episodes before each follow-up visit.

Results. From 2017 to 2020, a total of 301 patients were included. There were 167 males and 134 females. The mean age was 63 (SD, 11.9) years, and BMI, 29.5 (SD, 5.4) kg/m^2. At least 2 antiarrhythmic medications failed prior to PVI. Median for CHA2DS2-VASc – 2, HAS-BLED – 1. EHRA scale - 3.2 (SD, 0.1). On average, there were 56 (SD, 15) AF episodes per year.

Hypertension was observed in 68.9% of the cases; heart failure, 27.5%; diabetes mellitus, 12%; coronary artery atherosclerosis, 23.2%; and ischaemic stroke, 20%. More than half (59.2%) had one or more electrocardioversion. Persistent and paroxysmal AF was documented in 35.8% and 64.2% of the cases, respectively. The average length of atrial fibrillation before PVI was 5.4 (SD, 0.5) years. Majority of the cases AF length was 24 to 36 h. PVI by radiofrequency in 54% and cryoablation in 46% of the cases. Quality of life improved from 47.8% (SD, 2) to 75.29% (SD, 2.3) by a visual scale. At 12-month freedom of atrial fibrillation was 68%.

Conclusion. Efficacy of AF treatment by PVI in Latvian Cardiology centre is 68% after 1-year follow-up.

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The evaluation of *ex vivo* cell expansion process automation

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Background. The advanced therapy medicinal product development drives research towards more cost-effective, time and labour-effective human cell *ex vivo* cultivation methods. The bioprocess automation, traceability and repeatability are challenging issues in the field. To resolve these aspects this study is focusing on development of optimal bioprocess control protocol.

Aim. The current stage of study is aimed to assess *ex vivo* cell expansion process control automation protocol performance stability.

Methods. The automated bioreactor system constructed by research team was applied to evaluate cell expansion process. Disposable transfusion bioreactors were applied. The cell culture was seeded, and consumption of glucose and production of lactate were monitored. The glucose and lactate metabolism were monitored by multi-channel photometric device constructed in previous stages of the study. Same process control setpoints were applied for all cell expansion trial runs.

Results. Maximum average consumption of glucose reached 220.7 mg/d and minimum 17 mg/d. Maximum average production of lactate reached 107 mg/d and minimum 5 mg/d respectively. Average cultivation media used for feed during cultivation cycle reached 523.6 ml. Maximum daily cultivation media feed/overflow reached 96% of total volume in system. Average time to reach the end point of bioprocess were 16 days. The cell expansion media evaporation ratio did not exceed 1% of total media used. All performed trial runs (N=10) reached acceptance criteria. No bacterial contamination was observed during cell expansion.

Conclusion. The study results show minor differences between individual trial runs not exceeding acceptance criteria.

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Lipid extract of sea buckthorn seeds and blood fatty acid profile

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Background. A healthy lifestyle and balanced nutrition are key factors for good long-term general health, well-being and disease prevention. Nowadays there are a lot of risk factors affecting our health such as high-stress levels, unhealthy and irregular nutrition, inadequate rest as well as other social and environmental factors. One of the strategies to prevent disease is to supplement daily nutrition with plant-derived nutraceuticals. An important factor affecting nutraceutical efficiency is their absorption.

Aim. To determine the absorption of sea buckthorn seed lipid extract (SBS-LE) containing a combination of polyunsaturated, monounsaturated fatty acids, fat-soluble vitamins, and other bioactive compounds.

Methods. Five healthy subjects were recruited for this pilot study conducted at the University of Latvia. Five blood samples were taken from each subject in the following order: first, before administration of SBS-LE; second, one hour after administration of 4000 mg of SBS-LE; third, 6 hours after administration; fourth and fifth, 24 and 31 hours after administration respectively. During this bioavailability, study subjects were instructed to consume a diet containing fibres and proteins, but to avoid consuming any fat-containing products. SBS-LE 4000 mg was given in form of 4 standardized soft gelatine capsules in total containing 1480 mg of linoleic acid (LA), 1200 mg of alpha-linolenic acid (ALA), 680 mg of oleic acid and 28 mg of beta-sitosterol. SBS-LE was produced by supercritical fluid extraction with carbon dioxide. Blood samples were analysed with a gas chromatography–flame ionization detector (GC-FID).

Results. The mean age was 39.00±12.63 years and 3 (60%) were men. The increase of unsaturated fatty acid levels between baseline and 6 to 24 hours after SBS-LE administration was 10.50±9.68% (p=0.067); this increase was mostly by linoleic acid C18:2n6 (LA) 5.20±4.85% (p=0.077). There was a decrease in saturated fatty acid levels by 4.09±6.74% (p=0.122), mostly due to a decrease in palmitic acid C16:0 by 5.97±6.80% (p=0.110).

Conclusion. Results show that after administration of single dose of sea buckhorn seed lipid extract blood fatty acid profile changes and SBS-LE has the potential to be used as a nutraceutical in terms of absorption. Results show trends in unsaturated fatty acid increase and decrease in saturated fatty acid concentration after SBS-LE administration.

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INFECTIOUS DISEASES AND PULMONOLOGY

Variability of selected biochemical markers in hepatitis A – an 8-week observation study

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Background. Hepatitis caused by the hepatitis A (HAV) virus is one of the most common fecal-oral transmitted diseases. Its clinical manifestation can vary greatly from asymptomatic to life-threatening liver failure. Elevated serum activity of alanine aminotransferase (ALT), alkaline phosphatase (ALP), aspartate aminotransferase (AST), gamma-glutamylotransferase (GGTP), serum bilirubin (BIL) concentration with the presence of anti-HAV antibody is the main markers confirming the HAV infection.

Aim. This study estimates ALT, ALP, and BIL serum levels changes during 60-day observation.

Methods. We analyzed the results of 92 patients infected with HAV admitted to our department from January 2017 to December 2021.

We analyzed ALT, ALP, and BIL results on admission acting as point zero and used test results within a range of the subsequent 60 days to estimate value function.

We used Functional Data Analysis (FDA) to model the levels of ALT, ALP, and BIL as functions of time, with the only assumption being the smoothness of the underlying process. The mean level was established point-wise and confidence intervals were calculated assuming the normality of measurements.

Results. The mean ALT value on day 0 was 2196.8 IU/l (confidence intervals: 1606.9–3003.3) and dropped to 110.3 IU/l (confidence intervals: 50.0–243.2) on day 60 (ALT reference range: 10–40 IU/l). The mean bilirubin value on day 0 was 102.5 μmol/l (confidence intervals: 80.5–130.5) and dropped to 18.4 μmol/l (confidence intervals: 11.2–30.4) on day 60 (BIL reference range: 5.1–17.1 μmol/l). The mean ALP value on day 0 was 186.7 IU/l (confidence intervals: 158.4–200.1), dropping to 72.7 IU/l (confidence intervals: 56.4–93.7) on day 60 (ALP reference range: 30–120 IU/l). The mean value of each parameter declined rapidly until the 40th day when the decrease slowed. Out of all parameters, only ALP means value reached the normal range within 60 days, hitting 119.8 IU/l on day 18.

Conclusion. Elevated serum ALT activity and serum bilirubin concentration can be detected 8 weeks after the first symptoms of HAV infection.

Serum ALP activity normalizes faster than ALT and bilirubin concentration in 8-week observation.

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Stethoscopes as potential vectors in transmission of bacterial infections in three regional hospitals

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Background. Nosocomial infections are a major issue in hospital settings. It is widely known that medical employees and their equipment can spread infectious diseases.

Aim. This study aimed at evaluating the presence of bacterial contamination on the stethoscopes of medical personnel in three regional hospitals: A, B and C.

Methods. Together 48 stethoscopes from 23 different units belonging to physicians, residents and interns at the hospitals A, B and C were sampled for bacterial cultures by swabbing the entire surface of the diaphragm of the stethoscope with a sterile cotton-tipped applicator, placed in Amies transport medium. Next cultures were transferred to blood agar, mannitol-salt-agar and Levine EMB agar plates. The plates were incubated at 37°C for 24 hours and examined for colony growth, then Gram stained and microscoped. Next using streak plate method transferred to Trypticase Soy Agar to obtain a pure culture that could be used in VITEK-2 for accurate microbial identification. To obtain additional information, a questionnaire was given to respondents regarding their characteristics and stethoscopes.

Results. Bacteria were found on all of the sampled stethoscopes, except two. At hospital A, gram-negative cultures were isolated in 17 samples (94%), gram-positive cultures were not isolated in any and both in one (6%). At hospital B, all 13 cultures were gram-positive (100%). At hospital C, 13 gram-positive samples (93%) and both in one (7%). *Enterobacteriaceae* species were the most prevalent contaminants at the hospital A (95%), *Staphylococcus* species at hospital B (57%) and C (44%) with five cases of coagulase-positive *Staphylococcus aureus*.

Conclusion. Of 48 samples, 46 had bacterial contamination, which demonstrates how stethoscopes may act as vectors in transmission of various infectious diseases and should be considered as a serious problem.

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Morphological features of the lungs in post-COVID-19 period

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Background. The pandemic of coronavirus disease 2019 (COVID-19) caused by SARS-CoV-2 is an urgent problem of the modern health care system. The lungs are the main target organ for this infection. The pathogenesis and morphological changes in the lungs in COVID-19 are well understood. Morpho-functional state of the lungs in people who had recovered from COVID-19 is an under-researched issue.

The aim of the study was to identify the morphological features of the lung in individuals in post-COVID-19 period.

Methods. The material of the study was autopsy material: fragments of the lung tissue from 96 deceased (59 men and 37 women). The cause of death was acute or chronic cardiac/cardiovascular failure, dislocation of the brain stem, pulmonary artery thromboembolism, intoxication. The average duration of the post-COVID-19 period was 148.6±9.5 days. Slides were stained with hematoxylin and eosin, picrofuchsin according to van Gieson.

Results. Survey microscopy of the slides in all cases revealed pneumosclerosis of varying severity, characterized by excessive growth of connective tissue fibers perivascular and directly in the vessel's walls of various calibers, around the bronchi and bronchioles or directly in their walls, in the interalveolar septa with its thickening. In 46 cases (47.9%) pneumosclerosis was combined with atelectasis and emphysematous changes. In all cases, predominantly focal immune cells infiltration was detected in the fields of pneumosclerosis. In 23 cases (24.0%), metaplastic changes were detected in the foci of sclerosis, characterized by the presence of hyaline cartilage, bone and adipose tissues. In 20 cases (20.8%), metaplastic and dysplastic changes were detected in the epithelial layer of the deformed bronchial tree. In 17 cases (17.7%), areas of dystrophic calcification were noted in the foci of pneumosclerosis. In all cases, against the background of the above-described changes in the lung tissue outside the pneumosclerosis foci, hemodynamic disturbances (edema, vascular plethora, diapedetic hemorrhages, thrombosis) were noted.

Conclusion. Sclerosis, focal immune infiltration, emphysematous and atelectatic changes, metaplastic and dysplastic changes, dystrophic calcification and hemodynamic disturbances were detected in the lungs of individuals in post-COVID-19 period. The changes identified by the authors, firstly, help to explain pulmonary manifestations of post-COVID-19 syndrome, and secondly, should be the basis for the development of rehabilitation and treatment methods for such category of patients.

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Modelling of 3D printer emissions clearance and retention in the human respiratory tract

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Background. One of the primary components of air pollution is particulate matter (PM). Depending on its diameter, PM classifies as coarse ⩽10 μm (PM_{10}), fine ⩽2.5 μm ($PM_{2.5}$), and ultrafine ⩽0.1 μm ($PM_{0.1}$). The use of 3D-printers is extremely beneficial in modern medicine and other sectors. However, it has been established that during the 3D-printing process, PM is released, which can accumulate in tissues and result in health problems.

Aim. This study aims to investigate the clearance and retained mass of various PM diameters emitted during the 3D-printing in the tracheobronchial (TB) and pulmonary (PU) regions based on mass concentration (MC) and geometric mean diameter (GMD).

Methods. Using the Multiple Path Particle Dosimetry Model (MPPD-V3.04) and exposure to different particle sizes (PM_{10}, $PM_{2.5}$, and $PM_{0.1}$), clearance and retention in the human respiratory tract were modeled.

Results. In the first scenario, a single 5-hour exposure was followed by a 30-day post-exposure period, while in the second scenario exposure was 5-hour a day, five days a week, for one month, followed by a 1-year post-exposure period. The model estimated that retained mass in the first scenario would be ($PM_{0.1}$, PM_{10} – 0.8%, $PM_{2.5}$ – 1.0%) in the TB region, and ($PM_{0.1}$, PM_{10} – 84.8%, $PM_{2.5}$ – 84.7%) in the PU region. In the second scenario, the model estimated the retained mass to be ($PM_{0.1}$, PM_{10} – 1.2%, $PM_{2.5}$ – 1.5%) in the TB region and (54.5% for all PM) in the PU region. Additionally, the clearance rate for all PM sizes in the TB region was initially high and then decreased, whereas the clearance rate in the PU region was consistently low.

Conclusion. TB region clearance is more rapid than PU region clearance. According to the 2nd model, slower clearance rates would result in the retention of 54.5% of the PM mass in the PU region even after 1-year. The simulation of these scenarios demonstrates that persons using the 3D-printer regularly potentially risk developing chronic inflammation due to higher cumulative particle retention.

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The efficacy of combination of beclomethasone dipropionate and formoterol in HFA or DPI 100/6 µg inhalers as maintenance and reliever therapy in patients with moderate persistent uncontrolled asthma

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Background. Several international guidelines recommend using of ICS/formoterol as permanent maintenance treatment or as needed reliever therapy (MART). The MART strategy using the combination of BDP/F has been studied in several clinical trials in patients with moderate asthma and significantly reduced asthma exacerbation rate.

Aim. The aim of the study was to evaluate the efficacy of BDP/F MART in a real-life study in patients with moderate persistent uncontrolled asthma previously treated with continuous maintenance therapy and short-acting β2-agonists (SABA) per need. The primary objective was to determine the impact of the treatment on asthma control levels according to the Global Initiative of Asthma (GINA) criteria at week 12. Secondary objectives were an average change of FEV1 at weeks 12 and 24, level of asthma control at week 24, change of asthma symptoms score and assessment of patient satisfaction using the SatQ questionnaire at weeks 12 and 24.

Methods. This was a 24-week, prospective, multicenter, open-label, non-interventional observational study in 6 research centres in Latvia. Patients were treated with beclomethasone/formoterol 100/6 µg HFA pMDI or DPI inhaler 1–2 inhalations twice a day and 1 inhalation as needed up to a maximum number of 8 inhalations per day.

Results. A total of 61 patients were recruited in our study; 52 patients finished the study and arrived to visit 4 at week 24. The mean age of the recruited patients was 54.9 (95% CI 51.3; 58.5) years. There were 41 (67.2%) females. The average height was 1.68 (95% CI 1.66; 1.70) m, and asthma history, 10.4 (95% CI 7.5; 13.4) years.

Study treatment provided a shift of asthma control status from an uncontrolled to a partially controlled or well-controlled state at weeks 4, 12 and 24 (statistically significant only at week 4; $p<0.05$). We observed significant improvement of FEV1 by 100 to 160 ml ($p=0.001$; $p=0.031$) and reduction of average symptom score ($p<0.0001$) vs. baseline at weeks 12 and 24. At week 12, 17% of patients were very satisfied, 44% satisfied, and 8% were neutral about the study treatment. At week 24, 28% were very satisfied, 40% satisfied, 8% neutral, and 3% were unsatisfied.

Conclusion. Study treatment was associated with substantial improvement in asthma control status and lung function. Most patients were satisfied with the study treatment and got a significant reduction in asthma symptom scores.

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Molecular sensitization profile to dogs in residents of Latvia and its connection with asthma

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Background. The percentage of households keeping dogs indoors as pets in Europe is high and ranges from 5.4 to 25%. Studies show that the prevalence of sensitisation to dog allergens is increasing, and this sensitisation is associated with increased asthma development risk.

Aim. The aim of our study was to evaluate the dog allergen molecular sensitisation profile and its connection to the presence and control of asthma and the severity of allergic rhinitis in patients living in Latvia.

Methods. In dog-sensitised patients in whom Allergy Explorer (ALEX) test was performed, clinical history data were collected about age, gender, presence or absence of sensitisation to house dust mites, pollens and other furry animals, diagnosis of asthma, and dog or other furry animals at home. Additional data were collected about asthma control status according to Global Initiative for Asthma (GINA) criteria.

The data were analysed in IBM SPSS Statistics for Windows 20.0 software (IBM Corp., USA). The odds ratio of asthma presence and asthma control in patients with positive vs. negative dog allergen component-specific IgE were calculated using chi-square or Fisher's exact test. The result was considered statistically significant if the p-value was <0.05.

The study was approved by the Ethics committee of the University of Latvia.

Results. Of all 311 ALEX-tested patients, 91 were sensitised to dogs. Eighteen patients, or 9.8%, were exclusively monosensitised to dogs. Allergic rhinitis was diagnosed in 81 or 87.9% of patients, but 53 or 58.2% had a diagnosis of asthma.

More than half (63.7%) of patients were sensitised to Can f 1 molecule, 62% to Can f 5, 36.6% to Can f 6, 21.1% to Can f 4, 16.5% to Can f 2 and 15.4% to Can f 3.

Sensitisation to Can f 1 was associated with 3.41 times higher asthma risk ($p=0.006$). Positive specific IgE to Can f 2 was associated with 5.85 times higher asthma risk ($p=0.020$, Fisher's exact test; $p=0.015$, Chi-square test). There was a 4.17 times higher risk for asthma in patients sensitised to Can f 6 ($p=0.008$). Increased positive sensitisation count to various allergen molecules was associated with increased asthma risk ($p=0.006$). No correlation was found between sensitisation profile, positive sensitisation count and asthma control status.

Conclusion. Sensitisation count and specific sensitisation to dog allergen molecules, Can f 1, Can f 2 and Can f 6 may impact asthma risk.

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ONCOLOGY

The most common sites of metastases in sarcoma patients at Pauls Stradiņš Clinical University Hospital in 2018–2022

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Background. It is vitally important to indicate the right imaging studies at the diagnostic stage to evaluate spread of the disease, as it could alter the further action plan in the patients treatment.

Aim. The aim of the study was to analyze and summarize the most frequent metastasizing sites in patients with sarcomas.

Methods. This retrospective study was conducted at the Oncology Centre of Pauls Stradiņš Clinical University Hospital. From 2018 to 2022, patients with histologically confirmed sarcoma were enrolled in this study. All the patient data was collected from multitumour board reports including date of diagnosis, histological analysis, sites of metastasis, and performed imaging studies. Data was processed using IBM SPSS Statistics 22.

Results. The retrospective study included 55 patients off which 45% (n=25) patients had metastatic disease. 20% (n=5) of analyzed patients were men and 80% (n=20) were women. The average patient age was 55.17, for the women – 55.13, and for the men – 47.80. From all 25 patients 76% (n=19) had soft tissue sarcomas and 24% (n=6) had bone sarcomas. Of which more specifically – 24% (n=6) were leiomyosarcoma, 24% (n=6) were synovial, 20% (n=5) were undifferentiated pleomorphic sarcoma, 12% (n=3) were Ewing sarcoma, 8% (n=2) angiosarcoma, 4% (n=1) osteosarcoma, 4% (n=1) chondrosarcoma and 4% (n=1) were liposarcoma. By analyzing the most common location of metastases for each histologic type, 48 metastatic sites were found, and the results showed that for leiomyosarcoma most common metastatic site were lungs (n=2) and soft tissue (n=2), for synovial sarcoma were lungs (n=3), for undifferentiated pleomorphic sarcoma were lungs (n=2), soft tissue (n=2), bones (n=2), lymph nodes (n=2), for Ewing sarcoma were lungs (n=3), for angiosarcoma were lungs (n=1), bones (n=1), pleura (n=1), liver (n=1), for osteosarcoma were lungs (n=1), for liposarcoma were peritoneum (n=1), diaphragm (n=1). Undifferentiated pleomorphic sarcoma (p=0.0303), leiomyosarcoma (p=0.0262), and synovial sarcoma (p=0.0263) had the most metastasis comparing to other histologic sarcoma types. Overall, the most common localization of all types of sarcoma metastases were lungs (p=0.09; n=13), bones (p=0.10; n=6), pleura (p=0.11; n=5) and soft tissue (p=0.11; n=5).

Conclusion. Even though lungs occur as the most frequent metastatic site, it is not statistically significant. And analyzing each type of sarcoma results shows that undifferentiated pleomorphic sarcoma, leiomyosarcoma, and synovial sarcoma has the biggest potential to metastasize. But the results would be more precise, if each histologic type had the same number of patients, and total number of patients was larger.

Compliance of patients with head and neck cancer to specific anticancer therapy

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Background. Head and neck cancers (HNCs) represent the sixth most widespread malignancy worldwide. This type of cancer is generally associated with smoking, alcohol consumption, low socioeconomic status, age of the patient or infection with human papillomavirus. New treatments, gene mutations and their effect on the course of the disease are being studied more and more. The therapy includes surgical treatment, radiation therapy and also chemotherapy, which requires considerable patient support to achieve the desired result.

Aim. The aim of the study was to determine the compliance of patients with head and neck cancer to specific anticancer therapy and to determine how patient's age, gender, location of cancer and stage of disease affects their compliance.

Methods. This retrospective study included 115 patients with head and neck cancer who received specific anticancer or symptomatic therapy between 2020 and November 2022. Data was collected from ambulatory medical record cards of the Pauls Stradiņš Clinical University Hospital (PSCUH). Data processing was performed in BlueSky Statistics v10.2.0 by using Pearson chi-squared test. The p value of <0.05 was considered as statistically significant.

Results. Among 115 patients 37.4% (n=43) were women and 62.6% (n=72) were men. The mean age of patients was 64.81±12.63 years. 7.8% (n=9) of patients were diagnosed with stage I, 19.1% (n=22) – with stage II, 19.1% (n=22) – with stage III, 53.9% (n=62) – with stage IV. The most frequent locations of tumors were base of tongue (n=14, 12.2%), floor of mouth (n=13, 11.3%) and other and unspecified parts of mouth (n=13, 11.3%). A total of 51 patients (48.7%) received symptomatic treatment. Compliance was determined in patients who received specific anticancer treatment (surgery, radiation therapy or chemotherapy) (n=64, 55.7%). From this group 45.3% patients (6 women and 23 men) were treatment compliant, but 54.7% (11 women and 24 men) were not compliant to therapy. The associations between compliance, patient's age, gender and stage of disease was not significant ($p>0.05$). However, there was an association between compliance and tumor location ($p=0.040$).

Conclusion. The results showed that HNCs were more frequent in men with a mean age of 62.87 years. The IV stage accured more often and the most common HNC location was tongue. Most of the patients were not compliant to therapy. The association between patients' compliance and tumor location was found. But there was no association between compliance, age, gender, and stage of disease.

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Prevalence of mutations in homologous recombination repair genes and TP53 in patients with ovarian cancer in Latvian population

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Background. Ovarian cancer (OC) is the most common cause of death due to gynecological cancer. Homologous recombination deficiency (HRD) in one or more homologous recombination repair (HRR) genes increases the risk of OC. The most well-known cause of HRD is BRCA1/2 mutations. In high grade serous OC BRCA1/2 somatic mutations occur in 18%–30%, but mutations in TP53 are more frequent (96%). TP53m means that mutated cells uncontrollably proliferate, escape apoptosis and is associated with poor prognosis. An average risk of developing OC with a germline BRCA1m is 39% and with BRCA2m, 11%.

Aim. To evaluate a prevalence of HRRm in patients with OC in Latvian population and find the association between age and onset of OC and HRR gene mutations.

Methods. 138 OC patients treated at Riga East Clinical University Hospital, Oncology Centre of Latvia were included in a retrospective research study. Archived tumor tissue material was analysed by NGS to detect somatic mutations in HRR genes. Data was collected from ambulatory medical records of the Riga East University Hospital, Oncology Centre of Latvia. Data processing was performed in BlueSky Statistics v10.2.0 by using Pearson chi-squared test.

Results. A total of 138 patients with OC were included in this study. The mean age of patients was 58.22±10.52 years. 5.8% (n=8) of patients had stage I OC; 8.7% (n=12) of patients, II; 55.1% (n=76) of patients, III; 30.4% (n=42) of patients, IV. BRCA1 mutation was found in 52 patients (37.4%) and BRCA2m, in 26 patients (18.8%). Both BRCA1/2m were found in 9 patients (6.5%). 86 patients (62.3%) had mutations in TP53 gene. Other mutations in HRR genes were found in 119 patients (86.2%). A total of 38 women (27.5%) had mutations in BRCA1 and TP53, and 18 patients (13.0%) had mutations in BRCA2 and TP53 simultaneously. Ten patients (7.25%) had personal history of breast cancer before diagnosis of OC; 9 of them had BRCA1/2m ($p<0.01$). A statistically significant association between age of diagnosis and somatic BRCA1/2, TP53 mutations was not demonstrated.

Conclusions. In most cases ovarian cancer is diagnosed in advanced stages. The study shows that the prevalence of somatic HRRm is high in Latvian population. We suggest detecting germline mutations in women with personal/family history of ovarian/breast cancer to estimate the risk of cancer development. The association between age of diagnosis and HRRm was not significant.

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Percentage frequency of EGFR in non-small cell lung cancer patients treated in Pauls Stradiņš Clinical University Hospital in 2022

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Background. Lung cancer is the second most commonly diagnosed cancer, of which non-small cell lung cancer is the most common type. Epidermal growth factor receptor (EGFR) is a trans-membrane glycoprotein that regulates signaling pathways to control cellular proliferation. EGFR is found in epithelial, mesenchymal, and neurogenic tissue. Overexpression of EGFR can contribute to the pathogenesis of a variety of cancers, including non-small cell lung cancer. Worldwide EGFR mutation has been reported to occur at a rate of 20%.

Aim. The aim of the study was to find out how many patients with non-small cell lung cancer have a positive EGFR mutation.

Methods. The aim of the retrospective study was to evaluate the frequency of EGFR mutation in non-small cell lung cancer. Totally 111 patients were enrolled in the study. The data was taken from Pauls Stradiņš Clinical University Hospital oncological council reports and included data from 2022.

Results. The retrospective study included 111 patients with identified non-small cell lung cancer, of which 53% (n=59) were induced palliative treatment and 41.4% (n=46) of theme were tested for mutations. More than half (55.5%, n=5) of analyzed patients with mutations were men and 44.4% (n=4) were women. Average age of the patients was 66 (71 for women and 65 for men). Of all tested patients, 19.56% (n=9) patients had EGFR mutation. The most common non-small cell lung cancer in patients with mutations was adenocarcinoma 55.5% (n=5) and squamous cell carcinoma 44.4% (n=4) of which 33.3% stage 3 and 66.6% stage 4.

Conclusion. EGFR mutation in non-small cell lung cancer is more often in men with adenocarcinoma being the most common type. The obtained result corresponds to reported worldwide frequency.

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The value of assessment of BRAF and NRAS gene mutation status and its association with clinical and histopathological characteristics of in cutaneous invasive melanoma

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Background. Melanoma is the most aggressive type of skin cancer, which incidence is increasing during last years. Oncogenic BRAF and NRAS hotspot mutations are the most common genetic alterations in cutaneous melanoma. BRAF mutation is found in about 50% of skin melanomas, while NRAS accounts for about 20% of all mutations. Both mutations are associated with a more aggressive disease course with an increased risk of visceral and brain involvement. However, the association of BRAF and NRAS mutation with clinical and histopathological characteristics is still controversial.

Aim. The aim of our pilot study was to compare the associations of BRAF and NRAS mutation status with clinical and histopathological characteristics of melanoma.

Methods. 31 patients with primary invasive cutaneous melanoma who underwent surgical treatment in Riga East Clinical University Hospital from 2011 till 2018 were enrolled in the study. The study protocol was approved by the Central Medical Ethics Committee of Latvia (approval No. 01-29.1/2016-1-1; January 2016). The clinical and histopathological characteristics were assessed. Histopathological characteristics were evaluated based on the currently available World Health Organization (WHO) and The American Joint Committee on Cancer (AJCC) 8th guidelines. BRAF and NRAS mutations was analyzed by digital PCR method. Statistical analysis was performed by *GraphPad Prism* software.

Results. A total of 31 patients with melanoma were enrolled in the study. Five patients had stage IA, 4 patients had stage IB, 3 patients had stage IIA, 9 patients had stage IIB, and 10 patients had stage IIC. The median Breslow thickness was 4.5 mm (range 0.3–14 mm). BRAF mutation was observed in 16 patients (51.6%), NRAS mutation was observed in 7 patients (22.5%), and 8 patients (25.8%) had wild type melanoma genotype. The BRAF mutation in melanoma correlated with disease TNM stage (Rho=0.1008, p=0.02). The NRAS mutation did not correlate with disease TNM stage.

Conclusion. BRAF mutation status correlated with clinical diseases stage and should be used routinely in assessment of malignant melanoma.

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Correlation between haematological toxicity and pelvic bone marrow dosimetric parameters in cervical cancer patients during concurrent chemoradiotherapy

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Background. Cervical cancer is the second most common malignancy in women under 45 years in Latvia. Golden standard treatment for locally advanced stages is combined chemoradiotherapy. To achieve local and systemic control the treatment should be finished in 8 weeks, which is limited by haematological toxicity.

Aim. The aim of this study was to find a correlation between haematologic toxicity and dosimetric parameters of pelvic bone marrow to implement bone marrow sparing radiotherapy and improve chemotherapy feasibility.

Methods. This retrospective study included all cervical cancer patients (FIGO IB1-IVB) receiving combined chemoradiotherapy at the Oncology Centre of Latvia in 2020. Bone marrow was contoured on simulation CT scans and dosimetric parameters read from dose-volume histograms (DVH). Additional data (weekly complete blood count and biochemistry) was used to evaluate haematological toxicity according to CTCAEv5 grading (Common Terminology Criteria for Adverse Events). Spearman correlation and multiple linear regression was used with IBM SPSS Statistics 25.0 and Microsoft Excel Software. Results were statistically significant if $p<0.05$.

Results. The study included 63 patients with a median age of 50.9 years (range 28.0–74.0 years) with histologically verified cervical cancer and who underwent external beam radiotherapy (50–50.4 Gy in 25–28 fractions) with weekly concurrent Cisplatin (40 mg/m^2) chemotherapy and sequential brachytherapy (7 Gy to Point A in 4 fractions).

Only 6.3% of patients in study cohort received recommended 5 cycles of systemic treatment. Main limiting toxicity was leukopenia (Grade 3+), seen in 19.1% of cases, Grade 2 in 36.5%. Less common was neutropenia (Grade 3+), seen in 15.9% of cases, Grade 2 in 14.3%.

Strong correlation was shown between lymphopenia and D_{mean}, V_{30} and V_{40} ($r_s=0.36$, $r_s=0.42$, $r_s=0.47$; $p<0.05$), moderate correlation for thrombocytopenia and V_{10} ($r_s=0.32$, $p<0.05$).

Multiple linear regression analysis showed strong effect between lymphopenia and D_{mean}, V_5, V_{10}, V_{20} ($F(4,49)=12.19$, $p<.001$, $R^2=0.5$, $R^2_{adj}=0.46$), moderate effect for thrombocytopenia and all dosimetric parameters ($F(5,48)=5.17$, $p<0.001$, $R^2=0.35$, $R^2_{adj}=0.28$). Weak effect was shown for neutropenia and D_{mean}, V_{10} ($F(2;51)= 3.54$, $p<0.05$, $R^2=0.12$, $R^2_{adj}=0.09$).

Conclusion. Results concluded that dosimetric parameters correlate with haematological toxicity. Lymphopenia is affected by D_{mean}, V_5, V_{10}, V_{20}, and thrombocytopenia, by all dosimetric parameters and there is a weak correlation between neutropenia and D_{mean}, V_{10}. In order to implement bone marrow sparing radiotherapy more studies are necessary with larger amount of data to evaluate feasibility.

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Reduction rate of FOLFIRINOX chemotherapy regimen in pancreatic cancer patients

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Background. FOLFIRINOX (folinic acid, 5-fluorouracil, irinotecan, oxaliplatin) is a combination of chemotherapy drugs used in the treatment of pancreatic cancer patients, including patients with metastatic pancreatic cancer. Research shows that the FOLFIRINOX chemotherapy regimen improves the overall survival of pancreatic cancer patients, however, the combination of these drugs increases the toxic effects of chemotherapy on the human body.

Aim. The aim of the current study was to obtain information on the rate of reduction of the FOLFIRINOX regimen in pancreatic cancer patients in order to consider the need for reduction and to understand the timing of chemotherapy reduction.

Methods. The study was conducted retrospectively, analysing patient data from years 2019 to 2021. Altogether 257 patients were enrolled in the study. Data used for this study was collected from Pauls Stradiņš Clinical University Hospital Council of oncologists, chemotherapy course prescription sheets and "Doctors office" platform. Data was processed using MS Excel.

Results. From years 2019 to 2021, the total of patients diagnosed with pancreatic cancer at Pauls Stradiņš Clinical University Hospital were 257. From those women 54.09% (n=139), men 45.91% (n=118), mean age at the time of diagnosis was 71 years. Stage I pancreatic cancer was diagnosed in 1.17% (n=3), stage IIA 7.78% (n=20), stage IIB 6.23% (n=16), stage IIIA 5.06% (n=13), stage IIIB 19.84% (n=51), stage IV 29.96% (n=77), stage unknown 29.96% (n=77). Surgical treatment was indicated in 14.4% (n=37), symptomatic therapy and palliative care was indicated in 31.13% (n=80), other – 30.35% (n=78). Chemotherapy was indicated in 24.12% (n=62), from those with FOLFIRINOX chemotherapy regimen 29.03% (n=18). Of those assigned to FOLFIRINOX chemotherapy regimen, the full course of chemotherapy received 33.33% (n=6), dose reduction during the first 6 cycles of FOLFIRINOX regimen – 55.6 % (n=10), dose reduction during the last 6 cycles of FOLFIRINOX regimen – 11.11% (n=2).

Conclusion. Study shows that it is most likely that pancreatic cancer patients receiving FOLFIRINOX chemotherapy regimen will need a chemotherapy dose reduction in some time of the course. The most frequent dose reduction timing in FOLFIRNOX chemotherapy regimen is seen during the first 6 out of 12 cycles of the chemotherapy course.

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Frequency of IDH in patients with glial tumors

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Background. Isocitrate dehydrogenase (IDH) is enzyme involved in many cellular processes, which is used as valuable biomarker for patients with glial tumors to diagnose disease, predict prognosis and find more targeted treatment for the patient. Especially important is to know IDH status in WHO Grade 2 and 3 tumors because IDH+ improves the prognosis.

Aim. The aim of the current study was to discover how many IDH positive and negative glial tumors in Pauls Stradiņš Clinical University Hospital had in 2021 and 2022.

Methods. The study was conducted retrospectively, analysing patient data from year 2021 and 2022. We used data from Pauls Stradiņš Clinical University Hospital Council of oncologists, platform "Doctor's office" and Vilnius University Hospitals National Centre of Pathology. Data were processed with MS Excel.

Results. Summarizing the results, in 2021 and 2022, the total of 195 patients had glial tumors in Pauls Stradiņš Clinical University Hospital. 6.67% (n=13) of them were WHO Grade 2 and 3 oligodendrogliomas. In Grade 2 there were 69.23% (n=9) oligodendrogliomas with IDH+ status in 77.78% (n=7) and IDH- status in 11.11% (n=1) of them. IDH status unknown was in 11.11% (n=1) of them. Grade 3 oligodendrogliomas were 30.77% (n=4) with IDH+ status in 75% (n=3) and IDH- status in 0% (n=0) of them. IDH status unknown was in 25% (n=1) of them. 10.26% (n=20) of all tumors was WHO Grade 3 anaplastic astrocytomas. In these tumors IDH+ status was in 20% (n=4), IDH– status in 25% (n=5) but unknown IDH status in 55% (n=11) of them. 12.82% (n=25) of all tumors was WHO Grade 2 diffuse astrocytomas. In these tumors IDH+ status was in 40% (n=10) but IDH– status was in 28% (n=7) of them. 32% (n=8) of them status is unknown. 2.05% (n=4) of all tumors was WHO Grade 2 and 3 oligoastrocytomas. In Grade 2 IDH+ status was in 33.33% (n=1) and IDH– status was in 66.66% (n=2) of them. In Grade 3 IDH+ status was in 100% (n=1) of them. 53.85% (n=105) of all tumors was glioblastomas with IDH- status in 100% (n=105) of them. 14.35% (n=28) was other glial tumors with IDH status unknown.

Conclusion. Most often, IDH+ tumors are Grade 2 and 3 oligodendrogliomas, diffuse astrocytomas. However, considering that the number of patients is small, additional studies are needed to know more precisely the frequency of IDH in different gliomas.

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The persistence of cervical intraepithelial neoplasia is closely associated with HPV positivity, while CIN1 regression characteristics for HPV negative patients

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Background. Persistent infection with human papillomavirus (HPV) is a known etiological factor in the development of cervical intraepithelial neoplasia (CIN) and cervical cancer. However, the precise role of interplay between HPV and host factors in progression or regression of CIN in different individuals is still under investigation.

Aim. The aim of our pilot study was to assess the role of HPV, tobacco smoking in the regression and progression of CIN.

Methods. 28 patients aged 19–50 years referred to the Department of Oncogynecology, Latvian Oncology Center at Riga East Clinical University Hospital were enrolled in the study. The study was approved by a local ethical committee. The patients underwent the colposcopy. The cytological examination and uterine cervix biopsy with subsequent histopathological examination was performed. All patients underwent HPV testing from cervical smears. The *Aptima* HPV assay is an *in vitro* nucleic acid amplification test for the qualitative detection of E6/E7 viral messenger RNA (mRNA) from 14 high-risk types of human papillomavirus (HPV) in cervical specimens. The high-risk HPV types detected by the assay include: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68. Each patient after 6-12 months underwent follow-up with cytological, histopathological and HPV testing.

Results. A total of 28 patients were enrolled in the study. The median patient age was 31 (19–50) years. The mean smoking history was 1.15 (0–11.25) pack-years. In the initial (first) examination CIN 1 was detected in 27 patients, 13 patients were HPV positive, and 14 patients were HPV negative. However, during second follow-up histopathological examination CIN 1 was detected in 11 HPV positive patients and 4 HPV negative patients. The CIN1 regression significantly associated with HPV status (p=0.0034). However, the association between CIN 1 regression and smoking pack-year history and patient age has not been observed. In addition, the significant association between the histopathological and cytological testing has not been demonstrated.

Conclusion. CIN 1 persistence is closely associated to HPV positivity. However, CIN 1 regression predominantly observed in HPV negative patients. The study results stressed the value of strategy to combat HPV for the treatment of CIN and prevention of cervical cancer.

Breast tenderness during mammography examination in women after breast-conserving surgery

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Background. Annually approximately 1,100 to 1,200 women get breast cancer in Latvia – it is the most frequent oncological disease and the most frequent cause of death caused by oncological diseases in women. Identifying causes of discomfort during mammography is essential to prevent them and thus increase the number of women attending both breast cancer screening and diagnostic mammography.

Aim. Prospectively evaluate the differences in discomfort/pain caused by mammography examination in post-surgery breast cancer survivors and women with no history of breast surgery or cancer.

Methods. The study was conducted at Riga East University hospital, Oncology Centre of Latvia. Patients with discomfort in the breast caused by mammography were assessed via interview questionnaire immediately after mammography examination (n=83). The study included breast cancer survivors (n=45) and women without histories of breast surgery or cancer (n=38). A total of 166 breasts were examined: 33 operated and 43 non-operated breasts were rated separately and compared with control group (n=76) with no history of breast cancer. Breasts that have undergone implant surgery (n=2) or mastectomy (n=12) were excluded. Patients had to evaluate discomfort experience in mammography in a 10-point scale for each breast separately. Interview included also physical parameters, menopausal status and the received therapy in the treatment of breast cancer. Data analysis was performed in IBM SPSS Statistics 20 program.

Results. Of 166 breasts, 152 were evaluated and 14 were excluded (mastectomy (n=12), implant surgery (n=2)). The mean age of all female patients was 63.3 (SD, 10.9) years. The Mann-Whitney U test revealed that the level of discomfort during mammography were significantly lower in patients with no cancer history (Md=2.00, n=78) compared to patients that had breast-conserving surgery (Md=3.00, n=33), U=1660.5, z=−2.45, p=0.013). The study found no correlation between discomfort during a mammogram and how many years ago breast-conserving surgery was performed (p=0.45) and weak, negative and statistically significant correlation between body mass index and discomfort was found (rs=−0.28; p<0.001). Data will be collected by evaluating mammogram positioning criteria in CC and MLO views and technical parameters - compression force measured in Newtons – will be obtained.

Conclusion. In women after breast-conserving surgery, discomfort during the mammography examination is more severe compared to those women who did not have combined therapy, regardless of the time after surgery. Greater BMI slightly decreases the discomfort during scanning.

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Relationships between breast cancer histological type and magnetic resonance Kaiser score

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Background. Breast cancer is the most common cancer of women in Latvia. MRI is an important tool in breast cancer diagnostics.

Aim. The aim of the study is to correlate magnetic resonance imaging (MR) findings with pathohistological results.

Methods. Retrospective study enrolled women who had histologically confirmed breast cancer in one breast and who had undergone breast MRI. Kaiser score includes five main MRI diagnostic criteria (lesion type, shape and margins, root sign, enhancement kinetics, and edema). The histological reports of breast cancer biopsy were analyzed for tumor type, histological grade, estrogen receptors, progesterone receptors, HER2, Ki-67 and E-cadherin.

Results. A total of 76 women (28–75 years old) were included in the study. Kaiser scores ranged between 6 and 11 which corresponds to BI-RADS 4 and 5.

There was statistically significant correlation between the enhancement curve and two histological indicators: Ki67 (p=0.05) and E-cadherin (p=0.034). E-cadherin was found positive in 59.6% of cases where tumor mass had wash-out enhancement curve. Out of 76 breast lesions, 85% were invasive breast carcinoma of no special type and 15% were lobular carcinoma. The most common MRI findings of invasive breast carcinomas of no special type were mass lesions (69.2%). Lobular carcinomas more often were no-mass lesions. There are signs of significant correlation between enhancement curve and histological grade (p=0.053). No-mass enhancements were observed in HER2 positive cancers more often than mass type enhancement (p=0.029). Tumors with higher histological grade more commonly had spicular margins (p=0.029). There was correlation between Kaiser score and Ki-67 results (p=0.014).

Conclusion. There are some breast MRI signs that can predict a higher histological grade. The signs are tumor spicular margins and edema. Patients with high Ki-67 also had a high Kaiser score. Mass lesions usually are invasive breast carcinomas of no special type rather than lobular carcinomas. A greater number of patients are needed to find out if there is a correlation between enhancement curve and histological grade and type, and association between histological grade and edema.

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MENTAL HEALTH

The prevalence of burnout syndrome among emergency medical service professionals after the COVID-19 pandemic

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Background. Burnout syndrome is a common disorder among medical workers, and its prevalence among emergency medical personnel seems to be relatively high, especially after the COVID-19 pandemic.

Aim. The study aims to obtain information on the prevalence of burnout syndrome among Emergency Medical Service staff from all major cities of Latvia after the COVID-19 pandemic.

Methods. Permission from the University of Latvia Ethics Committee was obtained for this cross-sectional study. An anonymous questionnaire – Maslach Burnout Inventory – was used to obtain research data online. Data were processed using SPSS v27.0, descriptive statistics.

Results. In total, 260 participants were enrolled in this study, of which 72.7% (N=189) were females and 27.3% (N=71) were males. The minimum age of the respondents was 22 years, the maximum was 66 years. Among all participants, 71.5% were doctor assistants, 7.3% were physicians, 5.4% were emergency physicians, 13.1% were medical assistants, 1.5% were trainee doctors, and 1.2% were dispatchers. Results show that 74.6% (N=190) of respondents reported a significant (above average or high) risk of burnout ($p<0.001$). To be more specific, 30% (N=74) of participants reported a high risk of burnout, 44.6% (N=116) reported above-average risk of burnout, and 25.4% (N=66) had a low risk ($p<0.001$). The highest risk of burnout was most common among trainee doctors (75%), then among dispatchers (66.7%), and physicians (63.3%) ($p=0.012$). More often, a significant risk of burnout was found in workers aged 31 to 40 years (51.2%) and 20 to 30 years (46.8%) ($p=0.023$). Based on total years of experience, the most burnout-prone group were workers with 8–10 years of experience (66.7%), then the employees with 5–8 years of experience (55.7%) ($p=0.012$). There was no statistically significant association between burnout and workload ($p=0.573$). The variables sex and burnout were also not related to each other ($p=0.711$).

Conclusion. The prevalence of burnout syndrome among Emergency Medical Service workers post COVID-19 pandemic is alarmingly high (74.6%). As the research has demonstrated, EMS workers are an extremely vulnerable group and need more social support such as preventive and intervention approaches to emotional stress and burnout.

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The role of subjective therapeutic mastery and perceived professional confidence in burnout prevalence among Latvian mental health professionals

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Background. The prevalence of burnout syndrome, characterized by exhaustion, depersonalization, and reduced performance, is increasing among mental health professionals amid the ongoing global pandemic and regional conflict. The relationship between subjective therapeutic mastery, professional confidence and burnout is understudied, particularly among Latvian specialists.

Aim. The aim of the current study was to illuminate burnout rates in mental health professionals and explore correlation between burnout dimensions and domains of perceived professional identity and therapeutic mastery.

Methods. A total of 131 Latvian mental health professionals (doctors and resident doctors in psychotherapy and psychiatry, and psychotherapy specialists) were enrolled in the cross-sectional study conducted through 6 professional associations, hospitals and online. Participants were surveyed with 3 scales: Maslach Burnout Inventory-General Survey consisting of Exhaustion, Professional Efficacy and Cynicism subscales (MBI-GS, Maslach, Jackson & Leiter, 1996); Subjective Therapeutic Mastery and Professional Identity Self-Assessment Questionnaire (developed by authors and validated in a pilot study (N=20) with Cronbach's alpha >0.7 in all relevant subscales); and sociodemographic survey. Statistical analysis was conducted using SPSS Statistics 22, with a significance level of $p<0.05$.

Results. Participants were 25–81 years old (M=44.6 years; SD=13.9); 80.9% were female (N=106). Psychiatrists accounted for 37.4%; psychotherapists, 32.1%; psychotherapy specialists, 23.7%, and psychiatrists/psychotherapists, 6.9%.

Almost half (45.8%) of professionals worked one job (N=60), and 25.1% worked more than full-time. Half (50.4%) had 11 or more years of experience. Psychotherapy specialists worked in 1.55 jobs with 24.4 patients a week on average – compared to 1.81 jobs in doctors: psychiatrists treated the most patients per week (34.4) compared to psychotherapists (24.3) and psychotherapy resident doctors (14.1).

In all groups Spearman correlation analysis revealed higher cynicism and exhaustion that comes with extended experience, higher weekly patient hours and working in person compared to online work ($r_s=0.496–0.597$; $p<0.05$). Higher exhaustion rates were seen in participants with greater therapeutic mastery and confidence ($r_s=0.707–0.743$; $p<0.001$).

Mann-Whitney U test revealed that median cynicism and exhaustion rates were 40%–60% lower in psychotherapists and psychotherapy specialists compared to psychiatrists ($p<0.001$). Psychotherapy specialists had 7% higher cynicism than psychotherapists.

Conclusions. Higher subjective therapeutic mastery and confidence may be associated with increased exhaustion in this population sample. Further research should investigate factors that may contribute to burnout, including work satisfaction. The results also indicate higher rates of exhaustion and cynicism among psychiatrists compared to psychotherapists and psychotherapy specialists. Unlike working in person, no statistically significant association was found between burnout and working online.

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A performance analysis of prediction of depression, anxiety and stress using supervised machine learning methods

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Background. Depression, anxiety, and stress are major public disorders nowadays. To prevent cardiovascular diseases (CVD) in an appropriate and cost-effective manner, the total-risk approach is recommended. The prevalence of depression and anxiety among people of several cardiovascular diseases is higher than in general population. The gold standard scales for diagnosing depression and anxiety are rarely used in busy clinical settings. Machine learning (ML) algorithms could be applied for developing better clinical diagnostic tools.

Aim. To evaluate machine learning algorithms for prediction of depression, anxiety, and stress.

Methods. Cross-sectional study included patients with cardiovascular diseases hospitalized in Riga, Latvia. Patients' depression was assessed by PHQ-9 questionnaire, anxiety was determined by HADS and stress was assessed by the PSS-4 instrument. In addition, patients completed 12 self-assessment questions (sleep, pain, fatigue, happiness, etc.). ROC curve analysis (area under curve, AUC) was used to determine the single best self-report question that characterized depression, stress, and anxiety. A model was created that included these 3 self-assessment questions, and using 10 most widely used binary supervised ML algorithms: logistic regression (LR), decision tree (DT), random forest (RF), neural network (NN), naive bayes (NB), support vector machine (SVM), linear discriminant analysis (LDA), K-fold cross validation (CV), gradient boosting (GB) and XGBoost, the top 3 models were evaluated to predict depression, anxiety, and stress. Altogether 865 patients were enrolled in the study. In order to evaluate ML models and prevent overfitting the patients were divided into 2 subsets: train and test with a ratio of 70:30. Statistical analysis was performed using R v.4.2.2 (R Core Team (2022). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria).

Results. The median age of patients was 66 years (IQR 59–73 years). The most accurate self-assessment question for predicting depression was depression (AUC=0.82); for anxiety, anxiety (AUC=0.83); and for stress, life satisfaction (AUC=0.62). After creating a ML model for prediction of depression from 3 self-esteem questions, the AUC for the NN model increased to 0.93, for anxiety NN, RF and LDA to 0.87; for stress, NB and GN increased to 0.73.

Conclusion. Rapid detection of depression, stress and anxiety is very important for further evaluation of treatment of cardiac patients and the use of ML algorithms creates significant increase in prediction.

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Antidepressant therapy in first-time patients hospitalized in a child and adolescent psychiatry ward with a depressive episode

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Background. Paediatric depression typically presents in primary care and is undertreated. This may lead to acute hospitalization. A selective serotonin reuptake inhibitor (SSRI) is the first-line pharmacological treatment. Fluoxetine has been more widely studied in children and adolescents. Literature data suggest that paediatric depression doubled during the first year of the COVID-19 pandemic.

Aim. The aim of this study was to compare hospitalized patients' treatment with antidepressants before their hospitalization and during hospitalization from 2016 to 2021 and evaluate the association between the COVID-19 pandemic and changes in hospitalization rates of these patients.

Methods. Altogether 183 patients' data were analysed, divided into two time periods: pre- and intra-pandemic (2016–2019; 2020–2021). The study included patients with a depressive episode (F32) who were admitted to a child and adolescent inpatient ward for the first time in their lives and spent there more than one day. Data were obtained from the Children's Clinical University Hospital's information systems. All data were analysed using IBM SPSS v26.0 and Excel 2020.

Results. From all patients 11% (n=21) were boys and 89% (n=162) were girls. The mean age was 14.5 years (SD=1.7). The mean length of stay was 17 days (SD=10). Half (51%, n=94) of patients were treated before hospitalization; 63% (n=59) of them received only one antidepressant (AD): 37% (n=22) received sertraline; 15% (n=15), fluoxetine; and 24% (n=14), fluvoxamine. Non-specific side effects occurred in 9% (n=8) of patients. During hospitalization 84% (n=154) of patients were treated with ADs. 79% (n=121) of them received only one AD. Fluoxetine was the first choice in 40% (n=62) of patients; sertraline, 25% (n=38); and fluvoxamine, 23% (n=35). Less than one-third (28%, n=26) of patients had their AD switched due to side effects or lack of effectiveness. Hyperprolactinemia occurred in 7% (n=7) of patients, 10% (n=9) of patients experienced non-specific side effects. In the pre-pandemic period, 20 patients per year were hospitalized with a depressive episode for the first time in their lives; in the intra-pandemic period, 50 patients per year.

Conclusion. Half of the patients were treated with antidepressants before hospitalization. Before and during hospitalization patients mostly received sertraline and fluoxetine, respectively. The most common specific side effect experienced by hospitalized patients was hyperprolactinemia. During the intra-pandemic period the number of patients with a depressive episode who were hospitalized for the first time increased 2.5 times.

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Legal framework for the protection of EU cross-border patients with limited decision-making capacity in Latvian hospitals

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Background. Patients who are citizens or residents of other EU states, hereinafter cross-border patients, have a right to access health care in Latvia; these rights are based on EU laws. Latvia is obliged to provide safe and high-quality health care. Safe and quality care requires proper protection of patients' rights. According to data published by the NHS of Latvia, the number of EU cross-border patients in Latvia grew from 12664 in 2018 to 16571 in 2021 (NHS). Patients with mental health difficulties and limited decision-making capacity are especially vulnerable in general, and cross-border patients. To safeguard the protection of patients in this group, it is necessary to apply Latvian law, the law of the patient's country of origin, both in line with EU law. It is not an easy task.

Aim. This study aimed to study challenges in protecting cross-border mental health patients with limited capacity by applying national and EU healthcare laws.

Methods. To complete the proposed aim, the doctrinal legal research method was applied. International human rights, EU and national laws regulating the rights of cross-border patients in Latvia were reviewed.

Results. There are no legal provisions addressing patients' rights if a cross-border patient with limited capacity when treated in Latvia. In Latvia, there are no provisions on how decision-making capacity in health care should be evaluated and addressed in case of limited abilities. There are also no provisions on how decisions of limited capacity taken by authorities in other EU member states should be applied. There are no provisions for how a support person, guardian or trusty appointed by a relevant authority of another EU member state could act in Latvia.

Conclusion. In general, the existing legal framework for protecting patients' rights with mental health difficulties and limited capacity needs to be revised. No provisions secure the rights of cross-border patients with limited capacity that can be respected and fulfilled sufficiently. In case there is a representative of a person, a support person or a guardian appointed in an EU member state, no procedural rules provide efficient and proper recognition of their representation and support rights in Latvia. There is a need to amend legal regulations to eliminate these insufficiencies.

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Towards implementation of article 12 (2) of the CRPD in Estonian, Lithuanian and Latvian patients' rights laws

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Background. The United Nations Convention on the Rights of People with Disabilities (CRPD), Article 12 part 2, requires that persons with disabilities enjoy legal capacity on an equal basis with others in all aspects of life. National patient rights laws must implement that principle concerning all healthcare matters.

Aim. The overall objective of this study was to review the conformity of Latvian, Estonian and Lithuanian patients' rights laws with the principle of legal capacity stated in Article 12 (2) of the CRPD.

Methods. The doctrinal legal research method to review Estonian, Lithuanian and Latvian statutes, regulations and case law regulating legal capacity and decision-making capacity in health care was used. A comparative research approach was applied concerning Latvian, Lithuanian and Estonian patients' rights laws on the matter.

Results. Estonian regulations of capacity do not differ in general and healthcare-related regulations. The legal capacity of a person can be limited, and substitute decision-making in health care should be applied in such cases.

In Lithuania, regulations of capacity are similar in general and healthcare-related matters. Lithuania has taken the first steps towards article 12 of CRPD implementation in family law matters but not health care.

Latvian Civil Law permits the limitation of a person's legal capacity only concerning material rights. It does not allow a limitation of legal capacity in personal rights, including patients' rights. However, there are provisions of the Patients' rights law (Art. 7 and 11) stating that there can be situations when a patient lacks legal capacity, and a guardian has a right to take a substitute decision.

Healthcare providers do not address the issue of limited capacity and guardianship sufficiently or not at all.

Conclusions: There is no conformity of Estonian, Latvian and Lithuanian patient rights laws with the principle of legal capacity stated in Article 12 (2) of the CRPD. Legislators should address this non-compliance by amending the regulations to safeguard the rights and protection of people with disabilities in health care.

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Can the workplace negatively impact mental health: Exploring differences in mental health and organization settings among hospitals in Latvia

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Background. Work of medical professionals is demanding cognitively, physically, and emotionally and this can have a negative effect on mental health. Support for medical staff is important to maintain a positive mental health condition. The aim is to evaluate if the work environment in certain hospitals contributes to more burnout and if risk factors in hospitals with a detrimental work setting can be mediated by social support and control as indicated by job demands-support-control theoretical model.

Aim. Three aims were developed to determine the variability of mental health between hospitals and the support structure for medical staff challenged in stressful situations.

H1: Is there variability in mental health of employees at different hospitals?

H2: Do psychosocial factors mediate the work in certain hospitals and burnout symptoms? H3: Do support relationships moderate the influence of risk factors on burnout symptoms?

Methods. Employees (medical staff, administration, support staff) from 30 hospitals in Latvia were invited to complete three online questionnaires: the Copenhagen Psychosocial Questionnaire (COPSOQ III), patient health questionnaire (PHQ-2), Generalized anxiety disorder scale (GAD-7) and Burnout assessment tool (BAT) and demographic questions. Two invitations to hospital staff gave a response rate of approximately 40%. A total of 4756 participants were included in the analyses (87% women, 13% men, mean age of 45). Data was analysed in SPSS by performing mediator and moderator analyses.

Results. There is small but significant variation among hospitals in terms of the mental health of their employees, including burnout levels ($F=14.23$, $p<0.001$). A full, complimentary mediator effect was found for excessive workload ($b=0.59$, $t=10.10$, $p<0.001$), negative impact on home life ($b=0.13$, $t=22.56$, $p<0.001$), attending work ill ($b=0.12$, $t=15.72$, $p<0.001$), emotional strain ($b=0.10$, $t=14.19$, $p<0.001$). This relationship was not moderated by the social support ($b=0.002$, $t=1.80$, $p=0.07$) or control ($b=0.00$, $t=0.25$, $p=0.80$). Age was a covariate ($b=-0.001$, $t=-3.13$, $p<0.005$).

Conclusion. These results show that employees in some hospitals in Latvia have higher prevalence of burnout due to excessive workload. Social support alone cannot alleviate this effect on mental health.

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Relationships between the severity of anxiety and stress-coping strategies among preclinical students at the Rīga Stradiņš University Faculty of Medicine

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Background. Stress-coping strategies are certain actions or thought processes to manage stress. These strategies are divided into adaptive and maladaptive based on whether they promote well-being. Anxiety disorders are prevalent, and medical students are known to have an even higher prevalence of anxiety than the general population.

Aim. Therefore, the aim is to investigate the correlation between the severity of anxiety and stress-coping strategies among 1st–3rd study year students of Rīga Stradiņš University (RSU) Faculty of Medicine (MF).

Methods. This quantitative, cross-sectional study was based on data collected from 162 RSU MF 1st–3rd study year students who voluntarily filled in an anonymous electronic self-report questionnaire. It consisted of 74 questions, including The Coping Orientation of Problem Experience Inventory (COPE), General Anxiety Disorder-7 (GAD7), and additional sociodemographic questions. The data was collected from December 2022 to January 2023. Statistical analysis (descriptive statistics, Pearson's chi-square test, Fisher's exact test, and Spearman's correlation) was performed using IBM SPSS 29.0.

Results. Severe generalized anxiety was observed in 46 (28.4%) participants, moderate anxiety in 43 (26.5%) participants, mild anxiety in 53 (32.7%) participants, and no anxiety symptoms in 20 (12.3%) participants.

- A statistically significant, very weak, negative correlation was observed between the severity of anxiety and the following stress-coping strategies: planning ($r=-0.166$, $p=0.034$) and acceptance ($r=-0.154$, $p=0.050$).
- A statistically significant, weak, negative correlation was observed between the severity of anxiety and positive reinterpretation & growth strategy ($r=-0.203$, $p=0.009$).
- A statistically significant, very weak, positive correlation was observed between the severity of anxiety and mental disengagement strategy ($r=0.193$, $p=0.014$).
- A statistically significant, weak, positive correlation was observed between the severity of anxiety and the following stress-coping strategies: denial ($r=0.275$, $p<0.001$), behavioural disengagement ($r=0.267$, $p<0.001$), and alcohol-drug use ($r=0.239$, $p=0.002$).

Conclusion. Students use a variety of stress-coping strategies, some of which are associated with beneficial and others with adverse effects. More pronounced anxiety is associated with: more frequent use of maladaptive stress-coping strategies, such as denial, behavioural and mental disengagement, and alcohol-drug use; and less frequent use of adaptive stress-coping strategies, such as planning, positive reinterpretation & growth, and acceptance.

INTERNAL MEDICINE

Evaluation of the effectiveness of metabolic factors in the use of glucagon-like peptide-1 agonists

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Background. Glucagon-like peptide-1 (GLP-1) agonists are a class of medication used to treat type 2 diabetes (T2DM). This medication is an incretin hormone that stimulates insulin secretion after oral glucose ingestion. Although metformin remains first-line therapy for T2DM, the addition of GLP-1 should be considered in patients who are contraindicated for metformin use or in patients with subtarget HbA1c. These medications are approved as a treatment option for obesity which are known to reduce metabolic syndrome risks, such as elevated levels of serum uric acid (SUA). Cardiovascular disease is the leading cause of death in patients with diabetes. GLP-1 agonists lower cardiovascular disease risk factors by improving endothelial function thus reducing atherosclerosis progression.

Aim. The aim of the current study was to analyse metabolic and other factors that could affect the effectiveness of GLP-1 medication in two groups of patients: with adiposity and with T2DM.

Methods. A retrospective study was performed. In total, 84 patients were enrolled in the study and divided into 2 groups: 50% of patients with adiposity (group A) and 50% of patients with T2DM (group B). Their medical histories were analysed. Data were processed and analysed using R (version 4.2.1.).

Results. It was determined that in both (A, B) groups the body mass index (BMI) was lower after GLP-1 treatment, with statistically significant differences ($p<0.005$). In group A, BMI was strongly influenced by HOMA-IR ($r=0.76$) and was weakly associated with baseline vitamin D levels ($r=0.27$). Positive correlation was found between HOMA-IR and changes in triglyceride levels ($r=0.89$). Positive influence was observed between age and SUA changes ($r=0.65$). In group B, significantly lower HbA1c was observed ($p=0.0002$). GLP-1 treatment duration affected SUA ($r=0.50$). Weak negative correlation was observed in between the age and changes in SUA ($r=-0.40$).

Conclusion. The study confirms the metabolic efficiency of GLP-1 both in reducing BMI in groups A and B and in reducing HbA1c in group B. The results show that several factors significantly affect the effectiveness of GLP-1 agonists. In group A, as HOMA-IR improves during therapy, a more pronounced reduction in residual triglyceride levels is expected. Vitamin D levels could play an important role in predicting changes in BMI during therapy. SUA changes during therapy are expected to be more pronounced in younger patients without T2DM, but the duration of GLP-1 therapy in SUA reduction is expected to be essential in patients with T2DM.

Habits of vitamin D supplementation in Latvia – first results from a larger observational study

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Background. Positive role of vitamin D (VitD) supplementation in human health has been confirmed in multiple studies. However, no known study has looked into details of habits of VitD supplementation in Latvia, linking VitD status with chronic and acute respiratory illnesses. We present first results from our observational study (as of 2 Jan 2023) as recruitment still continues.

Aim. To investigate habits of VitD supplementation in Latvia in the light of chronic illnesses and acute upper-airway infections, including COVID 19.

Methods. Study design: I. Recruitment, II. Survey, blood test (BT); III. Survey, BT in 6 months; in between – regular reporting of VitD use; study to last until May 2023.

348 adult patients from cities and regions analysed. VitD status defined as in 2022 and 2013 consensus documents on VitD supplementation (Pludowski et al, 2013, 2022): insufficiency (<30 ng/mL); target-level (>=30–50 ng/mL); high-intake (>50 ng/mL).

Results. The median age was 51 years. The men-to-female ratio was 1:5.82. The median body mass index was 25.6. 80.5% (n=280) of all participants supplemented VitD; of those 71.8% (n=201) – regularly, achieving significantly better VitD level – 37.6 ng/mL vs. 25.1 ng/mL in hectic VitD habits ($p<0.001$). Nevertheless, VitD level was insufficient in 48.2% (n=168) of all participants; in 28.4% (n=51) of those with regular VitD use. Best VitD level in those supplementing VitD regularly ⩾6 months (41.4 (51–32) ng/mL).

34% (n=119) of all had chronic illnesses.

We found a strong negative correlation between:

- BMI and VitD, particularly in PAH group, suggesting for higher VitD doses in overweight individuals ($p<0.001$);
- BMI and VitD in those who got COVID 19 infection ($p<0.001$).

Preferred forms of VitD supplements: capsules (30.7%), drops (28.8%), ampules (26.6%). Significantly higher VitD levels in ampule-users (28.7; 27.9; and 37.6 ng/mL, accordingly). VitD better in those not using drops (35.7 vs. 28.8 ng/mL) ($p<0.001$).

Conclusion. 20% of all did not supplement VitD. Only ½ of all used VitD regularly. There is urgent need to raise awareness about role of VitD on health.

Acknowledgements. Financial support from Orivas, Ltd.

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The impact of dual intervention with dexamethasone and high-dose vitamin D on 25-OH-D3 and inflammatory markers

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Background. SARS-CoV-2 infection can alter the immune response and cause a cytokine storm, increasing the risk of patient mortality. The aim of this study was to determine the dual effects of dexamethasone and micronised vitamin D3 spray therapy on 25-OH-D3 levels and inflammation markers in hospitalized COVID-19 patients.

Aim. In this randomized, placebo-controlled study, 80 inpatients with vitamin D deficiency and PCR-confirmed SARS-CoV-2 infection were divided into two groups (post-hoc analysis). Due to viral pneumonia 54 (67.5%) patients received dexamethasone therapy. The intervention group received 12,000 IU of vitamin D per day for five consecutive days, while the control group received a placebo. Changes in 25-OH-D3 levels and inflammation markers were monitored throughout the therapy.

Results. Dexamethasone did not affect rapid increase of 25-OH-D3 levels in COVID-19 patients. Decrease in CRP levels was 4.6 times faster ($p=1.17\times10^{-2}$) when the patient received both high-dose vitamin D and dexamethasone compared to the patients without dexamethasone therapy, and three times faster ($p=3.83\times10^{-2}$) compared to the dexamethasone monotherapy. Ferritin level decreased more markedly ($p=1.48\times10^{-2}$) when patients received both high-dose vitamin D but not dexamethasone.

Conclusions. The findings indicate that dexamethasone combination with high-dose vitamin D therapy is beneficial for rapid reduction of inflammation markers in hospitalized COVID-19 patients with pneumonia. Further research is needed to determine the optimal duration and dosing of this dual intervention.

Acknowledgements. The authors declare absence of conflict of interest.

COVID-19 infection impact on low density lipoproteins in the bloodstream on practically healthy individuals

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Background. The problem is of outstanding importance since the year of 2019 when the COVID-19 infection became a significant health problem all over the world. Since then, only some research has been made on low density lipoprotein changes in the bloodstream, moreover, most of it excluded practically healthy individuals. The virus has an impact on all the organ systems, mostly affecting the respiratory and the gastrointestinal system.

Aim. The aim of the study was to understand on a deeper level impact of COVID-19 pandemic on practically healthy individual low density lipoprotein level changes, comparing the levels in the bloodstream before and after recovering from the infection.

Methods. Altogether 48 patients were enrolled in the retrospective study since the October 2022 till January 2023. The study population were individuals over 18 years old and without any chronic illnesses. Patients were divided in 2 groups, first group who were physically active during the illness, did not consume fast foods and the other group who were not physically active and consumed fast foods. All the participants signed consent. For data collection the laboratory test data were taken from ambulatory cards in the family doctor practise. Patients filled survey of 11 questions about their lifestyle. The study was approved by ethics committee.

Results. 43% of first group participants were females (n=10) and 56% males (n=13), the average low density lipoprotein level before disease was 2.89 mmol/L, after the disease 2.64 mmol/L (on average decreased for 0.25 mmol/L). 56% of the second group were females (n=14) and 44% males (n=11), the average low density lipoprotein level before disease was 3.16 mmol/L, after the disease 3.42 mmol/L (on average increased for 0.26 mmol/L).

P(T<=t) one-tail in both cases is significantly higher than the threshold value (p=0.05), which means a statistically significant association between lack of physical activities, consumption of fast foods and increased low density lipoproteins is found.

Conclusion. The study results show a correlation between lifestyle habits during the disease and increase of low-density lipoproteins after the disease which increase the risk of cardiovascular events in the future.

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Results of continuous glucose monitoring in patients with type 1 diabetes and diabetic kidney disease – a pilot study

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Background. Type 1 diabetes (T1D) is the leading cause of kidney disease worldwide, including diabetic kidney disease (DKD). The major contributor to development of DKD is hyperglycaemic burden.

Aim. Evaluate the association between the glucose variability assessed by continuous glucose monitoring (CGM) in patients with stable and progressive DKD and T1D.

Design. Cross-sectional case-control study. This study took place in Riga, Latvia, at the University of Latvia. Sample consists of 78 patients with T1D.

Methods. Progression of DKD was defined as estimated glomerular filtration rate (eGFR) decline ⩾3 ml/min/year or increasing albuminuria stage over the last 3–6 years. FreeStyle Libre ProiQ Sensors were used to obtain CGM data.

Results. The mean age in the T1D group was 45.27±11.75 years, mean diabetes duration was 25.48±11.43 years, the mean HbA1c was 8.52±1.98%, the mean days CGM sensor used was 14.45±1.38 and the prevalence of progression of kidney disease was 44.87% (n=35). Groups of progressors and non-progressors did not differ in gender, age, anthropometric measures, smoking habits, and days CGM sensor was worn. Progressors had higher prevalence of cardiovascular hard endpoints (p=0.03), higher HbA1c (p=0.03) longer T1D duration (p=0.03), higher prevalence of arterial hypertension (p<0.01); severe retinopathy (p<0.01); end-stage renal disease (p<0.01) compared to non-progressors. Groups of progressors and non-progressors did not differ in the following CGM indicators: low glucose events, % in target, % above target, coefficient of variance. Average glucose median, GMI and estimated A1C were higher in progressors versus non-progressors at the significance level of 0.1 (average glucose: non-progressors 8.95 (7.53–11.20) mmol/l, progressors 10 (8.70–13.10) mmol/l; p=0.08; GMI non-progressors 7.15 (6.60–8.15)%, progressors 7.60 (7.00–28.90); p=0.08; estimated A1C non-progressors 7.25 (6.32–8.65)%, progressors 7.9 (7.10–9.90) %), p=0.09).

Conclusion. Several parameters of glucose variability were associated with DKD at a significance level of 0.1. Glucose variability may be a factor in the development of DKD, but further research is required.

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Accumulation of advanced glycation end products in patients with different severity of diabetic retinopathy

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Background. Diabetic retinopathy is a common complication of diabetes mellitus and the leading cause of blindness in the working-age population in the developed world. Novel biomarkers are needed to improve the screening efficiency for the prevention and early treatment of this condition. Accumulation of advanced glycation end products (AGEs) is considered as one of pathogenetic pathways in development of diabetic complications. AGEs are formed in the process of non-enzymatic glycation of proteins. AGE assessment could represent a long-term memory of prolonged hyperglycaemia and could be an important biomarker in diabetic retinopathy.

Aim. The aim of the current study was to compare AGEs risk groups in patients with different stages of diabetic retinopathy in Latvia.

Methods. Altogether 115 patients with type 1 diabetes and type 2 diabetes were enrolled in the study. Patients were stratified into three groups according to the severity of diabetic retinopathy: "no retinopathy" group; "non-proliferative retinopathy" included patients with mild, moderate, and severe non-proliferative retinopathy; group of "proliferative retinopathy" included patients with non-high and high-risk proliferative retinopathy and status post laser-photocoagulation. AGEs risk groups (0, normal; 1, mild risk; 2, intermediate risk; 3, high risk) were determined using AGE reader (Diagnoptics). Statistical analysis was performed using programme R: Wilcoxon, Kruskal-Wallis, Fisher, chi-square test, Kendall's test and Spearman's correlation.

Results. Subjects in the group of "no retinopathy" (n=57) compared to "non-proliferative retinopathy" (n=29) and "proliferative retinopathy" (n=29) were statistically significantly older, had shorter duration of diabetes, higher prevalence of type 2 diabetes, lower HbA1c and low-density lipoprotein levels, less diabetic maculopathy cases. In the group of proliferative retinopathy there was a higher albumin/creatinine ratio in urine. AGE risk groups were statistically significantly related with the presence and severity of diabetic retinopathy (p=0.000) and also moderately positively correlated with the progression of diabetic retinopathy (Spearman correlation=0.438).

Conclusion. In this study, we demonstrated differences in AGEs risk groups between patients with different stages of diabetic retinopathy in Latvia. AGEs risk group could be used as one of non-invasive biomarkers of diabetic retinopathy.

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Impact of menopause hormone therapy on the low-density lipoprotein levels in menopausal women

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Background. Menopause hormone therapy (MHT) also known as hormone replacement therapy (HRT) can affect the levels of low-density lipoprotein (LDL) cholesterol in menopausal women.

During menopause the levels of hormones such as estrogen and progesterone naturally decline. MHT involves replacing these hormones in order to alleviate menopausal symptoms such as hot flashes and night sweats. Some studies suggest that MHT may have a positive effect on LDL cholesterol levels in menopausal women.

Aim. The purpose of the study is to evaluate and compare low-density lipoprotein level changes during the menopausal period in women who take hormone replacement therapy with those not taking any hormones.

Methods. This is a cross-sectional study in which a survey was used as a research instrument. The target group of the study consists of 104 respondents who are women aged between 40 and 70 years. Two samples of respondents were included in the study: a control group of respondents not taking HRT and a study sample of women taking HRT. Data analysis was performed in IBM SPSS Statistics 20 program using Pearson chi-square and Pearson correlation.

Results. Among all study participants 52 women took hormone replacement therapy and 52 women did not take it. Combined oral therapy with the use of estrogens and progestins was the most popular choice and used by 77% (n=40). Other types of therapy were not so common: oral estrogens were used by 13% (n=7); transdermal estradiols, 4% (n=2); and vaginal estrogens, 6% (n=3).

Around 44% (n=46) of women taking hormone therapy had low-density lipoprotein of $\leqslant$3 mmol/l, but those who did not take the therapy had low-density lipoprotein with a value $\leqslant$3 mmol/l and account for only 9% (n=9). The level of low-density lipoprotein of $\geqslant$3 mmol/l was found in 41% (n=43) of women who did not take therapy and in 6% (n=6) of women who took therapy.

Pearson chi-square test showed a statistically significant association between the increased level of low-density lipoprotein and the use of therapy in menopausal women (χ^2=52.8, p<0.001).

Conclusions. Menopause hormone therapy reduces low-density lipoprotein levels in menopausal women, which reduces the risk of cardiovascular events. However, it is important to note that MHT may not have the same effect on all women.

Acknowledgements. The authors declare absence of conflict of interest.

The effect of vitamin D on inflammatory markers during remdesivir therapy

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Background. COVID-19, caused by the SARS-CoV-2, can affect the immune system, leading to a cytokine storm and increased risk of mortality. Vitamin D deficiency has been linked to an increased risk of severe forms of COVID-19, and vitamin D supplementation has been suggested as a potential treatment option. This study aimed to evaluate the effects of short-term high-dose vitamin D supplementation on circulating 25-OH-D3 levels and inflammatory markers in hospitalized COVID-19 patients receiving remdesivir therapy.

Aim. Our study was a randomized, placebo-controlled trial that enrolled 80 inpatients with vitamin D deficiency and PCR-confirmed SARS-CoV-2 infection. Patients were randomized to receive either 12,000 IU of vitamin D per day for five days (total dose of 60,000 IU) or a placebo. Remdesivir therapy was prescribed to 37.5% of patients. The primary outcomes were changes in circulating vitamin D levels and inflammation markers (C-reactive protein, fibrinogen, ferritin, IL-6) during therapy. The study followed local clinical guidelines and was conducted in a hospital setting.

Results. CRP levels decreased five times faster in COVID-19 patients who received both high-dose vitamin D and remdesivir compared to those who received remdesivir alone ($p=3.21\times10^{-2}$). No statistically significant differences were observed in IL-6, ferritin, and fibrinogen levels in patients receiving remdesivir with or without high-dose vitamin D supplementation.

Conclusions. Remdesivir therapy in combination with high-dose vitamin D spray therapy may be a useful approach for reduction of CRP levels in hospitalized COVID-19 patients.

Acknowledgements. The authors declare absence of conflict of interest.

Assessment of quality of life in patients with chronic spontaneous urticaria and acquired angioedema in Latvia

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Background. Chronic spontaneous urticaria (CSU) is an autoimmune non-allergic disease that is characterised by the occurrence of hives, isolated angioedema, or both, that last more than 6 weeks. This condition can significantly affect an individual's quality of life, including their well-being, social relationships, cognitive function and work performance, sleep, and mental health. In addition, the quality-of-life test results serve as an indicator of disease control for specialists and a tool to evaluate the efficacy of therapy.

Aim. The aim of the current study was to determine the quality of life in patients with chronic spontaneous urticaria and acquired angioedema in Latvia.

Methods. The quantitative research method was used in the study. The quality of life was determined by Chronic Urticaria Quality of Life (CU-Q2oL) and Angioedema Quality of Life (AE-QoL) questionnaires. In data analysis, descriptive statistics were used.

Results. We interviewed 75 patients with chronic spontaneous urticaria. Most of all patients were female (88%) and the mean age was 42.7±13.6 years (18–73). Out of all patients, 67 completed CU-Q2oL questionnaire (89.95% female, 10.05% male) and 8 completed AE-QoL questionnaire (85.7% of females, 14.3% of males). In CU-Q2oL questionnaire the highest score being 91.3 and the lowest – 4.35 across all participants. Analysis of the CU-Q2oL questionnaire revealed that the itching/embarrassment domain was the most affected with an average impact of 57%, followed by mental status (53%), sleep (52%), appearance (49%), functioning (45%), and swelling/eating (30%). Similarly, in AE-QoL questionnaire the highest score being 88.24 and the lowest – 11.76. The AE-QoL questionnaire revealed that the fears/shame domain was most affected with an average impact of 60%, followed by nutrition (53%), functioning (52%) and fatigue/mood (34%).

Conclusion. In summary, this study found that patients with CSU and angioedema (AE) in Latvia experience a significant negative disease impact on their quality of life, as assessed by the CU-Q2oL and AE-QoL questionnaires. The study revealed that chronic spontaneous hives are more common in females than in males, with an average age of 42 years. Across the six domains of CU-Q2oL and four domains of AE-QoL itching/embarrassment domain is most affected in patients with CSU and fears/shame domain in patients with AE, with an average impact of 57% and 60%, respectively, emphasizing the need for effective interventions to improve the patients' quality of life.

Acknowledgements. The authors declare the absence of the conflict of interest.

PUBLIC HEALTH AND EPIDEMIOLOGY

Is the health self-assessment delusion of reality? Cross-sectional study of cardiovascular risk factors in population of Latvia

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Background. Cardiovascular diseases (CVD) and cardiovascular risk factors (RF) are the leading in terms of morbidity and mortality in most developed countries. Health self-assessment (HSA) is strong predictor of morbidity and mortality of CVD.

Aim. To determine correlations of HSA and main RF using data of Population Based Cross-sectional Study of Cardiovascular Risk Factors in Latvia.

Methods. A total of 4070 inhabitants (age 25–74) of Latvia were enrolled in the study out of the initial statistical sample (6000), formed by random selection from the Housing Register of the Central Statistical Bureau (1'209'756 pers.). Data on the socio-economic status, prevalence of arterial blood pressure (BP), low density lipoprotein cholesterol (LDL-C) level, glucose (Glu) level, body mass index (BMI), smoking, health self-assessment was obtained in face-to-face interviews, physical measurements and blood tests. The data were processed using Microsoft Excel and R.

Results. Individuals with normal BP had better HSA (good 61%, fair 34%, poor 5%) than individuals with elevated BP (good 32%, fair 55%, poor 13%); $p<0.001$. Individuals with normal Glu level had better HSA (good 56%, fair 37%, poor 7%) than individuals with elevated Glu level (good 35%, fair 53%, poor 12%); $p<0.001$. Individuals with normal BMI <25 had better HSA (good 62%, fair 32%, poor 6%) than individuals with elevated BMI (good 44%, fair 47%, poor 9%); $p<0.001$. There was no statistically significant difference between individuals with normal and elevated LDL-C level as well as between non-smokers and daily smokers. Individuals with less RF (including five: elevated BP, LDL-C, Glu, BMI, daily smoking) had better HSA than individuals with more RF: 0 RF – good HSA in 74%; 1 RF – in 65%; 2 RF – in 50%; 3 RF – in 40%; 4 RF – in 30%; 5 RF – in 23%; $P<0.001$.

Conclusion. People with less RF have better HSA. At the same time there are a lot of daily smokers and people with several RF who assess their health inappropriate high. The precisely targeted educational campaigns on RF recognition and significance are needed.

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Do we know unmet non-medical health-related social needs in mental healthcare?

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Background. Environmental and social factors, also known as social determinants of health, are measurable and modifiable. The detrimental impact of deprivation, inequality, and unmet non-medical social needs (food insecurity, housing instability, a lack of access to transportation, an inability to afford utility bills, and exposure to interpersonal violence) on health and healthcare utilisation is well-established. Addressing them can reduce healthcare costs and predict the population's need for early mental health interventions. To ensure collaboration among scholars, policymakers/authorities and healthcare stakeholders, standardised measurements of non-medical social factors are used.

Aim. The current study aimed to explore the availability, eligibility, and use of the national statistical data on social factors of mental health patients and suicide victims for the organisation of mental health care in Latvia.

Methods. EU and national statistical data analysis (stat.gov.lv, Eurostat/EU-SILC, ministerial reports) on the matter was done. Literature on indices and screening tools concerning social needs for persons with mental health difficulties was studied. Analysis of The Plan for improving the organisation of mental health care 2023–2025 Latvia was performed.

Results. There are mainly demographic and some social data of mental health service users and suicide victims available in Latvia. Various measurements and indices (individual and population-based) are used for administrative purposes worldwide and in the EU. The need for novel indices that consider specific parameters is emphasised in the literature. The suicide audits (reviews) demonstrate their feasibility in generating recommendations for suicide prevention.

Conclusions. The data management in the Latvian healthcare system does not provide a comprehensive view of the social needs of psychiatric patients and suicide victims. The available indices comprise factors that are irrelevant to Latvia. For effective public health interventions planning and ensuring social cohesion, it is necessary to develop and implement a regular and systematised assessment and monitoring of social factors of psychiatric inpatients with a unified screening tool. An original social deprivation index should be developed, implemented, and monitored in Latvia. Audits of suicides should be conducted. Stakeholder partnership and cross-sectoral cooperation should be promoted.

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Cardiovascular disease risk factors in Latvia: trends in prevalence over 10 years

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Background. Cardiovascular diseases (CVD) are the leading cause of premature death in Latvia. The most important risk factors of CVD are high blood pressure (BP), high levels of low-density lipoprotein (LDL) cholesterol and glucose, high body mass index (BMI) and smoking.

Aim. The aim of the study was to assess the prevalence of the most common CVD risk factors and to find the overall trend by age and sex groups in the Latvian population comparing the data of two cross-sectional studies proceeded in 2009 and 2019.

Methods. Data from 3,807 participants of the 2009 year's study and from 2,218 participants of the 2019 year's study were analyzed. Descriptive statistics and the chi-square test were used to assess the number of risk factors and differences between them in each age group in both studies. We compared the age groups of the people that became 10 years older, for example, for 55–64 years olds in 2009 the comparison group was 65–74 years olds in 2019. A p value of $\leqslant 0.05$ was considered statistically significant

Results. The prevalence of mean number of CVD risk factors in the population has decreased over 10 years: in 2009 it was 2.36 (SD 1.19), and in 2019 it was 2.10 (SD 1.19). A higher level of all risk factors was observed among men and in age groups 55–64/65–74 for both genders.

Prominent changes in the prevalence of risk factors were observed in the high LDL cholesterol: a decrease of 7.6% between the age groups 45–54/55–64 and a decrease of 19.9% between the age groups 55–64/65–74 (for years 2009 and 2019, respectively).

Glucose levels and high BMI tended to increase with age in both genders, except between the age groups 55–64/65–74.

There was a significant decrease in men smoking: by 12.2% among the age groups 55–64/65–74. No statistically significant differences were observed in women smoking.

Statistically significant differences in the prevalence of high BP were observed in women: 6.7% increase in high BP in the age groups 35–44/45–54 (for years 2009 and 2019, respectively). In age groups 55–64/65–74 a statistically significant decrease of high BP by 10.2% was observed.

Conclusion. The CVD risk profile of the Latvian population has improved over 10 years.

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Self-reported parents' approach to the health competence in the family

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Background. Parents' health competences, the approach to their own and their children health can affect family lifestyle, and child health attitudes formation.

Aim. To analyse self-reported parents' approach to the health competence in the family.

Methods. In the survey participated 303 parents, who have primary schoolchildren and live in the Western Lithuania. Spearman correlation coefficient was applied.

Results. The survey revealed that about 1/4 parents rated their health as very good (24.1%), about half of parents – as good (56.1%), and 1/5 – as moderate (19.1%). Nearly half of parents rated health of their child as very good (40.6%), a half of parents – as good (51.5%), and 7.9 % – as moderate. Parents who rated better their own health, better evaluated their child health (R=0.524; p=0.000).

About half (52.1%) of parents assessed their health competence as good, and 8.6% – as satisfactory.

The study showed what ways parents use for self-education to improve their health competences. Most parents use information about the means tested by the relatives and friends (81.3%), and advice of a family doctor/paediatrician (81.1%). 62.7% of parents rely on the advice of a dentist or oral hygienist, and 46.5% – use scientific literature for self-education. Unfortunately, only minority of parents attend health education training (13.5%) and use the results of health projects (14.5%). One-third (29.7%) of parents rely on the advice of the teacher.

Almost all (99.7%) parents believe that they are responsible for the health of their children. The parents indicated that national policy should be focused on the health promotion, starting with restrictions for unsafe products or the development of infrastructure for bicycle paths.

Conclusion. More than half of parents rated their health as very good or good, and they tended to rate their children's health a little better. Parents believe that they have health competences: knowledge about health, skills, and abilities, and have formed positive attitudes towards health.

Parents improve their health competences in various ways, mainly using the means tested in their close environment or advised by a doctor. Parents still do not enough use the advice and observations of their child teacher, although the primary school teacher conducts individual monitoring of children every day, and knows the individual characteristics, behaviour, mood, etc. of the child. Almost all parents believe that they are responsible for their children health; however, they miss the general health promotion policy.

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Associations between subjective physical health and substance abuse

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Background. Physical health can be simply defined as systems of the body functioning properly, the lack of illness or disease. Abuse of alcohol, tobacco, and drugs could be one of the culprits of poor subjective physical health. Body dissatisfaction is as high as 69.5% and more in Western countries and it is known to cause eating disorders and substance abuse which often lead to health issues.

Aim. The aim of this study is to analyze associations between subjective physical health and substance abuse.

Methods. The current study was conducted as an online interview in 2021. A total of 1255 respondents were included in the final sample. Information on respondent's subjective physical health was obtained using the 36-Item Short Form Survey (SF-36). Respondents were asked about their smoking, drinking and drug consumption habits. Statistical analysis was performed using the IBM SPSS 23.00 Version program.

Results. The majority of our respondents (55.4%) had a normal body mass index (BMI). Poor subjective physical health was associated with both higher subjective body weight ($p<0.001$). Pain intensity in the last 4 weeks was related to consumption of wine ($p=0.033$). We found an association between subjective physical health and respondents' smoking status ($p=0.048$). Pain intensity in the last 4 weeks was also related to the respondents' status of smoking ($p=0.015$). Respondents who smoked were more likely to rate their subjective physical health as poor and experience higher pain intensity. Neither the subjective physical health nor the pain intensity in the last 4 weeks was related to the use of drugs ($p>0.05$).

Conclusion. Tobacco smoking tends to increase body pain intensity. Substance abuse has a deleterious effect on subjective physical health and general wellbeing.

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The occurrence of gynecomastia and its link to men's lifestyle choices and self-worth

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Background. Correlations between physical appearance, lifestyle and self-esteem became increasingly popular in the era of social media. Nevertheless, the topic of gynecomastia remains in the shadow of public health perception and its impact on men's self-esteem remains unknown.

Aim. The purpose of this article was to assess gynecomastia prevalence and the associations between gynecomastia signs, men's lifestyle, and self-esteem.

Methods. An anonymous voluntary survey was conducted online in 2020–2021. Overall, 675 male respondents assessed their gynecomastia signs, presence of thickened glandular tissue beneath the nipple, diameter of the thickened tissue and breast appearance by Simon's visual gynecomastia scale. Respondents answered Brief Tripartite Questionnaire – Gynecomastia (BTQ-G) and Chest Satisfaction Questionnaire (CSQ), questionnaire about harmful habits, lifestyle. The average age of respondents was 30.14±10.15 years.

Results. The majority of participants (44.3%) reported that their chest matched image 0, which is considered a normal chest. Additionally, more than a third (35.7%) selected an image on Simon's gynecomastia scale that marked a slight breast enlargement. The study found that older respondents tended to choose images with higher breast enlargement degrees on average ($p<0.001$, $r=0.22$). Statistically significant correlation was found between factors such as tobacco use, alcohol consumption, the use of medications, supplements, and a higher breast enlargement grade ($p<0.001$). It was found that those with higher breast enlargement grades were more likely to use cardiovascular medications, as indicated by a statistical significance ($p<0.001$). Furthermore, it was found that those who had a lower level of physical activity had a higher average breast enlargement grade ($p<0.001$). Bigger breast enlargement was found to be statistically significant correlated with poorer self-esteem as measured by the CSQ and BTQ-G questionnaires (respectively $r=-0.526$ and $r=-0.209$; $p<0.001$).

Conclusion. Approximately 20% of males have noticeable enlargement of breasts (2A, 2B and 3). Factors that have major influence on manifestation of gynecomastia signs are older age, lack of physical activity, alcohol abuse, tobacco smoking, use of medications and food supplements. Bigger breast enlargement is linked to poorer self-esteem.

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ANESTHESIOLOGY, REANIMATOLOGY & INTENSIVE CARE

Anaesthesia-related patient satisfaction: a survey

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Background. Patient satisfaction after anaesthesia plays a major role in the quality of health care.

Aim. The aim of this study is to evaluate patient postoperative satisfaction at the Hospital of Traumatology and Orthopaedics.

Methods. A prospective study involving patients who underwent different types of orthopaedic surgeries was conducted from September to November 2022. Patients were interviewed on the second day after the surgery. Satisfaction with anaesthesia was categorized from 0 to 10, 0 being completely unsatisfied and 10 being completely satisfied. Patients were asked to evaluate their side effects – nausea and vomiting, itching, sleepiness and discomfort. Patients were categorized based on the gender and the age group. Statistical analyses were performed using SPSS 27th version. Spearman's Rho test was used to assess the association between factors.

Results. A total of 102 patients were included in this study. The mean score of patient satisfaction was 9.42 in both female and male patients. Although women noted stronger side effects. Looking at mean satisfaction with anaesthesia by age groups, the lowest score was in the 18–39 age group – 9.17 and the highest in the 80–90 age group – 9.88. Factors that affected patient satisfaction – nausea and vomiting ($p=0.373$), itching ($p=0.616$), sleepiness ($p=0.262$) and discomfort ($p=0.324$).

Conclusion. Patient satisfaction with anaesthesia was high. Younger patients have more side effects and are less satisfied with anaesthesia. Factors that affected patient satisfaction may be prevented and improved.

Acknowledgements. There are no conflicts of interest.

Comparison of fascia iliaca compartment block and standard multimodal analgesia in patients with hip fracture in emergency department

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Background. Hip fracture is one of the most common injuries among the elderly because the population is aging. It is expected to remain a major clinical challenge and public health problem for the foreseeable future. Pain management is integral to the management of hip fracture. Patients with hip fracture experience moderate to severe pain in the emergency department.

Aim. Aim of our study was to compare fascia iliaca compartment block (FICB) and standard multimodal analgesia on the pain relief in patients with femoral fracture.

Methods. A prospective randomised study was conducted at the Hospital of Traumatology and Orthopaedics, Riga, Latvia (April 2022 to November 2022) and enrolled 80 patients with hip fractures. All subjects were randomised into the 2 groups, using the online tool on www.randomiser.org. Treatment group (R) was allocated to block with Ropivacaine (75–112 mg depending on the patient weight). Both groups (R and control (C)) received multimodal analgesia. Primary outcome was the intensity of pain. Pain level was evaluated using Numerical Rating Scale (NRS) in the admission period, then 15 min, 1 h, 2 h, 4 h and 8 h after FICB. The data were analysed using SPSS Version 27.0.

Results. 80 patients (60 females and 20 males) aged 40–96 years were included in the study. Pain in the emergency department after admission – in the C group 6.73 in the NRS scale, in the R group 7.13. 1 h after FICB pain in the C group was 4.48 [4.26; 4.69], in the R group 2.33 [1.97; 2.68], $p<0.0001$, 4 h after in the C group 4.13 [3.88; 4.37], in the R group 2.28 [1.95; 2.60], $p<0.0001$, 8 h after in the C group 4.30 [3.96; 4.64], in the R group 2.45 [2.16; 2.74], $p<0.0001$. Analysing patients with pain level >5 NRS points, in the C group, 25% of the patients did not exceed 5 points, while, in the R group, 97.5% did not exceed 5 points.

Conclusion. The results show that FICB alone is effective analgesia. FICB performed in the Emergency department is an effective method that significantly reduces pain in patients with hip fractures. Comparing FICB and standard multimodal analgesia showed that pain levels using FICB was lower and request for opioids was lower.

Plexus brachialis block in the axillary approach to forearm bone fractures using long and moderate – acting local anesthetics

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Background. The main unpleasant thing after forearm surgery done in the *Pl. Brachialis* block anaesthesia are numbness and pain.

Aim. The aim of our study was to find the most effective regional anaesthesia approach for forearm osteosynthesis.

Methods. A prospective randomised study was conducted at the Hospital of Traumatology and Orthopaedics, Riga, Latvia (August 2022 to January 2023) after Ethics Committee approval and enrolled 38 patients with forearm fractures. All subjects were randomised into 2 groups, using the online tool on www.randomiser.org. Treatment group (R) was allocated to *Pl. Brachialis* block with Lidocaine 1% – 25 ml in axillar approach and Bupivacaine 0.25% to 5 ml per nerve in elbow joint area (*n. ulnaris, n. radialis, n. medianus*). Control group (C) was allocated to *Pl. Brachialis* block with Bupivacaine 0.25% to 25 ml in axillar approach. Both groups received similar multimodal analgesia. Pain level was evaluated using Numeric Rating Scale (NRS) in the pre-operative period, then 1h, 18 h and 22 h after forearm osteosynthesis.

Results. 38 patients (30 females and 8 males) were included in the study with mean age of 56.2 years [95% CI 50.8–61.6] and BMI 26.2 [24.8–27.6]. The mean duration of the operation was 157.5 [143.5–171.44] min. Pain at rest and movements before surgery in both groups was similar.

1 h after surgery pain level during movement in fingers was 1.6 [1–2.3] in group C and 3.4 [2.3–4.6] in group R ($p<0.05$). Pain during movement in shoulder and elbow in control group has given better results: 1.8 [0.9–2.5], comparing to study group 3.0 [1.9–4.1], but the difference was not statistically significant. Movements did not differ in both groups as well as patient's overall satisfaction.

Conclusion. *Pl. Brachialis* block performed with medium acting local anaesthetic Lidocaine in the axillary approach and long-lasting anaesthetic Bupivacaine distally does not improve patient satisfaction, movement and pain level after surgery.

Adaptive pain mechanisms – small fiber responses in winter swimmers: results of a pilot study

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Background. Winter swimming is once again becoming popular activity in the Northern countries, sustain an overall opinion of being a health promoting activity and even as a way to treat many diseases such as rheumatoid arthritis, asthma, fibromyalgia, chronic muscular pain. This interest of what can cold water do to the body could be traced back even to Hippocrates, unfortunately studies on this topic are quite few.

Aim. The aim of the current study was to evaluate the responses to pain stimuli in winter swimmers in comparison to healthy controls and patients with chronic muscular pain.

Methods. For our pilot study we enrolled 6 participants (3 male and 3 female), age group 35–40 y.o. Two participants are winter swimmers for 4 years and participate in swimming championships, two participants suffer from recurrent myofascial pain for over 6 month and two are healthy controls. All participants underwent quantitative sensory testing (QST) using hot and cold modalities, thermal pain threshold while monitoring breathing and heart rate to measure analgesia nociception index (ANI).

Results. The base line ANI in winter swimmers (32 and 42) was lower than in healthy controls (55 and 56) and chronic pain participants (57 and 56). During the testing the highest ANI 73–85 was seen the control group, winter swimmers had 49–66 and chronic pain patients 43–61. On sensation test one of the winter swimmers, male, showed a quite quick response to cold stimuli (27.6, 29.3/ normal value lowest limit 29.4) and a late response to hot stimuli (47.1, 49.7/ normal value upper limit 49.9) that mimicked the response pattern of chronic pain participants. Also, the same swimmer did not report any pain on thresholds tests (ANI – 63–66). The second swimmer had a similar pattern tendency as the first one thus staying in normal value range (hot stimulus, female, hands 39.2–45.9/ legs 41.0–46.6).

Conclusion. Apart from being just a pilot study and the need to enquire much more participants in the study, the results arise questions considering beneficial effects of exposure to cold, as the results of QST testing show a C fiber damage and a quick activation of A delta fibers in the male swimmer and a similar pattern but borderline values in the female swimmer.

Also, additional investigations should be performed to understand why there would be a different parasympathetic tone in winter swimmers comparing to healthy controls.

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Nociceptive stimulation monitoring during intravenous lidocaine infusion and regional anesthesia

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Background. Regional anesthesia is the golden standard for perioperative analgesia and anesthesia, but it has its limits and complications. Intravenous lidocaine infusion for perioperative analgesia is a known alternative and the analgetic effect wears off slowly and exceeds its half-life, but is not widely used because of cardiovascular side effects, which are reduced when the appropriate dosage is used. In current clinical practice, only indirect markers can estimate a patient's pain level during the maintenance of general anesthesia.

Aim. The aim of the study is to objectively evaluate the intravenous lidocaine effect with remote photoplethysmography (rPPG) and analgesia *nociception* index (ANI) in high-risk anesthesia patient.

Methods. The pilot study reviewed one patient with oncological illness and pathological fracture of the humerus while inserting a humerus prosthesis. Before general anesthesia induction, the patient received an intravenous lidocaine infusion of 1,5mg/kg over a 10-minute period. During this period, we monitored heart rate, blood pressure, oxygen saturation and took measurements with rPPG and ANI. rPPG is a simple and cost-effective technique for contactless skin perfusion monitoring, using visible or near-infrared light and a video camera. The perfusion index was measured from different sites (1–5) on the dorsal side of the palm before induction of general anesthesia was started. During surgery, another lidocaine infusion was at the rate of 2 mg/kg/h till the end of the surgery, while measurements were taken.

Results. A paired samples t-test was conducted to determine the effect of lidocaine infusion. The mean perfusion index during baseline was 0.18 (SD=0.03), and the mean perfusion index after manipulation [infusion] was 0.374 (SD=0.07) [$t(4)=-4.289$, $p=0.013$].

Conclusions. There were statistically significant perfusion index changes during lidocaine infusion. To validate photoplethysmography findings in this pilot study more extensive research should be done.

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Evaluating the accuracy of patient state index for measuring anesthetic depth in patients undergoing propofol-sevoflurane anesthesia

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Background. Electroencephalography indices such as the Patient State Index (PSI) can be used to measure and monitor anesthetic depth during general anesthesia. To ensure accuracy, the accuracy of PSI needs to be validated against other measures of anesthetic depth, such as the isolated forearm technique (IFT).

Aim. The aim of this study was to evaluate the ability of electroencephalogram-based Patient State Index to assess changes in the level of unconsciousness in patients undergoing intravenous induction with propofol followed by sevoflurane anesthesia. We compared PSI with clinical loss of consciousness defined as loss of isolated hand movements to verbal command.

Methods. Standard anesthesia technique was applied for 17 ASA I–III patients by intravenous administration of medication in following order: Fentanyl 1–2 mcg/kg, Propofol 2–3 mg/kg and Atracurium 0.25–0.5 mg/kg. Anesthesia was maintained with sevoflurane 0.8–1.0 MAC and 1–2 mcg/kg/h infusion of Fentanyl. Forearm contralateral to intravenous cannula was isolated for 30 min using arterial tourniquet before each muscle relaxant administration. PSI, isolated hand movement to verbal command, exhaled and inhaled sevoflurane concentration were registered during the induction of anesthesia, intubation, before, during and after operation and before extubation. Hand movements were defined as specific, non-specific and absence of movement. PSI in the range of 25–50 was considered to indicate optimal hypnotic state for general anesthesia. Presence of intraoperative awareness was assessed using Bruce questionnaire.

Results. After induction of anesthesia and intubation specific hand movements were observed in 41% of patients, 12% showed non-specific hand movements and 47% showed no movement. Before incision specific hand movements were observed in 12% cases. During surgery and immediately after surgery no hand movements were observed. Median PSI after intubation was 34, 31 and 25 in patients who had specific, non-specific and no hand movement ($p=0.14$). Median PSI before incision was 42 vs 35 in patients with and without hand movement (0.26). No patients reported intraoperative awareness.

Conclusion. There was no significant correlation between PSI indicating adequate hypnosis and absence of movement after induction of anesthesia and before incision. PSI monitoring may not be fully reliable for determining depth of general anesthesia.

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OPHTHALMOLOGY

Efficiency of vitrectomy in case of first rhegmatogenous retinal detachment

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Background. Retinal detachment may result in various visual defects, including blindness. If in the past extracellular surgery predominated, vitrectomy is increasingly used today. Although this method is becoming more popular, it is still important to find out in which cases vitrectomy should be chosen as the treatment method.

Aim. To evaluate the efficiency of vitrectomy in case of first rhegmatogenous retinal detachment.

Methods. The study included data from 102 patients over the age of 18 who underwent surgery for the first time for rhegmatogenous retinal detachment between 2019 and 2021. MS Excel and the programming language "R"; were used for data collection and processing. Fischer and chi-square test was used to collect the data.

Results. Data on the localization of exfoliation show that 86 (84.3%) study participants had partial exfoliation and 16 (15.7%) had total exfoliation. Analysis of the duration of delamination shows that in most cases it lasts less than 30 days, but there are atypical cases. Of the patients enrolled in the study, 76 (74.5%) had a single rupture and 26 (74.5%) had multiple ruptures. Topical silicone sealant was used in 26 (25.5%) patients, scleral circulatory in 11 (10.8%) patients, silicone vitrectomy in 35 (34.3%) patients, and gas vitrectomy in 30 (29.4%) patients. Half a year after the operation, reablation occurred in 8 or 14.3% of women and 7 or 15.2% of men. Based on the results of the chi-square test, it can be concluded that the patient's sex has no significant effect on the state of the retina after half a year (p-value=0.895), Fisher's test also shows similar results (p-value=1). One year after the operation, 10 or 17.9% of women and 10 or 21.7% of men responded. Based on the results of the chi-square test, it can be concluded that the patient's sex has no significant effect on the state of the retina after one year (p-value=0.623), and Fisher's test shows similar results (p-value=0.627).

Conclusion. Although no statistically significant results have been obtained on the effect of the surgical method, there is a tendency between vitrectomy and gas vitrectomy to retain the retina better over the half-year and year and over the period, but more data are needed to confirm this hypothesis.

Risk of visual field changes' progression in different stages of primary open-angle glaucoma

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Background. Glaucoma is a chronic, progressive degenerative disease of the optic nerve that causes a characteristic, irreversible visual field defect. According to the World Health Organization, in 2020, an average of 76 million people worldwide had glaucoma. (World report on vision. Geneva: World Health Organization: 2019. License: CC BY-NC-SA 3.0 IGO). In glaucoma, continuous therapy is required to reduce intraocular pressure to prevent the progression of the visual field defect as much as possible. The progression of glaucoma has a very individual course, so it is difficult to predict how fast the existing visual field defect will progress. By determining the cumulative risk of visual field progression in different stages of glaucoma, it would be possible to predict faster progression of the defect and make appropriate adjustments to the existing glaucoma treatment plan.

Aim. To determine the cumulative risk of visual field defect progression in different stages of glaucoma.

Methods. This retrospective study included patients who in the period from 2017 until 2021 had been treated in the RAKUS Ophthalmology Clinic for primary open-angle glaucoma. Data collection: Microsoft Office Excel 2010. Statistical data processing: IBM SPSS Statistics 22.

Results. The study analyzed the health data of 96 patients: 76 women and 20 men. Patients were divided into three groups, depending on the initial stage of primary open-angle glaucoma: in the first group there were 35 people; in the second, 31 people; and in the third, 30 people. In patients with first-stage POAG, the progression of MD changes was –3.94 dB over five years, with a mean of –0.79 dB/year. In patients with second-stage POAG, the MD decreased by –4.84 dB over five years, and the progression of MD changes was –0.96 dB/year. In patients with third stage, the progression of MD changes was –4.44 dB over five years, with a mean of –0.88 dB/year.

Conclusion. The results showed that the fastest rate of progression of visual field changes due to primary open-angle glaucoma was in patients with second stage of disease. Patients with systemic disease have a higher rate of progression of visual field changes. Average intraocular pressure measurements over five years were higher in patients with second and third stages of POAG.

Retinal neurodegeneration in patients with type II diabetes without diabetic retinopathy

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Background. Diabetes mellitus is an increasingly common disease worldwide, thus diabetic retinopathy is the leading cause of blindness in patients of working age. Neurodegenerative processes are involved in the pathogenesis of diabetic retinopathy, so diagnostic and therapeutic strategies based on its detection could ensure timely detection of diabetic retinopathy, allowing for early treatment and protecting the patient from the possible threat of vision loss.

Aim. The aim of the current study was to evaluate neurodegeneration in the retina (ganglion cell, nerve fiber layer thickness) and peripapillary area in patients with type II diabetes without diabetic retinopathy.

Methods. 60 patients were included in the study. Two groups of patients were created: the first had 30 patients with type II diabetes without diabetic retinopathy; the second, 30 patients who did not have diabetes. All patients included in the study underwent an OCT examination to determine the thickness of the ganglion cell layer and the thickness of the retinal nerve fiber layer, as well as OCT-angiography to determine the density of peripapillary vessels. The obtained data were analyzed using the statistical program IBM SPSS Statistics version 25.0.

Results. A statistically significant difference was found between the diabetic patient and the control group – in the ganglion cell layer, the retinal nerve fiber layer, and the density of peripapillary blood vessels. Patients with diabetes have a thinner layer of ganglion cells – 74.52 on average, and 80.83 micrometers in the control group ($P_T=1.80\times10^{-7}$). Similarly, the nerve fiber layer of the retina in diabetic patients is on average 88.10, and in the control group – 96.30 micrometers ($P_T=2.54\times10^{-5}$). In patients with diabetes, the density of peripapillary blood vessels is reduced, more specifically – 44.45%, and in the control group – 45.40% ($P_{MV}=1.53\times10^{-3}$).

Conclusion. Neurodegenerative changes in the inner layers of the retina can be detected early in type II diabetes patients without diabetic retinopathy. A reduced thickness of the retinal ganglion cell layer, retinal nerve fiber layer, and a reduced density of peripapillary blood vessels were found.

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Changes in ganglion cell complex thickness and vascular density of macular capillary plexuses in early non-exudative age-related macular degeneration

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Background. Age related macular degeneration is an eye disease that is related to aging. It is the main cause of low vision in the elderly. Ganglion cell complex is directly affected in many potentially blinding eye diseases. Superficial and deep macular capillary plexuses are providing ganglion cell complex. Therefore, it is important to evaluate whether these structures are affected in case of this disease.

Aim. The aim of the study was to evaluate the changes in the thickness of the ganglion cell complex and the density of capillary plexuses in macula in patients with non-exudative form of age-related macular degeneration.

Methods. 100 patients were enrolled in the study, 50 of them in study group (64 eyes) and 50 in control group (60 eyes), dividing them according to the presence or absence of early age-related macular degeneration. A prospective study was carried out. It included patients who underwent cataract surgery in Riga East Clinical University Hospital. All patients underwent examination of best-corrected visual acuity, intraocular pressure, ganglion cell layer thickness, density of capillary plexuses in macula.

Results. The median best corrected visual acuity in the study group was 0.5, but in the control group, 0.8 ($p<0.001$). It was obtained that in case of initial age-related macular degeneration ganglion cell complex thickness is reduced, compared to control group. The results were statistically significant in all macular quadrants and in the whole image, dividing the macula according to the ETDRS grid ($p\leqslant0.05$). The vascular density of the superficial macular capillary network was statistically significantly different between the groups in most of the quadrants and the image as a whole ($p\leqslant0.05$), except for the parafoveolar temporal, superior and inferior quadrants ($p>0.05$). Similar results were obtained by analysing the vascular density of the deep macular capillary network. Statistically significant differences were found in most of the quadrants and the image as a whole ($p\leqslant0.05$), except for the parafoveolar temporal, upper and lower quadrants ($p>0.05$).

Conclusion. Reduced vascular density of macular capillary plexuses could affect the state of the ganglion cell complex. Reduced thickness of the ganglion cell complex can be the reason for reduced visual function and a narrowed central visual field. Early diagnosis of the dry form of age-related macular degeneration is important because in that case it would be possible to make lifestyle changes and use prophylactic treatment earlier.

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Femtosecond laser-assisted arcuate keratotomy at the time of cataract surgery for the management of preexisting astigmatism

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Background. The correction of preexisting corneal astigmatism and the prevention of surgically induced astigmatism are essential to successful surgical outcomes, a high quality of postoperative vision, and patients' satisfaction.

Aim. To evaluate the outcomes of femtosecond laser-assisted arcuate keratotomy combined with cataract surgery in eyes with low-to-moderate corneal astigmatism.

Methods. This prospective analysis included case records of patients with preexisting corneal astigmatism ranging from 0.5 to 3.0 diopter (D). Study parameters included corneal astigmatism and refractive astigmatism inspection with PENTACAM. Then these data were analyzed at 3 months postoperatively

Results. The study records of 15 patients. The postoperative refractive astigmatism was reduced significantly compared with preoperative corneal astigmatism to 0.14 D±0.23 (SD) from 1.76±0.24 D. Patient who has preoperative astigmatism less than 2.0 D demonstrated postoperative refractive astigmatism of 0.7 D or less. All patients have reduced postoperative astigmatism by at least 0.2 D. No intraoperative or postoperative arcuate keratotomy-related events were observed.

Conclusion. The results suggest that femtosecond laser-assisted arcuate keratotomy represents a safe and effective method for astigmatism correction at the time of cataract surgery.

Surgical outcomes of gonioscopy-assisted transluminal trabeculotomy with polypropylene suture in patients with advanced glaucoma

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Background. Glaucoma is one of the leading causes of blindness worldwide. Gonioscopy-assisted transluminal trabeculotomy (GATT) with polypropylene suture is one of the minimally invasive glaucoma surgical techniques for which there is insufficient data on its efficacy in advanced glaucoma. The trabeculotomy can restore flow of patients' natural drainage system.

Aim. To assess the efficacy and safety of GATT with 5.0 polypropylene suture in patients with advanced glaucoma.

Methods. A retrospective study included 53 eyes of 52 patients, with mean age of 69.0 (63.0–75.0) years, which underwent GATT with 5.0 polypropylene suture by a single surgeon between December 2020 and June 2022. Patients were followed up for at least 6 months. Visual fields, intraocular pressure (IOP), number of glaucoma medication, and ocular adverse events were analyzed. Patients were divided into two groups according to visual field defect, mild to moderate glaucoma MD ⩽12 dB and severe glaucoma visual field defect >12 dB. The postoperative outcomes between advanced and mild to moderate glaucoma were compared.

Results. A total of 26 (49.1%) patients with mild to moderate glaucoma and 27 (50.9%) with advanced glaucoma underwent 360-degree GATT with thermally blunted 5.0 polypropylene suture. Preoperative mean IOP was 25.0 (20.0–29.0) mmHg and it significantly decreased to 12 (10–15.0) mmHg 6 month after operation (p=0.0173). There was no statistically significant difference in IOP between mild to moderate glaucoma and advanced glaucoma before surgery, 24.0 (17.0–28.0) mmHg and 26.0 (21.0–32.0) mmHg, respectively, and 6 months after surgery, 13.0 (10.0–15.0) mmHg and 12.0 (9.0–15.5) mmHg, respectively. The average count of medication (Q1–Q3) used before operation was 3.0 (3.0–3.0), 6 months after the surgery the number of medications decreased to 0.0 (0.0–0.0) (p=0.0033). There was no difference between two groups, 88.68% of patients did not require any medications. One patient with mild glaucoma and one with advanced glaucoma had medically uncontrolled glaucoma after GATT, and they underwent further surgery. In 28 (54.5%) of patients, a hyphema was observed on the first post-operative day, two patients underwent anterior chamber lavage after three weeks due to persistent hyphema. No statistically significant differences in the frequency and degree of hyphema were observed between two groups.

Conclusions. GATT with polypropylene suture is a safe and successful option for treatment of advanced glaucoma. This conjunctival sparing and low-cost operation are effective in advanced glaucoma treatment.

Micropulse cyclophotocoagulation effect on ocular vascular flow – early post-operative results

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Background. Glaucoma is a chronic, progressive degenerative disease of the optic nerve that results in an irreversible visual field defect, therefore requiring ongoing therapy. Vascular risk factors have been identified as an important factor in primary open-angle glaucoma pathogenesis. Micropulse cyclophotocoagulation is a relatively new method for reducing intraocular pressure in glaucoma, and there are currently no recent studies evaluating its effect on ocular vascular flow.

Aim. To evaluate micropulse cyclophotocoagulation effect on the ocular vascular flow in the retina and optic nerve disc, and to compare it with the changes in the ciliary body.

Methods. A total of 10 patients (10 eyes) with primary open-angle glaucoma were enrolled in this study. All patients underwent micropulse cyclophotocoagulation procedure (Iridex G6) under a retrobulbar block. Patients underwent a full ophthalmic examination before the procedure and one month after the procedure. Changes in the ciliary body were detected using UBM, and ocular vascular flow was measured using OCTA. Statistical analysis was performed using RStudio.

Results. Intraocular pressure decreased from 23.0 mmHg (19.3–27.5) to 16.0 mmHg (14.0–18.8) (p=0.0080) one month after the procedure. A decrease in ganglion cell count (GCC) was detected from 65.5 mkm (58.5–69.8) to 63.0 mkm (55.3–68.8) (p=0.0325). Evaluating retinal nerve fiber thickness (RNFL) an increase was detected from 62.0 mkm (60.3–71.5) to 68.5 mkm (64.3–73.5) (p=0.0173). When analysing the trabecular iris angle and ciliotrabecular angle an increase was detected from 50.3° (32.5–56.3) and 93.0° (78.5–101.8) to 59.9° (55.0–75.2) and 103.0° (84.0–111.8) accordingly (p=0.0371; p=0.0488, respectively). Foveolar avascular zone decreased one month after the procedure from 0.18 mm^2 (0.14–0.21) to 0.11 mm^2 (0.09–0.15) (p=0.0019). A decrease in optic nerve head perfusion was detected from 40.2% (36.6–41.8) to 39.2% (35.7–42.5) (p=0.0365). A moderate negative correlation was detected between the change in the trabecular iris angle and the foveolar avascular zone one month after the procedure (–0.548). A moderate negative correlation was detected between the change in the ciliary body area and optic nerve disc perfusion (–0.630). A moderate correlation was detected between optic nerve disc flow index and angle opening distance (–0.555), angle recession zone (–0.652), the scleral spur to iris root insertion (0.664), and ciliary body area (0.568).

Conclusion: Micropulse cyclophotocoagulation is an effective procedure to lower IOP. Detected results suggests that micropulse cyclophotocoagulation has beneficiary effects on ocular vascular flow by affecting part of choroidal tissues. Further research is to be conducted.

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Changes in corneal thickness and endothelial cells after cataract surgery with the phacoemulsification method in patients with II type of diabetes

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Background. One of the most common comorbidities in people with cataracts is diabetes. Many foreign studies have shown that the corneal endothelium in diabetic patients is more prone to damage after cataract surgery with the phacoemulsification method. The diabetic cornea suffers from an impaired repair mechanism after surgical injury.

Aim. To find out whether diabetic patients are at higher risk of postoperative corneal decompensation. It examines epidemiological data in Latvia and the world, aetiology, risk factors, prognostic indicators and secondary prevention. Study tasks were:

- To measure the main parameters of corneal structure before and after cataract surgery.
- To determine whether parameters change in a group of healthy patients after cataract surgery, as well as how significantly they change after cataract surgery in diabetic patients.
- Compare the differences between the two groups.
- To evaluate the possibilities of prevention in diabetic patients to reduce postoperative complications.

Methods. 40 patients were analysed: 20 were included in the study group, 20 in the control group. The study and control groups included those patients who underwent cataract surgery with the phacoemulsification in Ophthalmology Clinic in 2022. The study was prospective, some corneal parameters were analysed before and after cataract operation.

Results. Data analysis showed that corneal structural and morphological changes are more important in diabetic patients than in the control group. Corneal thickness increased by 11.6% in the control group and by 21.65% in the first week after surgery in the diabetic group, which is a statistically significant result with $p<0.005$. There was a significant reduction in endothelium hexagonality in some similar studies as well as in our study. It turned out that for the diabetic group the percentage of hexagonality has fallen more markedly than for the control group ($p<0.005$). However, the results of this study did not show statistically significant difference in the loss of endothelial cells between two groups. Presumably, the patients who were in the diabetic group were less decompensated and certainly had better control of their disease. However, the initial endothelium cells count in diabetic was lower than in the control group (2420 vs. 2521 cells/mm^2).

Conclusion. Diabetic patients are more predisposed to postoperative corneal complications. This means that the surgeon should be more cautious during the operation.

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Changes in peripapillary microvascular density and visual field in patients with optic disc drusen

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Background. Optic disc drusen (ODD) are yellowish, semitranslucent, lobulated formations in the optic nerve papilla, occurring in up to 2.4% of the population. It is known that they cause visual field (VF) defects and sometimes even total vision loss in case of complications. Advances in non-invasive ophthalmic imaging have revolutionized the clinical diagnosis of ODD, and peripapillary vascular density (pVD) may be the most easily obtainable biomarker to predict which eyes with ODD will develop VF loss.

Aim. To investigate the effect of ODD on functional and structural parameters of the eye and to compare these results with age-matched control group patients.

Methods. The study included 19 patients (33 eyes) with ODD and 18 age-matched controls (33 eyes). The patients were further divided into three groups: the first group contained patients with ODD who had VF defects (22 eyes), the second group contained patients with ODD who did not have VF defects (11 eyes) and the third group, which contained age-matched controls (33 eyes). All study subjects underwent an automated static perimetry examination, optical coherence tomography (OCT) imaging of optic disc and macula, as well as OCT angiography (OCTA) imaging of optic disc.

Results. Eyes with ODD had significantly reduced pVD (46.0% [40.2–47.6]) compared to controls (49.7% [48.4–51.7]) ($p<0.001$). Eyes with ODD, regardless of VF loss, had significantly reduced peripapillary retinal nerve fiber layer (RNFL) thickness and pVD compared to controls. Macular ganglion cell complex (GCC) thickness analysis showed significant thinning in eyes with ODD who had VF loss (90.5 μm [84.3–97.5]) compared to controls (102.0 μm [97.0–108.0]) ($p<0.001$). There was a significant positive correlation between both peripapillary RNFL and GCC thickness and pVD ($r=0.60$, $p<0.001$ and $r=0.57$, $p<0.001$, respectively). A significant, negative correlation was observed between pVD and pattern standard deviation (PSD) ($r=-0.47$, $p=0.006$). The positive correlation between mean deviation (MD) and pVD was significant ($r=0.39$, $p=0.027$).

Conclusion. In this study we demonstrated that OCTA can be used as an objective method in the analysis of pVD changes in patients with ODD, which correlates with a decrease in GCC and RNFL thickness. OCTA mean pVD reduction in eyes with ODD can be used as a marker of structural axonal damage that significantly correlates with functional VF loss.

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NURSING

Psychosocial stress in nurses and its impact on health satisfaction

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Background. Psychosocial stress is defined as a mismatch between the work environment and the individual characteristics of employees. Psychosocial stressors at work make it difficult for nurses to perform their duties properly and provide quality care. In the long term, this translates into reduced work motivation, poorer physical and emotional health.

Aim. The aim of the current study was to investigate the psychosocial stress experienced by nurses and its impact on health.

Methods. The survey was done in Vilnius level II and III hospitals and online. The respondents were 314 general nurses. The research method chosen for the study was a quantitative research method using an anonymous questionnaire survey.

Results. The assessment of respondents' psychosocial factors at work showed that the highest average scores were given to work role, support from colleagues and relationships. Job demands, job control, changes, and support from the manager scored lower. It can be stated that job demands, and job control are the most stressful factors at work. Nurses mostly agree that they must work very hard or very fast. According to data when work is emotionally stressful, nurses do not feel supported by their manager and colleagues are reluctant to listen to their work-related problems.

In terms of nurses' subjective health, it was found that one third each rated their health as good (38.2%), 18.2% rated it as very good, while the remaining 6.4% rated it as excellent, and 5.4% rated it as bad. Only 8.9% of the nurses were completely satisfied with their current health, 45.2% were satisfied, while 11.8% were dissatisfied, and 1.6% were not satisfied at all with their health. Regarding physical health, 38.2% feel physically tired. Similar findings were obtained for nurses' emotional health, with 37.3% feeling emotionally tired. Nurses suffer from a wide range of health problems: most commonly musculoskeletal (48.7%), digestive (43%) and cardiovascular (39%).

Conclusion. The main psychosocial factors causing stress in nurses' work are related to job demands, job control and relationships in the work environment. Better income at work is associated with better physical and emotional health. Psychosocial factors in the work environment are directly related to nurses' levels of anxiety and depression and physical and emotional health.

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Seasonal variation of health-related quality of life in patients with coronary artery disease

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Background. Comparing data from different years and the time of year during which data are collected can affect respondents' answers or health status (Jia 2009). Although numerous studies have examined health-related quality of life (HRQoL), until now little is known about the impact of seasonality on health-related quality of life (Jia 2009, Ylivuori 2021).

Aim. The aim of the current study was to investigate the possible impact of seasonality in HRQoL assessment in patients with coronary artery disease (CAD) during cardiac rehabilitation (CR).

Methods. This cross-sectional study included 1026 individuals with CAD (mean age 57±9 years, 217 women, 21.2%) attending CR within two weeks of receiving treatment for the acute coronary syndrome. Within three days of admission for a CR program, all participants were assessed for socio-demographic and clinical (including New York Heart Association (NYHA) functional class) characteristic, medication use and CAD risk factors. During the same time patients independently completed 36-Item Short Form Medical Outcome Questionnaire (SF-36) to assess HRQoL and questionnaires assessing. Type D personality (DS14 scale), mental distress symptoms as measured by the Hospital Anxiety and Depression scale (HADS). All the participants taking benzodiazepines were not included in the analysis. One way analysis of variance (ANOVA) was used to compare means across the seasons. POST-HOC test was used for multiple comparisons. Multivariable linear regression was used to determine the possible impact of seasonality in assessment of HRQoL.

Results. We found significant differences between seasons and the mean score of SF-36 domains. The mental health ($p=0.007$) and general health perception ($p=0.015$) were better in summer compared to spring. Meanwhile, the role limitation due to pain was lower in winter compared to spring ($p=0.034$) and summer ($p=0.028$). Results of the multivariate linear regression after considering all measured variables: sex, age, NYHA, Type D personality, mental distress symptoms and seasons confirmed that the summer season ($\beta=0.059$, $p=0.02$) was associated with a better assessment of mental health and the winter season was associated with a better assessment of pain domain ($\beta=0.073$, $p=0.02$), i.e., lower pain intensity and fewer pain-related limitations in daily activities.

Conclusion. Our results confirm possible impact of seasonality in HRQoL assessment in individuals with coronary artery disease during cardiac rehabilitation.

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Changes in patients' well-being after spine surgery

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Background. Damage to the spine affects vital activities: the patient's activity decreases, the treatment period increases, soft tissue injuries become more frequent, pain limits daily activities and independence.

Aim. To assess the well-being of patients after spinal surgery.

Methods. The study was conducted in one of Vilnius University hospitals in 2021–2022. Used for: pain –Visual Analog Scales (VAS for Pain, Likert test, in which well-being is recorded with statements from "very good" to "very bad", to assess vital functions – Roper-Logan-Tierney: A Model for Nursing Based on Model of Living. Patients after elective spine surgery were studied. A total of 121 respondents participated in the study, 38.8% of females and 61.2% of males. The age of the patients ranged from 18 to 70 years and older.

Results. Frequency of traumas: 30.6% (n=37) of patients did physical (dynamic, mixed), 37.2% (n=45) physical (static, sedentary), 32.2% (n=39) mixed work. After the operation: 65.3% (n=79) patients sometimes felt severe pain when making small movements, 71.9% of patients had urinary problems, 9.1% complained of fever and weakness, 18.2% had changes in blood pressure. 7.4% of the subjects sometimes felt breathing difficulties, 14.9% could perform usual hygiene factors independently, 8.3% needed help of the staff. After the operation, 51.2% of the subjects began to move on the same day using assistive devices, 33.1% – after two days. Due to back pain, 16.5% of the subjects require staff assistance. 73.6% indicated fatigue and the need for longer rest, 10.7% of patients wanted to rest lying down. After the operation, 25.6% felt weak (1–3 points) pain, 69.4% moderate (4–5 points) pain, 5.0% of the patients felt great (6–8 points) pain. More than a third of patients, 37.2%, assessed their well-being as "very good", 33.1% assessed it as satisfactory, and one fifth, 16.5%, assessed it as "bad".

Conclusion. Causes of feeling satisfactorily or badly: pain in the operated area at the slightest movement, weakness, rapid fatigue, urinary problems, loss of independence. The staff's help, stabilization changes in blood pressure, absence of fever, slight pain contributed to good and very good well-being.

Acknowledgments. No conflicts of interest to declare.

Tuberculosis patients' knowledge and attitude toward the disease and its preventive measures

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Background. The morbidity of tuberculosis (TB) in Lithuania is more than four times greater than in other European countries. The application of preventive measures in hospitals is one of the most significant ways to ensure a safe environment for both staff and patients.

Aim. To reveal TB patients' knowledge and attitude toward the disease and its preventive measures.

Methods. In 2021, a study was conducted in one of Vilnius University hospitals. 40 patients undergoing treatment for resistant TB in the hospital participated in the study. The questionnaire survey consisted of the following groups of questions: causative agents of TB, symptoms, sources of infection, medications, preventive measures.

Results. 67.5% of the subjects correctly assessed *Mycobacterium* as the causative agent of TB. Subjects aged 46 to 55 and over 65 had opposing views and indicated that the virus is the main cause of the disease ($p<0.05$). Respondents aged 26–45 correctly noted that effective second-line TB polydrugs are prescribed in the treatment of multidrug-resistant tuberculosis. 47.5% of the respondents assigned all coughing persons as the source of infection but patients re-treated for TB viewed the source of infection differently and claimed that a patient who had an open form of TB is more dangerous ($p<0.05$). 52.5% of the subjects, aged 56–65 and over 65, pointed out incorrectly that patients with diabetes mellitus, high blood pressure and obesity are less likely to become infected with TB. One third, 32.5% of the subjects, indicated the following as the greatest risk of contracting TB: alcohol and drug abuse and improper diet, while 17.5% of the subjects aged 46 to 65 prioritized immunity, personal hygiene, contact with TB and HIV-infected people, half of the participants marked all the risk factors correctly ($p<0.05$). The majority (72.5%) of respondents who have been getting a longer treatment for TB disease, considerably more valued such preventive measures as respirators, hand and surface disinfectants; respondents with shorter illness experience preferred wearing medical masks, coughing and hand hygiene, personal hygiene, changing bed linen ($p<0.05$).

Conclusion. Subjects with longer treatment experience for TB can evaluate the causative agents, sources and risk of infection, preventive measures of TB better. Subjects with shorter duration of treatment and younger age groups have more extensive knowledge about the causative agents of TB and their resistance to medication.

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Poster presentations

BASIC MEDICAL SCIENCE AND PHARMACY

Green biosynthesis of silver nanoparticles by using fermented aqua extract of *Matricaria chamomilla* and *Calendula officinalis*

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Background. The expansion of multidrug-resistant (MDR) pathogens becomes a global health concern of this century. Indeed, the availability of MDR pathogens significantly endangers both the health of animals and consumers due to the potential risk of entering the food chain causing severe poisoning.

Green synthesis among other methods offers simplicity in operational and process conditions as its uses exclusively plant-derived metabolites and metallic salts while omitting the use of toxic reagents, catalysts, and solvents. The key point in the successful synthesis of nanoparticles, however, is the availability of functional groups in bioreductants such as carboxylic, hydroxyl, and carbonyl groups that are reported to be involved in the bioreduction of Ag ions to NP along with stabilizing properties.

Aim. The aim of this study was to expand the boundaries on AgNPs using a novel low toxicity and green approach to the biosynthesis of metallic NPs using *Matricaria chamomilla* (*M. chamomilla*) and *Calendula officinalis* (*C. officinalis*) fermented aqueous extracts.

Methods. The formation of AgNPs was confirmed by transmission electron microscopy (TEM) scanning electron microscopy and energy dispersive spectroscopy techniques. The effectiveness of biosynthesized AgNPs in quenching free radicals and inhibiting the growth of gram-positive and gram-negative microorganisms was supported by *in vitro* antioxidant activity assay methods and using the Kirby–Bauer disk diffusion susceptibility test, respectively.

Results. The antimicrobial effect of engineered AgNPs against selected test cultures was found to be substantially stronger than for plant extracts used for their synthesis. The analysis of AgNPs by TEM revealed the presence of spherical-shaped nano-objects. The size distribution of AgNPs was found to be plant-type-dependent.

Conclusion. The dispersed AgNPs were biosynthesized by the green, sustainable, and eco-friendly method using fermented medical plant extracts of *M. chamomilla* and *C. officinalis* as a capping and reducing agent. The AgNPs presumably inherited biological functions of *H. officinalis* and *C. officinalis* medical plants can provide a platform to combat pathogenic bacteria in the era of multi-drug resistance.

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Total phenolic content of *Potentilla anserina* (L.) Rydb. grown in urban and rural habitats

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Background. *Potentilla anserina* (L.) Rydb. is a low-growing herbaceous plant in the *Rosaceae* family and is prevalent in various types of habitats such as meadows and road-sides throughout Europe, Siberia, the Far East and Central Asia [1]. Tannins and flavonoids are compounds of interest with notable pharmacological activities [2]. The phytochemical content strongly depends on individual ontogenesis and the environmental growing conditions. Urban ecosystems differ from natural biocenoses and their peculiarities can significantly impact the profiles of bioactive compounds.

Aim. To evaluate the amounts of phenolic compounds of *P. anserina* both in urban and rural areas during different vegetation periods.

Methods. Aerial parts of *P. anserina* every two weeks in the months of June, July and August were collected from the Northern part of Lithuania in urban and rural areas. The total phenolic content was determined according to Folin–Ciocalteu method [3].

Results. The total phenolic content was in a range of 82.11±2.21 GAE mg/g – 112.51±4.72 GAE mg/g and 83.57±0.70 GAE mg/g – 108.74±0.53 GAE mg/g in urban and rural areas, respectively. The results have shown that the greatest amount of phenolic content was determined in during the massive flowering period in the samples collected from both habitat types. At the end of the vegetation period, in August, the total amounts of phenolics significantly decreased to 82.11±2.21 GAE mg/g.

Conclusion. *P. anserina* from urban habitats contained higher amounts of phenolic compounds. The species due to extensive distribution and adaptation could be a sustainable source of phenolic compounds for industrial applications

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Analysis of phenolic composition and antioxidant activity of raspberry (*Rubus idaeus* L.) stem powders

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Background. Stems of red raspberry (*Rubus idaeus* L.) are used in Lithuanian folk medicine for a variety of ailments [1]. The pharmacological activities are mainly attributed to the presence of polyphenolic compounds and variational peculiarities and determine the optimal time for harvesting.

Aim. The aim of this study is to determine polyphenolic content in red raspberry stems during divergent phenological phases.

Methods. The samples of raspberry stems were collected in 2021 in natural habitat at Sviliai forest, Lithuania. The extracts were made by ultrasonic extraction method. The phenolic compounds were determined using Folin-Ciocalteu. Antioxidant activity was tested by FRAP [2].

Results. The yield of total amounts of phenolic compounds were in a range of 24.36–42.80 mg/g of GAE. The significantly greatest amounts of phenolics were obtained during start of fruit development stage 42.80±16.80 mg/g and later during senescence of leaves from 36.17±7.28 mg/g, to 41.91±7.73 mg/g, respectively. All investigated raspberry stems showed reducing activity ranging from 731.93±80.16 µM TE/g to 1098.8±317.12 µM TE/g. Samples that contained the highest quantity of polyphenolic compounds showed correspondingly higher activity.

Conclusion. Raspberry stem powders are polyphenolic rich materials with notable antioxidant activity. The optimal time for collecting herbal material is during the fruit development phase or later from the senescence of leaves.

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The chemosensitizing impact of combination of DOX and apple extract on HT-29 and U-87 cells in 2D and 3D cultures

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Background. Cancer is one of the predominant reasons for mortality worldwide. Based on WHO data in 2020 were determined 18.1 million new cancer cases, among them colon cancer representing 10.7% of all cases, and brain and central nervous system cancer, which included 1.7% of all cases. Cancer cells' resistance to drugs represents a major clinical problem and the most relevant failure of cure. Combination chemotherapy is more effective than monotherapy due to additive or synergistic effects. A diet rich in biologically active compounds containing foods could be used as the first line of prevention, or as a component of traditional chemotherapy.

Aim. The aim of our study was to assess the effects of the combination of apple extract and doxorubicin on human colon adenocarcinoma (HT-29) and glioblastoma (U-87) cell lines in 2D and 3D cultures.

Methods. This study involved the apple cultivars 'Auksis', 'Kostele', 'Ligol', and 'Rubin'. The effect of the combination of apple extracts and doxorubicin (DOX) against HT-29 and U-87 cell viability was tested by the MTT assay [1]. Spheroids were formed by a magnetic 3D bioprinting method [2]. The change of EC_{50} (expressed in %) was found, which indicates how much DOX toxicity changed when the apple extracts were added to the medium.

Results. All apple extracts sensitized the cell line HT-29 to DOX in a similar manner (change of EC_{50} varied from –43.2%±2.2% to –53.0%±1.4%). Cell line U-87 became even less sensitive to DOX when treated with all apple extracts (change of EC_{50} varied from 7.2%±0.4% to 34.2%±1.7%). HT-29 spheroids' size was most reduced by the combination of apple peel extract of 'Auksis' cultivar and DOX (spheroids diameter reduced from –32.9% to –11.8%). The combination of apple peel extract of 'Auksis' cultivar and DOX reduced the viability of HT-29 spheroids the most (spheroid viability reduced from –19.9% to –10.9%). From all tested apple extracts, of 'Rubin' and of 'Kostele' cultivars combined with DOX slightly reduced the viability of U-87 spheroids.

Conclusion. The HT-29 and U-87 cells were sensitive to DOX when treated with apple extracts in 2D and 3D cultures. We found that the HT-29 cell line was most strongly sensitive to DOX when treated with all tested apple extracts, compared to U-87 cells.

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Changes in anthocyanins composition in cranberry (*Vaccinium macrocarpon* Aiton) raw material during fruit ripening period

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Background. Cranberries are an important source of anthocyanins, which have antioxidant, anti-inflammatory effects and favorably modulate the gut microbiota. Fruits of cranberry are used in the production of food supplements, which highlights the importance of qualitative and quantitative analysis of anthocyanins in cranberry fruit raw material. The composition of anthocyanins in cranberry fruits depends on the effects of genetic factors and climatic conditions. The quantity of anthocyanins depends on the stage of cranberry ripening too. Therefore, the study of the composition of anthocyanins during the ripening of cranberries is important and allows determining the optimal time of fruit harvesting.

Aim. The aim of the current study was to determine qualitative and quantitative composition variability of anthocyanins in cranberry samples grown in Lithuania during the ripening period.

Methods. We studied the cranberry sample of the 'Stevens' cultivar during the ripening period. The samples of cranberry were lyophilized, and 1 g (exact weight) of lyophilized cranberry powder was extracted with 20 mL of 70% (v/v) ethanol containing 1% hydrochloric acid in an ultrasonic bath for 15 min at room temperature. Anthocyanins analysis was performed by ultra-high performance liquid chromatography method.

Results. Analysis of the composition of anthocyanins in cranberry samples grown in Lithuanian during the ripening period were performed. Delphinidin-3-galactoside, cyanidin-3-galactoside, cyanidin-3-glucoside, cyanidin-3-arabinoside, peonidin-3-galactoside, peonidin-3-glucoside, malvidin-3-galactoside, peonidin-3-arabinoside, cyanidin, malvidin-3-arabinoside, peonidin, and malvidin were determined. The highest total amount of anthocyanins (3710.58±91 μg/g DW) was detected at the end of ripening period (the 295th day of the year), the lowest amount (38.12±2 μg/g DW) was determined at the beginning of ripening period (the 224th day of the year). Two anthocyanins have been identified at the beginning of cranberry fruit ripening: cyanidin-3-galactoside 67% and cyanidin-3-arabinoside 33%. Four anthocyanins predominated in cranberry fruit samples during cranberry fruit ripening period from the 240th day to the 295th day of the year: cyanidin-3-galactoside (24.62%–33.38%), cyanidin-3-arabinoside (24.50%–31.57%), peonidin-3-galactoside (16.74%–29.28%), and peonidin-3-arabinoside (8.40%–15.41%).

Conclusion. Cranberry raw material prepared at the end of the ripening period have high levels of anthocyanins. Cranberry raw material prepared at the end of September or beginning of October is suitable for producing high-quality food products and food supplements with a determined composition of anthocyanins.

Acknowledgements. The authors declare absence of conflict of interest.

N-((4-sulfamoylphenyl)carbamothioyl) amides: potential neuropathic pain attenuating agents

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Background. Neuropathic pain refers to pain that develops when the central or peripheral nervous system is damaged or not working properly due to diseases or injuries, which it is more difficult and complex to manage than many other types of chronic pain. This pain affects about 10% of the general population and can lead to a substantial reduction in the quality of life. Unfortunately, there are no medicines or other therapeutics presently approved to treat this pain. Although, the mechanism underlying this pain still is not completely known, isoform selective inhibitors of brain-associated human carbonic anhydrase, hCA VII, are recently demonstrated neuropathic pain reducing properties by animal models of the disease. Needless to say, that because of its strong zinc-binding affinity, sulfonamide moiety ($R\text{-}SO_2NH_2$) is the most appropriate "warhead" to target CAs and as a matter of course sulfonamide-containing compounds are the main class of carbonic anhydrase inhibitors (CAIs).

Aim. The goal of this study was to synthesis a series of novel *N*-((4-sulfamoylphenyl)carbamothioyl) amides and evaluate their inhibitory potency against the brain-associated hCA VII.

Methods. An applied photophysics stopped-flow instrument has been used for assaying the CA-catalysed CO_2 hydration activity. Phenol red was used as indicator, working at the absorbance maximum of 557 nm, with 20 mM Hepes (pH 7.5) as buffer, following the initial rates of the CA-catalysed CO_2 hydration reaction for a period of 10–100 s.

Results. A panel of twelve *N*-((4-sulfamoylphenyl)carbamothioyl) amides was designed, synthesized, and their inhibitory effects were explored against the neuropathic pain relevant CA VII isoform (as the target isoform) and two most ubiquitous cytosolic hCA isoforms I and II as the off-target isoforms. Interestingly, some of the newly developed compounds displayed better inhibitory potency against the target isoform (K_I<1.1 nM) compared to the standard drug acetazolamide (K_I of 2.5 nM).

Conclusion. Some of the newly developed sulfonamides exhibited better inhibitory activity against the brain-associated hCA VII than the reference drug. These results make the compounds of interest for investigations *in vivo* as potential neuropathic pain attenuating agents.

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Comparative analytical profiling of bioactive constituents in *Vaccinium vitis-idaea* L. cultivars

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Background. Fruits and leaves of lingonberry (*Vaccinium vitis-idaea* L.) are valuable raw materials for food and pharmaceutical industries, gaining notoriety as 'superfoods'. However, raw materials from natural habitats are characterized by the chemical heterogeneity of secondary metabolites. Cultivated plant materials are preferred for industrial purposes as it is easier to follow the quality standards and ensure the homogeneity of batches. The identification of medicinally relevant substances in specific cultivars increases their commercial value and perspectives in the production of added-value products and functional or pharmaceutical food. The quality of the plant raw material, especially the phytochemical reproducibility, is crucial for the pharmacological efficacy of the extract.

Aim. The aim of the present study was to determine and comparatively assess the phenolic and triterpenic compound profiles in lingonberry cultivars and intraspecific taxa, and to elucidate the added-value cultivars for commercial cultivation and the targeted application.

Methods. Ten lingonberry cultivars ('Erntedank', 'Erntekrone', 'Erntesegen', 'Koralle', 'Masovia', 'Sanna', 'Sussi', 'Kostromskaja rozovaja', 'Kostromička', 'Rubin') and two intraspecific taxa (*V. vitis-idaea* subsp. *minus* and *V. vitis-idaea* var. *leucocarpum*) were included in the study. Extracts from cultivated lingonberry leaves and fruits were analysed using validated UPLC/HPLC-PDA techniques.

Results. Fifty-nine constituents in total, belonging to subgroups of phenolics and triterpenoids, were determined in extracts of lingonberry leaves and fruits. Findings showed that phytochemical profiles were significantly different among tested lingonberry cultivars and lower taxa based on the levels of individual components and their distribution throughout lingonberry parts. In general, cultivated leaves were richer in the content of most phenolics, while fruits stood out with their higher levels of triterpenoids. Compositional differences and phytochemical markers were found to be related to lingonberry origin location, suggesting the strong impact of genotype and heritability on metabolite expression. Targeted application of tested lingonberry cultivars and intraspecific taxa could be proposed on account of different predominant phenolics and triterpenoids determined. In line with quantitative analysis results and profound richness of identified compounds, the superiority of lingonberry cultivars 'Kostromička', 'Kostromskaja rozovaja', 'Rubin', and *V. vitis-idaea* subsp. *minus* is suggested, supporting their selection for breeding programs, agricultural production, and application in the food and pharmaceutical industries.

Conclusion. This work contributes genetic variation data and provides important insights into the phytochemical properties of lingonberry cultivars and intraspecific taxa, which can be useful for improving and selecting lingonberry genotypes for commercial cultivation and targeted application.

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Antibiotic consumption in the paediatric hospital in Latvia

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Background. Growing resistance to antibiotics is one of major threats to public health. Compared with other countries the prevalence of antimicrobial resistance in Latvia in general is still low however; antibiotic consumption in hospitals is one highest around the Europe. Therefore, antibiotic usage analysis is important step in improvement of prescription practices in hospitals.

Aim. To investigate current consumption patterns of systemic antibiotic usage in the tertiary-care paediatric hospital in order to improve prescribing practices.

Methods. This study was conducted by using several methods: retrospective and observational study of patients' electronic records in 2021, the World Health Organization standardized methodology and AWaRe classification and the defined daily dose method. Data were extracted from patients' electronic records for the period of 1/01–31/12/2021 and were entered into the Excel form for analysis. Inclusion criteria: hospitalized patients less than 18 years old who had prescribed antibiotics. The collected data included patients' demographic information, i.e., age and gender, antibiotics prescribed to the patient (ATC classes J01 and P01AB), single dose and doses per day, route of administration, diagnosis, and empirical or targeted prescription. Exclusion criteria: day-centre patients and emergency patients.

Results. In 2021 there were 13578 patients in the hospital and 4395 (32.4%) patients treated with antimicrobials 2443 (55.6%) males and 1952 (44.4%) females). Number of beds was 272 and average bed occupancy 78.6%. In total there were 9007 antibiotic prescriptions. Thirty-five different antibiotics were used in 2021. The most commonly used antibiotic class was 2nd generation cephalosporins – 2148 (23.9%) prescriptions, which accounted for 93.7 DDD/100BD. The most prescribed antibiotic was cefuroxime (2148; 23.9%) prescriptions. There were 7388/9007 (82%) parenteral prescriptions, 1605 (17.8%) oral prescriptions and 16 (0.2%) prescriptions for inhalations during the 2021. Antibiotics were prescribed empirically in 2800/4263 (65.7%) cases. Eighteen (51.4%) antibiotics belonged to the Access group, 14 (40%) to the Watch group and 3(8.6%) to the Reserve group.

Conclusion. The study identified some positive aspects in antibiotic usage, e.g., the most prescribed antibiotic groups were 2nd generation cephalosporins and penicillins with extended spectrum, cefazolin used for surgical prophylaxis. However, there are still problem areas for improvement, i.e., predominant use of parenteral antibiotics and empirical treatment. Several methods should be used in hospitals to get information about antibiotic consumption and prescribing tendencies.

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Variation of total phenolic compounds of different plant organs from *Artemisia campestris* L. herb

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Background. *Artemisia campestris* is a perennial plant that grows throughout North Europe, Asia, North America, and North Africa. Due to a lack of study on this species in Lithuania and a lack of studies in Europe, it is relevant to quantify the amount of phenolic compounds in its various plant organs. High levels of phenolic compounds in human nutrition reduce the risk of chronic diseases because they have antioxidant properties and thus protect cells during oxidative stress by neutralizing the harmful effects of free radicals.

Aim. To determine the amount of total phenolic compounds in extracts of *Artemisia campestris* L. collected form natural habitats in Lithuania.

Methods. *A. campestris* samples were collected at the stage of flowering in the natural habitats from different regions in Lithuania. Plant material (0.2 g) was extracted with 20 ml 70% ethanol in an ultrasonic bath for 15 min and centrifuged at 6500 rpm for 15 min, then filtrated though filter paper and stored in the brown bottle to avoid the light. Total phenolic content was determined by the Folin–Ciocalteu method using gallic acid as a standard. The test sample was prepared by mixing 20 μL of extract with 5 ml of Folin–Ciocalteu working reagent and 4 ml of sodium carbonate (7.5%). After one hour absorbance was measured at 765 nm. Through the calibration curve with gallic acid, the total phenolic content of plant parts was calculated as gallic acid equivalents/g of dry material.

Results. The total phenolic content of different plant organs varies from 34.596±6.798 mg GRE/g up to 88.787±7.515 mg GRE/g. The greatest amount of phenolic compounds was determined in stems (88.787±7.515 mg GRE/g), less in leaves (83.787±17.852 mg GRE/g) and flowers (72.366±8.000 mg GRE/g). The roots contained the lowest amounts compared to other plant organs, 34.596±6.798 mg GRE/g.

Conclusion. Total phenolic content varies significantly among the different plant parts of *A. campestris*. The aboveground parts could be further used for targeted extraction.

The effect of oregano (*Origanum onites* L.) essential oil on glutathione and malondialdehyde concentrations in mice blood

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Background. In Turkey, *Origanum onites* L. is primarily found in the south and west regions. The essential oil extracted from this plant, whose primary constituents are carvacrol and thymol, has been associated with several biological activities, including antifungal, antibacterial, antioxidant, insecticidal, hepatoprotective, and cytotoxic action. The bioavailability and absorption of herbal medicines are lowered by problems like poor solubility. Since liposome formulations can boost the stability, solubility, and bioavailability of bioactive components found in essential oils, *Origanum onites* L. essential oil antioxidant and other capabilities can be enhanced.

Aim. The purpose of the subsequent investigation was to determine if liposomes containing *Origanum onites* L. essential oil had any potential antioxidant effects on glutathione (GSH) and malondialdehyde (MDA) concentrations in the blood of BALB/c laboratory mice following aluminium exposure.

Methods. BALB/c laboratory mice were used for the investigations. Liposomes containing *Origanum onites* L. essential oil were administrated for 21 consecutive days. Oxidative stress was induced by $AlCl_3$ solution. The concentration of GSH and MDA in the blood of mice was evaluated spectrophotometrically. The study was approved by ethics committee of Lithuanian University of Health Sciences.

Results. Results showed that aluminium statistically significantly decreased GSH concentrations and statistically significantly increased MDA concentrations in the blood of mice compared to the control group. Findings demonstrate that compared to the aluminium-treated group, liposomes containing *Origanum onites* L. essential oil statistically significantly increased GSH concentration in mice blood exposed to aluminium. Mice who received *Origanum onites* L. essential oil-containing liposomes after being exposed to aluminium did not differ statistically from mice that received solely *Origanum Onites* L. essential oil-containing liposomes. Meanwhile, MDA concentrations in blood showed controversial results. Liposomes containing *Origanum onites* L. essential oil statistically significantly increased MDA concentration in mice blood compared to the control group. Results also showed that administration of *Origanum onites* L. essential oil-containing liposomes after aluminium exposure statistically significantly increased MDA concentration in mice blood compared to the aluminium-treated group.

Conclusion. Administration of liposomes containing *Origanum onites* L. essential oil can increase GSH concentration in mice blood and the conclusion was made that liposomes containing *Origanum onites* L. essential oil may have an antioxidant effect. Further research is required to ascertain the reasons behind the elevated MDA concentrations caused by liposomes containing *Origanum onites* L. essential oil.

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The influence of β-cyclodextrin on the parameters of emulsion and sodium alginate microcapsules

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Background. Cyclodextrins have a hydrophobic cavity and form an inclusion complex with a hydrophobic guest molecule, which leads to their use in the pharmaceutical field. One of the uses can be an emulsifier in o/w emulsions. The emulsions that cyclodextrins create are Pickering emulsions. Emulsions can be used in microcapsules production, a pharmaceutical form that can be applied to protect compounds such as volatile compounds (essential oil, extracts, oils with additional vitamins etc.).

Aim. The aim of this research was to determine the influence of β-cyclodextrin on microcapsules emulsion stability, particle size and capsule hardness.

Methods. The primary solution was prepared with sodium alginate, aqueous red clover extract, β-cyclodextrin, almond oil and thickener. The emulsion was prepared using unguator Q. The stability was studied using a centrifuge (3000 and 7000 rpm), and dispersion was determined using MasterSizer3000.

Microcapsules were prepared by using a medical syringe and calcium chloride solution (3%) as a crosslinker. The product was stirred, filtered, and washed with water. The diameter was measured instantly after the preparation and after one day using a micrometre. The press force was measured with Texture Analyser (TA.XT.plus). The results are presented as mean±SD, results were significant when $p<0.05$.

Results. In this research, only different CD concentrations (from 0% to 17.6%) varied in emulsions formulation. Emulsions were stable when the minimum concentration of cyclodextrin was 12.5%. It was determined that particle sizes of oil did not have a significant difference between them. Its concentration had an influence on microcapsules' size, using higher cyclodextrin amount sizes decreased significantly (from lowest to highest concentration of emulsifier): 2.41±0.14 mm and 2.27±0.13 mm. The firmness of microcapsules increased, increasing cyclodextrin concentration.

Conclusion. β-CD concentration had an influence on stable emulsions formation, particle sizes, dried and non-dried microcapsules' diameter, and press force (firmness). The optimal concentration of cyclodextrin in the emulsion was determined to be 9.09%. This emulsion was stable, formed medium firmness microcapsules that were not too hard to crush and could be released in gut medium more efficiently.

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Evaluation of zinc effect on δ-aminolevulinate dehydratase activity and on metallothionein content in brain of nickel treated mice

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Background. The overexposure to nickel due the extensive use of it in modern technology remains a major public health concern. Studies evidence various Ni-induced neurological symptoms that might be caused by its interaction with macromolecules, interference with physiological processes of trace element Zn and generation of free radicals that mediate cellular damage. Redox-stable zinc acts as antioxidant, protects sulfhydryl groups of proteins from oxidation, replaces redox active metals at critical cellular or extracellular sites, and/or induces synthesis of cysteine rich proteins metallothioneins.

Aim. The present study was devoted to evaluating the effect of nickel on the oxidative state of the brain cells of mice and to ascertain, whether it could efficiently protect cells against Ni neurotoxicity. Therefore, responses of two biomarkers, i.e., content of metallothioneins and an activity of δ-aminolevulinate dehydratase (δ-ALAD), were examined.

Methods. 4–6-week-old out-bred white laboratory mice were used in these experiments. There were two models chosen with a different duration of mice exposure to the metals. For the acute single metal exposure, the exposure time was set at 24 hours and mice were injected once. For the acute repeated exposure, mice were i.p. injected for 14 days (once a day). Mice were randomly assigned into 4 groups, first of which received an i.p injections of $NiCl_2$; second group was treated with $ZnSO_4$; mice of third group were i.p. injected with $ZnSO_4$ and after 20 min with $NiCl_2$ solutions; the fourth – control group received i.p. injections of saline.

Results. Single exposure to neither metal had no impact on δ-ALAD activity; however, repeated Ni^{2+} administration suppressed enzymatic activity by 15%. Repeated $ZnSO_4$ administration, did not affect δ-ALAD activity, meanwhile repeated mice pre-treatment with Zn^{2+} before $NiCl_2$ administration returned enzymatic activity back to the control level. Brain metallothionein content significantly (by 30%) increased in animals once treated with Ni^{2+} while remained at the control level after $ZnSO_4$ administration. In mice once administrated with both metals the content of metallothionein was elevated by 30%. 14 days of Ni^{2+} treatment enhanced brain metallothionein content by 36%, Zn^{2+} treatment increased metallothionein content by 121%, while exposure to both metals increased metallothionein concentration by 81%.

Conclusion. Increased metallothionein content potentially protected δ-ALAD activity after single but not repeated exposure to Ni^{2+}. Continuous pre-treatment with Zn^{2+}, either through natural antagonism or through increased metallothionein expression, managed to protect δ-ALAD activity from Ni-induced inhibition.

Protective effect of vitamin C on the prevention of damage to isolated porcine kidneys stored in simple hypothermia

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Background. One of the challenges of modern transplantation is to develop a preservation fluid with high efficacy. Our team is intensively researching the development of an optimal fluid composition that will provide effective organ protection against ischemia-reperfusion damage. The selection of an appropriate antioxidant is significant. Vitamin C is a potent extracellular and intracellular antioxidant. It maintains the redox balance of the cell and protects against lipid peroxidation and the development of inflammation in organs. It counteracts oxidative stress in mitochondria.

Aim. The aim of this study was to analyze the effect of ascorbic acid as a component of preservative fluid on the storage efficiency of isolated pig kidneys.

Methods. Ten kidneys of Polish Large White pigs were used in the study. The kidneys were stored by simple hypothermia (4°C). All experimental procedures were carried out under the approval of II Local Ethics Commission for Animal Experiments in Cracow, Poland (No. 1046/2013). The composition of the Biolasol preservative fluid was supplemented with Vitamin C (0.5 mmol/L). The efficacy of the fluid was assessed by analyzing the activity of ALT (alanine aminotransferase), AST (aspartate aminotransferase), LDH (lactate dehydrogenase), and lactate concentration determined in perfusates taken after 2 h and 48 h of graft storage, respectively.

Results. It was found that the determined activities of the indices significantly decreased after 48 h vs. 2 h of kidney storage by respectively: ALT [U/L] 15%, AST [U/L] 12%, lactate [mmol/L] 25%. The results were statistically significant at $p<0.05$.

Conclusion. Ascorbic acid had a protective effect on maintaining cytoskeletal integrity in cells.

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Attractive face – do we love or hate our facial features?

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Background. It is assumed that individuals are more attracted to the self's features. However, there is a scarcity of studies, evaluating the prevalence of facial traits and their attractiveness.

Aim. The main aim is to assess the prevalence of facial features between young Lithuanian adults in relation to the most attractive facial traits.

Methods. The study included 115 females and 115 males, aged 18–40 years. The prevalence and attractiveness of 9 facial features were investigated using online questionnaires: five categories for each trait were generated using *FaceGen ModellerDemo3* program (average feature matched 3rd category). Respondents had to assign their facial feature to a certain category and select the most attractive one. Cross tabulation was performed (*IBM SPSS 23.00 Version*).

Results. Respondents with average traits mostly chose them as the most attractive (females/males, respectively): eyes' size (56/73%, $p<0.01/p<0.01$), protruding cheekbones (65/65%, $p<0.01/p<0.01$), accentuated cheeks (only males – 61%, $p<0.01$), length of nose (only males – 84%, $p<0.01$), nostril width (55/92%, $p<0.01/p<0.01$), lip thickness (54/78%, $p>0.05/p<0.01$); chin height (81/85%, $p=0.01/p<0.01$), width of lower jaw (66/74%, $p>0.05/p<0.01$) and protrusion of lower jaw (86/79%, $p<0.01/p<0.01$). In very similar proportions, respondents with smaller cheekbones, cheeks, nose and lower jaw (only males) chose their size traits as the most attractive, as well as the respondents with more expressed eyes, lips, and lower jaw (only males). Respondents, who chose opposite features as the most attractive: men with more expressed cheekbones and women with more expressed cheeks chose less expressed cheekbones/cheeks, respectively. Also, men with smaller eyes and thinner lips chose bigger eyes and thicker lips as the most attractive.

Conclusion. Most respondents (regardless of feature category) chose their facial features as the most attractive. Men were more likely than women to choose their traits as the most attractive. Also, there was a tendency for men to value less "masculine" traits.

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Changes of the levels of cytokines APRIL and IL-10 in type 1 diabetes mellitus patients with and without diabetic nephropathy

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Background. Diabetic nephropathy (DN) is the most common complication of Type 1 diabetes (T1D). DN is the leading cause of the development of end-stage renal disease and renal replacement therapy. Although DN is not classically associated with inflammation, the role of the immune system in the development of this complication cannot be ignored. The unique ability of B cells to produce antigen-specific antibodies, cytokines and to present antigens makes them an important component of the immune system, but their role in the development of DN is unclear. However, it is known that the viability, proliferation, and ability of B lymphocytes to produce cytokines and antibodies are directly affected by changes in glucose levels in the environment in direct and indirect ways. Changes in the levels of B-lymphocyte-associated cytokines could signal changes in B-lymphocyte function and involvement in the pathogenesis of DN.

Aim. To find out the associations between B-lymphocyte-associated cytokines and the development of type 1 diabetic nephropathy.

Methods. Patients were recruited using the LatDiane database. The study involved 72 people, including 19 individuals without impaired glucose tolerance, 36 patients with T1D without complications, 7 patients with T1D with microalbuminuria and 10 patients with T1D with macroalbuminuria. Participants donated serum and urine. During the visit, anthropometric, demographic, and clinical data were collected. The levels of cytokines (IL-7, IL-10, APRIL, BAFF, TNF-α, IL-6) in the blood serum were detected using Luminex MagPix. Statistical analysis was performed with SPSS Statistics 25.0 program.

Results. It was found that the level median of IL-10 is the highest in patients with T1D with microalbuminuria 0.68 pg/ml (control 0.04 pg/ml; $p=1.66\times10^{-2}$), as well as there is a significant increase in growth in all T1D groups. The level of median of APRIL in T1D patients with macroalbuminuria (1823.44 pg/ml; $p=3.44\times10^{-3}$) is 3 times higher than in healthy individuals (620.08 pg/ml) and shows a statistically significant difference with the other groups. Also, the highest significant correlation is observed between APRIL and Glomerular Filtration Rate, where the correlation is inverse and moderately close, or $\varrho=-0.48$ ($p=1.90\times10^{-5}$).

Conclusion. For the first time, to our knowledge, APRIL has been shown to be associated with diabetic nephropathy, which indicates the need for further study of the role of the immune system, including B-lymphocytes, in the development of diabetic nephropathy.

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Effect of acute mesenteric ischemia on rat lung and kidney mitochondria functionality

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Background. Acute mesenteric ischemia (AMI) is a life-threatening condition that needs rapid action to avoid the damage as partial or complete intestinal necrosis. Accumulating evidence suggests that mitochondria allow lung cells to adjust to changes after the pathological processes in other tissues [1]. In very rare cases AMI can lead to an acute kidney injury, but cellular mechanisms of are still unknown [2].

Aim. To investigate the kidney and lung mitochondria functionality in a rat experimental AMI model.

Methods. Experimental AMI was performed by the occlusion of the superior mesenteric artery and vein in male rats 250–330 g weight (17–18 weeks old, n=5–7 per group). Rats underwent vessels occlusion for 15, 30, 60, 90, 120 min and Sham group underwent laparotomy without mesenteric ischemia. Mitochondrial respiration and reactive oxygen species (ROS) production in the form of hydrogen peroxide were analysed in homogenates of kidney and lung mitochondria with high-resolution fluorespirometry. Experiments were performed in accordance of obtained permission of Ethical Committee.

Results. Superior mesenteric artery and vein occlusion for 15 and 60 min in all respiratory states and for 90 min in Complex I&II OXPHOS state resulted in lower mitochondrial oxygen consumption in lung tissue comparing with Sham group data, whereas in kidney samples statistically significant changes were seen only in 15-min long ischemia period. There were significant differences in kidney oxygen flux and ROS release between 15- and 30-min groups in Complex I OXPHOS state, where 15-min long ischemia resulted in a lower mitochondrial activity. No significant differences in ROS release and oxygen consumption data in lung samples between ischemia periods were seen.

Conclusions. Ischemia in the intestine influence kidney and lung mitochondrial functioning, showing the effect already after 15-min long mesenteric artery occlusion.

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Gliomagenesis associated lncRNAs *LINC00461*, *GAS5* and *NEAT1* are post-transcriptionally m6A modified in gliomas

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Background. Using Oxford Nanopore Technologies platform, direct sequencing of native molecules allowed scientists to thoroughly study the changes in RNA. Most of the conserved sites in the m6A region were located in the *RRACH*. Glioblastoma (GBM) is a highly aggressive disease associated with a poor prognosis despite advances in treatment in recent years. Some studies showed that *LINC00461*, *GAS5* and *NEAT1* long non-coding RNAs are associated with gliomagenesis but information on epitranscriptomics is still lacking.

Aim. The aim of the current study was to evaluate *LINC00461, GAS5* and *NEAT1* m6A modification at gene and pathology level.

Methods. Total RNA was extracted from 24 snap-frozen glioma tumor tissues. Enriched polyA RNA was used for the Nanopore dRNA-seq. Sequencing data were processed using pipeline "*nf-pore/nanoseq*". m6A modification (prediction) was computed from RNA-seq.

Results. First, we identified the *AAACT* motif as the most m6A modified out of 11 *RRACH* motifs in the analyzed genes. Then, we determined m6A differences at *AAACT* motif in lncRNAs between low- and high-grade gliomas which showed slightly higher methylation level in low malignancy grade tumors (average in LGG 65% and GBM 57.7%). The comparison of m6A level between GBM and LGG groups applying chi-square test revealed that individually only in one *AAACT* motif location (3184) in gene *NEAT1* the m6A rate significantly differed between pathology groups ($p<0.05$). Furthermore, there were association between m6A modification in *NEAT1* gene at 3184 motif position and gene expression level in analyzed glioma sample set. Survival analysis at the gene level showed *NEAT1* m6A modification was significantly linked with poor patient survival in gene ($p<0.05$).

Conclusions. Here we showed that lncRNAs *LINC00461, GAS5* and *NEAT1* were m6A modified in gliomas and modification level was gene and pathology dependent. m6A modification in analyzed lncRNAs were most likely related to the development of higher malignancy of glioma and also affected the survival time of glioma patients (genes *GAS5* and *NEAT1*). However, further molecular, and biological experiments are required to confirm m6A effect on functions of the key genes in gliomas.

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Changes of hepatic enzymes activities in the free fatty acids-induced fatty liver model *in vitro*

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Background. There are no single reliable blood serum or urine markers for non-alcoholic fatty liver disease (NAFLD). Aminotransferases (ALT, AST, GGT), alkaline phosphatase (AP) are elevated but are neither sensitive nor specific markers. Diagnosis is mostly determined by ultrasound or other radiological methods. The search for more accurate non-clinical models, as well as animal welfare concerns by reducing the time and cost of drug development, make the development of *in vitro* systems a priority in pharmaceutical industry. HepG2 cells are often used as liver *in vitro* model; however, comparison of different enzyme activities in this cell line is not done.

Aim. To investigate the utility of human HepG2 cells in search of the characteristic fatty liver enzymatic markers in the lipid- loaded cells.

Methods. HepG2 cells steatosis was caused by addition of 0.5 mM oleic acid and palmatic acid (FA) mix 2:1 for 24 h. Silymarin was used as test compound. Lipid accumulation in the cells was measured using Nile Red dye and fluorescence measured at Ex/Em=550/640 nm with a microplate reader. Enzyme ALT, AST, GGT and AP activities in cell supernatant were analysed using commercial kits according to the manufacturers' instructions. Absorbance or fluorescence in all experiments was recorded by Infinite 200 PRO plate reader and i-control software (Tecan Trading AG, Switzerland). Statistical analyses were performed by One-way analysis of variance (ANOVA) followed by Dunnett's Multiple Comparison test using GraphPad Prism 7 software (GraphPad Software Inc., San Diego, CA, USA).

Results. FA induced 7-8-fold lipid accumulation as well as increased enzymes activities in comparison with un-treated HepG2 cells. ALT was increased up to 147%, AST – 179%, AP – 500%, whereas GGT and acetylcholinesterase (AchE) up to 120%–126%. Silymarin decreased the activities of all FA-upregulated enzymes.

Conclusion. In the fatty liver model, HepG2 cells show elevated levels of the characteristic lipids and enzymes ALT, AST and AP whereas GGT and AchE are not significantly increased. Therefore, HepG2 fatty liver *in vitro* model could be useful for elucidating the effects of substances on lipid accumulation and changes in ALT, AST and AP enzyme activities or levels. The effects of substances on lipid accumulation and the activity of cell-secreted enzymes can be further investigated.

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Increased neuroblast proliferation and mature astrocyte count 6 months after experimental ischemic stroke in mice

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Background. Ischemic stroke produces both acute and chronic functional changes in the brain. These changes include acute neuronal death followed by regeneration, but limited studies show the dynamics of this process in the long-term, i.e., after 6 and more months. Investigation of long-term changes allows one to understand what processes are pivotal in the recovery following ischemic stroke.

Aim. The aim of the study was to determine whether experimental ischemic stroke induces long-term changes in neurogenesis (specifically neuroblast proliferation) and alters the number of mature astrocytes in the brain neurogenic niches of mice.

Methods. Adult male C57Bl/6 mice (n=5/group) were used in this study. Ischemic stroke was induced by 60-minute filament occlusion of the middle cerebral artery (fMCAo). In sham-operated animals the filament was immediately withdrawn after insertion. Naïve animals did not undergo any surgery. Six months after surgery, animals were intraperitoneally injected with bromodeoxyuridine (BrdU, 100 mg/kg) for 2 consecutive days, then euthanized 12 h after the last injection. Coronal sections (30 μm) of the whole brain were obtained and used to determine the number of BrdU- and doublecortin-positive ($BrdU^+$/DCX^+) cells and glial fibrillary acidic protein (GFAP) staining in the neurogenic niches of mice brain – the subventricular zone (SVZ) and hippocampal dentate gyrus (DG). Detection was performed using immunohistochemistry with immunofluorescent secondary antibodies (AlexaFluor™ 488 or AlexaFluor™ 594-conjugated). One-way ANOVA followed by Holm-Sidak's post-hoc test was used for statistical analysis.

Results. The percentage of $BrdU^+$/DCX^+ cells out of all $BrdU^+$ cells was higher in the ipsilateral, but not contralateral SVZ of mice from the fMCAo group. Hippocampal $BrdU^+$/DCX^+ cells were not detectable. Optical density of GFAP was significantly higher in the hippocampal DG of both brain hemispheres, but not in the SVZ of fMCAo group mice compared to both Sham and Naïve groups.

Conclusions. Transient ischemia induced long-term changes in the generation of newborn cells and, probably, increases their transformation into mature astrocytes. Further research is required to specify whether increased GFAP staining is not associated with neuroinflammation in the brain of fMCAo group mice.

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Mesenchymal-epithelial interactions during hair follicle morphogenesis

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Background. Hair follicle formation in developing embryonic skin requires stepwise signalling between the epithelial epidermis and mesenchymal dermis, and their specialized derivatives, the placode/germ/peg and dermal condensate/papilla, respectively. There are some studies claiming that hair inductive capacity lies within the dermis.

Aim. In order to determine an epidermal-mesenchymal interactions in developing skin we studied the expression of CD34 (stem-cell-associated biomarker) and S100 (cell-growth-associated calcium-binding protein) in the follicular epithelium and embryonic fibroblasts.

Methods. We have studied 6 embryos and 10 fetuses at the age of 5 to 24 developmental weeks from embryological collection of the Department of Anatomy and Histology. Gestational age was estimated from the crown rump length of the embryo/fetus. An archival collection of skin samples of the upper limb bud, lower jaw, finger palmar surface, dorsal and lateral surface of the trunk, the forearm of the embryo/fetus stained with hematoxylin-eosin, Masson's Trichrome and immunostained with antibodies against CD34 and S100 were reviewed.

Results. Study demonstrated that CD34 and S100-positive mesenchymal cells are restricted to the specific regions of the dermis, such as dermal cells in the dermo-epidermal junction and developing human hair follicles (HF) have a different pattern of biomarker expression. Thus, in early fetal HF, CD34 and S100 are expressed by the mesenchymal cells of the dermal condensate that appear around 10 weeks gestation and consists of spheroid-like mesenchymal cells. These cells reside underneath the small epithelial ingrowth into the dermis, called the hair placode (HP). Following formation of the HP, an extensive capillary bed is established below the dermal condensate. These capillaries are supplied by communicating vessels. As the HF matures, CD34 and S100 can be seen in the spheroid-like dermal papilla (DP) fibroblasts and in the sub-bulge portion of the connective tissue sheath (CTS). By about 24 week two types of S100-positive cells (melanocytes and Langerhans cells) can be found in outer root sheath (ORS) of HF. It should be pointed out that the cells of the lower portion of CTS, which enclose hair bulb (HB), were CD34 negative, but S100-positive. The above-mentioned area of CTS is richly vascularized.

Conclusion. It can be hypothesized that in fetal skin CD34 and S100 marks highly active dermal specialized fibroblasts which might play important roles in the activation of hair bulb and ORS stem cells at the onset of hair follicle development as well as at later stages of hair placode pattern formation.

Metabolite fingerprint of peripheral mononuclear cells as a marker of diabetic nephropathy

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Background. The relevance of diabetic complications in the world is increasing, despite improvements in the treatment and care of diabetes. Diabetic nephropathy (DN) is one of the most severe and frequent complications of diabetes mellitus, which can be fatal. The need for early diagnosis of the development of complications is vital for initiating specific therapy.

Aim. The aim of this study was to find out whether the metabolic fingerprint in the peripheral blood mononuclear cells (PBMC) of patients with type 1 diabetes mellitus (T1D) may indicate the presence of complications.

Methods. This study included 24 patients with T1D from the LatDiane database, of which 9 had diabetic retinopathy, 7 had diabetic nephropathy, and 3 had both complications. Mononuclear cells were isolated from blood and frozen in 1xPBS solution at –20°C. Metabolic fingerprint in cells was detected using Fourier transform infrared spectroscopy (FTIR) [1]. Afterwards FTIR data was analyzed using several machine learning (ML) algorithms and SPSS Statistics 25.0.

Results. Statistical analysis of obtained FTIR data from T1D patients with DN compared to patients without DN showed significant changes in peak heights at 970 cm^{-1} (p=0.008), 1090 cm^{-1} (p=0.012), 1398 cm^{-1} (p=0.033), 1545 cm^{-1} (p=0.011), 1656 cm^{-1} (p=0.049). Among several ML algorithms tested in preliminary analysis the "Boosted Decision Tree" showed the most promising results in early prediction of diabetic nephropathy. Using this algorithm together with 15 different random seed experiments a mean AUC of 0.69 was achieved.

Conclusion. Despite the extremely small number of patients with T1D participating in the study, it was possible to trace a trend indicating a relationship between the metabolic imprint in peripheral blood mononuclear cells and the presence of diabetic nephropathy. The next challenge will be to screen patients with type 1 diabetes included in LatDiane by FTIR followed by follow-up to find out which PBMC metabolite fingerprint may indicate the initial stage of the development of diabetes complications.

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The morphofunctional state of cerebral hemisphere's neuropil in rats with nitrite-induced experimental Alzheimer's disease after mesenchymal stem cells intravenous injections

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Background. An increasing incidence of Alzheimer's disease (AD) has been found in excessive accumulation of nitrosamines, which are formed in the body by the interaction of sodium nitrite with proteins after ingestion with water, food and after smoking tobacco. It is known that nitrosamines increase oxidative stress, cause endothelial dysfunction of brain vessels and atrophy of the brain white matter. The effect of mesenchymal stem cells (MSC) on brain neuropil in rats with experimental AD is being actively studied.

Aim. The aim of the current study was to investigate the effect of mesenchymal stem cells on the cerebral hemisphere's neuropil in rats with nitrite-induced experimental Alzheimer's disease.

Methods. The experiment was performed on 48 male WAG rats with 14- and 28-days nitrite-induced models of Alzheimer's type dementia. Half of the animals received a single intravenous injection of MSC in a dose of 500,000 cells for each rat after sodium nitrite injections (Nitr, 50 mg/kg). Control animals (gr. C) received 0.1 ml isotonic saline. The brain slices were stained with Congo-red, bromophenol blue (BPB), according to Einarson's method and studied using Zeiss Axiostar plus binocular microscope and software GIMP.

Results. In all experimental groups, there were signs of amyloid accumulation in the cerebral hemisphere's neuropil in the form of red homogeneous masses. The homogenization of the neuropil was accompanied by a decrease in its optical density, especially in rats with 28-days AD. Using BPB staining in rats with AD the state of the cerebral hemisphere's neuropil was close to the control state, close to dystrophy, atrophy, and amyloid formation. The introduction of MSC lead to an increase the RNA content in neuropil branches. Moreover, in the brain slices stained with BPB, there were areas with reduced optical density of neuropil proteins and a predominance of carboxylic groups over amino groups, as in gr. C. It could be interpreted as the presence of new proteins in the neuropil.

Conclusion. The intravenous injection of mesenchymal stem cells led to the emergence of new cerebral hemisphere's neuropil branches in rats with nitrite-induced experimental Alzheimer's disease.

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MENTAL HEALTH

Subjective therapeutic mastery and professional identity self-assessment questionnaire for Latvian mental health professionals: a pilot study

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Background. Subjective therapeutic mastery and perceived professional competence are factors affecting mental health outcomes (e.g. burnout) among mental health professionals. However, no known tool in Latvian currently exists for measuring these domains.

Aim. The aim of the study was to develop a questionnaire in Latvian for measuring subjective therapeutic mastery and domains of professional identity.

Methods. In the cross-sectional pilot study 21 Latvian mental health professionals (doctors and resident doctors in psychotherapy and psychiatry, and psychotherapy specialists) were voluntarily, anonymously surveyed online at a private healthcare center. 3 instruments were used: sociodemographic survey; Subjective Therapeutic Mastery and Professional Identity Self-Assessment Questionnaire (MIQ); a questionnaire for content validity. MIQ was developed via research on similar instruments (e.g. Minnesota study, Rønnestad *et al.*, 2018) and various research/theoretical aspects, as well as qualitative interviews with resident doctors on their professional identity. All MIQ items were interrelated. Questionnaire included 8 questions on therapeutic mastery/competence, professional confidence, professional identity, identification with stethoscope and the white coat, persuasion of the professional's immaculate mental health, professional phase, and impostor syndrome. 7 of those were scored on a 5-point Likert scale. For example: "How much would you identify with being called a psychotherapist?" On a Likert scale: from "0-not at all" to "5-fully". Validation survey included questions on how terms were perceived/understood, timing, context, and general impressions. Statistical analysis was conducted using SPSS Statistics 22 with a significance level of $p<0.05$.

Results. Participants were 26–51 years old (Me=32.00 years; IQR=29–36). 52.4% (N=11) were psychotherapists, 33.3% (N=7) were psychiatrists, 1 in both specialties, and 9.5% (N=2) were psychotherapy specialists. Most of the participants (52.4%) were resident doctors. MIQ demonstrated high reliability for psychotherapists and psychotherapy specialists (Cronbach's $\alpha=0.75$). In a subscale for psychiatrists' reliability was acceptable (Cronbach's $\alpha=0.62$). For all participants on a subscale (3 items) Cronbach's $\alpha=0.68$.

Conclusions. The results indicate good internal consistency and validity of MIQ, and subscales could be used as separate indicators for correlation purposes, as well as a joint survey. Validity may be verified in a greater sample. Considering validation questionnaire and reliability results, final MIQ was improved with explanation of terms. Further research should investigate other factors influencing perceived professional identity.

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The use of coercive measures in child and adolescent psychiatry practice in the Children's Clinical University Hospital, Riga, Latvia – a 10-year audit study

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Background. The use of coercive measures is still a part of the child and adolescent psychiatry practice, aimed to resolve the highest risk situations, but in itself bearing risks of being traumatizing to the patient, so it needs constant scrutiny and audit. This is the first clinical audit study that has been carried out looking at the use of coercive measures in child and adolescent psychiatry in Latvia.

Aim. To evaluate the use of coercive measures (types of measures used, reasons for coercion) in patients receiving inpatient psychiatric care in the Clinical University Hospital between 2013 and 2022.

Methods. A retrospective study was conducted using Children's Clinical University Hospital Child psychiatry clinic inpatient medical records and data from restraint and seclusion registries and protocols. IBM SPSS v.26 was used for statistical analysis.

Results. The total number of events of coercion between the years 2013 and 2022 was 237. In 149 (62.9%) cases, restrained and secluded patients were male. The most used coercive method was mechanical restraint with soft bandages (96.2%, N=228), while isolation was used only in 3 cases (1.3%). The mean duration of mechanical restraint was 50 minutes. Boys tended to be restrained for longer periods than girls (55 vs. 41 min on average). The most common reasons for the use of restraint were aggression (78.5%, N=186); psychomotor agitation/psychosis (48.1%, N=114); autoaggression (43.9%, N=104); damaging hospital property (14.3%, N=34); necessity to administer medication (7.2%, N=17). Boys were significantly more likely to be restrained due to psychomotor agitation/psychosis (p=0.000), while girls were more likely to be restrained due to autoaggression (p=0.000). The medication most often used for pharmacological restraint were i/m diazepam (32.1%, N=76) and i/m olanzapine (21.5%, N=51). The patients' parents were informed about the event of restraint straight after the event in only 14.8% of cases (N=35), which can be partially explained by the majority of cases of restraint being carried out in patients coming from out-of-home care (56.1%, N=133).

Conclusion. Most of the cases of seclusion and restraint that happened in the Children's Clinical University Hospital Child psychiatry clinic in a 10-year period were carried out according to existing normative regulations, but there is still a need for improvement to ensure the best possible standard of care for psychomotorly agitated, aggressive and autoaggressive patients.

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The standard of disclosure in emergency psychiatry when applying the MacArthur competence assessment tool-treatment (MacCAT-T) methodology

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Background. Information about the diagnosis, prognosis, treatment goals and options is essential for obtaining valid informed consent for any healthcare intervention. The amount of information physicians provide for persons with psychiatric emergencies may differ because of the individual's affected decision-making ability or to avoid an information overload. It is challenging to secure valid informed consent. Therefore, scholars proposed simplifying the information disclosed, repeating the information, and using visuals (Dalal, 2020). The appropriative adaptations for disclosure must be made based on the decisional capacity assessment. The MacCAT-T (Grisso & Appelbaum, 1998) provides a structured format for capacity determination to recognise when a patient lacks capacity; for acute settings test is modified by skipping the diagnosis part.

Aim. This study aimed to explore the application of the informed consent principle in emergency psychiatry and identify the information disclosure standard for adult patients in a mental health crisis.

Methods. A Doctrinal analysis of ethical and legal regulations of an informed consent principle and mental capacity laws was performed. A literature review concerning the application of the informed consent principle in emergency psychiatry and an analysis of disclosure methodology in applying the MacCAT-T method were carried out.

Results.

The disclosure issues of the following fields were addressed:

1. Dimension of disorder and treatment (incl. dangerousness to self and others due to untreated disease or medication side effects, if applicable).
2. Inpatient milieu and limitation of rights during a hospital stay.
3. Therapeutic privilege and other exceptions (excl. voluntary waived consent).
4. Disclosure to third parties and exceptions (e.g., warning a third party about an intended violence).
5. Self-disclosure – level of expertise and conflict of interest.

Conclusion. The legal standard of disclosure comprises much more components and information than the standard of disclosure during medical decision-making capacity assessment. The standard of information needed to consent is the most appropriate for patients in mental health crisis; in acute settings, the understanding and appreciation of the diagnosis are less essential to decide about offered treatment. Other ethical and legal aspects of inpatient treatment also must be addressed.

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Depression levels in relation to glycaemic control in patients with type 1 and type 2 diabetes mellitus

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Background. Currently, there are 422 million people in the world suffering from diabetes. The International Diabetes Federation predicts that in 2030 the amount of people living with diabetes will account for 643 million, and in 2045 this number will reach 783 million. In 2021 there were 94,448 people living with diabetes in Latvia which has increased by 19,933 since 2011. It has been proven that depression is associated with the development of diabetes, and that people with diabetes are at a higher risk of having depression. However, research on associations between depression and glycaemic control is lacking.

Aim. The aim of this study was to determine whether there are correlations between glycaemic control and levels of depression.

Methods. This was a cross-sectional study including adults aged 18 or older with the diagnosis of diabetes presenting in a hospital or clinical setting. PHQ-9 which is a self-report questionnaire was used to measure levels of depression. HbA1c was used to determine glycaemic control. Spearman's rank correlation and descriptive data analysis were used to interpret the data.

Results. The participants of this study were 21 women (50%) and 21 men (50%) either with type 1 (19%) or type 2 (78.6%) diabetes, and the mean age of 58.39 years (±17.75). The duration of diabetes was 10.63 years (±8.43) on average. Mean HbA1c was 7.90% (±2.34) and the average PHQ-9 score was 5.90 (±5.69). The correlation between PHQ-9 score and HbA1c was not statistically significant ($r=0.248$, $p=0.123$).

Conclusions. A higher HbA1c was not associated with a higher level of depression, vice versa. This could indicate that poor glycaemic control does not affect nor is caused by high levels of depression.

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PUBLIC HEALTH AND EPIDEMIOLOGY

Uncovering the disparities: comparison of therapeutic approaches for late-stage non-small cell lung cancer patients at Riga East University Hospital, Latvia and Shaare Zedek Medical Center, Israel

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Background. Lung cancer is a leading cause of cancer-related death worldwide. According to data from the World Health Organization, the age-standardized mortality rate from lung cancer for 2020 was 19.7 per 100,000 in Latvia and 16.0 per 100,000 population in Israel. Treatment of lung cancer involves a combination of surgery, chemotherapy, immunotherapy, radiation therapy, and personalized molecular target therapy.

Aim. The aim of the current study was to compare the treatment strategies for non-small-cell lung cancer (NSCLC) patients in Latvian and Israeli hospitals, investigate the potential relationship between the mortality rate difference and treatment strategies.

Methods. This retrospective study enrolled patients diagnosed with stages III and IV NSCLC between 2017–2020 from Shaare Zedek Medical Center (Israel) and Riga East University Hospital (Latvia). Demographic data and therapy information were collected from electronic and manual patient records. Socio-demographic parameters of patients and the therapies they received were compared using Pearson's chi-square test and Fisher's exact test.

Results. The study population consisted of 42 grade III and 44 grade IV Israeli and 100 grade III and 100 grade IV Latvian patients. Patients did not differ by age at diagnosis ($p=0.73$) and the post-operative type of cancer (most were adenocarcinoma, $p=0.35$). Most of the patients did not have any other cancer (77.4% and 85.5% for Israeli and Latvian patients, respectively, $p=0.10$). In both countries, most of the patients did not receive surgery (74.4% and 79.5% for Israeli and Latvian patients, respectively, $p=0.34$). However, among Israeli patients 53.5%, 24%, and 9.3% received 1st, 2nd, and 3rd lines of chemotherapy; 25.6%, 7%, and 7% received 1st, 2nd, and 3rd lines of immunotherapy; and 26.7%, 14%, and 5.8% received 1st, 2nd, and 3rd lines of targeted therapy. Among Latvian patients, these numbers were lower: 19.5%, 4.5%, and 1.0% for 1st, 2nd, and 3rd lines of chemotherapy; 2.5%, 1.0%, and 0% for 1st, 2nd, and 3rd lines of immunotherapy; and 4.0%, 0.0%, and 0.0% for 1st, 2nd, and 3rd lines of targeted therapy. Differences between Israeli and Latvian patients were $p<0.01$ for all kinds of therapies.

Conclusion. This study revealed significant differences in the treatment strategies for NSCLC patients between Latvian and Israeli hospitals. The results suggest that the lack of appropriate therapy might be a distinguishing factor in the survival outcomes of NSCLC patients in Latvia. These findings highlight the need for improved patient care and therapeutic strategies in Latvia to enhance survival outcomes for NSCLC patients.

Changes in the prevalence of the intestinal diverticular disease during 2001–2021

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Background. Diverticulosis is a condition that is described as the presence of small pouches or pockets (diverticula) in the wall or lining of any portion of the digestive tract, most frequently occurring in the intestinal part of the digestion system.

Aim. To assess the changes in diverticulosis prevalence in Lithuania during 2001–2021.

Methods. Data on the prevalence of diverticular disease (ICD-10 code K57) in Lithuania during 2001–2021 were collected from the Institute of Hygiene, where the data were systematized from the Mandatory Health Insurance Information System. Changes in the prevalence trends in adult men and women during 2001–2021 were assessed using Joinpoint regression analysis.

Results. In 2021, the overall prevalence of diverticular disease of the intestine was 314/100 000 of the population. The prevalence of diverticulosis was higher in women than men, with an average ratio of 2.1:1. During 2001–2021, the average annual increase among adults was 10.7% per year ($p<0.001$). Comparing the results among different age groups, the prevalence of the diverticular disease greatly increased in elders: in the 18–44 age group the average prevalence was 23/100 000, in the 45–64 age group was 309/100 000, and in the 65+ age group 509/100 000. Analyzing the results among different age groups and sex, the most significant increase in the prevalence of diverticulosis was observed between age groups 18–44 and 45–64 for both females and males.

Conclusion. The results show that the prevalence of diverticulosis in adults is steadily increasing in Lithuania. Women are more prone to develop diverticulosis of the intestine compared to men.

Acknowledgments. The authors declare that there is no conflict of interest.

Changes in the mortality from cerebrovascular disease in Lithuania during 1993–2019

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Background. Cerebrovascular diseases are described as a group of medical conditions that affect the blood vessels of the brain and cerebral circulation. This alteration of blood flow can sometimes impair the brain's functions on either a temporary or permanent basis. The most common presentation of cerebrovascular disease is an ischemic stroke and a haemorrhagic stroke.

Aim. This study aimed to examine trends in mortality from cerebrovascular disease between 1993 and 2019 and to compare Lithuanian data with mortality trends in the EU.

Methods. Data on the mortality of cerebrovascular disease (ICD-10 code I60–I69) per 100 000 population in Lithuania and EU countries over the period 1993–2019 were collected from the World Health Organization (WHO) mortality database. Joinpoint regression was used to calculate the average annual percentage change (AAPC) and to identify changes in mortality trends in Lithuania and EU Member States, males and females.

Results. In 2019 mortality rates for cerebrovascular disease were significantly higher than the EU average mortality rate, with a ratio of 2.3:1. The mortality rate in Lithuania steadily decreased over the period from 129/100 000 in 1993 to 83/100 000 in 2019, while the average mortality rate of the European Union decreased from 98/100 000 to 36/100 000. Comparing the results among Lithuania and the EU, mortality rates in Lithuania reached an average mortality decrease of 1.5% per year ($p<0.001$), while in the EU rates decreased by 3.7% per year ($p<0.001$). During the 1993–2019 period men had a higher mortality rate than women both in Lithuania and in the EU (with an average ratio of 1.4:1). The mortality rate of cerebrovascular disease surpassed the average mortality rate of the EU during the whole study period.

Conclusion. The results show that the prevalence of cerebrovascular disease is steadily decreasing in Lithuania. However, it should be noted that the mortality rate is still significantly higher compared to the average mortality rate in other EU countries.

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Women's opinion and awareness about physical activities during pregnancy

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Background. Regular physical activities during pregnancy helps to maintain a normal body mass index, eases the pregnancy period, reduces the risk of various diseases and complications, and it is easier to return into previous physical shape after childbirth.

Aim. The aim of the study was to find out women's opinion about physical activities during pregnancy and awareness of health benefits.

Methods. From September 5th until October 25th, a survey was published. In the survey participated 295 pregnant and postpartum women. Respondents were not divided into categories to objectively reflect the trends in the population of Latvia. The questionnaire was done by women with various BMI and various lifestyles.

Results. The age was ranged from 17 to 44 years, weight from 49 kg to 105 kg, height from 165 cm to 181 cm.

In the question whether a woman should engage in physical activities during pregnancy, 73.2% answered yes, 16.3% answered occasionally, 1.4% answered no, 9.1% answered only moderate activities. In summary 26.8% of respondents believes that pregnant women should not engage in physical activities regularly.

In the question whether your gynecologist has informed you about the benefits of physical activities during pregnancy, 55.6% answered yes, 32.6% answered no, 9.8% do not remember and 2% answered it does not matter. 66.4% have seen information on social media, while 33.6% did not saw any information.

In the question how physical activities affects pregnancy, 92.5% answered positively both for the woman and new-born, 3.7% answered healthy for the pregnant woman, but negatively affects the new-born, meanwhile 3.7% had a different opinion.

Conclusion. Most of the women are aware how physical activities effect pregnancy and knows that physical activities have positive effects both on mother and fetus.

However, there are still stereotypes in society, that pregnant women should not engage in physical activities during pregnancy and there is not enough information about the health benefits from health care specialists and social media.

It is necessary to inform society more actively and encourage women to engage in physical activities and clarify the benefits of an active lifestyle.

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Frequency of physical activities of reproductive age women in Latvia

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Background. WHO recommendations for adults are 150–300 minutes of moderate-intensity aerobic physical activity or 75–150 minutes of high-intensity aerobic physical activity in a week. By following the recommendations, it is possible to improve health and well-being, so it is important to update the topic and promote an active lifestyle.

Aim. The aim of the study was to find out the frequency of physical activities of women of reproductive age and evaluate whether women of reproductive age in the Latvian population perform physical activities in sufficient level.

Methods. From September 5th until October 25th, a survey was published. In the survey participated 295 reproductive age women. Respondents were not divided into categories to objectively reflect the trends in the population of Latvia.

Results. The age was ranged from 17 to 44 years, weight from 49 kg to 105 kg, height from 165 cm to 181 cm.

In the question of whether you regularly engage in physical activities, assuming that one session is at least 30 minutes, 50.2% answered they engage regularly, 28.1% occasionally, 21.7% do not engage at all.

In the question of how many times a week you engage in physical activities, 32.9% answered 1–2 times a week, 23.1% 2–3 times a week, 14.9% 3–4 times a week, 7.8% 4–6 times a week, 2% more than 6 times a week, 19.3% do not participate at all.

In the question what kind of physical activities you do, 68.8% answered walking, 31.2% exercising, 29.5% active leisure time, 17.6% running, 15.6% lifting weights, 13.9% swimming, 8.5% do yoga, and 10.2% do not participate at all.

Only 9.8% engage in physical activities in amount that is based on WHO recommendations. 80.7% engage in physical activities at least once a week or more often, while the remaining 19.3% do not engage in physical activities at all.

Conclusion. There is an insufficient amount of physical activities, and society should strive for WHO recommendations, because regular physical activities helps maintain a normal body mass index, reduces the risk of cardiovascular diseases, strengthens the bone and muscle system, enhances cognitive abilities, and promotes many other health benefits.

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Prevalence of uterine cancer in Uzbekistan

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Background. Uterine cancer (UC) is one of the most frequent malignant tumors of the reproductive organs among women in economic developed countries. However, in Uzbekistan a gradual increase of UC is observed, and this pathology is on the second place after cervical cancer.

Aim. The aim of the current study was to study prevalence of uterine cancer in Uzbekistan.

Methods. The statistical indicators for analyses were taken from official report from 2019 to 2021 years.

Results. Analyzed data of UC in 2019–21 years showed that 640 (1.9), 609 (1.8) and 789 (2.3) UC patients were identified in the Republic, respectively. The age of patients varied from 18 years to over 80. In group from 18 to 35 years, UC was diagnosed in 14 patients; from 36 to 50 years, in 119; from 51–65 years, in 337; and older than 65 years, in 170 patients. The patients were into stages as follows: stage I – 34.8%, stage II – 41.7%, stage III – 14.4%, – IV 3.1%. The mortality rate in 2019 was 0.8 (256 patients) and the 5-year survival rate consisted of 49.5%. In 2020 the patients were into stages as follows: stage I – 35.8%, stage II – 41.7%, stage III – 11.3%, stage IV – 4.4%. The 5-year survival rate was 48.7%. The mortality rate in 2020 was 0.8 (274). In 2021 in age group from 18 to 35 years, UC was diagnosed in 20 patients; from 36 to 50 years, in 145; from 51–65 years, in 412; and older than 65 years, in 212 patients. 400 patients were from rural areas. The patients were into stages as follows: stage I – 31.8%, stage II – 42.3%, stage III – 16.9%, stage IV – 4.8%. The mortality rate in 2021 was 1.8 (304 patients) and the 5-year survival rate was 48.0%.

Conclusion. The morbidity of UC in Uzbekistan has tended to increase and requires primary care physicians to promote a healthy lifestyle, a more careful approach to all types of uterine bleeding at women of both fertile and menopausal age. Timely putting diagnosis and treatment of endometrial hyperplastic processes will significantly reduce the number of women at risk for UC.

Acknowledgements. The first author confirms that there is no conflict of interest included in this abstract and have been approved by all co-authors.

Incidence trend of colorectal cancer in Uzbekistan

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Background. Colorectal cancer in the overall structure of oncological morbidity and mortality ranks 3rd and 2nd all over the world. Approximately 880 000 people die from colorectal cancer each year worldwide. In Uzbekistan, in turn, colorectal cancer ranks 4th place in the structure of morbidity and mortality.

Aim. The aim of the current study was to study prevalence of colorectal cancer in the Republic of Uzbekistan.

Methods. The material of the study was official cancer report in Uzbekistan according state form – "Information on diseases of malignant neoplasms".

Results. Colorectal cancer takes the 4th place in the structure of the general oncological morbidity, accounting for 7.0% of all newly cancer cases. At the same time, in the structure of oncological morbidity among male, colorectal cancer ranks 3rd (9.3% of all new cancer cases), and 4th among female (5.4% of all new cancer cases). Men are almost 2 times more likely to develop colorectal cancer than women. In 2021 year, there were diagnosed 1787 new colorectal cancer cases in the Republic, and the incidence rate was 5.2 per 100 000 population. Moreover, 33.6% of cancer patients were diagnosed with stages I–II cancer; 41.5%, stage III; and 20.6%, stage IV. By the end of 2021, there were 6374 patients with colorectal cancer. In 2021, 1008 patients died from colorectal cancer, while the mortality rate per 100 000 population was 2.9. At the same time, in the overall structure of oncological mortality, colorectal cancer also takes the 4th place (6.9% of all deaths from cancer). Moreover, in the structure of oncological mortality among male, colorectal cancer ranks 3rd (8.5% of all deaths from cancer), and only 5 among female (5.6% of all deaths from cancer).

Conclusion. The analysis showed that colorectal cancer takes a leading position in the structure of oncological morbidity and mortality in the Republic of Uzbekistan and colorectal cancer occurs more often among male than female. About 1/3 of new colorectal cancer cases were diagnosed at early stage I–II, and just over 20% of patients were diagnosed in stage IV disease.

Acknowledgements. The first author confirms that there is no conflict of interest included in this abstract and it has been approved by all co-authors.

Economic analysis of non-small-cell lung cancer patients' treatment in Riga East Clinical University Hospital, Latvia: retrospective data from the Value-Based Healthcare project

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Background. According to the World Health Organization, lung cancer mortality in Latvia was ranked first among all types of cancer, reaching 2020 the age-standardized mortality rate of 19.7 per 100,000 population. Treatment of lung cancer involves a combination of surgery, chemotherapy, immunotherapy, radiation therapy, and personalized molecular target therapy. Target therapy mostly is not reimbursed by the state for patients in Latvia.

Aim. In this study, we aimed to assess the time trends in the treatment of non-small-cell lung cancer (NSCLC) patients' costs for the Latvian health system according to the stage of cancer.

Methods. 400 patients of the Riga East Clinical University Hospital (100 patients of each stage of NSCLC) were retrospectively enrolled in the study. We assessed the time between the diagnosis and different stages of treatment, as well as the economical effectiveness of treatment of patients in different stages of NSCLC. We received information from the National Health Service on governmental reimbursement for staying in the hospital, medical procedures, and drug compensation for all patients enrolled in the study for two years before their diagnosis and for the years of life after the NSCLC diagnosis. We calculated pre- and post-diagnostic costs of patients by stage of cancer.

Results. The mean time of treatment for all patients was 501 days after the diagnosis: the longest was found for stage I patients (mean of 764 days) with subsequent reduction (mean of 281 days for stage IV patients). Reimbursement for two years before the diagnosis was 22.6% of all patients' costs. Total post-diagnostic reimbursement for all patients was 2,854,643 euros. The highest costs were found for staying in the hospital (>1.8 million euros for all patients) and the lowest for medical procedures (491,932 euros). Reimbursement for medical procedures was the lowest among all three categories of costs. Stage IV patients had the highest reimbursement for medications in comparison with other stages. However, total post-diagnostic reimbursements were divided almost equally between patients with different stages of NSCLC: 23%, 29%, 23%, and 24% for stages I, II, III, and IV, respectively.

Conclusion. The increase in reimbursement of medical procedures making the target diagnostic for NSCLC available might not only improve patients' outcomes and increase their survival, but also reduce the costs for other categories of medical procedures.

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Vaccination: the booster of immunity

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Background. The development of vaccine is a pioneering achievement in the history of mankind. It has led to the successful eradication of a deadly disease like smallpox. Trepidation surrounding vaccinations in the 1970s, had led to a significant increase in the previously eradicated disease, pertussis, in countries like the UK, Japan and Sweden. In 1990, Soviet Union cut down immunisation against diphtheria leading to an epidemic.

Aim. The purpose of our study is to understand the stigma around vaccination.

Method. A quantitative study was conducted utilising a survey, containing 19 questions, pertaining to vaccination and their subsequent effect on the individual.

Result. 100 participants took part in the research, of these participants, 67 were of mean age 22.5(σ=3.23), 18 were of mean age 40(σ=5.32) and 15 were of mean age 63.2(σ=3.45).They were vaccinated with the following: BCG (49%), OPV (70%), DPT (42%), MMR (36%), influenza vaccine (32%), hepatitis-B vaccine (43%), varicella vaccine (18%) and COVID-19 vaccine (100%). Despite vaccination, 3% suffered from chicken pox, 1% from hepatitis, 1% from typhoid and 11% from COVID-19. Out of those who got infected with COVID-19, 7 experienced mild, 3 experienced moderate and 1 experienced severe symptom. 55% participants experienced fever, headache, pain on injection site and general body weakness after vaccination. 50% were concerned about vaccination compromising fertility. 41% suspected that vaccination leads to severe damage and death. Vaccines were considered unnecessary by 6% and 2% were unsure. 35% considered vaccine as a cure. 15% would not choose to be vaccinated after encountering a disease. 11% would avoid taking any precautions after vaccination. 36% considered infant's immunity to be fragile for vaccines.

Results show that although 66% of our sample deemed vaccinations a success, still a lot of work should be done to ensure that the remaining 34% of the sample population receive adequate education and information regarding the importance of vaccines for the prevention and eradication of diseases.

Conclusion. As reported by the survey 16% were affected post-vaccination, out of those, 63% experienced mild symptoms. 45.5% had the notion that vaccination can affect fertility and can cause death.

This information should prove valuable when educating the public about diseases and reducing mortality with vaccines.

Anti-vaccination protests and misinformation should be addressed by evidence-based education and transparency regarding pros and cons of such interventions. This may reduce the hesitancy of the general public when presented with life-saving interventions, such as vaccines.

The necessity to improve section No 149 of the Latvian civil procedure law

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Background. Article No 149 of the Latvian Civil Procedure Law (thereinafter – Article No 149) determines the actions of a judge upon preparing a case for trial. Paragraph 8 of this article directly states that in cases regarding the reinstatement of an employee in work and in cases regarding the annulment of an employer's notice of termination, the date of the court hearing shall be determined not later than 15 days after receipt of explanations or after other preconditions mentioned in this paragraph. A similar 15 days' time limit is set for cases regarding claims arising from the alienation of immovable property for public needs (see para. 9 article No. 149). Paragraph 10 of Article 149 sets some time limits in cases regarding insolvency, but in other cases, article No 149 does not oblige the judge to set the date and time of the court hearing within the specific time limit. Therefore, existing regulation do not provide the opportunity for patients, persons, or plaintiffs – in claims arising from personal injuries that have resulted in mutilation or other damage to health, to get some advantage in terms of scheduling court hearings. This is a notable misregulation.

Aim. The aim of this research is to analyze Article No 149 and expand its understanding towards its modernization in the nearest future to provide a much better opportunity for patients to get judicial protection within a reasonable time. Existing regulation forms an impression of a course of questionable value. Much declared care for citizens, persons in need and children's rights are not provided well enough in terms of prioritizing cases via the court hearing scheduling process in Latvia.

Methods. The author has used grammatical, historical, systemic, and teleological methods for the interpretation of Article No 149. For a broader understanding of this topic, the author has analysed historical and recent court practice, legal literature of various periods.

Results. Research results are: 1) a more precise understanding of the Article No 149; 2) the recognition that the existing regulation does not provide a consistent approach to the process of scheduling court hearings and prioritizing civil cases in this regard; 3) identified a way of action for the legislator to improve regulation – revise the case prioritization system.

Conclusion. Article No 149 should be completely revised to prioritize civil cases via the court hearing scheduling process. This legislatorial task should be considered not only examples of exceptions to general provisions regarding court expenses but the value-based approach in civil litigation as well.

Is early diagnosis and therapy of children's development a part of basic level of medical assistance guaranteed by the constitution of the Republic of Latvia?

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Background. Article No 111 of the Constitution of the Republic of Latvia (thereinafter – Article No. 111) directly states, that: "The State shall protect human health and guarantee a basic level of medical assistance for everyone." The Constitutional Court of the Republic of Latvia, interpreting this article, pointed out, that respect, protect and an obligation to ensure a person's right to health is provided by it (see case No. 2002-04-03, para 1, No. 2003-15-0106, para 6, case No. 2008-37-03, Para 12.1.2). Clearly, the legislator has the discretion to select the most appropriate means for fulfilling its obligations (see case No. 2012-14-03, para. 15), the state is not obliged to provide all medical or other healthcare services even to children regardless of costs (see case No. 2009-12-03, para. 16) and "[..] basic level of medical assistance [..]" is an open concept of law. Nevertheless, it cannot be dismissive of or ignoring prevention in medicine as a part of the medical assistance system as a whole. The preventive approach of the protection of public and individual health should be debated within the understanding of basic level of medical assistance.

Aim. The aim of this research is to unveil the understanding of "[..] basic level of medical assistance [..]" set by Article No 111 in the context of the prevention (e.g., early diagnosis of children's development) and its impact on legislators' discretion for selecting appropriate means of fulfilling its duties.

Methods. The author has used teleological grammatical, historical and systemic methods for the interpretation of Article No 111. To unveil facets of the open concept of law and understand the connection of prevention with existing health system guarantees, the author analysed legal literature of various periods, and court practice as well.

Results. Research results are: 1) widened understanding of the concept "[..] basic level of medical assistance [..]" which: a) includes not only the availability of medical services, treatment and medicines to some degree but no less important or even primarily – to a set of proper preventive measures, b) is designed for provision of sequential medical assistance, where the diagnosis is the first step and treatment is the second one; 3) recognition that the content of the concept of law, in this case, is primarily formed by the principles of medical science.

Conclusion. Clearly, there could be a wide range of objections, but early diagnosis and therapy of children's development is rather a part of the basic level of medical assistance guaranteed by the Constitution of the Republic of Latvia, than not.

SURGERY

Vascularisation patterns of solid renal tumours: differentiation of benign and malignant renal masses

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Background. Renal masses present a wide histological spectrum ranging from benign oncocytoma and angiomyolipoma to malignancies as high grade renal clear cell carcinoma.

Aim. The task was to study imaging patterns, including vascularization of renal masses on Computed tomography (CT) to increase the accuracy of visual differentiation to further facilitate the decision of choosing treatment tactics.

Methods. The retrospective study with 100 patients with total renal tumour resection. Qualitative and quantitative parameters were evaluated to determine benign and malignant prognostic imaging parameters, including contrast uptake and wash-out patterns in the lesion, and relative to the renal parenchyma in preoperative CT examinations.

Results. 100 patients with solid renal lesions, male 49% vs. female 51% (31–84 years, mean 64 years). From 100 lesions benign were 22 (14% oncocytoma, 6% AML, 2% adenoma), malignant were 78 (RCC 72%, urothelial 6%).

Statistically significant association between benign masses and homogeneity in non-enhanced CT was found (p=0.019). Positive, mild correlation (r=0.236; p=0.020) of homogeneity and benign lesions in non-enhanced scans and AML and homogeneity in the arterial phase using chi^2 test was found (p=0.031). AML showed mild wash-out (p=0.018), and adenoma showed moderate wash-out (p=0.018) in venous phase.

Malignant lesions uptake contrast more than benign (U=465.5; p=0.013), also malignant masses uptake more contrast relative to the cortex compared with benign lesions (U=545.5; p=0.38).

In the delayed phase malignant masses washed-out slower than benign (U=1123; p=0.002).

Conclusions. Qualitative and quantitative evaluation reveals differences between benign and malignant renal masses, showing that benign masses are predominantly homogeneous on non-enhanced CT examination, whereas malignant masses retain the contrast longer in the venous phase and wash-out slowly in the delayed phase.

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CARDIOVASCULAR AND REGENERATIVE MEDICINE

Dabigatran concentrations in blood samples with various storage conditions as assessed with chromogenic assay

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Background. Chromogenic assays for direct oral anticoagulant concentration assessment are more applicable for clinical practice than liquid chromatography-tandem mass spectrometry. Nevertheless, both assays have minimal availability due to high equipment costs. Manufacturer-provided storage recommendations are also problematic for blood sample transportation because of insufficient infrastructure.

Aim. This study aimed to evaluate various storage durations and temperatures which do not affect dabigatran concentrations in citrated blood or citrated plasma compared with baseline values.

Methods. This was an *in vitro* study performed at Pauls Stradiņš Clinical University Hospital, Riga, Latvia, from August to December 2022. One sample consisting of two 2.7 mL tubes was obtained from patients taking dabigatran. Functional anti-IIa assay (Siemens Healthineers) was used. Concentrations were determined immediately after blood collection (baseline value). The first tube was left unprocessed and stored at refrigerator (2–8°C). The second tube was centrifugated, and the plasma was separated into three parts – one for storage at refrigerator (2–8°C) and two at frozen conditions (–20°C). The repeated assay was done for one sample of each temperature regime on days 3 and 7 of storage. Acceptable stability was defined as 80%–120% of the baseline result. Descriptive analysis is presented as median and interquartile range (IQR). IBM SPSS Statistics (27.0) Wilcoxon paired signed-rank test were used.

Results. A total of 11 samples were analysed for this study. Table 1 provides an overview of the stability results. Only one sample aliquot stored as an unprocessed whole blood sample with a respective low baseline concentration level (24 ng/mL) exceeded >20% deviation on the 3rd (+38%) and 7th day (+33%).

Table 1. Summary of concentrations at specific time points under different sample storage conditions.

Sample (temperature)	Median [IQR], ng/mL, p-value		
	Baseline value	Day 3	Day 7
Whole blood (2–8°C)	139 [96]	133 [76], p=0.286	130 [96], p=0.305
Plasma (2–8°C)		137 [74], p=0.247	134 [98], p=0.722
Plasma (–20°C)		126 [83], p=0.722	118 [96], p=0.721

Conclusion. Our data suggest that dabigatran concentrations do not change significantly if plasma is stored at 2–8°C or at –20°C for up to 7 days. More examinations of low-concentration dabigatran samples are needed to make conclusions about the storage conditions of the whole blood samples. If confirmed, these findings have practical implications for improving accessibility to the clinical use of the assay.

Application of implantable loop recorder in identifying the most prevalent causes of recurrent syncope in different age groups

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Background. Syncope is a common clinical problem affecting people of all ages. The median age of first syncope is about 15 years, and it increases further in people over 70 years of age. The cause of syncope often remains undiagnosed even after an extensive investigation. Patients with syncope of uncertain origin may be diagnosed earlier with implantable loop recorder (ILR) implantation. The ILR is a device that continuously records the heart rhythm for several years. Nevertheless, more scientific data are needed to understand the characteristics of the ILR findings and advantages of this device in different age groups.

Aim. To define the most common causes of syncope using ILR in two different age groups (patients 50 years old or younger, and patients over 50 years old).

Methods. This study included 38 patients who underwent ILR implantation at Hospital of Lithuanian University of Health Sciences Kaunas Clinics between February 2017 and July 2022 due to syncope of unknown origin. The research data was retrieved from electronic medical records. Statistical Package for the Social Sciences version 27.0 software was used for statistical analysis. Shapiro–Wilk test, Mann–Whitney U test and chi-square test were performed. Research was approved by the Bioethics Center of the Lithuanian University of Health Sciences.

Results. The mean age of patients was 49.2±16.7 years. There were 17 (44.7%) older than 50 years old. Arrhythmias associated with syncope were more often recorded in older patients than in younger patients (7 (41.2%) versus 2 (9.5%) patients, p=0.02). The follow-up duration (from implantation until confirmed rhythm disturbance) did not statistically significant differ between groups (younger – 233 [191–275] days and older – 136 [30–452] days, p=0.50). In younger patients' group ILR revealed third-degree atrioventricular (AV) block in 1 (4.8%) and sinus node dysfunction (SND) in 1 (4.8%) patient related with syncope. In older group third-degree AV block in 1 (5.9 %), ventricular tachycardia in 1 (5.9%), SND in 4 (23.5%) and high-rate supraventricular tachycardia in 1 (5.9%) patient. Therefore, a pacemaker was recommended for all patients in younger group, but in the older group the recommendations were more varied (cardiac pacemaker implantation, ablation, and electrophysiological study).

Conclusion. In the older group arrhytmogenic cause of syncope was identified in one-third of patients, and faster than in the younger group.

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The study of chronic coronary artery total occlusion – a single center registry

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Background. Chronic coronary artery total occlusion (CTO) is an aggressive form of coronary artery disease characterized by heavy atherosclerotic plaque burden resulting in complete occlusion of the vessel. CTO carries a high risk of heart failure and arrhythmia development as well as negatively impacts quality of life.

Aim. The aim of the study was to detect presence of CTO in patients ongoing elective coronary angiography and to summarize treatment strategy.

Methods. A total of 1000 patients admitted for elective coronary angiography to Latvian Cardiology Center were included. Data on patient's demographics and risks factors was obtained. Coronary angiograms as well as treatment strategy were analyzed. All patients signed informed consent. Statistical analysis performed in SPSS software.

Results. Mean age of included subjects was 66.4 (SD 11.5) years. There were 523 (52.4%) males. In total 656 (65.7%) patients had at least 50% stenosis in coronary artery greater than 2.5 mm. Three-vessel disease was found in 23 (52.3%) cases. Main branch (LAD, LCX or RCA) CTO was observed in 44 (6.7%) cases. CTO location was as follows: 10 (0.22.7%) LAD, 8 (18.2%) LCX and 20 (45.5%) RCA. Six (13.6%) patients had multiple-vessel CTO. Most common cardiovascular risk factors in CTO patients were: arterial hypertension (n=42, 95.5%), dyslipidemia (n=44, 100%) and diabetes mellitus (n=12, 27.3%). Previous revascularization was observed in 21 (47.7%) patients, among them previous percutaneous coronary intervention (PCI) in 19 (90.5%) and coronary artery bypass graft surgery (CABG) in 2 (9.5%). The recommended treatment strategy was medical treatment (n=13, 29.5%), PCI (n=22, 50%) or CABG (n=9, 20.5%). Age was found to be statistically significantly associated with type of revascularization (CABG vs. PCI 68.9 (SD 8.7) years vs. 63.1 (SD 13.6), p=0.04).

Conclusion.

1. CTO is observed in 6.7% patients;
2. Most common treatment strategy in CTO patients was PCI;
3. Older age is a predictor of selection of CABG as a revascularization method.

Acknowledgements. The authors have nothing to declare.

Low-density lipoprotein cholesterol profile and response to treatment in familial hypercholesterolemia patients in relation to presence of monogenic mutations

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Background. Monogenic familial hypercholesterolemia (FH) patients are at higher risk for premature coronary heart disease and have poorer therapeutic response to lipid lowering medications (LLM) compared to polygenic hypercholesterolemia patients.

Aim. To evaluate whether there are significant differences in lipid profile and response to LLM between patient groups with or without identified monogenic mutation in Latvian FH patients.

Methods. Data from the Latvian Registry of Familial Hypercholesterolemia (LRFH) was used. Whole genome sequencing (WGS) was performed in selected FH probands. The diagnosis was defined according to the Dutch Lipid Clinic Network criteria. Variants were annotated as pathogenic / likely-pathogenic (P/LP) using the FH Variant Curation Expert Panel guidelines for LDLR and adaptations were used for APOB and PCSK9. Patients were classified as monogenic (FH/M+) and genetically undefined (FH/M–) Lipid profile and response to LLMs was compared between the two patient groups.

Results. By November 2022 a total of 1155 patients were included in LRFH. Clinical FH was diagnosed in 513 patients. WGS was performed in 164 probands. Causal monogenic mutation was found in 34 patients (21.0%, FH/M+ group). Two patients were excluded from this analysis (one with compound heterozygous FH resistant to injectable PCSK9 interfering therapies, and one with suspected secondary causes). Coronary artery disease was diagnosed in 16 (47.1%) and 86 (67.2%) patients, respectively (p=0.045). The main lipid results are summarized in Table. Only 6 patients had reached their LDL-cholesterol goal at the last follow-up visit (one patient or 2.9% in FH/M+ group and five patients or 3.9% in the FH/M– group; p=0.999). The received LLMs did not differ significantly between the two patient groups.

Table.

	FH/M+ (n=34)	FH/M– (n=128)	p value
Highest LDL-cholesterol at baseline, mmol/L	7.4 (6.6–8.5)	7.0 (6.5–8.1)	0.240
LDL-cholesterol at follow-up, mmol/L	4.3 (3.0–6.0)	3.8 (2.3–5.6)	0.085
LDL-cholesterol absolute reduction, mmol/L	3.2 (1.8–4.3)	3.7 (1.4–4.9)	0.517
LDL-cholesterol relative reduction, %	47.2 (22.6–59.0)	52.7 (19.7–67.8)	0.164

Values are median (interquartile range).

Conclusion. Although statistically not significant, our data suggest that in Latvian cohort patients with monogenic FH have higher baseline LDL-C levels than those without identified P/LP variants and reduction of LDL-C is less effective in monogenic FH patients.

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Predictors of the complicated course of arterial hypertension

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Background. The frequency of arterial hypertension (AH) in the Republic of Kazakhstan varies from 15.2 to 27%. AH is a risk factor of the cardiovascular complications.

Aim. The aim of the study was to identify predictors of cardiovascular complications in patients with AH.

Methods. A total of 60 patients aged 28 to 89 years with AH were examined; all patients underwent 24-hour monitoring of blood pressure. By gender, females (55%) predominated. All patients underwent ECG, echocardiography, and a lipid profile. According to the nature of the nocturnal decrease in systolic blood pressure, patients were divided into three groups: dippers (nocturnal BP decreases more than 10%), non-dippers (the nocturnal BP has less than 10%), and night-pickers (increasing of nocturnal BP). The objective obtained data were analyzed in easymedstat.com program. Correlation analysis was performed to identify the relationship between several variables and the trends of arterial hypertension.

Results. 33.3% of patients were classified as dippers. 55% and 11.7% of the subjects are classified as non-dippers and night-pickers. In the group of non-dippers and night-pickers, higher values of mean systolic daytime and night pressure (148 mmHg and 150 mmHg) were observed. In this group women predominated (59%) and had more pronounced circadian blood pressure indicators. In this group hypercholesterolemia (12.5%), smoking (25%), positive family history (over 50%), left ventricular hypertrophy (25%) were more often detected. Correlation analysis revealed high positive correlation (0.715) with age (more than 60 years) and left ventricular hypertrophy (0.722), high triglyceride levels (0.922), with concomitant endocrine diseases (0.87) among the night pickers.

Conclusion. In the group of non-dippers and night-pickers, higher values of mean systolic daytime and night pressure (148 mmHg and 150 mmHg) were observed. Night-pickers characterized by high positive correlation link between old age and LV hypertrophy (0.722), high triglyceride levels (0.922), with concomitant endocrine diseases (0.87). Understanding the meaning of the circadian variations of BP identifies the patients at higher risk of cardiovascular mortality.

Acknowledgements. The authors declare the absence of conflict of interest.

INFECTIOUS DISEASES AND PULMONOLOGY

Tuberculosis incidence trends in Lithuania during 1991–2021

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Background. Tuberculosis (TB) is a highly contagious infectious disease, which is caused by mycobacterium tuberculosis (MTB). Tuberculosis may infect any body part, but most commonly occurs in the lungs. Extrapulmonary TB occurs when the disease develops outside of the lungs, such as the central nervous system (CNS), lymphatic system, and gastrointestinal system.

Aim. This study aimed to examine trends in the incidence of tuberculosis cases between 1991 and 2021 and to compare Lithuanian data with the EU trends.

Methods. Data about TB incidence (ICD-10 code A15–A19) per 100 000 population in Lithuania and the EU countries over the period 1991–2021 were obtained from the Lithuanian Institute of Hygiene.

Results. Tuberculosis in Lithuania has always been a severe health concern. In 1998, due to rapidly increasing incidence, a directly observed treatment short course (DOTS) was established. Due to this reason, we examined two time periods: new tuberculosis cases before 1998 and after 1998. In 2021, 550 new cases of tuberculosis were registered in Lithuania. Analyzing the results among Lithuania and the EU, the crude incidence rate (CIR) reached 19.6/100 000 of the population while the EU average CIR was 11.5/100 000. Lithuania ranked second in the EU by CIR, with only Romania ranking first with 45/100 000 new cases of the population. During 1991–1998, the CIR in Lithuania increased by 13.8% per year ($p<0.001$), while during 1999–2021 a decrease of 5.8% per year ($p<0.001$) was observed. Comparing the results among different sex, the CIR was greatly higher in men than in women, with an average ratio of 2.1:1. According to National Public Health Center experts, the newly diagnosed TB cases in 2022 is expected to rise by 33% compared to 2021.

Conclusion. The newly diagnosed number of patients with tuberculosis is slowly decreasing. It should be noted that the CIR in Lithuania remains one of the highest in the EU. The implementation of this study results is recommended in managing tuberculosis prevention activities in Lithuania.

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Long-term outcomes of high dose vitamin D treatment in COVID-19 patients

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Background. Outcomes of COVID-19 could be divided in prolonged, long-lasting, post-acute, long-term, or chronic effects. There is growing interest to profoundly investigate the role of vitamin D and its deficiency in pathogenesis of these symptoms.

Aim. The aim of the current study was to investigate long-term outcomes of high dose vitamin D treatment in COVID-19 patients.

Methods. In a randomized, placebo-controlled study 80 inpatients with vitamin D deficiency and PCR-confirmed SARS-CoV-2 infection were divided into two groups. The intervention group (group B) received 12,000 IU of vitamin D per day for five consecutive days, while the control group (group A) received a placebo. All patients were contacted and interviewed via telephone for 2-year follow-up standardized questionnaire that consisted of symptoms graded according to 4-point Likert scale (no problems, mild problems, moderate problems, or severe problems).

Results. According to the data obtained from the death registry of Latvia 25 of 100 patients (25 %) passed away during 20 months since hospitalization due to COVID-19. The majority (60%) had at least 2 or more comorbidities prior to hospitalization. When analyzing all long-term symptoms, none of the cases showed a statistically significant difference between group A and group B. Most frequent symptoms of post COVID-19 were fatigue (69.4%), dyspnea (53.2%), cough (51.6%), anxiety and depression (48.4%) and dizziness (48.4%).

Conclusion. A large part of people living in Latvia still do not consider maintaining vitamin D at an optimal level to be important. Almost all patients with COVID-19 and vitamin D deficiency suffer from post-COVID-19 symptoms. The symptoms that most often persist for over 20 months are fatigue, anxiety, and depression.

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NEUROLOGY AND ONCOLOGY

Difficulties in integrating virtual reality technologies into clinical practice for patients with multiple sclerosis in Latvia

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Background. Recently, many attempts have been made to apply virtual reality technology with the aim of improving the rehabilitation and diagnosis of multiple sclerosis patients. Unfortunately, the lack of experience and understanding of the integration of such technologies complicates the process of introducing them into real clinical practice.

Aim. The main goal of the work is to identify and evaluate the main difficulties in introducing virtual reality technologies into clinical practice.

Methods. This study was prospective. The total number of respondents was 36 respondents between 24 and 50 years old. 50% (n=18) were patients with multiple sclerosis and 50% (n=18) were the control group. As part of the study, software for virtual reality glasses Oculus Quest 2 was created using programs like Unity3D, Blender3D and Substance painter. During the study, patients were asked to undergo four multiple sclerosis diagnostic tests created as part of the study. After that, all patients underwent an anonymous questionnaire about user experience.

Results. According to the results of the survey, 75% (n=27) of all respondents noted positive impressions from the use of virtual reality glasses and 83% (n=15) of all patients with multiple sclerosis noted that they would like to undergo rehabilitation based on virtual reality technologies in the future. 44% (n=16) of all respondents noted discomfort during the diagnostic tests, of which 22% (n=8) were patients and 22% (n=8) were the control group. The most frequent causes of discomfort in the surveys were: eye fatigue 22% (n=8), of which 11% (n=4) were patients and 11% (n=4) were the control group. Discomfort after wearing glasses was found in 22% (n=8) of all respondents of which 11% (n=4) were patients and 11% (n=4) were the control group. Problems with coordination was found in 33% (n=12) of which 22% (n=8) were patients and 11% (n=4) were the control group.

Conclusion. Despite the general positive attitude after using virtual reality, the question of reducing the discomfort associated with the prolonged use of virtual reality glasses, especially among neurological patients, with dysfunctions of the vestibular apparatus, remains open.

Acknowledgements. The authors declare absence of conflict of interest.

The experience of dementia caregivers at the beginning of the disorder

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Background. According to the World Health Organization, more than 55 million people live with dementia worldwide, and there are nearly 10 million new cases every year. This clinical syndrome can manifest with various cognitive and emotional symptoms that interfere with daily functioning and quality of life.

Aim. To assess the experience of dementia caregivers at the beginning of the disorder.

Methods. A survey was conducted from January to May 2022 at the Hospital of Lithuanian University of Health Sciences *Kauno klinikos*. The questionnaire included 35 questions and was completed by caregivers who had a relative with dementia. Altogether 32 people were enrolled in the study. The data was analyzed using IBM SPSS 27.0. Statistical significance was determined to be $p<0.05$.

Results. 43.8% (n=14) of respondents' relatives were diagnosed with Alzheimer disease, 15.6% (n=5) with vascular dementia and 40.6% (n=13) etiology was unknown. The average age of dementia diagnosis was 72 years (SD=7.68).

Respondents indicated that disease of their relatives started with the following cognitive symptoms: deterioration of memory (81.3%, n=26), disorientation in time and space (37.5%, n=12), lack of concentration of attention (31.3%, n=10), behavioral and emotional changes (28.1%, n=9), impairment of speech (12.5%, n=4), disability to make correct decisions (15.6%, n=5), impaired smooth performance of sequential actions (15.6%, n=5).

In the beginning of disease behavioral and emotional symptoms were more common among female patients than men (p=0.035). Referral to a doctor was more often initiated by patients' relatives, as patients themselves had no complaints (56.3%, n=18).

Patients with impaired speech as the reported first symptom by their relatives were more likely to require assistance with daily activities at initial physician visit (p=0.004). Similar findings were observed for disorientation in time and space (p=0.023), impaired decision-making (p=0.005), and difficulty performing sequential actions (p=0.001) as initial symptoms.

Conclusion. According to the caregivers, cognitive dysfunction was more often noticed by relatives than reported by patients. The most common first symptom was memory impairment. The need for assistance with daily activities at the initial physician visit was associated with the firstly reported cognitive symptoms.

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Efficacy of cholinomimetic medications in patients with vascular dementia compared to Alzheimer's dementia

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Background. Vascular dementia (VaD) and Alzheimer's disease (AD) are the two most common causes of dementia overall. While they share some clinical features, key eliciting pathological events are different, and so does the therapeutic response to specific medications.

Aim. The aim of the current study was to compare the progression of cognitive deterioration in patients with AD and VaD while being treated with cholinomimetic and NMDA antagonists.

Methods. Histories of 100 patients were selected and analyzed retrospectively. Cognitive functions were assessed using the MoCA test in the outpatient setting. The observational period was determined by the researchers to be 3 years.

Results. The mean age of 50 patients diagnosed with AD was 71.4 years and 73 years for remaining 50 patients with VaD. The mean MoCA score for the patients with AD at the initial consultation was 12.4 (SD, 3.5), and the same value for VaD was 10.1 (SD, 6.1), at the end of 1st, 2nd and 3rd years – 12.2 (SD, 4.1), 12 (SD, 4.5), 10.3 (SD, 3.2) for AD, and 9.5 (SD, 5.5), 8 (SD, 4.3), 6.2 (SD, 7.1) for VaD, respectively.

Conclusion. Relative decline of MoCA test for VaD was 3 times more prominent during 1st year, 5.25 during 2nd year and 1.85 during 3rd year compared to AD, indicating that the patients with moderate to severe AD remained relatively unchanged in terms of cognitive decline, while the latter was steeper for VaD. This underlies the need to develop new strategies aimed at cognitive deterioration for patients with VaD.

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Impact of primary arterial hypertension and CHD on the level of functioning of patients with primary CNS lymphoma

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Background. Often, patients do not understand the seriousness of primary arterial hypertension and chronic heart disease on the functioning of the whole body, including the development and progress of oncological diseases.

Aim. The aim of the current study was to find out how the development and treatment of primary central nervous system disease is affected by primary arterial hypertension or chronic heart disease known in the anamnesis and to compare the primary ECOG assessment in patients with and without a known cardiovascular system disease in the anamnesis, on which the choice of this oncology therapy further depends.

Methods. This study was retrospective. Considering that primary central nervous system lymphoma is rare against the background of other oncological diseases, the number of respondents was N=49, their median age Md=68 [IQR: 61–75] years, more than half were women N=30 (61.2%). Patient medical histories with the ECOG scale and cardiovascular disease data were used for statistical data analysis, processed in Excel, and analyzed in PSPP. The Mann-Whitney test was used for analysis and data processing.

Results. Mann-Whitney test found a statistically significant (p=0.01) ECOG difference between patient without primary arterial hypertension (Md=2 [IQR: 1–3]) and patient with primary arterial hypertension in anamnesis (Md=3 [IQR: 2–4]), also found a statistically significant (p=0.001) ECOG difference between patient without CHD (Md=1.50 [IQR: 1–3]) and those with CHD (Md=3.50 [IQR: 2.70–4]).

Conclusion. Analysing the statistical data, diseases of the cardiovascular system are an important indicator in the process of disease development, significantly affect the general condition of patients, which is classified using the ECOG scale, and it can be concluded that patients with a history of cardiovascular disease have a higher ECOG scale rating, indicating faster disease progression, minimal treatment options and a poor survival prognosis.

Acknowledgements. The authors declare absence of conflict of interest.

The role of the sentinel lymph node biopsy in melanoma patients: A retrospective single center analysis

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Background. Sentinel lymph node biopsy (SLNB) is a sensitive, minimally invasive operative method for lymph node evaluation and staging in patients with melanoma without clinical and radiological lymph node findings. Precise SLNB identification is crucial for accurate diagnosis of micro-metastasis at an early stage of melanoma progression and treatment initiation.

Aim. A retrospective single center study was performed to compare and analyze the patients who have undergone sentinel lymph node biopsy vs. observation.

Methods. The retrospective study was conducted at the Latvian Oncology Center. 134 patients with histologically confirmed melanoma in the 2015, were involved in the study. Patient data: diagnosis, age, sex, histological parameters, stage, treatment tactics, disease progression were collected. Kaplan-Meier analysis was performed to compare patients who have undergone SLNB vs. observation. The observation time was 7 years. Progression-free survival curves were created by R-studio program.

Results. Patients who have undergone SLNB had shown improving progression-free survival comparing to an observational group. The significance given by Fisher's test is p=0.05, which is on the edge. There was 22% (N=30) of patients who have undergone SLNB and 78% (N=104) patients who have undergone observation only. In SLNB group there was 7% (N=2) SLNB of metastases positive patients and 93% (N=28) of metastases negative patients. There were 11% (N=11) of patients who did not undergo SLNB due to technical reasons.

Conclusions. Data analyses revealed that in necessary cases SLNB was not taken accordingly to NCCN recommendations and there may be several reasons for that. In Latvia Pembrolizumab was included in the list of compensable medications only from 2018 and we suppose that there was no need to perform SLNB, because the treatment was not available. That is why SLNB plays crucial role in cases without clinical and radiological lymph node findings in melanoma patients, therefore we recommend SLNB in this group for making alterations to disease staging and change of treatment strategy. Another possible reason is that Latvian Oncology center's dermato-oncology unit got established in 2019 only.

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Liquid biopsy pattern in non-oncological patients

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Background. Liquid biopsy is an evolving method based on examination of the circulating genetic material in blood. It is mainly used for the diagnosis of malignant tumors and evaluation of treatment effect. However, mutations in circulating blood DNA also are detectable in people without oncological disease. Significance of these mutations is not clear and such result could lead to incorrect clinical decisions. The occurrence of such mutations and the contributing factors for the population of Latvia are also unknown.

Aim. The aim of this study is to determine most common genetic alterations in non-oncological patients and account probable contributing factors.

Methods. This is a prospective study. Patients with various non-oncological lung diseases were selected. Liquid biopsy results were analyzed in correlation with clinical data to clarify the incidence of mutations, identify most common mutations and possible contributing factors.

Results. Biopsy results and clinical data of 76 patients were collected. For 23 patients, the examination was inconclusive. 53 patients were selected (average age 65 years, 60% were males). Most patients were obese (40%) or overweight (30%). 32% of patients were smokers with an average smoking exposure of 25 pack/years. TMB (Tumor mutation burden) was detected in 96% patients with a mean value of 1.26 (0.00, 2.53), indicating low TMB status. The most frequently mutated genes were: DNMT3A (49.1%), ATM (30.2%), TSC1 (26.4%), MSH3 (18.9 %) and CHEK2 (17.0%), while the most common allelic variations were: TSC1 M322T (22.6%), FLT1 S733del (11.3%), JAK2 V617F (11.3%), MSH3 A62_P63insAAAPAA (7.5%) and ZNF703 A401_H402insPTHLGGSSCSTCSA (7.5%).

Conclusions. Study population is overweight and obese, which, together with smoking and greater age can weaken the body's immune system and damage cell's DNA, promoting pathogenesis, including accumulation of DNA mutations. While TMB status is low in all non-oncological patient biopsies, we have identified allelic variations that may be related to promotion of pathogenesis, such as JAK2 V617F and ZNF703 A401_H402insPTHLGGSSCSTCSA and found a positive correlation of smoking with MSH3 mutational status. Multifactorial approach is needed for a more accurate liquid biopsy assessment, incorporating clinical and genetic data.

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Frequency and severity of thrombocytopenia in patients with breast carcinoma during neoadjuvant chemotherapy in 2021

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Background. Bone marrow suppression and chemotherapy-induced thrombocytopenia (CIT) are common complications of chemotherapy. At the same time, thrombocytopenia may have other differential diagnoses that should be excluded. Severe CITs are rare, but in such cases chemotherapy adjustment may be necessary, which could affect both tumour response and subsequent surgical treatment after neoadjuvant chemotherapy.

Aim. The aim of the study was to determine the frequency and severity of thrombocytopenia during neoadjuvant chemotherapy in patients with breast carcinoma.

Methods. After patient selection, altogether 76 patients were enrolled in the study. The selection of patients was carried out using Pauls Stradins Clinical University Hospital Oncology clinic oncological councils in 2021. Then, using the treatment sheets of the selected patients and the DataMed system, the chemotherapy used by the patients and the dates of its administration were determined, combining them with the results of blood tests and specifically changes in the platelet count before each chemotherapy course. After selecting the necessary data, statistical methods were used in program R and the analysis was performed on the first 5 chemotherapy courses to preserve the number of patients as possible.

Results. Thrombocytopenia ($<150\times10^9$/l) was present in patients only in 12 cases, which included 5 patients, meaning that 3 of the patients had 3 or 4 consecutive episodes of thrombocytopenia while the remaining 2 patients each had one isolated episode of thrombocytopenia. Overall changes in platelet count were within normal limits ($150–410\times10^9$/l), however, statistically significant changes in platelet count were obtained in general between chemotherapy courses. Without thrombocytopenia, a statistically significant increase in the platelet count within the normal range was observed between the pre-neoadjuvant chemotherapy platelet count and the platelet count before four next chemotherapy courses.

Conclusions. In general, changes in patients' platelet counts during neoadjuvant chemotherapy remain within normal limits and thrombocytopenia was found only in isolated cases, therefore, to assess the effect of specific chemotherapy drugs on the reduced platelet count, the research groups and study design should be adjusted, and further studies should be conducted. Also, contrary to the aim of the study, a statistically significant increase in the number of platelets between chemotherapy courses was observed, which also would require additional research.

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The importance of histopathological reporting in melanoma patients

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Background. NCCN and CAP guideline principles of pathology include recommendations about the essential and additionally recommended parameters that should be reported to the histological protocol. The reporting of additional parameters can play a crucial role in melanoma patient staging and treatment tactics.

Aim. To determine the percentage of patients having no complete histological information with minimally recommended parameters and to analyze the description frequency of each additional parameter.

Methods. 134 histopathological reports of year 2015 were analyzed during one center retrospective study. The following patient data was collected: diagnosis, age, sex, stage, Breslow thickness, Clark level, ulceration, mitotic rate, margin status, location, lymphovascular and perineural invasion, regression, tumor-infiltrating lymphocytes (TIL), vertical growth phase (VGP), histologic subtype, desmoplasia, disease progression. This data was analyzed with descriptive statistics method using R-studio software.

Results. Complete histological information with minimally recommended parameters was not found in 5% (N=7) of investigated patients. In seven cases the ulceration was not described. The most frequent additionally recommended parameter of histological protocol was location of melanoma found in 99 % (N=133) of patients. While tumor mitotic rate was described in 32% (N=43), lymphovascular and perineural invasion in 45% (N=60); TIL, in 56% (N=75); cutaneous melanoma subtype, in 43% (N=57); cell type, in 78% (N=104); and desmoplasia, in 2% (N=3). Regression and VGP were not documented in any case.

Conclusions. A correct melanoma staging and change of treatment strategy depend on complete and properly performed histopathological protocol and that is why we want to emphasize the importance of additionally recommended parameters. Lymphovascular and perineural invasion, tumor mitotic rate, regression can play crucial role for making a decision about sentinel lymph node biopsy in thin (<1 mm) melanoma cases.

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Compliance of breast and colorectal cancer survivors to participate in digital surveillance activities

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Background. The quality of life of cancer survivors following the completion of the cancer treatment is an important healthcare aspect having been elucidated by the EU Beating Cancer Plan and the EU Cancer Mission Board recommendations. Here we are reporting the preliminary results of an international collaborative project PERSIST, part of which was conducted in Latvia.

Aim. To evaluate the compliance of breast and colorectal cancer survivors to participate in a surveillance strategy by using smart technologies.

Methods. Breast and colorectal cancer patients who have survived curative cancer treatment (surgery±radiation therapy±chemotherapy) 3–12 months before the enrolment.

The participants were offered a smart bracelet that collect health data such as heart rate, blood pressure, steps etc. and a mobile phone with a special mHealth application. Patients were interviewed three times and filled the Questionnaires.

Results. The first patient was enrolled April 2021. Altogether 46 patients were recruited to the study in Latvia Oncology Center, Riga East University Hospital. Among the recruited patients, 24 have had breast cancer and 22 colorectal cancer. The average age of the patients at the time of inclusion was 54 years. There were 7 male and 39 female patients included in the study.

As for July 2022, about one third (30.4%, n=14 patients, 11 women, 3 men) of the patients have stopped their participation in the study. The mean age for this group was 54 years. In total 8 breast cancer and 6 colorectal cancer patients stepped out of the study. The most frequent reason for stopping the participation was difficulty to use device. The remaining patients continue to participate, from whose bracelets data is still being collected.

Conclusion. A significant proportion of survivors are not motivated to participate in health surveillance activities by using digital wearable devices. Difficulty to use the devices and lack of immediate benefits for the patients seem to be the key factors responsible for the situation. Simplification of the device to make it easy-to-use could be among the solutions for the situation.

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PAEDIATRICS

Species-level analysis of core breast milk microbiota of Latvian mothers' milk samples

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Background. Human breast milk microbiota has interindividual variability and is influenced by many factors, but few studies have been done to evaluate it at the species level.

Aim. To analyse breast milk microbiota composition at species level in Latvian mothers' milk samples and analyse the factors influencing it.

Methods. This study is a sub-analysis of breast milk microbiota within a study "Breast milk composition and association with faecal microbiota and influencing factors". Breastfeeding mothers were asked to donate a milk sample of 50 ml and fill out a questionnaire (answering questions about pregnancy, lactation, anthropometric data, illnesses, antibiotic use). Totally, 46 milk samples were analysed. Mothers' mean age was 28 years (SD, 3.54), and their infants' mean age was 4.7 months (SD, 1.45); 53% (20/38) of infants were males, and 47% (18/38) were females. Bacterial taxonomic units were determined by isolating DNA from milk samples and performing 16S gene sequencing. Mean abundance of bacterial taxonomic units up to species level was analysed. Statistical analysis: descriptive statistics; Spearman correlation.

Results. Ten most abundant bacterial genera (mean±SD) to which the most common species belong were *Bifidobacterium* (0.42±2.09), *Rhodococcus* (0.19±0.25), *Actinobacteria* (0.18±1.18), *Pseudomonas* (0.13±0.18), *Stenotrophomonas* (0.12±0.17), *Acinetobacter* (0.06; ±0.21), *Brevibacterium* (0.05; ±0.09), *Enterobacteriaceae* (0.04±0.12), *Bacillus* (0.03±0.10), and *Brevundimonas* (0.02±0.04). Mean abundance of *Staphylococci* was 0.09±0.003.

The most often identified species were *Bifidobacterium longum*, *Bifidobacterium breve*, *Streptococcus thermophilus TH1435*, *Streptococcus salivarius subsp. salivarius*, *Ruminococcus* spp., *A254*, *MGS-108*, *Acinetobacter ursingii NIPH 706*.

The abundance of *Bifidobacterium* correlated with *Lactobacillus*. The abundance of *Brevibacterium* correlated with *Pseudomonas luteola* ($p<0.0036$), and *Pseudomonas psychrophila* ($p=0.0001$). *Bacillus* correlated with *Staphylococcus* ($p=0.0001$), and *Streptococcus* ($p<0.0001$). There was also a correlation between *Actinomyces* and unidentified bacteria ($p<0.0001$).

Conclusions. The most abundant bacterial genera in our sample – *Bifidobacterium*, *Pseudomonas*, *Stenotrophomonas*, *Acinetobacter*, *Enterobacteriaceae*, *and Brevundimonas* – have been previously reported as being part of predominating genera of human breast milk microbiota, while neither *Staphylococcus* nor *Streptococcus* were identified among the ten most abundant entities in our sample.

Factors, associated with increased abundance of *Rhodococcus*, observed in our human milk samples, should be studied further.

Certain correlations between bacterial genera and species suggest interaction of specific taxa and factors modulating bacterial composition that should be analysed in multifactorial analysis.

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Vitamin D and total IgE in childhood bronchial asthma in Latvian population

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Background. Low circulating vitamin D (VD) levels have been associated with a risk of asthma and elevated immunoglobulin E (IgE), therefore it may represent a marker of other factors that may lead to increased asthma severity. Genetic variants in genes related to VD have also been studied as potential contributors to autoimmune and allergic diseases.

Aim. To determine the relationship between genetic variations in the vitamin D receptor (*VDR*) gene, vitamin D, and total IgE levels in blood serum in a BA childhood cohort and in healthy individuals.

Methods: The examined cohort included 72 children diagnosed with asthma; the control group included 48 healthy individuals. VD and IgE levels in blood serum were determined by means of enzyme-linked immunosorbent assay (ELISA). *VDR* gene SNPs (rs731236 and rs1544410) were genotyped in 148 cases and 253 controls from the Latvian population using the restriction enzyme site polymorphism method. Statistical analysis was performed with SPSS.25 Statistical Package.

Results: The level of vitamin D was below normal (<20 ng/ml, Lips et al., 2019) in 32% of BA patients and only in 15% of healthy individuals; it was found to be statistically significant ($p=3.16\times10^{-2}$) associated with vitamin level supplementation among patents, divided into groups with <20 ng/ml (deficiency) and >20 ng/ml (normal). The level of IgE in 23% of BA patients does not correspond to the norm and significantly differs between the BA groups and the Control group ($p=3.94\times10^{-4}$); IgG levels that do not correspond to the norm were almost twice as common in the male patients as in female (21.21% and 40%, respectively).

In the BA cohort, a statistically significant association was found between IgE levels and *VDR* gene rs731236 and rs1544410: associative analysis revealed two protective heterozygous genotypes: rs*731236* (TC, $p=3.95\times10^{-3}$) and rs1544410 (GA, $p=0.075$) and two risk homozygous genotypes for common alleles: rs731236 (CC, $p=2.47\times10^{-3}$) and rs1544410 (AA, $p=4.99\times10^{-2}$)

Conclusions: no correlation was found between vitamin D and IgE levels in BA and Control groups. A case/control study of selected *VDR* genetic polymorphisms illustrates the potential of using them as possible genetic markers associated with BA in the Latvian population.

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Is it possible to anticipate the type of acute appendicitis in children without extensive testing?

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Background. Over recent years acute appendicitis has been shown not to always progress towards perforation as in cases of complicated disease, but sometimes have the possibility to resolve in uncomplicated acute appendicitis, thus prompting that an operation may not always be needed. However, we have noticed that some cases exist, where the gangrene and perforation seem to happen very rapidly, which in turn requires prompt surgical treatment and this has not yet been addressed in literature.

Aim. We aimed to see if we can predict if patients with rapidly progressive complicated acute appendicitis can be anticipated in children, as well as cases where the disease is uncomplicated and could have the possibility to resolve only using routinely performed and readily available tests.

Methods. We retrospectively analyzed patients' records from the Pediatric Surgery departments in two hospitals with surgically treated acute appendicitis over a 15-year period. The patients' groups were formed according to type of appendicitis (uncomplicated or complicated) and duration of disease: the early (duration of symptoms <24 hours) and the late (>24 hours duration). Phlegmonous appendicitis was categorized as uncomplicated appendicitis. Gangrenous or perforated appendicitis was categorized as complicated. Patient anamnestic data and routinely performed tests were analyzed. The early uncomplicated appendicitis (A1) group was compared to the late uncomplicated (resolving) appendicitis (A2) group. Also, the early complicated, or as we called it, rapidly progressive acute appendicitis (B1) group was compared to the late complicated appendicitis (B2) group.

Results. Overall, 456 patient records were analyzed. We found that the resolving A2 group in comparison to A1 showed a lower median white blood cell count 12.3 [3.9; 34.3] vs. 16.5 [4.1; 29.5], $p<0.0001$, neutrophil percentage 74.6 [38.7; 91] vs. 83.8 [42.7; 94.6], $p<0.0001$ and neutrophil to lymphocyte ratio (NLR) 4.3 [0.8; 24.7] vs. 9.6 [0.9; 33.7], $p<0.0001$ but a higher CRP 20 [0.9; 177] vs. 5 [0.7; 227], $p<0.0001$. In the complicated groups' comparison, we found that rapidly progressive B1 showed a higher median basophil count 0.068 [0; 0.629] vs. 0.017 [0; 0.151], $p<0.0001$, but a lower CRP 17 [0.9; 155] vs. 35.5 [4.2; 431.3], $p=0.003$. There were no other differences in anamnestic and demographic data in the groups.

Conclusions. Resolving acute appendicitis appears to show signs of deteriorating inflammation by a lower white blood cell count, neutrophil percentage, and neutrophil to lymphocyte ratio, while CRP stays elevated. Rapidly progressive acute appendicitis on the other hand could be suspected with an early higher basophil count.

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When can we anticipate multisystem inflammatory disorder in children with symptoms of acute appendicitis?

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Background. Multisystem inflammatory disorder (MIS-C) currently presents a diagnostic challenge to suspect it early, especially in patients who present like acute appendicitis.

Aim. We aimed to compare patients with MIS-C and patients with regular acute appendicitis and those with a positive COVID-19 test at the time, and find factors which can help suspect MIS-C.

Methods. We analyzed patient data from over a six-month period in 2021 of patients admitted to the PICU, Pediatric Department of Pediatric Surgery Department, and selected patients operated for acute appendicitis as well as unoperated MIS-C cases. Patients were categorized into 4 groups: Group A- patients with acute appendicitis; Group B – patients operated for acute appendicitis with a later confirmed MIS-C, Group C – either solitary MIS-C cases or patients operated for acute appendicitis found to have minimal or no inflammatory changes on pathology with later confirmed MIS-C and into group D- patients operated for acute appendicitis with a positive COVID-19 PCR antigen test on admission or during the stay at the hospital. We compared demographic values, symptoms, other organ system involvement and laboratory test values to find the most prominent differences between these groups. The most prominent differences were added into a makeshift score to see how it could help differentiate MIS-C cases from appendicitis cases.

Results. Overall, 76 patients were analyzed: 36 in Group A, 6 in Group B, 29 in Group C, and 5 in Group D. The most significant differences were found in duration of disease (1.4, 4.5, 4, and 4 days in Groups A, B, C and D, respectively, $p<0.0001$), CRP values (19.3, 112.5, 143.8 and 141 mg/l, respectively, $p<0.0001$), presence of febrile fever (13.9%, 66.7%, 96.6% and 40%, respectively, $p<0.0001$) and other system involvement (0%, 100%, 100% and 20%, respectively). Other differences were found in symptoms, WBC, platelet count, hemoglobin and pediatric appendicitis score. A makeshift score was made using the most significant differences and found to predict MIS-C in a ROC curve with an AUC=0.983 ($p<0.0001$) and a sensitivity of 94.3%, a specificity of 92.7% when at least 3 criteria were met.

Conclusions. Our current data suggests that MIS-C could be suspected if patients with acute appendicitis symptoms have at least 3 out of 4 symptoms: a CRP count higher than 55.8 mg/l on admission, a fever of at least 38 degrees Celsius, show other organ system involvement and have symptoms lasting at least 3 days.

Acknowledgements. This study was conducted without funding and presented no conflict of interest.

Characteristics of preschool children with functional constipation

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Background. Functional constipation is a functional gastrointestinal order that is often observed in preschool children. Although most common causes of constipation are dietary mistakes, variety of emotional aspects are also important factors in the etiopathogenesis of functional constipation. An attendance of kindergarten is also supposed as an important emotional factor.

Aim. To analyze clinical characteristics of children with functional constipation and to compare the data in relation to attendance of kindergarten.

Methods. Retrospective cross-sectional study was performed, selecting children (aged 3–8 years) who were undergoing investigation and treatment in Children Clinical University Hospital (Riga) due to constipation (SSK K59.0) during 2017.

The following data were analyzed: age, gender, hospitalization history, kindergarten attendance (yes/no), side diagnoses (yes/no); clinical tests; prescribed treatment and recommendations.

Statistical analysis: descriptive statistics; chi^2 test.

Results. In total, 105 children (51.4% of girls) with the mean age of 4.47 years (SD, 1.0837) were included in the study. Out of them, 74.3% were hospitalized due to constipation for the first time; in 56% of patients, constipation was the main diagnosis. Majority of children (81.7%) were attending kindergarten. In total, 42% (44/105) of children had some abnormalities in clinical tests; 10.5% (n=11) had changes in allergy tests.

Overall, prevalence of deviations of clinical tests/investigations did not differ between children attending kindergartens or not. However, children who did not attend kindergarten had more often constipation as a side/secondary diagnosis compared to children who attended kindergarten: 73.7% (14/19) vs. 37.6% (32/85) (p=0.004). Children attending kindergartens had non-significantly more often changes in urine analysis compared to children who did not attend kindergarten: 5% (4/85) vs. 0% (0/19) (p>0.05).

The main treatment prescribed was macrogolum (n=20); only 11 patients were treated with enema. Dietary changes were recommended for all patients at discharge, while psychological help for treating of constipation was advised only in 10% cases.

Conclusion. A relatively large number of children with functional constipation had some deviations in clinical investigations, raising questions about the interpretation of the definition of functional constipation.

Functional constipation seems to be also a problem in severely ill children. In primary care more attention could be paid to possible urinary tract infection among children with constipation.

Although treatment corresponds to the latest guidelines for the treatment of functional constipation, psychological treatment possibly should be recommended more often.

GASTROENTEROLOGY AND GASTROINTESTINAL ONCOLOGY

Prognostic criteria for the course of GERD

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Background. The prevalence of GERD is steadily increasing, since 1995 there has been a 50% increase in patients worldwide.

Aim. The aim of the study is to identify the correlation between clinical and pathogenetic variants of GERD with age, gender, and risk factors.

Methods. A total of 69 patients with gastroesophageal reflux disease were examined. In all patients, the diagnosis of gastroesophageal disease was confirmed endoscopically. The analysis of obtained data was performed on *easymedstat.com* to identify the relationship between several variables and correlation.

Results. The study included patients with GERD aged 19 to 83 years; the majority of patients were older than 50 years (86%). In the study, GERD was more often diagnosed in women over 50 years of age, in the menopausal period, the median age was 58 years. Erosive reflux disease was detected in 43.5%, and non-erosive reflux disease (NERD) was detected in 56.5%. NERD is more common in women. There was a high positive correlation between erosive esophagitis and age (0.589) and concomitant gallbladder diseases (0.725), a high positive correlation between erosive esophagitis and frequent coffee consumption (0.546) and treatment of hypertension with calcium channel blockers (0.628). Non-erosive gastro-esophageal reflux disease revealed a high positive correlation with the age (0.529) of patients and concomitant gallbladder diseases (0.702). A high positive correlation was found between NERD with smoking (0.512) and frequent consumption of fatty foods. No correlation was found between GERD and HP.

Conclusion. GERD is more often detected in women in the menopausal period, which suggests the role of estrogen in the development of the disease and suggests that the postmenopausal period increases the risk of GERD. A high positive correlation was revealed between erosive esophagitis and age (0.589), concomitant gallbladder diseases (0.725), frequent coffee consumption (0.546), and treatment of hypertension with calcium channel blockers 2+ (0.628). Patients with GERD showed a high positive correlation with age (0.529), concomitant gallbladder diseases (0.702), smoking (0.512), and frequent consumption of fatty foods.

Macroscopic and histological findings in patients with type I diabetes mellitus and indications for endoscopic gastrointestinal examination, preliminary data

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Background. Diabetes mellitus is associated with various gastrointestinal complaints, but their pathophysiology in type I diabetes (T1DM) is not clear enough.

Aim. The aim of this study was to analyse histopathological findings of biopsy material obtained during endoscopy investigations of patients with T1DM and upper and lower gastrointestinal (GI) manifestations.

Methods. A total of 21 T1DM patients with GI symptoms according to questionnaire and/or elevated faecal calprotectin (>50 μg/g) underwent upper GI endoscopy and/or colonoscopy. Endoscopic biopsy specimens were examined by histopathologist. *H. pylori* was identified by rapid urease test (RUT) and histological staining (Giemsa).

Results: 14 (67%) female and 7 (33%) males with T1DM were included. The indications for endoscopic examination were abdominal pain (n=17; 81%), bowel movement disorders (n=9; 43%) and elevated faecal calprotectin (>50 μg/g) (n=5; 24%).

10 individuals underwent upper GI endoscopy, 7 (70%) of them had abnormal macroscopic findings and were analysed for *H. pylori* infection by RUT. None of the patients had positive RUT, however histopathology identified 2 patients with *H. pylori* visible on special stain. Most of the gastric lesions were minor endoscopic findings: hyperaemic gastropathy (n=4, 40%), hyperaemic duodenopathy (n=1, 10%), oesophageal candidiasis (n=1, 10%) and gastric intestinal metaplasia (n=1, 10%). Of the 5 endoscopically reported hyperaemic gastropathy and duodenopathy patients, two had active gastritis and two, chronic atrophic gastritis. Of the three patients with macroscopically normal upper GI endoscopy, one had chronic atrophic gastritis and one had active gastritis with erosive gastropathy.

Five (24%) of the colonoscopies performed showed abnormal findings, the main were polyps (n=2, 9.52%), diverticulosis (n=1, 4.76%), erosion of sigmoid colon (n=1, 4.76%) and perianal papilloma (n=1, 4.76%). The most reported histopathological findings were eosinophilic infiltration (n=8, 38%), lymphoid follicles/lymphoid aggregates (n=7, 33%) and lymphoplasmacytic infiltration (n=5, 24%). Other histopathological findings were lymphocyte and macrophage infiltration (n=3, 14 %), stromal fibrosis (n=3, 14%), mononuclear cell infiltration (n=2, 9.5%) and tubular adenomas (n=2, 9.5%). Active inflammation was found in a total of 6 (29%) cases, and 1 (4.76%) patient had active colitis on histopathological examination. No malignancies were found.

Conclusion. The most frequent upper GI endoscopic and histopathological findings were hyperaemic gastropathy, but during colonoscopies – polyps. Eosinophilic infiltration was the most common histopathological colonoscopic finding, but without confirmed clinical diagnosis.

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Breath fingerprint preliminary results of colorectal based on the gas chromatography-mass spectrometry analysis

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Background. Effective screening for colorectal cancer can reduce mortality by early detection of tumours. An altered pattern of volatile organic compounds (VOCs) in exhaled breath has been proposed as a potential noninvasive diagnostic tool for detection of cancer.

Aim. The aim of this task was to characterize the VOCs chemical patterns associated with breath of colorectal cancer patients and controls.

Methods. Altogether 141 study subjects were enrolled in the study: 56 colorectal cancer and 85 control patients. Breath samples were collected in clean rooms free from chemicals, cleaning agents, drugs, solvents or kitchen waste, with stable temperature to avoid as much as possible the influence of the external factors, including hospital contaminants. Prior to the sampling the study subject were advised to following an overnight fast, refraining from smoking, alcohol consumption, using chewing gum, and avoiding physical activities at least 2 hours prior to sampling. Breath samples were taken using an in-house made breath sampler. The breath sampler consisted of a disposable mouthpiece (Intersurgical, 1931000) installed on a disposable elbow (Intersurgical, 2714000S) and the CO2 sensor cell (Masimo, IRMA), USA) connected to the other end of the elbow. A two-stage thermal desorption of VOCs was performed using a commercial thermal desorber (UNITY, Markes International Limited, UK) and autosampler (TD100, Markes International Limited). The VOCs analysis relied on an Agilent 7890A/5975C GC-MS system (Agilent, USA).

Results. Preliminary results have been obtained. It was found that the following volatile organic compounds in breath are elevated in patients with colorectal cancer: two ketones (2-pentanone and 6-methyl-5-hepten-2-one), two aromatic compounds (p-xylene and ethylbenzene), three alcohols (1-octanol, 1-decanol and 2-ethyl-1-hexanol), three hydrocarbons (decane, dodecane and tridecane) and two aldehydes (hexanal and decanal).

Conclusion. These compounds can be potentially responsible for the breath fingerprint changes related to colorectal cancer. The educated detection of these changes can support the development of non-invasive tests for future early detection of colorectal cancer. Further analysis is needed to verify possible markers of colorectal cancer and gain better insight into their origin in the human body.

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OPHTHALMOLOGY

Determination of association between cardiovascular disease and glaucoma using OCT-angiography

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Background. Glaucoma is irreversible and the leading cause of blindness in the world. New methods of diagnostics should be developed, as 50% of cases of glaucoma stay undiagnosed. Arterial hypertension is a chronic disease, which affects not only the heart and kidneys, but is also associated with glaucoma. Optical coherence tomography angiography (OCT-A) is a new, noninvasive, and fast technology, which allows visualizing retinal and papillary vascularization. In previous research, it has been concluded that in the case of glaucoma, OCT-A shows decreased vascular density.

Aim. To investigate the correlation of mean arterial pressure and macular and peripapillary microvasculature changes detected by OCT-A in patients with glaucoma and cardiovascular disease.

Methods. A total of 58 glaucoma patients – 38 with cardiovascular disease and 20 control group patients – were enrolled in this study. OCT-A (Angiovue Imaging System, Optovue Inc.) was done for each patient, and correlations of measurements of radial peripapillary capillary (RPC), macular vessel density (mVD), and mean arterial pressure were compared among groups. Vessel density was calculated using Angiovue Angiometrix software.

Results. Analyzing measurements of macular superficial capillary density, it was the highest in control group (46.3%), but lowest in PAH (45.0%) and PAH with diabetes group (45.5%) (p=0.031). Deep mVD was the highest in control group (50.7%), but the lowest in the PAH group (48.7%) (p=0.024). A statistically significant difference was concluded when comparing measurements of deep mVD in patients with cardiovascular disease (48.5%) and the control group (50.0%) (p=0.007). The thickness of the ganglion cell layer was higher in control group than in the cardiovascular disease group, but not statistically significant (p=0.407). Measurements of mean arterial pressure were the lowest in the control group (114.0 mmHg), but the highest in PAH and DM II group (120.0 mmHg), but the difference was not statistically significant (p=0.186).

Conclusion: High arterial pressure has an impact on superficial macular vessels. Adequate and regular control of arterial pressure is essential in patients with primary arterial hypertension, and even more critical in patients with PAH and diabetes, as the retinal microvasculature density decreases with increasing mean arterial pressure, which could lead to glaucoma progression. OCT-A can detect microstructural defects and offers the potential to facilitate the diagnosis of glaucoma. Longitudinal studies must elucidate the relationship between retinal microvasculature dropout and systemic and ocular factors.

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