1653 doses of vaccines were injected. The three- dose schedule was given to 414 (66%) girls and 2-dose schedule to 198 (31.6%) girls. Vaccine safety assessment was carried out. Adverse effects were observed in 9.6% of cases and were mainly characterized by dizziness and pain at the injection site. Vaccination was well tolerated.

When calculating socio-economic feasibility of the proposed technology , not only the economic damage caused by the high mortality of women from cervical cancer, but also the cost for treatment of precancerous cervical lesions were taken into account, as out of 25 women with undetected CINII-III, 10 will develop cervical cancer. There have been calculated the estimated damage from cervical cancer in the Tomsk region, which takes into account not only the cost of diagnosis and treatment of cervical pre-cancer, but also losses associated with temporary permanent disabilities (social benefits, including disability pension before the age of 55 years). Calculations show that the total economic damage caused by the management of patients with cervical cancer, can be from 20 to 40 mln. rubles per year.

Thus, primary prevention of cervical cancer, taking into account the prevalence of HPV infection and economic impact of cervical cancer can be considered as an effective technology for public health that will allow preservation of not only reproductive but also employment potential of women of Tomsk region.

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P49

Macrophages with JAK2V617F mutation are activated by platelets: Role in pathogenesis of myelofibrosis

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Background: Myelofibrosis with myeloid metaplasia is chronic mieloproliferative disease with ineffective erythropoiesis, dysplastic-megakaryocyte hyperplasia, and an increase in the ratio of immature granulocytes to total granulocytes. The incidence rate is 3.7–5.7 per 100,000 and the median survival is estimated to be between three and six years. The pathogenesis of myelofibrosis might be explained, in part, by a somatic point mutation on exon 14 (V617F) of the JAK2 kinase gene that is located on chromosome 9p24. However, mechanisms of regulation of microenvironment in bone marrow fibrosis are not clear. Considered that main producents of pro-fibrotic factors are the megakaryocytes and the macrophages. In this study we investigated influence of platelet factors on the cellular characteristics of the macrophages with oncogenic mutation JAK2 V617F.

Materials and Methods: THP-1 cell line was modified by lentiviral modification. Two cell lines established: one with expression of JAK2 with oncogenic mutation JAK2 V617F, another wild type JAK2. Cells were cultured in RPMI-1640 containing 0.1% gentamicin, 1% l-glutamine and 10% fetal bovine serum (FBS) or 5%, 10% and 15% platelet lysate (PL). Cell lines were differentiated into the macrophages using 50 ng/ml phorbolmyristate acetate (PMA). Level of expression transforming growth factor β (TGFβ), galectin-3, matrix metalloproteinases (MMP) 2, 9, 12, 13 and tissue inhibitors of matrix metalloproteinase (TIMP) accessed by specific RT-qPCR.

Results: We observed increased expression level of galectin-3, MMP-2, MMP-13 and TIMP-3 in JAK2 V617F expressing macrophages cultured with 10% FBS compare to ones with WT JAK2. Also macrophages containing mutation JAK2 V617F has increased level of expression of MMP-2, TIMP-3 and TIMP-4 when cultured with 10% platelet lysate. Also, cell line containing mutation JAK2 V617F has increased levels of expression of MMP-12 for cells cultured with 15% PL. We did not observe any significant difference in expression of TGFβ, MMP-9, TIMP-1 and -2.

Conclusion: Increased levels of expression of galectin-3, MMP-12 and -13 suggest that platelet factors induce macrophages with JAK2 V617F mutation to release of profibrotic factors. On the other hand, increase of antifibrotic MMP-2 and TIMP-3 revealing complex nature of melofibrosis development and need to be analyzed further.

This study confirms the idea of interaction between macrophages and platelets in pathogenesis of primary myelofibrosis.

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T145

Comprehensive flow cytometry tracking of regulatory T cells and other lymphocyte subsets during HD IL-2 therapy for melanoma

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High dose IL-2 (HD IL-2) has been extensively used as an immunotherapy against metastatic melanoma However, why HD IL-2 is effective only in a subset of patients and whether predictive biomarkers, before or early during the course of therapy, can be used to improve response rates remain unresolved. In addition, it has been found that IL-2 therapy potently expands CD4+CD25+Foxp3+ T-regulatory cells (Tregs) but how Treg cell levels, phenotype, and function change and whether specific subsets of Tregs are activated and expanded during HD IL-2 therapy is remain unclear. In this study, we performed comprehensive multi-parameter FACS analysis of patient blood before and two days after the last bolus of IL-2 infusion during cycle 1 of HD IL-2 therapy. Two lymphocyte subsets were found to expand the most during the first cycle of IL-2 therapy: CD4+CD25+Foxp3+ Tregs expressing an activation marker, inducible costimulator (ICOS), and CD3-CD56hiCD16loPerforin+ NK cells. ICOS+ Tregs expressed significantly higher levels of CD25, Foxp3 and had a more activated phenotype than ICOS− Tregs as indicated by lower levels of CD45RA and CD127 expression. Further phenotypic characterization revealed a more suppressive phenotype on ICOS+ Treg with higher expression levels of CD39, CD73, and TGF-β/LAP than ICOS− Treg. ICOS+ Tregs were also the predominant Treg cells that secreted IL-10 and have potent T-cell suppressor function. Majority of ICOS+ Tregs from HD IL-2-treated patients were Ki67+ and exhibited an enhanced proliferative response to

IL-2 ex vivo relative to ICOS− Tregs. Functional analysis revealed that ICOS+ Tregs secreted little IFN- and IL-2 in comparison to CD4+Foxp3 – cells. Furthermore, analysis on 38 IL-2-treated patients at MD Anderson, we found that non-responders had a significantly higher degree of ICOS+ Treg expansion than responders during the first cycle of IL-2 therapy, while no significant changes in the ICOS− or bulk Treg population. In conclusion, our data suggests that tracking changes in ICOS+ Tregs early during the course of HD IL-2 therapy may be a new predictive biomarker of clinical outcome.

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P48

Integrated analysis of genomic and transcriptomic data in clear-cell renal cell carcinoma

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Renal cell carcinoma (RCC) is the most widespread kidney tumor, which originates mostly from distal kidney tubules. RCC is the major mortality cause in excretory system cancer in adults, and constitutes about 80% of various kidney cancers. The 5-year survival rate is 60–70%, but lowers significantly in case of metastasis formation. The tumor is relatively resistant to chemotherapy and radiation therapy, but responds to immunotherapy. Target drugs (e.g. sunitinib, bevacizumab, α-interferon, sorafenib) are more preferable in RCC therapy. In most cases RCC is caused by obesity, smoking, arterial hypertension and hereditary factors.

4 patients of Voronezh Regional Clinical Oncology Center, aged from 50 to 75 years old were enrolled in this study. All were diagnosed with renal cell carcinoma confirmed by immunohistochemical analysis.

Gene expression profiling was performed using Affymetrix GeneAtlas system with Affymetrix Hunam Gene 1.1 ST DNA-microarrays. Data moralization, statistical analysis and differentially expressed genes (DEG) list creation were performed using Partek Genomics Suite v. 6.6. DNA samples were sequenced on Ion Proton sequencer with Comprehensive Cancer Panel primer pool. Subsequent pathway analysis and biological interpretation were conducted using Ingenuity Pathway Analysis system and Ion Reporter Suite.

3528 genes were differentially expressed with expression change more than 2 times, and 351 genes changed their expression more than 5 times (p-value <0.05).

Only mutations causing either amino-acid substitution in corresponding protein, open reading frame shift, or truncated transcript formation were taken into account. Targeted DNA sequencing revealed 99 common mutations in normal tissues of all test subjects. 14 mutations were localized in SDHB, TRIM33, PDE4DIP, PBX1, ABL2, MTR, VHL, ROS1, PRKDC, CSMD3, MLLT10, TRIP11, PER1, genes and were tumor-specific in all patients. 33 mutations were localized in promoter regions of EGFR, PDGFRA and HNF1A genes.

The most representative metabolic and signaling pathways according to Ingenuity knowledge base were "FXR/RXR Activation", "Atherosclerosis Signaling", "LXR/RXR Activation", "Production of NO and ROS in Macrophages", "Cell migration and adhesion". Five most downregulated genes among all patients were CALB1 (−187), HPD (−133), KNG1 (−126), SLC36A2 (−126) and PAH (−122). Five most upregulated genes were TNFAIP6 (+34), ANGPT2 (+23), SERPINE1 (+22), CP (+20), HILPDA (+20).

Genes with mutations and differentially expressed genes were simultaneously included in pathways analysis in order to generate gene networks with possible upstream regulators (mutated genes) included. This allowed observing a number of gene interactions not present in existing reports.

This method of genomic and transcriptomic data integration allowed us to determine the sources of mRNA level variation. A number of mutations and specific DEGs discovered in this study are not registered in existing databases of annotated mutations, which allows us to propose population heterogeneity of RCC causes. These DNA mutations and mRNA level changes could be used as predictive biomarkers of renal cell carcinoma.

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A29

Melanoma B16F10 causes redistribution of bone marrow cells on model of tumor–host interaction

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Background: Cancer cells attractive and activate many non-tumoral cells, including bone marrow-derived cells, as a result stimulate migration of this cells. Many studies demonstrate that tumor causes active redistribution of bone marrow cells during specific stages of cancer progression and metastasis. However, to date how the bone marrow derived cells at primary sites is hijacked to support tumor growth and how tumor progression influence of bone marrow cells distribution is not understood.