Questionnaire

Summary of the main activities of a scientific Organisation of the Slovak Academy of Sciences

Period: January 1, 2003 - December 31, 2006

I. Formal information on the assessed Organisation

1. Legal name and address

Institute of Molecular Physiology and Genetics
Slovak Academy of Sciences
Vlárska 5, SK-833 34 Bratislava
Slovakia

2. Executive body of the Organisation and its composition

Directoriat	name	age	years in the position
director	Ing. Albert Breier, DrSc.	50	1998
deputy director	RNDr. Ľubica Lacinová, DrSc.	48	2002
	doc. Oľga Križanová, DrSc.	47	2006
scientific secretary	Ing. Dagmar Zbyňovská, CSc.	42	2002

3. Head of the Scientific Board

RNDr. Ivan Zahradník, CSc. 2003 - 2004
Doc. Ing. Oľga Križanová, DrSc. 2005 - 2006

4. Basic information about the research personnel

4.1 Number of employees with a university degree (PhD students excluded) engaged in research and development and their full time equivalent work capacity (FTE) in 2003, 2004, 2005, 2006 and average number during the assessment period

4.2 Organisation units/departments and their FTE employees with the university degree engaged in research and development

Research staff	2003		2004		2005		2006		average	
	No.	FTE	No.	FTE	No.	FTE	No.	FTE	No.	FTE
organisation in whole	38	25.9	40	26.5	34	24.8	41	24	38.25	25.3
Laboratory of Biophysics	1	1	1	1	3	2	3	2.3	2	1.575
Laboratory of Biochemistry of Transport Systems	3	2.4	5	3.2	5	3.9	5	2.4	4.5	2.975
Laboratory of Cell Morphology	2	1.5	2	2	2	2	3	2.3	2.25	1.95
Laboratory of Electrophysiology	4	3.4	4	3.2	4	3.1	4	2.9	4	3.15
Laboratory of Biochemistry and Cytochemistry	3	3	4	3.3	4	3.6	4	4	3.75	3.475
Laboratory of Genetics	4	2.9	4	2.9	2	2	3	2	3.25	2.45
Laboratory of Protein Chemistry	6	4.9	6	4.9	4	3.8	4	2.2	5	3.95
Laboratory of Molecular Biophysics	2	1	1	1	1	1	1	1	1.25	1
Laboratory of Intracellular Ion Channels	5	4.2	5	3.6	4	3.2	2	1.4	4	3.1
Laboratory of Ion Channel Function	0	0	0	0	0	0	2	2	0.5	0.5
Integrated Team	8	1.6	8	1.4	5	0.2	10	1.5	7.75	1.175
	38	25.9	40	26.5	34	24.8	41	24	38.25	25.3

5. Basic information on the funding

5.1 Total salary budget[1] of the Organisation allocated from the institutional resources of the Slovak Academy of Sciences (SAS) in 2003, 2004, 2005, 2006, and average amount for the assessment period

Salary budget	2003	2004	2005	2006	average
total salary budget (millions of SKK)	9.955	10.380	10.806	11.369	10.628

6. URL of the Organisation's web site

http://www.umfg.sav.sk/

[1] Sum of the brutto salaries without the fund contributions.

II. General information on the research and development activity of the Organisation

1. Mission Statement of the Organisation as presented in its Foundation Charter

Scientific orientation is focused on the molecular basis of elementary physiological functions, with main orientation on cardiac muscle physiology, membrane transport and genetics.

Modern methods of research now employed at the institute often have been introduced as first or unique in our country (microelectrode techniques, voltage clamp, patch clamp, planar lipid bilayers, isolated single skeletal and cardiac muscle cells, cell culturing, optical methods of intracellular ion detection, electron microprobe analysis of elemental compositions in cellular compartments, fast freezing techniques for electron microscopy, PCR techniques, monoclonal antibody production, radioisotope techniques, etc.). The institute introduced the method of DNA fingerprinting in Slovakia and it is currently being used in forensic practice.

The institute, although established as pure basic research facility, engages also in education, mainly at undergraduate and graduate level. Institute is publisher of the physiologically oriented journal - General Physiology and Biophysics.

2. Summary of R&D activity pursued by the Organisation during the assessed period, from both national and international aspects and its incorporation in the European Research Area (max. 10 pages)

Institute of molecular physiology and genetics pursues its research in various closely related fields concentrating on the basic properties of living cells. Laboratory of genetics deals mostly with human genetics describing genetics basis of inheritable diseases in Slovak population. Laboratory of biochemistry and cytochemistry and Laboratory of protein chemistry concentrate mainly on subcellular mechanisms underlying emergence of multidrug resistance in cancerous cells. Laboratory of biochemistry of transport systems applies contemporary methods of biochemistry and molecular biology to assessment of expression and regulation of various cellular transport proteins and signaling cascades under physiological and pathophysiological conditions. Laboratory of biophysics, Laboratory of electrophysiology and Laboratory of molecular

biophysics analyze function of cellular transport proteins, particularly ion channels and exchangers, using physical methods, mostly ion currents measurements and fluorescence assays assessing concentrations of free intracellular ions. Laboratory of intracellular ion channels and Laboratory of ion channel function complement all other groups by analysis of the function of ion channels incorporated into membranes of intracellular organelles like mitochondria or sarcoplasmic reticulum using bilayer lipid membrane method. Finally, Laboratory of cell morphology and Laboratory of biochemistry and cytochemistry use methods of electron microscopy and immunohistochemistry to assess cellular ultrastructure and subcellular localization of proteins of interest. All laboratories collaborate with other laboratories within the institute, with other Slovak research and educational institution, with institutions in EU member states and with institutions outside of EU. Individual laboratories were recipients of bilateral international grants from funding institutions in USA, Germany and France and several multilateral European grants within 6th framework program. The institute maintains also collaboration with newly independent states, e.g., Ukrainian, which may be in the future associated members of European Union and should be integrated into European research area. International recognition of their work can be expressed in terms of number of papers published in international peer reviewed journals and in terms of citations referring to papers authored by members of individual laboratories.

The research activity of **Laboratory of genetics** is concentrated on the study of human genome at the DNA level with special emphasis on the regions, which are involved in inherited pathologies frequent in the population of Slovakia. Results from this laboratory contribute to the general knowledge on the structure and organization of human genome and to molecular pathology of inheritable disorders. Practical outcome represent development of diagnostic tests based on direct mutation detection for purposes of more effective diagnostics, prevention, and treatment. Part of this work was carried out in frames of international consortium within European Research Area. In the evaluated period main attention was focused on the following monogenic disorders: cystic fibrosis, Huntington's disease, alkaptonuria, nonsyndromic hearing loss and retinitis pigmentosa.

For the clinical phenotype of cystic fibrosis is characteristic a broad range of variability in terms of disease severity. In frames of an international consortium we were involved in studying the role of TG polymorphism within *CFTR* gene on this variability. Our results shows that individuals with 12 or 13 TG repeats are more likely to exhibit an abnormal phenotype than those with 11 repeats. This result represents direct evidence that, structural elements within the gene, even if not coding (localization in introne) can impact significantly gene expression.

Huntington's disease also shows considerable variability in terms of the age of onset of first symptoms, ranging from early childhood to late adulthood. The variability of the length of disease causing expansion of CAG repeat in IT-15 gene explains only partially (40 – 70 %) this broad range. In frames of an international consortium we were involved in studying the modifying role of several candidate genes. Modifying effect of genes *GRIK2, TBP, BDNF, HIP1,* and *ZDHHC17* has been ruled out. On contrary it has been confirmed that theS18Y polymorphism of *UCHL1* gene decreases the age of onset by as many as 9 years. This result is of practical value in assessing the age of onset in

presymptomatic individuals in clinical praxis, thus due preventive interactions can be applied sooner.

The incidence of alkaptonuria in Slovakia (1:19000) is the second higher one in the world. Complex mutation analysis of involved gene (*HGO*) revealed the scale of disease causing mutations in Slovak patients. For all identified mutations simple and rapid DNA- based tests were developed for every-day use in diagnostic praxis.

Non-syndromic hearing loss represent the most frequent inborn error of sensoric system in humans (1:1000 newborns). For more than 50 % of cases mutations in *GJB2* gene are responsible. Thus we subjected this gene to complex mutation analysis in Slovak patients and obtained the whole mutation scale. Our results show that there are significant differences between patients of Roma and non-Roma ethnic origin, in term of mutation scale. This difference must be considered when providing mutation-based diagnostic testing. Simple and rapid tests for direct identification of prevailing mutations in both populations were developed.

Blindness in some subpopulations of Slovak Roma shows significantly increased incidence. Earlier we confirmed that some of them are due to primary congenital glaucoma, and are caused by founder mutation E387K in *CYP1B1* gene. Recently we have identified a second gene, mutations of which cause degeneration of retina (retinitis pigmentosa). This gene is the gene *RDH12*, and the disease causing mutation in all patients of Roma ethnic origin is a founder mutation R106X. Simple and rapid DNA-based test was developed for direct identification of this mutation, both in heterozygote and homozygote state for every-day diagnostic use.

Members of this laboratory published 6 papers in international journals. During evaluated period were their works cited 223 times.

Main research activity in **Laboratory of biochemistry and cytochemistry** was focused predominantly on studies of P-glycoprotein (P-gp) mediated multidrug (MDR) resistance of cancer tissue. It was found that this multidrug resistance of L1210/VCR cells is associated with:

- Dramatical depression of transglycosylation reactions that was linked with decrease in levels of UDP-sugars, glycogen, intracellular glycoproteins and cell surface sialic acid.
- Elevation of cells sensitivity to hypoxia that induced predominantly necrotic way of cell dead in resistant cells,
- Elevation of cells sensitivity to an increase of external calcium concentration that was linked with changes in intracellular calcium homeostasis.

Multidrug resistance of L1210 cells could be effectively depressed by pentoxifylline and its analogues that are associated with the depression of P-gp expression. Doxorubicin similarly as vincristine induced the multidrug resistance phenotype in L1210 cells characterized by P-gp expression without changes in expression of glutathione-S transferases and other drug transport pumps. Expression of P-gp in L1210/VCR cells seems to be at least indirectly regulated by PI3K/Akt kinase pathway. All these data were published in nine papers indexed in WOS and CC (Biochim. Biphys. Acta, Biochem. Biophys. Res. Commun., Eur. J. Pharmaceut. Sci twice, Toxicology in vitro, Gen. Physiol. Biophys. four times). Quality of research in this topic may be documented by fact that Dr. Breier and his colleagues were invited to submit a review paper in Current Cancer Drug Targets.

Beside this main research activity research group found that during reperfusion after liver ischemia several peptides were released from liver tissue. These peptides were found to exert protective effect on heart tissue against ischemia and reperfusion injury. These data were published in two papers in Gen. Physiol. Biophys.

The Ca^{2+}-binding S100A1 protein displays a specific and high expression level in the human myocardium and is considered to be an important regulator of heart contractility. Using immunostaining of electron microscopy samples prepared from biopsies of human heart a co-localization of a calcium binding protein S100A1 and the cardiac sarcoplasmic reticulum Ca^{2+}-ATPase 2a (SERCA2a) at the elements of sarcoplasmic reticulum was described. This finding helped to elucidate the S100A1 signaling pathway in the human heart. These data were obtained in cooperation with group of Professor Claus Heizmann from Switzerland and were published in Biochem. Biophys. Res. Commun. Papers published during the years of 2003-2006 by the members of Laboratory of Biochemistry and Cytochemistry has been cited 28 times. All papers authored by members of Laboratory of Biochemistry and Cytochemistry were cited 209 times during the years of 2003-2006.

Research of the **Laboratory of protein chemistry** (LPCH) during the assessed period (years 2003-2006) was focused mostly on multidrug resistance (MDR) of neoplastic cells. The research program of LPCH was oriented on study of mechanisms involved in P-glycoprotein (P-gp)-mediated multidrug resistance. Investigated were:

- protein systems involved in Pgp-mediated multidrug resistance
- mechanisms of reversal of multidrug resistance by several drugs
- principles that secures the binding of ligands to the specific binding sites
- protein kinase signaling pathways and their role in modulation of multidrug resistance

The research was carried out on the molecular and cellular level using mouse leukemic parental L1210 and multidrug resistant L1210/VCR cell lines as an experimental model. The research program of the LPCH was during the assessed period realized through several grant projects. At the national level LPCH actively participated on 2 projects of Agency for the Promotion of Research and Development (APVV) „*Signal and transport function of biological membranes under normal and pathological conditions*“ APVV-51-01013802 and APVV-51-027404, ii) State Program „*Genomic of cardiovascular diseases*“ and iii) project ordered by government of the Slovak Republic „*Cardiac adaptation to the pathological conditions (regulatory mechanisms)*“. Moreover, research workers from the LPCH were during the assessed period principal investigators of 3 projects of Scientific Grant Agency of the Slovak Academy of Sciences and the Ministry of Education (VEGA grants). From 2006 LPCH participates on international NATO grant *„P-glycoprotein mediated multidrug resistance in radiation-associated hematological malignances following the Chernobyl accident.“*.

During project realization several principally new observations were found and some of the obtained results may find application also in clinically oriented research and in clinical practice. The most important results are:

-out of all methylxanthines only analogues of pentoxifylline (PTX) were able to depress the P-gp- mediated multidrug resistance

-based on the comparison of physico-chemical properties of PTX analogues we were able to recognize properties of substances that are important for their reversal efficiency

-LY294,002, a specific inhibitor of PI3K/Akt kinase pathway reversed the P-gp- MDR and this is connected with inhibition of P-gp transport activity and Akt kinase activation

-development of MDR in L1210/VCR cells is associated with changes in systems involved in regulation of cell death (apoptosis) and the presence of LY294,002 significantly stimulated vincristine-induced apoptosis in these MDR cells

Results of research work of this team were published during years 2003-2006 in 9 articles in both foreign and domestic international peer reviewed journals. Works of laboratory members were cited 151 times during this period.

Main field of interest of **Laboratory of biochemistry of transport systems** are intracellular Ca^{2+}-transporting proteins, particularly IP3 receptors. Inositol 1,4,5-trisphosphate receptors (IP3 receptors) belong to the intracellular channels, releasing calcium from the intracellular calcium stores. Although most predominant in neuronal tissue (especially the type 1), these channels are expressed in variety of tissues, e.g. stellate ganglia, heart, kidney, skeletal muscle, etc. Although the exact physiological relevance of IP3 receptors is not clear, several papers suggested their involvement in apoptosis.

Team under the leadership of Olga Krizanova has shown that stress affects the gene expression and/or protein levels of the type 1 and 2 IP3 receptors in different tissues. While in the heart single immobilization stress increases gene expression of the type 1 and 2 IP3 receptors, in kidney no changes were observed. Nevertheless, in both these tissues and also in stellate ganglia repeated immobilization stress reduces the gene expression of the type 1 and 2 IP3 receptors.

Transcriptional regulation of IP3 receptors can be modulated through the responsive elements, localized in the promoter region of genes coding these receptors. We tested the effect of retinoic acids, thyroid hormones, catecholamine depletion on the gene expression of IP3 receptors in different tissues. All these compounds were able to affect gene expression and/or protein levels of individual types of IP3 receptors differently in various tissues.

During the period of years 2003-2006 results on localization and modulation of IP3 receptors were published in 11 papers in recognized international and domestic journals.

Other part of research of this laboratory was dedicated to catecholaminergic regulation and the effect of various stressors on the gene expression of catecholamine synthesizing enzymes in various tissues, with the special impact on heart, since catecholamines are known to possess inotropic and also chronotropic effect on cardiac function. In respect to cardiovascular diseases occurrence of some polymorphisms on genes coding renin-angiotensin system in Slovak population was determined. This research was conducted in collaboration with many Slovak basic research institutions and with clinical departments. Its results were published in 30 articles during the period of years 2003-2006. Works of laboratory members were cited 108 times during assessed period.

Research conducted in **Laboratory of biophysics** was focused on structure, function and regulation of voltage-dependent calcium channels, particularly L-type and T-type channels.

Work on L–type calcium channels was concentrated on the contribution of $Ca_v1.2$ voltage-gated calcium channel to the excitability of the hippocampal CA1 region. This was investigated in **H**ippocampus $\alpha 1$**C K**nock-**O**ut (HCKO) mice. Approximately 90% decrease in the amount of $Ca_v1.2$ protein was demonstrated by Western blots and by electrophysiological essay in both hippocampus and neocortex of young mice between 8 and 15 weeks old. The resting membrane potential was not altered by inactivation of the $Ca_v1.2$ gene. The input resistance of CA1 pyramidal neurons measured at a membrane potential of –70 mV was slightly, but not significantly, increased. As expected, the knock-out had no effect on the shape of single action potentials (AP). Maximal slope of the ascending and the descending phase, half-maximal width as well as threshold and amplitude single AP induced by brief 5 ms current pulse were not significantly different between the two genotypes. The threshold for generating a series of APs from the resting membrane potential of –70 mV was significantly enhanced and the AP frequency within an AP series was lowered in CA1 neurons from HCKO mice compared with the control. The $Ca_v1.2$ channels facilitate initiation of burst firing but are less important for steady state activity of hippocampal neurons. They do not participate in settling of resting potential.

Further, structure, function and regulation of the T-type calcium channel and the role of voltage-gated calcium channels in neuronal excitability were investigated.

Properties of the charge movement measured from the $Ca_v3.1$ channel expressed in HEK 293 cells were characterized. Threshold of its activation was by 10 mV more negative than the threshold for current activation. Slope of voltage dependence of charge movement was extremely shallow compared to voltage dependence of current activation. Prolonged depolarisation did not immobilise the charge movement. Coexpression of $\alpha_2\delta$-2a subunit or the γ_5 subunit improved charge movement – channel opening coupling probably by facilitation of transitions between individual closed states and the transition between last closed state and an open state. Gating of the $Ca_v3.1$ channel is modulated by Ca^{2+} ions, which facilitate the transition of the channel from conducting, i.e. open channel state into non-conducting, i.e., closed and inactivated states and backwards transition from non-conducting states into conducting state. Channel is not regulated by phoshorylation through the protein tyrosine kinase (PTK) dependent pathway. Nonspecific PTK inhibitor genistein effectively inhibits the channel by PTK-independent mechanism involving specific interaction with the voltage sensor of the channel together with the channel pore occlusion. Acute inhibition of the $Ca_v3.1$ channel both organic (methylmercury, MeHg) and inorganic (Hg^{2+}) mercury as well as its potentiation by chronic exposure to nanomolar concentrations of MeHg may contribute to pathology of acute and chronic mercury poisoning. When uppermost arginines in S4 segments of domains I to IV were replaced by neutral cysteines all aspects of channel gating were altered. With the exception of mutation in domain IV all other mutations significantly shifted activation towards more negative membrane voltages and increased slope factor of channel activation. Similarly, inactivation was shifted towards more negative

potentials and its slope factor was increased. When mutations in two neighboring domains were combined, effects on channel activation were only slightly enhanced, while effects on channel inactivation were additive. Recovery from the inactivation was slowed down by mutations in IS4 or IIIS4 segments. S4 segments in individual domains contributed differently to channel activation and inactivation with S4 segment in the domain III playing the most important role and S4 segment in the domain IV having the smallest impact.

Results of this research were published in 9 research reports and 3 review papers in foreign and domestic international peer reviewed journals during the years 2003-2006. Work of laboratory members was cited 373 times during this period.

Research in **Laboratory of electrophysiology** followed several lines that delineated transition of the research in LEF from the well-established area of the role of calcium ions in cardiac excitation and excitation-contraction coupling towards the integral level. The specific expertise of LEF led to invitation for collaboration in the integrated project and the Specific target research project of 6. Framework Program of EU starting in the year 2006. Our part is a system characterization of muscle cells of failing hearts. The research in LEF was further supported by grants VEGA and APVT from Slovakia, FIRCA and HHMI from USA.

Line 1 - Membrane noise as an integral measure of the functional state of the cell: An original method (so-called the Q-method) for very exact and reliable measurement of electrical parameters of isolated cardiac myocytes was developed in order to resolve changes of membrane capacitance and membrane resistance at their theoretical limits. The Q-method revealed for the first time that membrane capacitance fluctuates spontaneously, hypothetically due to very fast changes of the active surface area of myocytes. The Q-method also revealed that membrane resistance fluctuates only partially due to fluctuations of membrane capacitance. In resting myocytes, these fluctuations display fractal dynamics that arise by a large part from unstable activity of the background sodium conductance. These findings support the potential of the Q-method to provide an integral view of molecular processes within the complex system of the functional myocyte.

Line 2 – Cardiac excitation-contraction (E-C) coupling: In close collaboration with the LMB processes of E-C coupling at the level of elementary release units in isolated cardiac myocytes were characterized. It was shown that physiological concentration of magnesium ions determines responsiveness of ryanodine receptor (RyR) channels to calcium ions *in situ*, and that the triggering of calcium release depends on the recent history of calcium signalling. Next, the mechanisms that govern activation and termination of calcium release *in situ* were identified. In this context sensitivity of RyRs mutated in the putative calcium binding site was analysed. It was shown that the mechanism of RyR activation is based on allosteric interactions of its four independent calcium binding subunits. Group has identified parameters that gauge the rate and duration of calcium release, and determined their values in control cells. These parameters make possible to characterize changes of calcium release in diseased myocardium.

Line 3 - Reaction-diffusion model of the tubulo-reticular junction in cardiac myocytes: Local character of excitation-contraction coupling in cardiac

myocytes represents experimentally and theoretically a very difficult problem that relays on interpretation of indirect observations. A full 3D reaction-diffusion model was developed, in which recent understanding of the physico-chemical principles of the function of cardiac dyads in E-C coupling was integrated. This allowed showing the dynamics of calcium ions and verifying consequences of possible mechanisms of calcium signalling within cardiac dyads. (In collaboration with the Faculty of Natural Sciences, Comenius University, Bratislava, and intramural collaboration with the LMB).

Line 4 – Quantification of structural aspects of striated muscle cells:

In close collaboration with the LCM structural differences among different types of striated muscle cells were quantitatively characterized with the aim to understand the role of myocyte architecture in its function. Using transgenic models, it was found that cytoarchitecture reacts logically to non-structural stimuli like defects in energy transduction and, in this way, it influences transduction of internal signals, e.g., between mitochondria and other organelles. For this, a new method of analysis of electron micrographs of muscle cells was developed.

Line 5 - Architectural determinants of myocyte function: Internal architecture of cardiac myocytes and other striated muscle cells is believed to be optimized for their performance and to have an important role in the contractile and the signal transduction functions. To capture these complex phenomena a set of computer modelling tools was developed that allows to design the architecture of myocyte from its organelles in terms of geometry. At present, the model makes possible to test various structural hypotheses, to compare models of different myocyte types and changes of their structure due to various influences including pathology. (In collaboration with the Faculty of Mathematics, Physics and Informatics, Comenius University, Bratislava, and intramural collaboration with the LCM)

During assessed period laboratory members has published 14 articles in international and domestic peer-review journals and/or proceedings from conferences. Their works were cited 96 times.

The main topic of research in the **Laboratory of molecular biophysics** is biophysics of calcium signalling in cardiac myocytes. The laboratory has a well-established position in the European Research Area in this field, due to its use of cutting-edge experimental techniques (recording electrical activity of single ion channel molecules; temporally and spatially resolved measuring of intracellular calcium concentration using confocal microscopy) in combination with advanced mathematical modelling, and due to the long-standing tradition of excitation-contraction coupling research at the Institute. The Laboratory is considered a serious partner also in the USA. As a result, in the reported period the laboratory pursued research funded by two US grants (Howard Hughes Medical Institute International Research Scholar Award, 2001 – 2005 (A. Zahradníková, principal investigator); National Institutes of Health Fogarty International Research Collaboration Award, 2001 – present; A. Zahradníková – international collaborator) and two 6th Framework Programme grants (2006 – present; integrated project EUGeneHeart; A. Zahradníková – group leader; specific targeted research project CONITCA; A. Zahradníková – group leader). Additionally, the laboratory participated in three VEGA grants and one APVV grant (Slovak resources).

In the reported period laboratory has studied molecular mechanisms of calcium-induced calcium release in isolated cardiac myocytes. This central point between cell excitation and contraction, mediated by ryanodine receptor calcium release channels (RyRs) of intracellular calcium stores, is impaired in cardiac diseases such as heart failure and certain types of arrhythmias. In collaboration with the Laboratory of Electrophysiology, methods for relating activity of isolated RyR channels to calcium release activity inside living cells were developed and used to characterize the stimulus – response relationship in voltage-clamped cells. It was shown for the first time that physiological concentration of magnesium ions, which act as competitive inhibitors of the activating calcium ions, dramatically limits the responsiveness of RyR channels to brief calcium stimuli such as those occurring in the cell, and that, as a result, the ease of triggering the calcium release process is dependent on the recent history of calcium signalling at individual calcium release sites. The analysis of calcium sensitivity of RyRs mutated in the putative calcium binding site has shown that the mechanism of RyR activation is based on allosteric interaction of its four independent calcium binding sites with the channel-opening module. Additional closed channel conformations that limit its maximum open probability are required. Work of this group culminated in determination of the time course of calcium release at individual calcium release sites in the cardiac myocyte. By combining electrophysiological methods and confocal microscopy recordings of local calcium release signals with unprecedented spatio-temporal resolution were obtained. By applying the originally developed new methods of analysis and mathematical modelling, it was showed for the first time that the kinetics of local calcium release is reliably traced with the low-affinity fast fluorescent indicator OG-5N. The mechanisms that govern activation and termination of calcium release *in situ* that are in agreement with the concept of steep calcium-dependent activation and fateful inactivation of calcium release flux were identified. Parameters that gauge the rate and duration of calcium release were identified and their values were determined in cells from healthy animals. These parameters will enable quantitative characterization of the nature and extent of changes caused by cardiac disease.

During evaluated period laboratory members published 9 articles in recognized foreign journals as well as in domestic international peer reviewed journals. Their work was cited 145 times.

Laboratory of intracellular ionic channels has studied electrophysiological properties and modulation of intracellular ion channels using the method of bilayer lipid membrane (BLM). Particularly we studied the properties of the intracellular Ca^{2+} channels, ryanodine and inositol 1,4,5-trisphosphate receptors, and mitochondria channels (chloride, and potassium channels) and their involvement in pathology and cardioprotection.

Scientific production of the laboratory is nationally and internationally recognized and incorporated in the European Research Area. In addition to VEGA grant its work was supported by two major APVT/APVV grants (51-027404 and 51-01-013802) and two international cooperation grants FIRCA grant 2R03-TW00949-04A1 (Coupled gating between intracellular calcium release channels - collaboration of Karol Ondrias, DSc. and prof. Andrew R. Marks, M.D., Columbia University, New York, USA) and NATO grant LST.CLG979217 (Role of mitochondrial channels in cardioprotection -

collaboration of Karol Ondrias, DSc. and prof. Adam Szewczyk, Institute of Experimental Biology, Warsaw, Poland).

During the evaluated period 6 research articles in international scientific journals were published. Three articles were produced in cooperation with the Nencki Institute of Experimental Biology, Warsaw, Poland, and one article with Department of Neurosurgery, St. Luke's Roosevelt Hospital Center, New York, USA. Works of laboratory members were cited 479 times during this period.

Research of **Laboratory of ion channel function** during 2003-2006 was mainly focused on the role of intracellular store Ca^{2+} ions in the modulation of functional properties of cardiac ryanodine receptor (RYR2). Ca^{2+} released from the sarcoplasmic reticulum (SR) via RYR2 channels is the key determinant of cardiac contractility. Although, activity of RYR2 channels is primary controlled by Ca^{2+} entry through the plasma membrane, there is growing evidence that Ca^{2+} in the lumen of the SR can also be effectively involved in the regulation of the RYR2 channel function. At present, it is not known whether Ca^{2+} acts directly at lumenal side of the channel or flows through the channel pore and acts at cytoplasmic Ca^{2+}-binding sites. Most papers reporting the effect of luminal Ca^{2+} on RYR2 channels focused on the determination of the differences in channel activity induced by elevating luminal Ca^{2+}, whereas a detailed examination of channel gating kinetics was not of particular interest. Laboratory conducted series of experiments in which luminal Ca^{2+} concentration was kept constant and the concentration of caffeine and Ca^{2+} applied to the cytosolic face of the RYR2 channel was changing. Planar lipid bilayer method was employed in this study. Luminal Ca^{2+} effectively shifted the EC_{50} for caffeine sensitivity to lower concentration, but did not modify the response of RYR2 channels to cytosolic Ca^{2+}. Importantly, luminal Ca^{2+} remarkably slowed down channel gating kinetics. Both, the open and closed dwell times were considerably prolonged over the whole range (response to caffeine) or the partial range (response to cytosolic Ca^{2+}) of open probability. This effect was not induced by direct addition of Ca^{2+} to the cytosolic face of the channel in the absence of luminal Ca^{2+}. Thus, these findings indicate that the effect on gating kinetics is likely attributed to the action of luminal Ca^{2+} on the luminal face of the channel providing further evidence in support of the existence of distinct intraluminal Ca^{2+} sensing sites, which regulate the behaviour of the RYR2 channel. This study provides the framework for precise characterization of disease-linked RYR2 mutations from the functional point of view. Enhanced sensitivity to luminal Ca^{2+} represent common defects of RYR2 mutations associated with ventricular tachycardia – the leading cause of sudden death. Currently, members of the laboratory are part of European research team supported by grant CONTICA attempting to elucidate the molecular mechanisms linking defective RYR2 function to the generation of arrhythmias.

During evaluated period laboratory members have published 3 articles in renowned international journals. Their works were cited 333 times.

Research activities in **Laboratory of cellular morphology** (LCM) were devoted to understanding of structural aspects of adaptational mechanisms of striated muscle cells, of both the skeletal and the cardiac muscle types, to complex stimuli under normal and pathological conditions. Due to the complexity of the system, research strategy was based on comparative study of ultrastructural changes in phenotypically different muscle fibres adapting to very specific stimuli like invalidation of a single protein of transgenic animal models.

Laboratory focused its work on evaluation of changes in the overall ultrastructure and in mutual relations between organelles. This required a new quantitative approach to analysis of reorganisation of the muscle cell architecture in addition to morphological studies. To reach this goal, a new stereological parameter - the organelle environment - was introduced, which was instrumental in characterizing changes of muscle cytoarchitecture. Using quantitative arguments, it was shown for the first time that spatial relations among organelles of muscle cells undergo adaptation in response to non-structural stimuli like metabolic deficiency due to lack of creatine kinase (the creatine kinase null mouse model, $CK^{-/-}$). The adaptational response of the ultrastructure that generally compensated for energy transport deficiency was found substantially different for muscle fibres of the fast glycolytic muscle, of the slow oxidative muscle and of the cardiac ventricular muscle. Additionally, the environment of mitochondria in the three studied muscle types was quantitatively characterized and compared and for the first time provided structural equivalent of various oxidative phenotypes.

Using the model of the MLP-null mouse (MLP - the muscle LIM protein, a cytoskeletal protein type) subcellular disorganization affecting direct transfer of energy from mitochondria to the sarcoplasmic reticulum calcium pump was revealed. It means that perturbation of cardiac cytoarchitecture may impair the direct energy transfer and contribute to energy wastage during contractile dysfunction.

In addition to these specific changes several types of unspecific changes occurring in cardiac muscle fibres of transgenic animal models with null mutations of CK, MLP, AMPKα2, and PMCA4 were revealed.

These studies were made possible through the informal cooperation with the INSERM Unit 446, Faculte de Pharmacy, University Paris Sud, Chatney-Malabry, France, and supported by the program Stefanik, the Slovak-France collaboration program of the ministries of education. French partners contributed by providing animal models and by functional measurements on isolated muscles.

The unique expertise of LCM led to invitation for collaboration in the Integrated project EuGenHeart of the 6. Framework Program of EC, the partner group led by A. Zahradniková, head of Laboratory of molecular biophysics. Our part is ultrastructural characterization of muscle cells of failing hearts in several animal models.

Research group was involved in 4 other domestic collaborative projects. These included ultrastructural studies of atrial myocytes of mice exposed to forced running with the Institute of Experimental Endocrinology SAS, morphological description of Inka cells of epitracheal glands in pupae of insects, with the Institute of Zoology SAS, and intramural collaborations with laboratories of Dr. I. Zahradník, development of computer model of striated muscle cells, Dr. K. Ondriaš, electron microscopic characterization of mitochondrial fractions, and Ing. O. Križanová, ultrastructural changes in atrial myocytes due to immobilization stress.

3. Concept of R&D activity of the Organisation for the next four years

3.1 Present state of knowledge and status of ongoing research related to the subject of the Concept, from both international and national perspective

The main focus of the research conducted on the IMPG SAS is modulation of the calcium concentration in cells, mainly through the calcium transport systems. Also, processes regulated by altered calcium handling are studied, mainly in the excitable cells.

The local character of calcium signalling participating in the transduction of electrical excitation into cardiac muscle contraction has been clearly established at the cellular level. The LMB group contributed to understanding of ryanodine receptors behaviour in the context of excitation-contraction coupling. Using techniques of molecular biology, genetics and diagnostics, other groups in the USA, Europe and Japan have developed concepts of failing and arrhythmogenic heart related to specific functional and genetic defects of the ryanodine receptor. In Slovakia, significant effort is devoted to understanding of cardiac function in ischemia and reperfusion injuries including molecular signalling. At our institute, expertise towards molecular biology of the ryanodine receptor and ultrastructure of cardiac muscle cells has shown significant progress within the last ten years.

Membrane channels seem to be a precondition for all living matter. For this reason, a better understanding of their function constitutes an important basis for understanding many disease states. Disturbances in ion channel function can lead to serious diseases of the nervous system, skeletal and cardiac muscle, and intestine. This makes ion channels important drug targets for the pharmaceutical industry. Calcium currents through T-type and L-type voltage-gated Ca^{2+} channels (L-VGCCs) play a key role in the cellular signalling in brain. The pore forming α1-subunits $Ca_v1.2$ and/or $Ca_v1.3$ as well as $Ca_v3.1$ conduct Ca^{2+} currents that are required for their function. As $Ca_v1.2$ and $Ca_v1.3$ show an overlapping expression pattern in a variety of tissues and subunit-specific antagonists or agonists are not available so far, genetic tools are needed to dissect the specific roles of both $Ca_v1.2$ and $Ca_v1.3$ in normal and pathological function. Collaboration with laboratories from universities in Marie Curie Research Training Network CavNet will give us an access to various knockout mouse models. Comprehensive electrophysiological analysis of Ca^{2+} currents in neuronal cells isolated from such models will allow an analysis of the differential roles of $Ca_v1.2$ and $Ca_v1.3$ in neuronal plasticity. For understanding of the role of low-voltage-activated $Ca_v3.1$ channel in neuronal excitability structure-function relation should be established. We will analyse mutated channels, in which charged arginine residues in their putative voltage sensors were replaced by neutral cysteines. Such replacements have dual effects: i), innate charge of voltage sensor will decrease, ii), newly introduced cysteine residues may enable formation of disulfidic bridges, giving additional information on tertiary structure of channel pore. On the basis of analysis of macroscopic inward currents theoretical model of channel transition between closed, open and inactivated states will be designed.

Inositol 1,4,5-trisphosphate receptors (IP3Rs) a intracellular calcium channels, which are able together with ryanodine receptors to release calcium from the intracellular stores. Although the function of these receptors has been just proposed, experiments in this field are oriented towards understanding the mechanisms, by

which these channels are modulated in normal, physiological state and during pathophysiological conditions.

Structure of striated muscle cells is well known from the morphological concept of view at the ultrastructural level. Relatively less progress was made, however, towards quantitative aspects of the cell structure that are very specific for various muscle fibre types and various species. This was related to the overall difficulties in assessing quantitative aspects of the cell structure that led to divergence of general interest towards molecular-biological aspects of the cell construction. At the microscopic level, most studies are focused on the fluorescence and immunocytochemical techniques that are more commercially feasible with less laboratory expertise. Our group has contributed in the previous period to revitalization of the electron microscopic and stereologic approaches towards understanding of the cellular architecture and its adaptation to changed demands on the cell function, e.g., due to ontogenesis, pathogenesis, or simply the changes in overall environment.

P-glycoprotein (P-gp) –mediated multidrug resistance (MDR) of neoplastic tissue represents a serious obstacle in effective chemotherapy of cancer. This problem has a global character and so several research institutions over the world intensively investigate the ways how to regulate and block the function and expression of P-gp. The Institute of Molecular Physiology and Genetics (Laboratory of Protein Chemistry, LPCH and Laboratory of Biochemistry and Cytochemistry, LBC) is accepted at both national and international level as Institution playing important role in reserach related to multidrug resistance.

3.2 Organisation's role or significance in the overall research effort within the field of the Concept on both the national and international scales

IMPG SAS belongs to most renowned institutions of Medical Research in SAS, ranked on the first place by the independent rating agency ARRA. Scientists from the Institute have wide and fruitful cooperation with research teams from other scientific institutions in Slovakia (e.g. Faculty of Natural Sciences Comenius University, Faculty of Chemical and Food Technology Slovak Technical University, Institute for Heart Research SAS, Institute of Experimental Endocrinology, SAS, Institute of Virology, SAS), Czech Republic (e.g. Medical Faculty of Masaryk University, Brno, Medical Faculty, Karlova University, Prague, Institute of Physiology Academy of Sciences of Czech Republic, etc.) and other recognized teams from Europe and USA.

The Institute belongs to the leading institutions in the field of calcium transport systems globally. Therefore, our goal for the next four years is to keep the momentum in elucidating ryanodine receptor function at the molecular level and to develop our expertise towards integrated and complex aspects of RyR function in disease at the cellular level.

At our institute, the LMB and LEF focus on cardiac muscle cell function in close collaboration with LCM. The three laboratories of our institute are coupled to European research area through the 6. FP projects, where ultrastructural expertise is required. Moreover, as a part of our APVV project with LEF, we have started recently

collaboration with the group of Dr. J. Bartunek and Dr. M. Vanderheyden, at the Cardiovascular Center, OLV Hospital in Aalst, Belgium, who have outstanding expertise in human heart pathology, including surgery, transplantation and biopsy, and renown scientific excellence.

In the previous period, group of LEF has contributed by conceptually original approaches to understanding of cell contractility and excitation–contraction coupling at higher level of integration by promoting interdisciplinary approach. This included combination of electrophysiology with confocal and electron microscopy, signal analysis, image analysis, mathematical and computer modelling. The ambition of LEF is to take advantage of this progress and participate in the overall effort towards application of strong basic research in human physiology and medicine. At the level of the institute, four laboratories work in close collaboration toward this goal – LEF, LMB, LICF and LCM. The four laboratories are coupled together and to the European research area through the 6. FP projects EUGeneHeart and CONTICA that started recently and partially through grants of APVV and VEGA.

3.3 Objectives of the Concept

The research in Laboratory of Molecular Biophysics will focus on explanation of the mechanisms responsible for the pathological changes in activation of calcium-induced calcium release in cardiac ryanodine receptor channels of diseased cardiac myocytes. We will focus on the mechanisms that govern activation of local calcium release and the transition from local calcium release to arrhythmogenic calcium waves, with accent on the role of regulation of the calcium-binding and channel-opening processes by calcium in the lumen of calcium stores.

We will focus on the tissue specific organization of striated muscle cells with emphasis on cardiac myocytes to reveal the role of the cell architecture in specific functions especially excitation – contraction coupling. To this aim we will employ our original morphological and stereological approaches to elucidate cellular mechanism of E-C coupling and their alteration under pathological conditions of heart failure and transplantation. The focus on the muscular architecture in animal models of heart failure and in human diseased hearts and its adaptation would help in the understanding of the physiopathology of cardiac and skeletal myopathies.

The objective of LEF will be to understand the process of excitation–contraction coupling in cardiac myocytes and its relation to the failing heart disease and arrhythmias in a wider frame of its determinants and consequences at the cellular level using interdisciplinary approach. Specifically:

1/ To understand fluctuations of membrane capacitance and resistance in relation to calcium signalling in cardiac myocytes of selected mice models including experimentally induced heart failure.

2/ To understand integral myocyte functions using mathematical modelling of the action potential generation with included calcium signalling in and between known cellular constituents and compartments, and mathematical modelling of elementary and integral calcium signals of selected mice models.

3/ To understand key aspects of spatial cell organization related to calcium signalling in health and disease with the use of stereologic analysis and geometrical modelling.

In the next 4 years the research program of LPCH and LBC will be oriented on study of mechanisms involved in P-gp-mediated MDR. Parental and multidrug

resistant cell lines will be used and will be characterized on overexpression of P-gp and other drug transporters. Knowledge of factors that determine the substrate specificity of P-gp is crucial for successful drug targeting and rational design of new effective drugs. For this reason new derivatives of pentoxifylline (P-gp antagonist) will be synthesized. Also on the basis of our previous observations the role of intracellular signaling (protein kinase pathways) and systems involved in regulation of apoptotic responses in modulation of MDR will be investigated. By application of specific modulators (activators, inhibitors) of protein kinase pathways and using systems characterized by overexpresion of some protein kinases (MAPK) the ways of inhibition of development of MDR and modulation of resistance in case of increased P-gp expression will be studied. The consequences on apoptotic responses will be also assessed. Association of retinoids and retinoids nuclear receptors in expression and regulation of P-gp activity will be intensively studied with the aim to contribute in knowledge about mechanisms involved in P-gp expression. Interplay between P-gp mediated MDR and transglycosilation reactions will be also studied. This will be focused predominantly on differences in cell surface saccharides between sensitive and resistant cells that may be monitored by interaction with different lectines.

The main aim of our work is to contribute to the deeper understanding of this important phenomenon using the method of reconstitution of RYR2 channels into artificial lipid membrane. This method provides detail information about functional properties of ion channels by observing electrical current (~pA) flowing through these channels. Particularly, we will examine whether oxidation/reduction of –SH groups of cysteins of RYR2 channel play role in coupled gating phenomenon and we will examine a potential impact of luminal Ca2+ on stability and gating kinetics of coupled RYR2 channels.

3.4 Proposed strategies and methods to be applied, and time schedule

Strategies and time schedule is highly dependent on the amount of finances granted to the Institution.

Ultrastructural adaptation of cardiac muscle cells will be studied by the computer aided image analysis of electron microscopic images, including quantitative stereological methods that will be used for analysis of volume, surface and environmental parameters of organelles in experimental models. The complex data sets will be integrated using advanced computer modelling we have recently developed with colleagues in LEF.

The methodological repertoire used in the laboratory will be expanded by simultaneous measurement of cytosolic and luminal calcium transients, probing of regulatory domains of the ryanodine receptor using synthetic peptides and planar lipid bilayer measurements, and modelling and simulation of the cellular action potential and calcium handling based on molecular and local models of processes and proteins participating in excitation-contraction coupling. We will train about 2 doctoral students in biophysics. Our plan of activities is in accordance with the recently awarded grants of the 6th FP of the EU that will end in 02/2009 and 12/2010, respectively.

The central method will be comparative analysis of data from selected animal models and their integration to heuristic integral models. Experimental techniques will include patch clamp combined with high resolution laser scanning confocal

microscopy applied to isolated cardiac myocytes, signal and image analysis, and electron microscopy with stereology. Mathematical and computer modelling will focus on action potential variability, calcium signals and cell architecture.

During the period of next four years we will try to establish the short-term cell culture from the hippocampal neurons, analyze the excitability of hippocampal neurons from various transgene models and analyze the expression of calcium transporting proteins in the absence of various L-type calcium channel subtypes. Also, we will analyze the expression of calcium transporting proteins in the absence of various L-type calcium channel subtypes.

Another project will be to establish the method of gene silencing and overexpression of selected calcium transport systems, especially IP3 receptors, to be able to deduce functional role of this calcium transport system in the heart.

III. Partial indicators of the main activities

1. Research output

1.1 List of the selected publications documenting the most important results of basic research.

(Total number of publications in the whole assessed period should not exceed the average number of the research employees)

1. MIČUTKOVÁ, L. – KVETŇANSKÝ, R. – KRIŽANOVÁ, O. Repeated immobilization stress reduces the gene expression of the type 1 and 2 IP3 receptors in stellate ganglia. In *Neurochemistry International,* 2003, Vol. 43; p. 557-561. (2.902 – IF 2002)
2. ZAHRADNÍKOVÁ, A. – DURA, M. – GYÖRKE, I. – ESCOBAR, A.L. – ZAHRADNÍK, I. – GYÖRKE, S. Regulation of dynamic behaviour of cardiac ryanodine receptor by Mg^{2+} under simulated physiological conditions. In *American Journal of Physiology - Cell Physiology,* Vol. 285, no. 5, 2003, p. C1059–C1070. (3.936 – IF2002)
3. FIALA, R. – SULOVÁ, Z. - EL-SAGGAN, A.H. – UHRÍK, B. – LIPTAJ, T. - DOVINOVÁ, I. – HANUŠOVSKÁ, E. – DROBNÁ, Z. – BARANČÍK, M. – BREIER, A.: P-glycoprotein-mediated multidrug resistance phenotype of L1210/VCR cells is associated with decreases of oligo- and/or polysaccharide contents. In Biochimica et Biophysica Acta, Vol. 1639, no.3, 2003, p. 213-224. (3.300 – IF2003)
4. KREPSOVA K. - MICUTKOVA L. - NOVOTOVA M. - KUBOVCAKOVA L. - KVETNANSKY R. - KRIZANOVA O.: Repeated immobilization stress decreases mRNA and protein levels of the type 1 IP3 receptors in rat heart. In *Annals of the New York Academy of Sciences,* Vol. 1018; (2004), p. 339-344, (1.892 – IF2003)
5. LACINOVÁ, Ľ. - KLUGBAUER, N. Modulation of gating currents of the $Ca_v3.1$ calcium channel by $\alpha 2\delta 2$a and $\gamma 5$ subunits. In: *Archives of Biochemistry and Biophysics* Vol. 425, no. 2 (2004), p. 207-213 (2,338 - IF2003)
6. ZAHRADNÍKOVÁ, A. – KUBALOVÁ, Z. – PAVELKOVÁ, J. – GYORKE, S. – ZAHRADNÍK, I. Activation of calcium release assessed by calcium release-induced inactivation of calcium current in rat cardiac myocytes. In *American Journal of Physiology – Cell Physiology,* Vol. 286, (2004), p. C330-C341. (4,103 - IF2003) LACINOVÁ, Ľ. – HOFMANN, F. Ca2+- and voltage-dependent inactivation of the expressed L-type Cav1.2 calcium channel. In *Archives of Biochemistry and Biophysics*. Vol. 437, no. 1 (2005) p. 42 - 50. (2,657 - IF2004)
8. LI, X. – MALATHI, K. – KRIŽANOVÁ, O. – ONDRIAŠ, K. – SPERBER, K.- ABLAMUNITS, V. – JAYARAMAN, T. Cdc2/cyclin B1 interacts with and modulates inositol 1,4,5-trisphosphate receptor (type 1) functions. In *Journal of Immunology.* Vol 175, no. 9 (2005), p. 6205-6210 (6,486 - IF2004)
9. MINÁRIK, G. – FERÁKOVA, E. – FICEK, A. – POLÁKOVÁ, H. – KÁDASI, Ľ. GJB2 gene mutations in Slovak hearing-impaired patients of Caucasian origin:

spectrum, frequencies and SNP analysis. In: *Clinical Genetics,* Vol. 68, no. 6 (2005) p. 554-557. (2,367 - IF2004)

10. SULOVÁ, Z. – ORLICKÝ, J. – FIALA, R. – DOVINOVÁ, I. – UHRÍK, B. – ŠEREŠ, M. - GIBALOVÁ L. – BREIER. A. Expression of P-glycoprotein in L1210 cells is linked with rise in sensitivity to Ca^{2+}. In *Biochemical and Biophysical Research Communications,* Vol. 335, (2005) p. 777-784. (2,904-IF2004)
11. ŠTEFÁNIK, P. – MACEJOVÁ, D. – MRAVEC, B. – BRTKO, J. – KRIŽANOVÁ, O. Distinct modulation of a gene expression of the type 1 and 2 IP3 receptors by retinoic acid in brain areas. In *Neurochemistry International* Vol.46, (2005) p. 559-564, (3,211 – IF2004)
12. BARANCIK, M. - BOHACOVA, V. - SEDLAK, J. – SULOVA, Z. - BREIER,A. LY294,002, a specific inhibitor of PI3K/Akt kinase pathway, antagonizes Pglycoprotein- mediated multidrug resistance. In: *Eur J Pharm Sci.* Vol. 29(5),(2006), p. 426-34. (IF 2.347)
13. GABURJAKOVA, J. - GABURJAKOVA , M. Comparison of the effects exerted by luminal Ca2+ on the sensitivity of the cardiac ryanodine receptor to caffeine and cytosolic Ca2+. In *Journal of Membrane Biology*. Vol. 212(1), (2006), p. 17-28 DOI: 10.1007/s00232-006-7018-z (2,208 – IF2005)
14. JURKOVICOVA, D. – KUBOVCAKOVA, L. – HUDECOVA, S. – KVETNANSKY, R. – KRIZANOVA, O. Adrenergic modulation of the type 1 IP3 receptors in the rat heart. In: *BBA - Molecular Cell Research,* Vol. 1763, (2006), p. 18-24. (4.844– IF2005)
15. KUBOVCAKOVA, L. – MICUTKOVA, L. – BARTOSOVA, Z. – SABBAN, EL. – KRIZANOVA, O. – KVETNANSKY, R. Identification of phenylethanolamine N-methyltransferase gene expression in stellate ganglia and its modulation by stress. In: *J Neurochem* Vol. 97, (2006), p. 1419- 1430. (4.604– IF2005)
16. NOVOTOVÁ, M. - PAVLOVICOVÁ, M. - VEKSLER, V.I. - VENTURACLAPIER, R. – ZAHRADNÍK, I. Ultrastructural remodeling of fast skeletal muscle fibers induced by invalidation of creatine kinase In *American Journal of Physiology - Cell Physiology* Vol. 291, no. 6 (2006), p. C1279-1285. (3,942 - IF2005)
17. PROKS, P. – LIPPIAT, JD. Membrane ion channels and diabetes. In: *Curr Pharm Design* Vol. 12, (2006), p. 485-502 (4.829– IF2005, 1.889 - Median IF)
18. SLAVIKOVA J, - DVORAKOVA M, - REISCHIG J, - PALKOVITS M, - ONDRIAS K, - TARABOVA B, - LACINOVA L - KVETNANSKY R - MARKS A, - KRIZANOVA O. IP3 type 1 receptors in the heart: their predominance in atrial walls with ganglion cells. In: *Life Science.* Vol. 78, no 14 (2006), p. 1598-602. (2.512 – IF2005)
19. TARABOVA, B. - KUREJOVA, M. - SULOVA, Z. - DRABOVA, M. - LACINOVA, L. Inorganic mercury and methylmercury inhibit the Cav3.1 channel expressed in human embryonic kidney 293 cells by different mechanisms. In: *Journal of Pharmacology and Experimental Therapeutics* Vol. 317, no. 1 (2006), p. 418-427. (4,098 – IF2005)

20. TILLINGER, A. – NOVAKOVA, M. – PAVLOVICOVA, M. - LACINOVA, L - ZATOVICOVA, M. – PASTOREKOVA, S. – KRIZANOVA, O. – KVETNANSKY, R. Modulation of gene expression of the PNMT in a heart during immobilization stress by 6-hydroxydopamine. In: *Stress*, Vol. 9(4), (2006), p. 207-213 (2,962 – IF2005)

1.2 List of monographs/books published abroad

1.3 List of monographs/books published in Slovakia

1. KÁDAŠI, Ľ. *Molekulárna genetika vybraných monogénne dedičných ochorení*. Bratislava: Veda, 2005, ISBN 80-2240869-7

1.4 List of other scientific outputs specifically important for the Organisation

1. BREIER, A. - BARANČÍK, M. - SULOVÁ, Z. – UHRÍK, B. P-Glycoprotein – Implications of metabolism of neoplastic cells and cancer therapy. In *Current Cancer Drug Targets,* Vol. 5, (2005) p. 457-468 - Dr. Breier and his colleagues were invited to submit a review paper in Current Cancer Drug Targets. This journal is indexed in WOK starting from the year 2004 and thus IF will be available in the year 2007. Therefore it could not be included to the table concerning renormalized publications. Nevertheless the paper was 8 times cited.
    - Schafer ZT, Parrish AB, Wright KM, Margolis SS, Marks JR, Deshmukh M, Kornbluth S Cancer Research 66 (2006) 2210-2218 (WOK)
    - Prados J, Melguizo C, Fernandez JE, Carrillo E, Marchal JA, Boulaiz H, Martinez A, Rodriguez-Serrano F, Aranega A Neoplasma 53 (2006) 226-231 (WOK)
    - Leon C, Sachs-Barrable K, Wasan KM Drug Development and Industrial Pharmacy 32 (2006) 779-782 (WOK)
    - Solazzo M, Fantappie O, Lasagna N, Sassoli C, Nosi D, Mazzanti R Experimental Cell Research 312 (2006) 4070 (WOK)
    - Rohr J ACS CHEMICAL BIOLOGY 1 (2006) 747-750 (WOK)
    - Bu L-M, Sun S-H, Hua J-P, Han Y, Lai J, Bao W-Y World Chinese Journal of Digestology 14 (2006) 2082-2086 (SCOPUS)
    - Holmes AR, Tsao S, Lamping E, Niimi K, Monk BC, Tanabe K, Niimi M, Cannon RD Japanese Journal of Medical Mycology 47 (2006) 275-281 (SCOPUS)
    - Pawelek J, Chakraborty A, Lazova R, Yilmaz Y, Cooperc D, Brash D, Handersone T Co-Opting Macrophage Traits in Cancer Progression: A Consequence of Tumor Cell Fusion? In Dittmar T, Zaenker KS, Schmidt A (eds): Infection and Inflammation: Impacts on Oncogenesis. Contrib Microbiol. Basel, Karger, 2006, vol 13, pp 138–155 (in monography)

2. LACINOVÁ, Ľ. Voltage gated calcium channels. In *General Physiology and Biophysics.* Vol. 24, Supplement 1 (2005), p. 1-82. (0,694 - IF2004). This paper represent monography like publication that was published in a separate supplement of General Physiology and Biophysics. This publication was 7 times cited.
    - Weiergraber M, Kamp MA, Radhakrishnan K, et al. NEUROSCIENCE AND BIOBEHAVIORAL REVIEWS 30 (2006): 1122-1144
    - Chu ZG, Moenter SM JOURNAL OF NEUROSCIENCE 26 (2006): 11961-11973
    - McKeown L, Robinson P, Jones OT ACTA PHARMACOLOGICA SINICA 27 (2006): 799-812
    - Wolfe DM, Pearce DA NEUROMOLECULAR MEDICINE 8 (2006): 279-306
    - Felix R NEUROMOLECULAR MEDICINE 8 (2006): 307-318
    - Krieger A, Radhakrishnan K, Pereverzev A, et al. CELLULAR PHYSIOLOGY AND BIOCHEMISTRY 17 (2006): 97-110
    - Gill S, Gill R, Xie Y, et al. ASSAY AND DRUG DEVELOPMENT TECHNOLOGIES 4 (2006): 65-71
3. VALENT, I. – ZAHRADNÍKOVÁ, A. - ZAHRADNÍK, I. An implementation of the VLUGR-3 solver for 3D-simulation of the reaction-diffusion processes in the cardiac dyad. In Capasso V. *Mathematical Modelling and Computing in Biology and Medicine.* Bologna: Esculapio, 2003. ISBN 88-7488-055-3. p. 213-218
4. ZAHRADNÍK, I. – GABURJÁKOVÁ, J. – KUBÍNOVÁ, L. – NOVOTOVÁ, M. Stereologicanalysis of isolated rat ventricular myocytes using the method of vertical sections. In *Science, technology and education of microscopy: An overview* Ed. By A. Mendez-Vilas, Badajoz, Spain, 2003. ISBN Vol. 1 84-607-6698-5. p. 413-419
5. LACINOVÁ Ľ. Geneticky modifikované živočíchy. In: TIMKO, Jozef - SIEKEL, Peter *Geneticky modifikované organizmy.* Bratislava: Veda, nakladateľstvo SAV, 2004. ISBN 80-224-0834-4 p. 39-50
6. LACINOVÁ, Ľ. Interaction of L-type calcium channels with dihydropyridines. In: Bachárová Ljuba, Kyselovič Ján, Slezák Ján *Experimental hypertension and ischemic heart disease.* Bratislava: Veda, 2005. ISBN 80-224-0856-5. p. 9-22.
7. LACINOVÁ Ľ. From a Simple code to the Complex Trait. In: Kerra Denisa, Sedlák Pavel *entermultimediale*^{2} Praha: CIANT – International Centre for Art and New Technologies, 2005. ISBN 80-239-4927-6. p. 74.

1.5 Table of research outputs

*Table **Research outputs** shows research outputs in number of specified entries; these entries are then divided by FTE employees with a university degree (from Tab. Research staff) for all Organisation at the respective year; finally these entries are divided by the total salary budget (from Tab. Salary budget).*

Research outputs	2003			2004			2005			2006			total			
	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	averaged number per year	av. No. / FTE	av. No. / salary budget
chapters in monographs, books published abroad	2	0.08	0.20	1	0.04	0.10	1	0.04	0.09	0	0.00	0.00	4	1.0	0.04	0.09
chapters in monographs, books published in Slovakia	1	0.04	0.10	0	0.00	0.00	1	0.04	0.09	0	0.00	0.00	2	0.5	0.02	0.05
CC publications	21	0.81	2.11	18	0.68	1.73	12	0.48	1.11	29	1.21	2.55	80	20.0	0.79	1.88
scientific publications indexed by other databases (specify)	0	0.00	0.00		0.00	0.00		0.00	0.00		0.00	0.00	0	0.0	0.00	0.00
scientific publications in other journals	0	0.00	0.00	2	0.08	0.19	3	0.12	0.28	3	0.13	0.26	8	2.0	0.08	0.19
publications in proc. of international scientific conferences	37	1.43	3.72	62	2.34	5.97	52	2.10	4.81	35	1.46	3.08	186	46.5	1.84	4.38
publications in proc. of nat. scientific conferences	9	0.35	0.90	6	0.23	0.58	12	0.48	1.11	39	1.63	3.43	66	16.5	0.65	1.55
active participations at international conferences	37	1.43	3.72	62	2.34	5.97	52	2.10	4.81	34	1.42	2.99	185	46.3	1.83	4.35
active participations at national conferences	9	0.35	0.90	6	0.23	0.58	12	0.48	1.11	37	1.54	3.25	64	16.0	0.63	1.51

1.6 Renormalized publications[2]

Renormalized publications = number of CC publications in the given year times authorship's portion of the Organisation times the journal impact factor in 2005 divided by the median impact factor in the research field

Renormalised publications	2003			2004			2005			2006		
	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget
	8.300	0.320	0.834	12.970	0.489	1.250	7.050	0.284	0.652	19.640	0.818	1.728

1.7 Standard manuscript page count[3]

Standard manuscript page count	2003			2004			2005			2006		
	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget
page count	0	0.0	0.0	0	0.0	0.0	0	0.0	0.0	0	0.0	0.0

1.8 List of patents and patent applications

[2] This information is required only from the Organisations of the Section 2 of the Slovak Academy of Sciences.
[3] This information is required only from the Organisations of the Section 3 of the Slovak Academy of Sciences.

1.9 Supplementary information and/or comments on the scientific output of the Organisation

During the years 2003-2006 researchers of IMPG SAS published 80 papers in journals indexed in CC or in WOK. From these papers 22 were published in General Physiology and Biophysics i.e., in a journal that is published by IMPG SAS. This journal represents an international scientific platform for publication of papers concerning molecular and cellular physiology and biophysics with stable impact factor between 0.5-0.9. The scientists from our institute were asked to publish high quality papers in this journal to keep its quality comparable to other international journals. Quality of papers published by authors from our institute during the years 2003-2006 in General Physiology and Biophysics may be documented by fact that they were cited 31 times according WOK. These publications are in starting phase of citation because cited half-life in our Journal is according to WOK 6.3 years.

Papers of IMPG SAS researchers were cited 2453 times during the years 2003-2006. These scientometric data are documenting the high quality of research done in IMPG SAS.

We have computed the descriptor “renormalized publications” (chapter 1.6 in this questionnaire) according to rules that we obtained from Accreditation committee for Section 2 of Slovak Academy of Sciences. Unfortunately this descriptor is of very low adequacy for consideration of quality of journal in which papers were published. This may be documented by the following facts:

1. Considering journals generally believed to be (more or less) of similar quality we obtained for: Circulation – Impact factor (IF) 11.632, median impact factor (MIF) for the subject category: Cardiac & Cardiovascular Systems 1.559 i.e., ratio IF/MIF=**7.471**; Proc. Nat. Acad. Sci.USA – IF 10.231, MIF for the subject category Multidisciplinary Sciences 0.445 i.e., ratio IF/MIF=**22.991**; Am. J. Human Genet. – IF 12.649, MIF for the subject category Genetics & Heredity 2.626 i.e., ratio IF/MF=**4.817**.
2. When we consider journals that are believed to be of different quality we obtained for: Coll. Czech. Res. Commun. – IF 0.949, MIF for subject category Chemistry, Multidisciplinary 0.921, i.e., the ratio IF/MIF=**1.030**; Biochem. Biophys Res Commun. – IF 3.000, MIF for Subject Category Biophysics 2.193 i.e., the ratio IF/MIF=**1.368**.

Ratio IF/MIF is the basic feature of “renormalized publication” descriptor that is believed to be useful for comparison of journals in which papers were published.

Unfortunately this ratio will give unrealistic values without any real meaning for this purpose as it is documented on the above examples.

Doc. RNDr. Ľudovít Kádasi, DrSc. has published the following three papers as a member of international team of researchers that describe fundamental features of Huntington disease and cystic fibrosis:

- METZGER, S. – BAUER, P. – TOMIUK, J. – LACCONE, F. – DIDONATO, S. – GELLERA, C. – SOLIVERI, P. – LANGE, H.W. - WEIRICH-SCHWAIGER, H. – WENNING, G.K. – MELEGH, B. – HAVASI, V. – BALIKO, L. – WIECZOREK, S. – ARNING, L. – ZAREMBA, J. – SULEK, A. - HOFFMAN-ZACHARSKA, D. – BASAK, A.N. – ERSOY, N. – ZIDOVSKA, J. – KEBRDLOVA, V. – PANDOLFO, M. – RIBAI, P. – KADASI, L. – KVASNICOVA, M. – WEBER, B.H. – KREUZ, F. – DOSE, M. – STUHRMANN, M. - RIESS, O. The S18Y polymorphism in the UCHL1 gene is a genetic modifier in Huntington's disease. In Neurogenetics, Vol. 7, no. 1 (2006), p.27-30.
- METZGER, S. – BAUER, P. – TOMIUK, J. – LACCONE, F. – DIDONATO, S. – GELLERA, C. – MARIOTTI, C. – LANGE, H.W. - WEIRICH-SCHWAIGER, H. – WENNING, G.K. – SEPPI, K. – MELEGH, B. – HAVASI, V. – BALIKO, L. – WIECZOREK, S. – ZAREMBA, J. - HOFFMAN-ZACHARSKA, D. – SULEK, A. – BASAK, A.N. – SOYDAN, E. – ZIDOVSKA, J. – KEBRDLOVA, V. – PANDOLFO, M. – RIBAI, P. – KADASI, L. – KVASNICOVA, M. – WEBER, B.H.F. – KREUZ, F. – DOSE, M. – STUHRMANN, M. – RIESS, O. Genetic analysis of candidate genes modifying the age-at-onset in Huntington's disease. In Human Genetics, Vol. 120, no. 2 (2006), p. 285-292. 26.
- GROMAN, J.D. - HEFFERON, T.W. - CASALS, T. - BASSAS, L. - ESTIVILL, X. - DES GEORGES, M. - KOUDOVA, M. - FALLIN, M.D. - NEMETH, K. - FEKETE, G. - KÁDASI, L. - FRIEDMAN, K. - SCHWARZ, M. - BOMBIERI, C. - PIGNATTI, G.F. - KANAVAKIS, E. - TZETIS, M. - SCHWARTZ, M. - NOVELLI, G. - D APICE, M.R. - SOBCZYNSKA-TOMASZEWSKA, A. - BAL, J. - STUHRMANN, M. - MACEK, M.JR. - CLAUSTRES, M. - CUTTING G.R. Variation in a repeat sequence determines wheter a common variant of the cystic fibrosis transmembrane conductance regulator gene is pathogenic or benign. In American Journal of Human Genetics. Vol. 74. no. 1 (2004), p. 176-179.

Memberships in consortiums such as these have to be considered as an explicit international acceptation of scientific quality. However in expression of renormalized publication, these papers give only little contribution due to quantity of authors necessary for obtaining the data from several parts of world.

2. Responses to the scientific output

*Table **Citations** shows specified responses to the scientific outputs; these entries are then divided by the FTE employees with a university degree (from Tab. Research staff) for all Organisation at the respective year; finally these entries are divided by the total salary budget (from Tab. Salary budget).*

Citations	2002			2003			2004			2005			total			
	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	No. / FTE	No. / salary budget	number	averaged number per year	av. No. / FTE	av. No. / salary budget
Web of Science	583	22.5	58.6	559	21.1	53.9	641	25.8	59.3	673	28.0	59.2	2456	614.0	24.3	231.1
SCOPUS	0	0.0	0.0	3	0.1	0.3	15	0.6	1.4	13	0.5	1.1	31	7.8	0.3	2.9
out of Database	1	0.0	0.1	3	0.1	0.3	2	0.1	0.2	5	0.2	0.4	11	2.8	0.1	1.0
in monographs, conf. proceedings and other publications abroad	0	0.0	0.0	0	0.0	0.0	0	0.0	0.0	2	0.1	0.2	2	0.5	0.0	0.2
in monographs, conf. proceedings and other publications in Slovakia	0	0.0	0.0	0	0.0	0.0	0	0.0	0.0	0	0.0	0.0	0	0.0	0.0	0.0

2.1 List of 10 top-cited publications and number of their citations in the assessment period

1. MARX,S.O. - GABURJAKOVA,J. - GABURJAKOVA,M. - HENRIKSON,C. - ONDRIAŠ,K. - MARKS,A.R. COUPLED GATING BETWEEN CARDIAC CALCIUM RELEASE CHANNELS (RYANODINE RECEPTORS). IN *CIRCULATION RESEARCH*. VOL 88 (2001), P. 1151-1158. **90 times cited during years 2003-2005**

2. MARX, SO. – ONDRIAŠ, K. - MARKS,AR. COUPLED GATING BETWEEN INDIVIDUAL SKELETAL MUSCLE CA2+ RELEASE CHANNELS. IN *SCIENCE*. VOL 281 (1998), P. 818-821. **80 times cited during years 2003-2005**

3. HAMOSH, A. (COORDINATOR) - KADASI, L. THE CYSTIC FIBROSIS GENOTYPE-PHENOTYPE CONSORTIUM. STUDY. CORRELATION BETWEEN GENOTYPE AND PHENOTYPE IN PATIENTS WITH CYSTIC-FIBROSIS. IN *NEW ENGLAND JOURNAL OF MEDICINE***.** VOL 329, NO. 18, (1993), P. 1308-1313 .**60 times cited during years 2003-2005**

4. GROMAN, JD. – HEFFERON, TW. – CASALS, T. – BASSAS, L. – ESTIVILL, X. - DES GEORGES, M. – KOUDOVA, M. – FALLIN, MD. – NEMETH, K. – FEKETE, G. – KÁDASI, L. – FRIEDMAN, K. – SCHWARZ,M. – BOMBIERI, C. – PIGNATTI, GF. – KANAVAKIS, E. – TZETIS, M. – SCHWARTZ, M. – NOVELLI, GD. – APICE, MR. - SOBCZYNSKA-TOMASZEWSKA, A. – BAL, J. – STUHRMANN, M. – MACEK, MJR. – CLAUSTRES, M. – CUTTING, GR. VARIATION IN A REPEAT SEQUENCE DETERMINES WHETER A COMMON VARIANT OF THE CYSTIC FIBROSIS TRANSMEMBRANE CONDUCTANCE REGULATOR GENE IS PATHOGENIC OR BENIGN. IN *AMERICAN JOURNAL OF HUMAN GENETICS*. VOL 74, (2004) P. 176-179**. 30 times cited during years 2003-2005**

5. KLUGBAUER N., LACINOVÁ Ľ., FLOCKERZI V., HOFMANN F. (1995): STRUCTURE AND FUNCTIONAL EXPRESSION OF A NEW MEMBER OF THE TETRODOTOXIN-SENSITIVE SODIUM CHANNEL FAMILY FROM HUMAN NEUROENDOCRINE CELLS. EMBO J. 14: 1084-1090. **27 times cited during years 2003-2005**

6. ZAHRADNÍKOVÁ A, MINAROVIČ I, VENEMA RC, MÉSZÁROS LG: INACTIVATION OF THE CARDIAC RYANODINE RECEPTOR CALCIUM RELEASE CHANNEL BY NITRIC OXIDE. CELL CALCIUM 22: 447-454, 1997 27 **times cited during years 2003-2005**

7. KLUGBAUER N., MARAIS E., LACINOVÁ Ľ., HOFMANN F. (1999): A T-TYPE CALCIUM CHANNEL FROM MOUSE BRAIN. PFLÜGERS ARCH., 437: 710 - 715. **22 times cited during years 2003-2005**

8. LACINOVÁ Ľ., KLUGBAUER N., HOFMANN F. (2000) LOW VOLTAGE ACTIVATED CALCIUM CHANNELS: FROM GENES TO FUNCTION. GEN. PHYSIOL. BIOPHYS., 19: 121 - 136. **19 times cited during years 2003-2005**

9. MOJZISOVA A., KRIZANOVA O., ZACIKOVA L., KOMINKOVA V., ONDRIAS K EFFECT OF NICOTINIC ACID ADENINE DINUCLEOTIDE PHOSPHATE ON RYANODINE CALCIUM RELEASE CHANNEL IN HEART..: PFLUGERS ARCH. 441, 674-677, 2001 **18 times cited during years 2003-2005**

10. BARANCIK M, BOHACOVA V., KVACKAJOVA J., HUDECOVA S., KRIZANOVA O., BREIER A.: B203580, A SPECIFIC INHIBITOR OF P38-MAPK PATHWAY, IS A NEW REVERSAL AGENT OF P-GLYCOPROTEIN-MEDIATED MULTIDRUG RESISTANCE. EUR.J. PHARMACEUT SCI. 14, 29-36, 2001. **14 times cited during years 2003-2005**

2.2 List of top-cited authors from the Organisation (at most 10 % of the research employees) and their number of citations in the assessment period

1. RNDr. Karol Ondriaš, DrSc. - 479
2. RNDr. Ľubica Lacinová, DrSc. - 370
3. Mgr. Marta Gaburjáková, PhD. - 333

2.3 Supplementary information and/or comments on responses to the scientific output of the Organisation

Papers of other following researchers: RNDr. Miroslav Barančík, CSc., Ing. Albert Breier, DrSc., Mgr. Jana Gaburjaková, PhD., RNDr. Ľudovít Kádasi, DrSc., RNDr. Marta Novotová, CSc., RNDr. Peter Proks, CSc., Ing. Alexandra Zahradníková, CSc. were cited more than 100 times during the years 2003-2006.

3. Research status of the Organisation in the international and national context

- **International/European position of the Organisation**

3.1 List of the most important research activities documenting international importance of the research performed by the Organisation, incl. major projects (details of projects should be supplied under Indicator 4). Collective membership in the international research organisations, in particular within the European Research Area

Projects:

Molecular mechanisms of calcium signaling in cardiac excitation-contraction coupling Ing. Alexandra Zahradníková, CSc. Howard Hughes Medical Institute International Scholar's Award. HHMI 55000343, Howard Hughes Medical Institute, Bethesda, MD, USA

Calcium signaling in cardiac excitation-contraction coupling S. Györke, Department of Physiology, TTU HSC, Lubbock, TX, USA Ing. Alexandra Zahradníková, CSc., RNDr. Ivan Zahradník, CSc., ÚMFG SAV, Fogarty International Research Collaboration Award (FIRCA), 1 R03 TW05543-01, NIH, USA.

Role of L-type and T-type Ca^{2+} channels in neuronal excitability, RNDr. Ľubica Lacinová, CSc., Volkswagen grant, in cooperation with Prof. Franz

Hofmann, Technische Universität München; Institut für Pharmakologie und Toxikologie; Biedersteiner Straβe 29; 80802 München; SRN, 01/2002-12/2004

Reorganization of calcium signaling in heart failure. S. Györke, Department of Physiology, TTU HSC, Lubbock, TX, USA Ing. Alexandra Zahradníková, CSc., RNDr. Ivan Zahradník, CSc., ÚMFG SAV, Fogarty International Research Collaboration Award (FIRCA), 2R03TW005543-05, NIH, USA

P-glycoproteín Mediated Multidrug Resistance in Radiation-Associated Hematological Malignancies Following the Chernobil Accident. Coordinator: Ing. Albert Breier, DrSc. NATO Grant PDD(CP)-(CBP.NUKR.CLG 982646)

Marie Curie Research Training Network – projekt CAVNET, MRTN-CT-2006-035367, RNDr. Ľubica Lacinová, DrSc., Marie Curie Research Training Network – projekt CAVNET, 12/2006 – 12/2010

Integrated Project 6FP LifeSciHealth **Genomics of Cardiomyocyte Signalling to Treat and Prevent Heart Failure (EUGeneHeart),** Coordinator: Prof. Gerd HASENFUß (Georg-August-University, Göttingen), Researchers: Ing. Alexandra Zahradníková, CSc., RNDr. Ivan Zahradník, CSc., ÚMFG SAV

STREP Project 6FP LifeSciHealth **Control of intracellular Calcium and Arrhythmias (CONTICA),** Coordinator: Prof. Dr. med. Burkert Pieske (Universität Göttingen, Göttingen), Researchers: Ing. Alexandra Zahradníková, CSc., RNDr. Ivan Zahradník, CSc., ÚMFG SAV

Coupled gating between intracellular calcium release channels. Andrew R. Marks, M.D., Clyde and Helen Wu Professor of Molecular Cardiology, Professor of Medicine and Professor of Pharmacology Director, Molecular Cardiology Program, Columbia University College of Physicians & Surgeons P&S 9-401, Box 65, 630 West 168th Street, New York, NY 10032, 6/2002-6/2005, RNDr. Karol Ondriaš, DrSc., IMPG SAS, Fogarty International Research Collaboration Award (FIRCA), 2R03TW000949-04A1, NIH, USA.

Role of mitochondrial channels in cardioprotection. RNDr. Karol Ondriaš, DrSc., NATO grant SA (LST.CLG.979217) Cooperation: Prof. Szewczyk Adam, PhD., DSc., Nencki Institute of Experimental Biology, Polish Academy of Sciences, Pasteura 3, 02-093 Warsaw, Poland, 07/2002-12/2004

Modulation of compounds affecting the excitation-contraction couling of slow and fast muscles of the rat. Doc. Ing. Oľga Križanová, DrSc., NATO grant 979876 Cooperation: RNDr. Tomáš Soukup, PhD., Institute of Physiology, Academy of Sciences of the Czech Republic, Videnska 1083, CZ-142 20 Praha

„BIOMEMBRANES: cross-sectional educational program for graduate students and young scientists in life sciences. European Social Fond Project. JPD 3 2005 1-010 (Code 13120200072) Coordinator: ÚBGŽ SAV,

RNDr. Ivan Hapala, CSc. Responsible in IMPG SAS: Ing. Albert Breier, DrSc., RNDr. Ľubica Lacinová DrSc.

3.2 List of international conferences (co-) organised by the Organisation

1. XXII. Xenobiochemical Symposium, Smolenice Castle, June 9-11, 2003
2. 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice Castle, June 5-9, 2004
3. XX. Biochemical Symposium, Hotel Sorea, Piešťany, September 12-14, 2006
4. Workshop on Cellular and Molecular Aspects of Cardiac Function, Smolenice Castle, March 1-3, 2006

3.3 List of international journals edited/published by the Organisation

1. General Physiology and Biophysics

3.4 List of edited proceedings from international scientific conferences and other proceedings

1. XXII. Xenobiochemical Symposium, Smolenice Castle, June 9-11, 2003, Program and Abstracts
2. 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice Castle, June 5-9, 2004, Program and Abstracts
3. XX. Biochemical Symposium, Hotel Sorea, Piešťany, September 12-14, 2006, Program and Abstracts
4. Workshop on Cellular and Molecular Aspects of Cardiac Function, Smolenice Castle, March 1-3, 2006, Program and Abstracts

• National position of the Organisation

3.1 List of selected most important national projects (Centres of Excellence, National Reference Laboratories, Agency for the Promotion of Research and Development (APVV/APVT), National Research Programmes, Scientific Grant Agency of the Slovak Academy of Sciences and the Ministry of Education (VEGA), and others)

Projects:

Genomics of cardiovascular diseases for healthier human population. doc. Ing. Oľga Križanová, DrSc, 05/2003-12/2005, ŠPVV SP51/0280800/0280802

Transport and signal mechanisms of biological membranes under normal and pathological conditions, RNDr. Karol Ondriaš, DrSc., 9/2002-9/2005, APVT-51-01-013802

Signaling and transport function of biological membranes under normal and pathological conditions, RNDr. Karol Ondriaš, DrSc., 10/2005-12/2007, APVT-51-027404

Mechanism of excitation-contraction coupling in normal and failing mammalian myocardium, RNDr. Ivan Zahradník, CSc., 01/2005-12/2007, APVT-51-31104

Building of Biotechnological Centrum – BITCET. Contract 337/2003. Coordinator: prof. RNDr. Jaroslav Pastorek, DrSc., Institute of Virology SAS, Responsible in IMPG SAS: Ing. Albert Breier, DrSc.

Adaptation of heart undrer pathological conditions. Regulation mechanisms. Coordinator: Ing. Monika Strnisková,PhD., 05/2003-12/2005, Responsible in IMPG SAS: Mgr. Marta Gaburjáková, PhD.

3.2 List of national scientific conferences (co)-organised by the Organisation

1. Drobnicov memoriál, 2. ročník, Hotel Senec, Senec, November 12-14, 2003
2. Seminár “Genetické technológie: hrozba alebo nádej? IV”, České centrum v Bratislave, Január 20, 2003
3. Drobnicov memoriál, 3. ročník, Makov, June 15-17, 2005
4. 17. Izakovičov memorial, Bratislava, October 5-6, 2006
5. Kurz “Biomembrány”, Blok II: Funkcie biologických membrane v bunkách živočíchov, UMFG SAV, Bratislava, October 6-7, 2006

3.3 List of national journals published by the Organisation

3.4 List of edited proceedings of national scientific conferences/events

1. Drobnicov memoriál, 2. ročník, Hotel Senec, Senec, November 12-14, 2003, Program and Abstracts

2. Drobnicov memoriál, 3. ročník, Makov, June 15-17, 2005, Program and Abstracts
3. 17. Izakovičov memoriál, Bratislava, October 5-6, 2006, Program and Abstracts

- **International/European position of the individual researchers**

3.1 List of invited/keynote presentations at international conferences, documented by an invitation letter or programme

Ing. A. Zahradníková, CSc.	- Human Proteome Organization (HUPO) 2nd Annual & International Union of Biochemistry and Molecular Biology (IUBMB) XIX Joint World Congress, Montreal, Canada, October 8-11, 2003 - MBI workshop on "Synapses and Muscle", March 8-12, 2004, Columbus, OH, USA

3.2 List of employees who served as members of the organising and/or programme committees for international conferences

1. Ing. Albert Breier, DrSc.	Member of Programm Commettee XXII. Xenobiochemical Symposium, Smolenice Castle, 2003
2. doc. Ing. Oľga Križanová, DrSc.	Member of Programm Commettee XXII. Xenobiochemical Symposium, Smolenice Castle, 2003
3. Ing. Dagmar Zbyňovská, CSc.	Member of Programm Commettee XXII. Xenobiochemical Symposium, Smolenice Castle, 2003
4. Ing. Albert Breier, DrSc.	Member of Organizing Commettee XXII. Xenobiochemical Symposium, Smolenice Castle, 2003
5. PhDr. Zuzana Klimešová	Member of Organizing Commettee XXII. Xenobiochemical Symposium, Smolenice Castle, 2003
6. doc. Ing. Oľga Križanová, DrSc.	Member of Organizing Commettee XXII. Xenobiochemical Symposium, Smolenice Castle, 2003
7. Silvia Marková	Member of Organizing Commettee XXII. Xenobiochemical Symposium, Smolenice Castle, 2003
8. Andrej Opálek	Member of Organizing Commettee XXII. Xenobiochemical Symposium, Smolenice Castle, 2003

9. Ing. Dagmar Zbyňovská, CSc.	Member of Organizing Commettee XXII. Xenobiochemical Symposium, Smolenice Castle, 2003
10. doc. RNDr. Ľudovít Kádasi	Member of Organizing Commettee 7th Symposium for Cystic Fibrosis, Bratislava, 2003
11. doc. Ing. Oľga Križanová, DrSc.	Member of Organizing Commettee 8th Symposium on Catecholamines and Other Neurotransmitters in Stress, Smolenice, 2003
12. Ing. Alexandra Zahradníková, CSc.	Member of Organizing Commettee Local Calcium Signaling, Montreal, Canada, 2003
13. Ing. Albert Breier, DrSc.	Member of Programm Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
14. Mgr. M. Gaburjáková, PhD.	Member of Programm Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
15. doc. Ing. Oľga Križanová, DrSc.	Member of Programm Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
16. RNDr. Ľubica Lacinová, DrSc.	Member of Programm Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
17. RNDr. Marta Novotová, CSc.	Member of Programm Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
18. RNDr. Karol Ondriaš, DrSc.	Member of Programm Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
19. Ing. Albert Breier, DrSc.	Member of Organizing Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
20. PhDr. Zuzana Klimešová	Member of Organizing Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
21. doc. Ing. Oľga Križanová, DrSc.	Member of Organizing Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
22. RNDr. Ľubica Lacinová, DrSc.	Member of Organizing Commettee 4th International Symposium on Membrane

	Channels, Transporters and Receptors, Smolenice, 2004
23. RNDr. Marta Novotová, CSc.	Member of Organizing Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
24. Ing. Dagmar Zbyňovská, CSc.	Member of Organizing Commettee 4th International Symposium on Membrane Channels, Transporters and Receptors, Smolenice, 2004
25. Ing. Alexandra Zahradníková, CSc.	Chair of Ryanodine receptors, 49th Annual Meeting of the Biophysical Soiety, Long Beach, California, 2005
26. Ing. Albert Breier, DrSc.	Coordinator, Workshop on Cellular and Molecular Aspects of Cardiac Function, Smolenice, 2006

3.3 List of employees who served as members of important international scientific bodies (e.g. boards, committees, editorial boards of scientific journals)

MUDr. B. Uhrík, CSc.	- Coordinated editor of journal General Physiology and Biophysics (GPB)
Ing. A. Breier, DrSc.	- editor of journal GPB
RNDr. K. Ondriaš, DrSc.	- Member of editorial boards of journal GPB
RNDr. M. Barančík, CSc.	- International Society for Heart Research, Member
Ing. A. Breier, DrSc.	- European Academy of Sciences, Member - International Society for Heart Research, Member - European Calcium Society, Member - European Federation for Pharmaceutical Sciences, Member - Member of editorial board of Journals Recent Patent on Anticancer Drug Discovery, Bentham Science Publishers, Ltd. http://www.bentham.org/pra/EBM.htm
doc. RNDr. Ľ. Kádaši, DrSc.	- Európska spoločnosť humánnej genetiky, Member - Genetická spoločnosť Gregora Mendela, Member
doc. Ing. O. Križanová, DrSc.	- International Society for Heart Research, Member - European Peptide Society, Member
RNDr. Ľ. Lacinová, DrSc.	- Member of editorial board of journal Sedmá generace, Hnutí Duha v Brne
RNDr. M. Novotová, CSc.	- European Muscle Club, Member

RNDr. J. Orlický, CSc.	- International Brain Research Organization, Member - The New York Academy of Sciences, Member
RNDr. P. Proks, CSc.	- British Diabetic Association, Member - Physiological Society Affiliate, Member - Biophysical Society, Member
Ing. Z. Sulová, CSc.	- European Calcium Society, Member
MUDr. B. Uhrík, CSc.	- International Brain Research Organization, Member - European Calcium Society, Member - International Society for Heart Research, Member
RNDr. I. Zahradník, CSc.	- Biophysical Society (USA), Member - International Society for Heart Research, Member - Česká a Slovenská Neurochemická spoločnosť, Member - European Society of Cardiology, Member - European Working Group for Cardiac Cellular Electrophysiology, Member
Ing. A. Zahradníková, CSc.	- Biophysical Society (USA), Member - International Society for Heart Research, Member - Česká a Slovenská Neurochemická spoločnosť, Member - European Society of Cardiology, Member - European Working Group for Cardiac Cellular Electrophysiology, Member - Editor of journal Central European Journal of Biology, VERSITA in partnership SPRINGER http://www.versita.com/science/lifesciences/cejb/editors/alexandra_zahradnikova/

3.4 List of international scientific awards and distinctions

RNDr. Peter Proks, CSc.	- Wellcome Trust Integrative Physiology Initiative Poster Prize

• National position of the individual researchers

3.1 List of invited/keynote presentations at national conferences documented by an invitation letter or programme

Ing. A. Zahradníková, CSc.	- 40. Kuželov seminár, 27.11. 2003, Prírodovedecká Fakulta UK, Bratislava

	- Workshop on Cellular and Molecular Aspects of Cardiac Function, Smolenice Castle 1-3 March 2006
doc. Ing. Ol Križanová, CSc.	- Workshop on Cellular and Molecular Aspects of Cardiac Function, Smolenice Castle 1-3 March 2006

3.2 List of employees who served as members of organising and programme committees of national conferences

Ing. Albert Breier, DrSc.	- Member of Organizing Committee, Drobnicov memorial 2003, Senec
PhDr. Zuzana Klimešová	- Member of Organizing Committee, Drobnicov memorial 2003, Senec
Silvia Marková	- Member of Organizing Committee, Drobnicov memorial 2003, Senec
Ing. Zdena Sulová, CSc.	- Member of Organizing Committee, Drobnicov memorial 2003, Senec
Ing. Dagmar Zbyňovská, CSc.	- Member of Organizing Committee, Drobnicov memorial 2003, Senec
Ing. Albert Breier, DrSc.	- Member of Organizing Committee, Študentská vedecká konferencia, 2003, Bratislava
doc. RNDr. Ľudovít Kádaši, DrSc.	- Chairperson of Organizing Committee, 15. Izakovičov memorial, 2004, Nitra
doc. RNDr. Ľudovít Kádaši, DrSc.	- Chairperson of Organizing Committee, 16. Izakovičov memorial, 2005, Štrbské Pleso
Ing. Albert Breier, DrSc.	- Member of Organizing Committee, Drobnicov memorial 2005, Makov
PhDr. Zuzana Klimešová	- Member of Organizing Committee, Drobnicov memorial 2005, Makov
Ing. Zdena Sulová, CSc.	- Member of Organizing Committee, Drobnicov memorial 2005, Makov
Ing. Dagmar Zbyňovská, CSc.	- Member of Organizing Committee, Drobnicov memorial 2005, Makov
doc. RNDr. Ľudovít Kádaši, DrSc.	- Chairperson of Organizing Committee, 17. Izakovičov memorial, 2006, Bratislava
Ing. Albert Breier, DrSc.	- Member of Program Committee, XX. Biochemical symposium, 2006, Piešťany
Doc. Ing. Oľga Križanová, DrSc.	- Member of Program Committee, XX. Biochemical symposium, 2006, Piešťany
Ing. Albert Breier, DrSc.	- Member of Organizing Committee, XX. Biochemical symposium, 2006, Piešťany
Doc. Ing. Oľga Križanová, DrSc.	- Member of Organizing Committee, XX. Biochemical symposium, 2006, Piešťany
Silvia Marková	- Member of Organizing Committee, XX. Biochemical symposium, 2006, Piešťany

Andrej Opálek	- Member of Organizing Committee, XX. Biochemical symposium, 2006, Piešťany
Ing. Zdena Sulová, CSc.	- Member of Organizing Committee, XX. Biochemical symposium, 2006, Piešťany
Ing. Dagmar Zbyňovská, CSc.	- Member of Organizing Committee, XX. Biochemical symposium, 2006, Piešťany

3.3 List of employees serving in important national scientific bodies (e.g. boards, committees, editorial boards of scientific journals)

RNDr. Ľ. Lacinová, DrSc.	- Member of editorial boards of journal Mosty - Member of editorial boards of journal General Physiology and Biophysics
Ing. A. Breier, DrSc.	- Slovenská spoločnosť pre biochémiu a molekulovú biológiu, Scientific Secretary - Slovenská fyziologická spoločnosť, Member - Member of Komisia experimentálnej kardiológie pri českej a slovenskej fyziologickej spoločnosti
RNDr. V. Boháčová, CSc.	- Slovenská spoločnosť pre biochémiu a molekulovú biológiu, Member
Ing. P. Dočolomanský, CSc.	- Slovenská spoločnosť pre biochémiu a molekulovú biológiu, Member
doc. RNDr. Ľ. Kádaši, DrSc.	- Slovenská spoločnosť lekárskej genetiky, President
doc. Ing. O. Križanová, DrSc.	- Slovenská spoločnosť pre biochémiu a molekulovú biológiu, Member
RNDr. Ľ. Lacinová, DrSc.	- Slovenská fyziologická spoločnosť, Member - Slovenská spoločnosť pre neurovedy, Member - Slovenská biochemická spoločnosť, Member
Mgr. Ľ. Lenčešová, PhD.	- Slovenská spoločnosť pre biochémiu a molekulovú biológiu, Member
Ing. P. Novák, PhD.	- Slovenská biofyzikálna spoločnosť, Member
RNDr. M. Novotová, CSc.	- Slovenská fyziologická spoločnosť, Member
RNDr. J. Pavelková, CSc.	- Slovenská fyziologická spoločnosť, Member
MUDr. B. Uhrík, CSc.	- Slovenská fyziologická spoločnosť, Member
RNDr. I. Zahradník, CSc.	- Slovenský komitét pre biofyziku, vice-president - Slovenská biofyzikálna spoločnosť, vice-president - Slovenská fyziologická spoločnosť, Member n
Ing. A. Zahradníková, CSc.	- Slovenská fyziologická spoločnosť, Member - Slovenská biofyzikálna spoločnosť, Member
Ing. D. Zbyňovská, CSc.	- Slovenská spoločnosť pre biochémiu a molekulovú biológiu, Member

3.4 List of national awards and distinctions

Mgr. M. Gaburjáková, PhD.	- The winner of Competition of young scientists for the best publication on the occasion of the 50th anniversary of the Slovak Academy of Sciences, 2003
Mgr. J. Gaburjáková, PhD.	- After her PhD thesis defense she won the competition for a postdoctoral research position issued for the best PhD graduates by the Presidium of the Slovak Academy of Sciences, 2003
Mgr. M. Pavlovičová, PhD.	- After her PhD thesis defense she won the competition for a postdoctoral research position issued for the best PhD graduates by the Presidium of the Slovak Academy of Sciences, 2003
Mgr. G. Minárik	– The prize of the Slovak Medical Society for the best publication of young researchers in 2003 in the field medical genetics, 2004
Ing. A. Zahradníková, CSc.	- The bronze medal of the Slovak Medical Society, 2004
Doc. Ing. O. Križanová, DrSc.	- The prize of the Ministry of Education of the Slovak Republic for Science and Technique in 2004, in the category Research and Development, 2004
MUDr. B. Uhrík, CSc.	- Commemorative Plaque of the Slovak Academy of Sciences, 2005
doc. RNDr.Ľ. Kádaši, DrSc.	- Izakovič Medal, the prize granted by the Slovak Medical Society for the development of medical genetics in Slovakia, 2006
doc. RNDr.Ľ. Kádaši, DrSc.	- The prize of the Slovak Medical Society for the best publication in the field of Medical Genetics in 2006
Ing. A. Breier, DrSc.	
RNDr. M. Barančík, CSc.	
MUDr. B. Uhrík, CSc.	- The prize of the Slovak Medical Society for the best publication in the field of Medical Physiology in 2006

- **Supplementary information and/or comments documenting international and national status of the Organisation**

IMPG is recognised as a standard scientific institution of international significance. Main criteria documenting international status of IMPG SAS are number of publications in renowned international journals and their extensive citedness.

4. Project structure, research grants and other funding resources

- **International projects and funding**

4.1 List of major projects within the European Research Area – 5th and 6th Framework Programme of the EU, European Science Foundation, NATO, COST, INTAS, CERN, etc. (here and in items below please specify: type of project, title, grant number, duration, funding, responsible person in the Organisation and his/her status in the project, e.g. coordinator, principal investigator, investigator)

Molecular mechanisms of calcium signaling in cardiac excitation-contraction coupling Ing. Alexandra Zahradníková, CSc. Howard Hughes Medical Institute International Scholar's Award. HHMI 55000343, Howard Hughes Medical Institute, Bethesda, MD, USA

Calcium signaling in cardiac excitation-contraction coupling S. Györke, Department of Physiology, TTU HSC, Lubbock, TX, USA Ing. Alexandra Zahradníková, CSc., RNDr. Ivan Zahradník, CSc., ÚMFG SAV, Fogarty International Research Collaboration Award (FIRCA), 1 R03 TW05543-01, NIH, USA.

Role of L-type and T-type Ca^{2+} channels in neuronal excitability, RNDr. Ľubica Lacinová, CSc., Volkswagen grant, in cooperation with Prof. Franz Hofmann, Technische Universität München; Institut für Pharmakologie und Toxikologie; Biedersteiner Straβe 29; 80802 München; SRN, 01/2002-12/2004

Reorganization of calcium signaling in heart failure. S. Györke, Department of Physiology, TTU HSC, Lubbock, TX, USA Ing. Alexandra Zahradníková, CSc., RNDr. Ivan Zahradník, CSc., ÚMFG SAV, Fogarty International Research Collaboration Award (FIRCA), 2R03TW005543-05, NIH, USA

P-glycoproteín Mediated Multidrug Resistance in Radiation-Associated Hematological Malignancies Following the Chernobil Accident. Coordinator: Ing. Albert Breier, DrSc. NATO Grant PDD(CP)-(CBP.NUKR.CLG 982646)

Marie Curie Research Training Network – projekt CAVNET, MRTN-CT-2006-035367, RNDr. Ľubica Lacinová, DrSc., Marie Curie Research Training Network – projekt CAVNET, 12/2006 – 12/2010

Integrated Project 6FP LifeSciHealth **Genomics of Cardiomyocyte Signalling to Treat and Prevent Heart Failure (EUGeneHeart),** Coordinator: Prof. Gerd HASENFUß (Georg-August-University, Göttingen), Researchers: Ing. Alexandra Zahradníková, CSc., RNDr. Ivan Zahradník, CSc., ÚMFG SAV

STREP Project 6FP LifeSciHealth **Control of intracellular Calcium and Arrhythmias (CONTICA),** Coordinator: Prof. Dr. med. Burkert Pieske (Universität Göttingen, Göttingen), Researchers: Ing. Alexandra Zahradníková, CSc., RNDr. Ivan Zahradník, CSc., ÚMFG SAV

Coupled gating between intracellular calcium release channels. Andrew R. Marks, M.D., Clyde and Helen Wu Professor of Molecular Cardiology, Professor of Medicine and Professor of Pharmacology Director, Molecular Cardiology Program, Columbia University College of Physicians & Surgeons P&S 9-401, Box 65, 630 West 168th Street, New York, NY 10032, 6/2002-6/2005, RNDr. Karol Ondriaš, DrSc., ÚMFG SAV, Fogarty International Research Collaboration Award (FIRCA), 2R03TW000949-04A1, NIH, USA.

Role of mitochondrial channels in cardioprotection. RNDr. Karol Ondriaš, DrSc., NATO grant SA (LST.CLG.979217) Cooperation: Prof. Szewczyk Adam, PhD., DSc., Nencki Institute of Experimental Biology, Polish Academy of Sciences, Pasteura 3, 02-093 Warsaw, Poland, 07/2002-12/2004

Modulation of compounds affecting the excitation-contraction couling of slow and fast muscles of the rat. Doc. Ing. Oľga Križanová, DrSc., NATO grant 979876 Cooperation: RNDr. Tomáš Soukup, PhD., Institute of Physiology, Academy of Sciences of the Czech Republic, Videnska 1083, CZ-142 20 Praha

„BIOMEMBRANES: cross-sectional educational program for graduate students and young scientists in life sciences. European Social Fond Project. JPD 3 2005 1-010 (Code 13120200072) Coordinator: ÚBGŽ SAV, RNDr. Ivan Hapala, CSc. Responsible in IMPG SAS: Ing. Albert Breier, DrSc., RNDr. Ľubica Lacinová DrSc.

4.2 List of other international projects incl. funding

„Štefánik“ – č. 11, **Architecture cellulaire et transferts d´energie dans les cellules cardiaques et musculaires**, RNDr. Marta Novotová, CSc., in cooperation with PhD. R. Ventura-Clapier, INSERM, Unite 446, Laboratoire de Cardiologie Cellulaire et Moléculaire, Faculté de Pharmacie, Université de Paris-Sud, 5 rue Jean Baptiste Clément, 92 296 Chatenay-Malabry, France.
2004 - 2005

4.3 List of other important projects and collaborations without direct funding

Localization of S100A1 in human heart muscle cells. MUDr. Branislav Uhrík, CSc., in cooperation with Division of Clinical Chemistry and Biochemistry, Department of Pediatrics, University of Zurich, Zurich, Switzerland

• National projects and funding

4.1 List of projects supported by the Agency for the Promotion of Research and Development (APVV/APVT), National Research Programmes, and their funding

Transport and signal mechanisms of biological membranes under normal and pathological conditions, RNDr. Karol Ondriaš, DrSc., 9/2002-9/2005, APVT-51-01-013802

Signaling and transport function of biological membranes under normal and pathological conditions, RNDr. Karol Ondriaš, DrSc., 10/2005-12/2007, APVT-51-027404

Mechanism of excitation-contraction coupling in normal and failing mammalian myocardium, RNDr. Ivan Zahradník, CSc., 01/2005-12/2007, APVT-51-31104

Genomics of cardiovascular diseases for healthier human population. doc. Ing. Oľga Križanová, DrSc, 05/2003-12/2005, ŠPVV SP51/0280800/0280802

Adaptation of heart under pathological conditions. Regulatio mechanisms. Ing. Monika Strnisková, RNDr. Miroslav Barančík, CSc., 05/2003 – 12/2005, in cooperation with UVS SAV

Building of Biotechnological Center – BITCET. Zmluva o dielo 337/2003. Head: doc. RNDr. Jaroslav Pastorek, DrSc. In cooperation with VÚ SAV

The use of variability of mitochondiral and Y chromosome speciphic DNA in the personal identification, Orderer: Ministerstvo vnútra SR, 01/2002-12/2003, Cooperation: ÚMFG SAV, PriF UK

4.2 Number of projects supported by the Scientific Grant Agency of the Slovak Academy of Sciences and the Ministry of Education (VEGA) for each year, and their funding

VEGA	2003	2004	2005	2006
number	10	15	15	14
funding (millions of SKK)	1.260	1.834	1.681	1.870

- **Summary of funding from external resources**

External resources	2003	2004	2005	2006	total	average
external resources (millions of SKK)	15.360	15.280	18.240	8.520	57.400	14.350
external resources transfered to coooperating research organisations (millions of SKK)	0.000	0.000	0.000	0.000	0.000	0.000
ratio between external resources and total salary budget	1.543	1.472	1.688	0.749	5.452	1.363
overall expenditures (millions of SKK)	29.902	33.628	36.622	32.147	132.298	33.074

Founding from VEGA and from MVTS (international scientific and technology cooperatin) were included to the institutional budget.

- **Supplementary information and/or comments on research projects and funding resources**

5. Organisation of PhD studies, other pedagogical activities

5.1 List of accredited programmes of doctoral studies (as stipulated in the previously effective legislation as well as in the recently amended Act on the Universities)

In the previous period the Institute was a training centre in the subject field 11-57-9 biophysics.

Under the current law IMPG SAS is accredited in the following programs:

4.1.22 biochemistry

4.1.12 biophysics

4.2.10 animal physiology

5.2 Summary table on doctoral studies (number of internal/external PhD students; number of students who completed their study by a successful thesis defence; number of PhD students who quitted the programme)

PhD study	12/31/2003			12/31/2004			12/31/2005			12/31/2006		
number of potential PhD supervisors	26			27			27			31		
PhD students	number	defended thesis	students quitted	number	defended thesis	students quitted	number	defended thesis	students quitted	number	defended thesis	students quitted
internal	7	3	1	8	1	1	10	2	1	10	2	3
external	0	0	0	2	0	0	2	0	0	2	0	0
supervised at external institution by the research employees of the assessed organisation	3	0	0	3	0	0	3	3	0	0	0	0

5.3 Postdoctoral positions supported by

a) external funding (specify the source)

b) internal funding - the Slovak Academy of Sciences Supporting Fund of Stefan Schwarz

In 2003 we gained two postdoctoral positions supported from the Schwarz' fond. New rules introduced in the Schwarz' fond, an namely an impossibility to draw the support for a doctorand during his stay abroad, we refrained from applications to gain support from this fond, because our best doctorands after completing the studies and defending PhD thesis apply generally for postdoctoral positions abroad, their applications being successful in most cases.

5.4 Summary table on pedagogical activities in undergraduate programmes for each year

Teaching	2003	2004	2005	2006
lectures (hours/year)	62	45	96	144
practicum courses (hours/year)	382	86	213	168
supervised diploma works (in total)	8	10	13	8
members in PhD committees (in total)	7	7	8	5
members in DrSc. committees (in total)	1	2	4	4
members in university/faculty councils (in total)	0	1	1	1
members in habilitation/inauguration committees (in total)	1	2	2	0

5.5 List of published university textbooks

5.6 Number of published academic course books

„Funkcie biologických membrán v bunkách živočíchov“ (Function of biological membranes in animal cells)

5.7 List of joint research laboratories/facilities with the universities

Joint Research Laboratories: Laboratory of Genetics IMFG SAS and Department of Molecular Biology Faculty of Natural Sciences Comenius University in Bratislava

Joint Research Laboratories with Department ob Biochemistry and Microbiology Faculty of Chemical and Foot Technology Slovak Technical University in Bratislava

5.8 Supplementary information and/or comments on doctoral studies and pedagogical activities

IMPG SAS is external education institution in Animal Physiology (main garant is doc. Ing. O. Križanová, DrSc.), Biochemistry (main garant is Ing. A. Breier, DrSc.) and Biophysics (main garant is RNDr. K. Ondriaš, DrSc.). Accreditation for Molecular Genetic (main garant will be RNDr. Ľ. Kádaši, DrSc.) is under preparation. Researcher in IMPG SAS are very active in third stage of university education and were supervisor of 11 PhD students that were defence their thesis in years 2003-2006.

6. Direct output to the society

(Applications of results, popularisation and outreach activities)

6.1 List of the most important results of applied research projects

Study of genetic factors, affecting the age-at-onset of Huntington's disease

The expansion of a polymorphic CAG repeat in the IT-15 gene encoding huntingtin has been identified as the major cause of Huntington's disease, and determines 42-73 % of the variance in the age-at-onset of the disease. Polymorphisms in huntingtin interacting or associated genes are thought to modify the course of the disease. In frames of a large international study this modifying effect has been ruled out for genes GRIK2, TBP, BDNF, HIP1, and ZDHHC17. On the contrary, it has been shown that polymorphism S18Y in UCHL1 gene decreases the age-at-onset on average by as many as 9 years. This result is of importance in prediction of the age-at-onset in presymptomatic stage of disease, so the preventive interactions can be applied in the due time.

Molecular genetic analysis of severe monogenic disorders in Slovakia

Severe forms of hearing impairment with the frequency of 1:1000 newborns represents the most common disorders of the sensory system in humans. More than one half of cases is caused by damages of the DNA. The complexity of hearing apparatus and processes influencing hearing is directly proportional to the number of genes involved. As a consequence of this, deafness as a clinical entity is characterized by extreme locus and allelic heterogeneity. At present more than one hundred genes distributed all over the human genome are known to be linked to deafness. In Slovakia, up to present, no molecular genetic studies have been carried out, thus methods of DNA diagnostics could not had

been applied in the everyday clinical praxis, which in other countries belong to the standard medical care in patients affected with deafness.

Our study was focused on the gene GJB2, mutations of which affect more than 60 % of patients with deafness. The mutation spectrum has been identified in this gene in patients of both Roma and non-Roma ethnic origin. The results obtained, according to our expectation, disclosed profound differences both in spectrum and prevailing mutations in these two subpopulations of Slovakia. This result is important for developing effective strategies for DNA diagnostics, using simple tests, based on DNA analysis. Our results and tests developed represent the basis for differential diagnosis of hearing impairment, which is the prerequisite of more effective therapy and prevention of this severe disorder of sensory system in Slovak patients.

Results obtained in the field of molecular genetic analysis of severe monogenic disorders in Slovakia, with direct impact concerning their DNA based diagnostics have been summarized in a monography.

Multidrug resistance of neoplastic tissue

Multidrug resistance of neoplastic tissue represents a heavy obstacle in effective chemotherapy of cancer. Overexpression of P-glycoprotein – membrane transport protein with the function to efflux effectively drugs from intracellular space – represents a dominant mechanism responsible for MDR. To block function and expression of PGP represents the way how to treat MDR cancer diseases. In our laboratory we brought the evidence that analogues of pentoxifylline enable to depress MDR. Based on the comparison of physico-chemical properties of analogues with their effectivity we were able to recognize such properties of substance that are important for its effect to be an MDR reversal agent. With this knowledge we were able to propose a new analogue of pentoxyfilline for which a more pronounced effect as MDR blocker was verified.

Rapid detection methods for five HGO gene mutations causing alkaptonuria.

Alkaptonuria represents the very first genetic disorder in man for which autosomal recesive inheritance was proved as early as in year 1902, shortly after rediscovery of Mendel's laws. The disorder is caused by defect in activity of enzyme homogentisate-1,2-dioxygenase (HGO). Though alkaptonuria does not significantly shorten life expectancy of affected individuals, patients suffer from serious arthritic pains. The affection of large joints leads to premature invalidism. The incidence of alkaptonuria in Slovakia (1:19000) belongs to the highest ones in the world. This was one of the main reasons while we subjected the HGO gene to profound analysis in Slovak alkaptonuric patients. As a result of this analysis we identified the complete scale of HGO mutations in Slovak patients. From more than 40 mutations described worldwide, in Slovak patients we found 10. They are however distributed not evenly. In almost 80 % of patients occur only four mutations, the proportion of others is significantly lower. For three mutations Slovak origin can be considered with high probability on the basis of their high prevalence, and almost exclusive occurrence in Slovakia. From the analysed sample of patients as high as 85 % come from one region of Slovakia, Kysuce. On the basis of these results it is highly probable that, for the increased incidence of alakaptonuria in Slovakia is caused by founder effect and genetic drift as a consequence of genetic isolation and endogamy of this

region in the past. Taking into account the results of molecular studies, however, it can be concluded, that the spectrum of HGO mutations in Slovakia was formed by several founder effects. In addition, some mutations have come to Slovakia by migration, and some arose as new mutation events in the relatively not very far past. For each mutation identified in Slovak alkaptonuric patients a simple and rapid detection method based on DNA analysis was developed. These detection methods enable unequivocal diagnosis of the disorder even in the preclinical stage making the prevention more successful and more effective.

Detection of Polymorphisms Associated with Cardiovascular Diseases Methodology Letter No. 1

(Factor V-Leiden, Factor V H1299R, Prothrombin G20210A, Factor XIII V34L, Beta-fibrinogen 455G/A, PAI-t4G/5G, HPA1 a/b, MTHFR C877T, A1298C, ACE I/D, ApoB R3500Q, Apo E2, E3, E4)

6.2 List of the most important studies commissioned for the decision-making authorities, the government and NGOs, international and foreign organisations

doc. RNDr. Ľ. Kádaši, DrSc. - Court expert in genetics specialization: DNA analysis

RNDr. Ľ. Lacinová, DrSc. - Expert for Commision for biosafety, an advisory organ of Ministry of Environment of Czech republic

RNDr. H. Poláková, - Court expert in genetics specialization: DNA analysis

6.3 List of the most important popularisation activities

doc. RNDr. Ľudovít Kádaši

Human genome

TV:	TA3 – 15.4.2003,
	TV JOJ – 21.4.2006
Radio:	Radio Devin – 14.3.2003
	Radio SR1 – 21.6.2004
	Slovenský rozhlas – 20.8.2003
Printed media:	Plus 7 dní – 10.3.2003
	Báječná žena – 24.3.2005, 18.8.2005
	Emma – November 2005

Genetic disorders, genetic testing

TV:	STV1 – 17.4.2005, 31.7.2005
	TV JOJ – 5.10.2005, 5.2.2006
Radio:	SR1 – 25.2.2003, 22.1.2004
	Radio Regina – 25.1.2004
Printed media:	Zdravie – February 2003, August 2003

Új Szó – 12.11.2005, 12.11.2005

Cloning

TV:	TV Markiza – 20.2.2003
	TA3 – 13.2.2004, 2.12.2005
	STV1 – 16.2.2004, 18.2.2004
Radio:	Radio Slovakia – 13.1.2003
	Radio Twist – 12.8.2004
Printed media:	Moment – 9.1.2003
	Súvislosti – 6.2.2003
	Express – No.26, 2003
	Národná obroda – 14.2.2004

Forensic use of DNA testing (paternity testing)

TV:	TV Markiza – 26.9.2004, 12.7.2006, 14.7.2006
	TV JOJ – 30.10.2006, 1.12.2006,
	STV1 – 12.7.2006
Radio:	Radio Express – 12.7.2006
Printed media:	Súvislosti – 16.1.2003,
	Pravda – 15.3.2003
	Markiza – No. 19 2004
	Zdravie – March 2006,
	Šarm – 18.7.2006, 26.9.2006

Genetics and ethics

Printed media:	Hospodárske noviny – 22.2.2004,
	Týždeň – 12.6.2006

GMO

TV:	STV2 – 8.11.2006

RNDr. Ľubica Lacinová, DrCs.

Gene technologies and their industrial application (GMOs)

TV:	TA3, 11. 5. 2003 21:30
Radio:	SRo, 23. 5. 2003 7:00 „Dobré ráno“
	Radio Twist, 5. 12. 2003, repeated brodacasting in weeks 20. and 21. 2004
	07. 10. 2004 Twist, Slovenský rozhlas, Rádio Lumen
Printed media:	„Plus 7 dní“ issue 30, 2003, pp. 76-78
	Monthly magazine „Sedmá Generace“, issues 1–12,2003
	Új Szó, Pravda
	Monthly magazine „Sedmá Generace“, issues 1–12,2004
	Hospodárske noviny, 4. 4. 2005, p. 8.
	Bi-monthly magazine „Sedmá Generace“, issues1–6,2005
	Bi-monthly magazine „Sedmá Generace“, issue 3 2006
www.changenet.sk	7.1.; 7.2.; 7.3.; 12.4.; 11.5.; 21.5.; 21.6.; 7.9.; 24.9.; 20.10. 2004
www.magazin.station.sk	6. 9. 2005
www.changenet.sk	17. 2.; 15. 3.; 2. 5.; 29. 6.; 25. 8.; 2. 11. 2005

Bioethics

Radio:	SRo, Slovensko 1, 22. 03. 21:00-22:00

SRo, Slovensko 1, 23. 3. 11:00
Rádio Devín, 27. 7. 2004
Printed Media: Mosty issue 8., 12. 4. 2005
týždeň issue 20, 16. 5. 2005

Gentics and human health

TV:
Radio: SRo, „Nočné dialógy“, 4. 4. 2003 24:00-02:00
SRo, Rádio Slovensko, 28. 02. 11:00-11:30
Printed Media: Domino fórum, issue 41, 2003, str. 20
Knihy a Spoločnosť issue 3, 2006
http://www.inzine.sk : 3.11.2003; 30.9.2003; 3.7.2003; 5.5.2003; 11.4.2003; 26.3.2003; 18.2.2003; 12.2.2003; 5.2.2003; 8.1.2003;

Biosciences in our life - generaly

TV: Markíza, Správy Markíza, 12. 5. 2003 19:00

Radio:
Printed Media: Mosty issue 12, 08. 06. 2004
„24 hodín“ 7. 10. 2005
Mosty issue 24, 23. 11. 2005
Bi-monthly magazine „Sedmá Generace“, issue 1 and 6, 2006
Mosty issue 19, 2006
www.magazin.station.sk 6.2.; 15.3.; 20.7.; 17.8.; 24.8.; 3.10.; 23.11.2006

Ľubica Lacinová: Transgénne živočíchy. In: Konferencia o geneticky modifikovaných organizmoch. Zborník. Editori Jozef Timko a Branislav Peťko, VEDA, vyd, SAV Bratislava 2003. pp. 33-42

Ľubica Lacinová: Myslia ženy inak? Príroda versus výchova. In: Ružový a modrý svet. Rodové stereotypy a ich dôsledky. Editorky: Jana Cviková a Jana Juráňová. Aspekt, Bratislava, 2003.

Ing. Alexandra Zahradníková, CSc.

TV: STV2, Pod Lampou „The miracle called a cell“, 8. 12. 2005, 21:45

RNDr. Marta Novotova, CSc.

Film: “Proffesion - researcher", Coproduction TV ARTE Production Claud Mourier 2004
TV: TV Markíza 5. 5. 2005, Interwiev about film

6.4 List of patents issued abroad, incl. revenues

6.5 List of the patents issued in Slovakia, incl. revenues

6.6 List of licences sold abroad, incl. revenues

6.7 List of licences sold in Slovakia, incl. revenues

6.8 List of contracts with industrial partners, incl. revenues

6.9 List of research projects with industrial partners, incl. revenues

6.10 Summary of outreach activities

Outreach activities	2003	2004	2005	2006	total
studies for the decision sphere, government and NGOs, international and foreign organisations	72	94	81	73	320
articles in press media/internet popularising results of science, in particular those achieved by the Organization	26	18	25	18	87
appearances in telecommunication media popularising results of science, in particular those achieved by the Organization	13	13	7	11	44
public popularisation lectures	3	1	1	4	9

6.11 Supplementary information and/or comments on applications and popularisation activities

Healthcare should be considered as main acceptor of IMPF SAS research activities for applications. Several methods and methodologies were offered to the clinical institutions for diagnosis predominantly in monogenic hereditary diseases. Solution of state program concerning genomic of cardiovascular diseases revealed data that are applicable in early diagnosis of cardiovascular diseases and in this time are tested on National institute of cardiovascular diseases. Several researchers from IMPG SAS were active in cooperation with media like TV, radio, etc. Documentation of major activities are summarized above.

7. Background and management. Staffing policy and implementation of findings from previous assessments

7.1 Summary table of personnel

Personnel	2003	2004	2005	2006
all personel	61	61	58	66
research employees from Tab. Research staff	38	40	34	41
FTE from Tab. Research staff	25.9	26.5	24.8	24
averaged age of research employees with university degree	40	41	40	39

7.3 Professional qualification structure

Number of	2003	2004	2005	2006
DrSc.	3	5	5	5
PhD / CSc.	23	22	22	27
Prof.	0	0	0	0
Doc./Assoc. Prof.	1	1	1	2

7.4 Status and development of research infrastructure incl. experimental, computing and technical base (description of the present infrastructure, premises, and material and technical resources. Infrastructure, instrumentation and major technical equipment necessary for the achievement of the objectives specified in the research Concept)

During years 2003-2006 in IMPG Laboratory of confocal microscopy was completed that was supported by the project HHMI 55000343. Laboratory of bio-imaging methods based on Typhoon 9210 instrument was built using funding from the project Building of Biotechnological Centre – BITCET. Besides these major activities several other methods and instruments were completed. In the next time period acquisition of patch clamp apparatus, instrumentation for molecular biology, molecular genetics and proteomics as well as instrumentation for electron and light microscopy would be necessary for methodical development of IMPG.

7.5 Status and development of bibliographic resources, activities of the Organisation's library and/or information centre

Activities of the Library of IMPG SAS in the years 2003-2006

State of the book-fund	until Dec 31, 2003	until Dec 31, 2004	until Dec 31, 2005	until Dec 31, 2006
Books	6.770 book units	6.816 book units	6.821 book units	6.829 book units
	7	46	5	8
Periodicals	8	8	8	8
Readers	From SAS/56	53	51	52
	Outside of SAS/28	32	28	29
Interlibrary borrowing service				
To other libraries	131	154	147	167
From other libraries	415	521	612	587
Borrowing between libraries abroad	31	68	52	56

Institute of Molecular Physiology and Genetics SAS has a sufficient amount of books in its library which are used both by Academy staff and by employees of other organizations. Our Information Centre ensures an extensive interlibrary borrowing service since our stock of periodicals comprises journals which are unique in Slovakia. Financial resources of the Institute do not allow a continuous build-up of the Information Centre by acquisition of new titles of books and periodicals at the optimal level. The remedy to this shortage may be found partially in the access to databases of online journals, which was provided by SAS by means of activities of the Central Library of SAS.

7.6 Describe how the results and suggestions of the previous assessment were taken into account

Institute were evaluated in category A without any suggestions for future development.

7.7 Supplementary information and/or comments on management, research infrastructure, and trends in personnel development

Members of the institute were very successful in application for international research funds and therefore were able to enhance significantly their set-ups. Another valuable instrument was purchased with help of domestic programs supporting build-up of research infrastructure (BITCET). State-of-the art experimental equipment not only enables high quality training of diploma and PhD students, but also motivates them to return to our institute after spending several years in top European and overseas universities. Therefore the institute is able to maintain low average age of independent researchers and also principal researchers can reach DrSc degree well before age of 50.

- **Other information relevant to the assessment**