Name of the Center: CENTRO INTERDISCIPLINARIO DE NEUROCIENCIA DE VALPARAÍSO	
Acronym	CINV
Code	P09-022-F
Reported period	August, 2011 to December 2012
Starting date of the Center	08/08/2011
Address	Pasaje Harrington 287, Playa Ancha, Valparaíso.
Telephone(s)	056-032-2508040
Fax	056-032-2508047
Web Page	www.cinv.cl
Host Institution(s)	Centro Interdisciplinario de Neurociencia de Valparaíso, Universidad de Valparaíso, Pontificia Universidad Católica de Chile, Centro de Modelamiento Matemático de la Universidad de Chile, Universidad de Talca
Contact Information	
Scientific Contact	*Ramón Latorre de la Cruz* *CINV Director*
Electronic Address	*ramon.latorre@uv.cl*

Institute / Nucleus Principal Researcher Name	*Institute / Nucleus Deputy Principal Researcher Name*
Ramón Latorre de la Cruz	*Alan Neely Delgueil*
Principal Researcher's Signature	*Deputy Principal Researcher's Signature*

Index

1.1 Executive Summary

The main aim of the **Centro Interdisciplinario de Neurociencia de Valparaíso (CINV) Millennium Institute** was to transform the CINV into an **outstanding multidisciplinary, interregional science center.** In its first year of activities, the CINV shows qualitative and quantitative progress in all the lines of research. In particular, the Cross-cutting line on **Molecular Simulations and Computational Biology** and the laboratory of Dr. J.C. Sáez at the Universidad Católica, Santiago, has improved the associativity and productivity of the CINV.

The strengthening of our network of international collaborations has also resulted in exciting research projects. These include: i) the unveiling of the genome of the acarid, *Tetranychus urticae*, an advance of potential importance for the control of this pest, which attacks different plants including grapes (Grbić et al., *Nature* 479:487, 2011); ii) the detailed mechanisms by which the Na^+/K^+ pump translocates ions (Castillo et al., *PNAS* 108:20556, 2011); iii) the validation of the rodent endemic to Chile, *Octodon degus*, as a model for Alzheimer's disease (AD) (Ardiles et al. PNAS 109:13835, 2012); and iv) the mechanisms allowing rapid dynamic control of the polarity and gain of NMDAR-dependent synaptic plasticity (*Neuron* 73:497, 2012).

The CINV deals with several aspects of a fundamental scientific question: ***How does the Nervous System respond to Stimuli in Health and Disease?*** We are addressing this question along four research lines and one cross-cutting theme. **1. Structure and Function of Molecular Sensors.** The studies performed during the first year of ICM funding dealt with the key question in understanding ion channel and pumps function: ***how do the protein domains involved in sensing stimuli (sensors) and opening the pore (gates) communicate?*** In this regard, we published exciting results on the molecular workings of the voltage sensors of voltage-dependent K^+, Ca^{2+}- and voltage-activated K^+ channels (BK), voltage-dependent Ca^{2+} channels, and proton channels. We also studied the characteristics of the voltage and temperature sensors in two temperature-sensitive transient receptor potential channels (TRPV1 and TRPM8). This was possible thanks to the incorporation of state-of-the art techniques such as **L**anthanide **R**esonance **E**nergy **T**ransfer (**LRET**) (the first in Chile) and voltage-clamp fluorometry. **2. Cellular Signaling.** One aim of this research line was to study the role of different dynamins in secretion. Using amperometry in chromafin cells of adrenal glands, we found that cortical actin organization and *de novo* formation of F-actin filaments depend on the GTPasa dynamine-2 and hindering the actin function increases the quantum size and delays the expansion of the fusion pore. A second aim was to study the role of connexin (Cx)- and pannexin1 (Panx1)-based channels in neurodegenerative diseases. We found a sequential activation of glial cells including activation of Cx and Panx1 hemichannels in microglia that leads to TNF-α/IL-1 release. The latter activate Cx43 hemichannels in astrocytes through which ATP and glutamate are released and induce neurodegeneration. A third aim was to study the molecular organization of Cx based channels. Additional motifs involved in oligomerization of the gap junction protein Cx26 were identified. Using the TOXCAT assay for interactions of isolated transmembrane α-helices, TM1, a Cx26 pore domain, was found to have a strong propensity to homodimerize. **3. Genetic and Developmental Neuroscience.** We continue to make exciting discoveries in environment/ genome interactions within the developing nervous system, and endocrine regulation of neural networks underlying behavior. We generated a fluorescent reporter research tool valuable for studies of neural progenitors in the developing nervous system allowing us to track neural progenitors in vivo. We have for the first time in zebrafish isolated Neural Stem Cells from the adult brain and shown their capacity to regenerate the neuroendocrine cells essential for puberty. Using genetically modified calcium indicators we have shown that neural activity initiated by peptide hormones is correlated with observed behaviors of the animal. Finally we have progressed in the understanding how the circadian clock imposes a daily rhythm on behavior.

4. Systems Neuroscience. We are studying the neural capacity (mechanism and circuits) involved in learning and memory in mammals, during normal and pathological states. We have reported that during aging the rodent *O. degus* shows the main hallmarks of AD. Our findings validate *O. degus* as a suitable natural model for studying how sporadic AD may be initiated. We have also revealed the importance of neuromodulators (Gs and Gq11) through G-protein receptors as pull-push mechanisms determining the switch between LTP or LTD neural plasticity and we explored and published in subjects related to sensory neural coding and neural plasticity. **Cross-cutting**: ***Molecular Simulations, and Computational Biology.*** During the period we are reporting and in association with the Universidad Nacional Andrés Bello, we set-up one of the largest computer facility in Chile through the installation of a SGI ICE 8200 server with 1,536 cores and 3 TB of RAM. This facility together with those implemented in Dr. P. Orio and Dr. T. Pérez-Acle laboratories are at present being used to integrate data from genes to structures. As we suggested previously, using the crystallographic structure of the β2 adrenergic, we confirmed the existence of two ligand binding microdomains in the GPCR molecular architecture. We also provided structural evidence to support the role of transmembrane domain 1 in Cx26 in regulating channel oligomerization, gating and permeability. We implemented molecular models of ion channels including the pore K^+ channels (Shaker and BK), TRP channels (TRPV1 and TRPM8) and Ca^{+2} channels, which are being validated experimentally.

Advanced training and new investigators: The Ph.D. program in Neuroscience of the Universidad de Valparaiso, created in 2002 by members of our Center, at present has 24 Ph.D. students. Seventeen students have graduated, of which 6 received their degree during this period. Most students have visited laboratories abroad learning new techniques and/or doing experiments that could not be performed in our center due to the lack of the appropriate equipment. Our bilateral graduate student research program with the International School for advanced studies (SISSA) in Trieste (Italy) has been particularly successful. During this year's recruitment period we received 26 applications and admitted 13 students, representing a 2-3-fold increase in the number of applicants relative to our historical average. All the details for the new Ph.D. program in Biophysics and Computational Biology proposed in the original grant have been worked out and we plan to start accepting students to the program by September 2013. The CINV also includes the majority of the faculty involved in the Neuroscience M.Sc. Program, which has 30 students. The incorporation of Dr. C. González to our faculty, an expert in proton channels, added strength to the scientific endeavors of the Molecular Sensors research line and to our graduate programs.

Outreach: The members of CINV delivered lectures in high schools within the Valparaíso Region and in other institutions reaching about 3,000 students. We also took Neuroscience to the general public with the inauguration of the series "***Tertulias Porteñas***". These panel discussions approach a particular neuroscience subject (e.g., dreams, consciousness) from different perspectives. We also developed laboratory workshops for high school students with the purpose of attracting future talents to a scientific career, and secured funding to publish the book, "***La Alegria de Ciencia****"* containing the workshops of "***Ciencia Al Tiro"***.

Networking: We have co-organized the International practical course, *Small Brains, Big Ideas* (in Chile), the *International Meeting of Zebrafish Development and Genetics* (in USA), and organized the *Synthesis of Scents* international symposia and *Evolution & Olfaction II* international workshop in Chile. We have organized the International Symposium: "*Intercellular Communication Via Pannexin and Connexin-based Channels in Health and Disease*" and the international meeting "*Neuroscience Meets Valparaíso*".

1.2 Resumen Ejecutivo

El principal objetivo del **Instituto Milenio Centro Interdisciplinario de Neurociencia de Valparaíso (CINV)** es convertirse en un **destacado centro de ciencia interregional y multidisciplinaria**. En su primer año de actividades, el CINV mostró progresos cualitativos y cuantitativos en todas las líneas de investigación. En particular, la línea transversal de **Simulación Molecular y Biología Computacional** y el laboratorio del Dr. Sáez en la Universidad Católica de Chile han mejorado la asociatividad y productividad del CINV.

El fortalecimiento de nuestra red de colaboraciones internacionales también ha traído importantes proyectos de investigación, tales como: i) el genoma del ácaro *Tetranychus urticae*, con potencial impacto en el control de esta plaga que ataca diferentes plantas incluyendo la uva (Grbić et al., *Nature* 479:487, 2011); ii) el mecanismo de transporte de iones en la bomba Na^+/K^+ (Castillo et al., PNAS 108:20556, 2011), iii) la validación del roedor endémico en Chile *Octodon degus*, como modelo de la enfermedad de Alzheimer (EA) (Ardiles et al. PNAS 109:13835, 2012) y iv). Los mecanismos que controlan la polaridad y ganancia de la plasticidad sináptica dependiente de NMDA-R (*Neuron* 73:497, 2012).

El CINV trabaja empleando diversos enfoques con el objetivo de responder una pregunta científica fundamental: ***¿Cómo responde el sistema nervioso a estímulos, en condiciones fisiológicas y patológicas?*** Abordamos esta pregunta en cuatro líneas de investigación y una línea transversal. **1. Estructura y función de Sensores Moleculares**. Los estudios realizados durante el primer año de financiamiento ICM tratan de entender la función de canales de iones y bombas: ***¿Cómo se comunican entre sí los dominios proteicos responsables de sentir estímulos (sensores) y de abrir el poro (compuerta)?*** Respecto a esto, publicamos interesantes resultados sobre el funcionamiento del sensor de voltaje en canales de K^+ dependientes de voltaje, canales de K^+ activados por voltaje y Ca^{2+} y canales de protones. Estudiamos también las características de los sensores de temperatura y voltaje en dos canales TRP dependientes de temperatura (TRPM8 y TRPV1). Todo esto fue posible gracias a la incorporación de técnicas de última generación como la Transferencia de Energía Resonante de Lantánidos (LRET) (el primero en Chile) y fluorometría con fijación de voltaje. **2. Señalización Celular.** Un objetivo de esta línea fue el estudio del papel de diferentes dinaminas en la secreción. Usando amperometría en células cromafines de glándula adrenal, encontramos que la organización cortical de la actina y la formación *de novo* de filamentos de F-actina dependen de la GTPasa dinamina-2 y que interferir con la función de actina retarda la expansión del poro de fusión y aumenta el tamaño cuantal. Un segundo objetivo fue estudiar el papel de canales formados por Conexinas (Cxs) y panexina 1 (Panx1) en enfermedades neurodegenerativas. Observamos una activación secuencial de células gliales que involucra la activación de canales de Cx y Panx1 en microglia y que conduce a la liberación de TNF-α/IL-1. Este último activa hemicanales de Cx43 en astrocitos a través de los cuales se libera ATP y glutamato induciendo neurodegeneración. Un tercer objetivo fue estudiar la organización molecular de canales de Cx. Utilizando el ensayo TOXCAT para ensayar la interacción de α-hélices transmembranales, identificamos en dominio TM1 de Cx26 como un motivo con alta tendencia a la homodimerización. **3. Genética y Neurociencia del Desarrollo.** Hemos realizado interesantes descubrimientos en la interacción ambiente/genoma durante el desarrollo del sistema nervioso y en la regulación endocrina de redes neuronales que controlan comportamientos. Generamos una herramienta de marcaje fluorescente útil para estudios de progenitores neurales permitiendo su seguimiento *in vivo*. Fuimos los primeros en aislar células progenitoras neuronales del cerebro adulto y mostramos su capacidad de regenerar células neuroendocrinas esenciales para la pubertad. Usando indicadores de calcio genéticamente modificados, mostramos que la actividad neuronal iniciada por hormonas peptídicas se correlaciona con comportamientos observados en el animal. Finalmente, avanzamos en nuestra comprensión de cómo el reloj circadiano impone un ritmo diario al comportamiento.

4. Neurociencia de Sistemas. Estamos estudiando la capacidad neuronal (mecanismos y circuitos) involucrada en aprendizaje y memoria en animales, en condiciones normal y patológica. Reportamos que en el envejecimiento el roedor *O. degus* muestra los principales signos de la EA. Nuestros hallazgos validan al *O. degus* como un modelo natural adecuado para estudiar cómo se puede iniciar la EA esporádica. También mostramos la importancia de neuromoduladores que actúan a través de proteínas G (Gs y Gq11) en la selección de plasticidad tipo LTP o LTD; también publicamos en tópicos relacionados con la codificación sensorial y plasticidad neuronal. **Transveral: *Simulación Molecular y Biología Computacional.*** Durante este periodo, y en asociación con la Universidad Nacional Andrés Bello, montamos una de las más grandes instalaciones de cómputo en Chile consistente en un servidor SGI ICE 8200 con 1536 núcleos y 3 TB de RAM. Esta instalación, junto con las instaladas en los laboratorios de los Dres. P. Orio y T. Pérez-Acle están siendo usadas para integrar datos de la estructura a la función. Usando la estructura del receptor adrenérgico β2, confirmamos la existencia de dos microdominios de unión de ligando en la arquitectura de GPCRs. También entregamos evidencia estructural para apoyar el papel del dominio TM1 de Cx26 en la oligomerización, permeabilidad y apertura del canal. Implementamos modelos moleculares de canales iónicos incluyendo el poro de canales de K^+ (Shaker y BK), canales TRP (TRPV1 y TRPM8) y canales de Ca^{2+}, modelos que están siendo validados experimentalmente.

Entrenamiento avanzado y nuevos investigadores: El programa de Doctorado en Neurociencia de la Universidad de Valparaíso, creado en 2002 por miembros de nuestro Centro, cuenta actualmente con 24 estudiantes. La mayoría de ellos ha visitado laboratorios extranjeros para aprender nuevas técnicas y/o realizar experimentos que no podrían realizarse en nuestro Centro por no contar con equipamiento específico. Durante las últimas postulaciones al programa recibimos 26 postulantes y aceptamos 13, más del doble de nuestro promedio histórico. Se han graduado 17 estudiantes, 6 de ellos en este periodo. Nuestro programa bilateral con la Escuela Internacional de Estudios Avanzados (SISSA) en Trieste (Italia) ha sido particularmente exitoso. Respecto al nuevo programa de Doctorado en Biofísica y Biología Computacional, propuesto en el proyecto original, esperamos empezar a aceptar estudiantes en Septiembre 2013. El CINV también alberga a la mayoría del claustro académico del programa de Magister en Neurociencia, que cuenta con 30 estudiantes. La incorporación del Dr. C. González, experto en canales de protones, ha sumado fuerza al trabajo de la línea de Sensores Moleculares y a nuestros programas de postgrado.

Proyección al medio: Los miembros del CINV dieron charlas en escuelas y liceos de la Región de Valparaíso y otras instituciones llegando a alrededor de 3000 estudiantes. También llevamos la neurociencia al público general con la inauguración de las **"Tertulias Porteñas"**. Estas conversaciones se acercan a un tópico de la neurociencia (p. ej. Sueños, conciencia) desde diferentes perspectivas. Hicimos talleres de laboratorio para estudiantes de educación Media para acercar a futuros talentos a una carrera científica, y aseguramos financiamiento pare el libro "La Alegría de la Ciencia" basado en los talleres de **Ciencia al Tiro.**

Redes: Co-organizamos el curso práctico internacional *Small Brains, Big Ideas* (en Chile) y el *International Meeting of Zebrafish Development and Genetics* (en USA), y organizamos el simposio internacional *Synthesis of Scents* y el taller *Evolution & Olfaction II*; además del simposio internacional *"Intercellular Communication via Pannexin and Connexin-based Channels in Health and Disease"* y la reunión internacional *"Neuroscience Meets Valparaíso".*

2 Introduction

a) Description of the Institute:

Our Center deals with several aspects of a fundamental scientific question: *How does the Nervous System respond to Sensory Stimuli in Health and Disease?* In an interdisciplinary environment, we are addressing this question at several levels, spanning from the atomic to the organismal. The central themes of our research are molecular transduction, intercellular communication, neural correlates of perception, the generation of appropriate behavioral responses, and the genetic pathways controlling development and complex behaviors. Our research is organized along four broad lines and one ***cross-cutting theme***. We want to emphasize that most investigators (both principal and associates) are involved in more than one research line, and that specific research questions will be answered through efforts carried out by different research teams.

b) Research Lines:

Structure and Function of Molecular Sensors: This line of research seeks to understand, at a molecular level, how ion channel proteins are able to sense stimuli such as temperature and voltage, and how the Na^+/K^+ pump transports Na^+ and K^+ across the cell membrane. During the period we are reporting and using a variety of different experimental techniques that include electrophysiology, molecular biology and fluorescence we have made progress in the understanding how the voltage sensor of different ion channels work and how the Na^+/K^+ pump translocate ions (*e.g*., *PNAS*. **108**:20556,2011; *PNAS*. **109**:18577,2012; *Neuron* 77:288, 2013).

This line of research is composed by the P.I. (R.Latorre), the Co-PI (A.Neely), Assoc. Invs. D. Naranjo, F. Bezanilla and M. Holmgren working together with C. Gonzalez, a recent recruit to CINV. Associativity within the group (e.g., *Nature Comm*. 2:436, 2011; *Comprehensive Physiol.* **2**:2087, 2012; *PNAS*. **109**:18577,2012) and collaboration with other lines of research (2 & 5; e.g., *FEBS Let*t: **586**:2287, 2012; *Biophys. J.* **103**:1198, 2012) is thriving. The interpretation of many experimental results has been possible through the interaction with the Cross-cutting line.

2. Cellular Signaling: All cells express proteins for communicating with other cells and important players are gap junction channels and hemichannels composed of connexins or pannexins, and proteins involved in secretory mechanisms that are controlled by dynamin. This group investigates the physiological mechanisms that regulate these proteins, and their alterations in neurodegenerative diseases such as Alzheimer disease, stroke and brain trauma. The goal is to find therapeutic targets and to design molecules that inhibit or activate them.

This research line is composed by the Assoc. Investigators J.C. Saez and A.M. Cardenas, and the Adjunct Inv. A.D. Martinez. Associativity within the group (e.g., *Mol Biol Cell*. 23:3299, 2012; *Biochim Biophys Acta.* 1818:1169, 2012; *Mol Biosyst.* 2012 8:685) and collaboration with other lines of research (1 & 5) (e.g., *FEBS Lett*: **586**:2287, 2012; *Mol Biol Cell*. 23:3299, 2012) is thriving. The interaction with the Cross-cutting line has been relevant to develop molecular models of hemichannels bearing mutations associated to deafness.

3. Genetic and Developmental Neuroscience. This line of research includes K.Whitlock, J.Ewer, and R. Greenspan. We use modern molecular-genetic tools to visualize the differentiation of cells in vivo (Harden et al., 2012) and visualize the pattern of neuronal activity induced by neuropeptides as they elicit specific behaviors in the animal (Wilson and Ewer, in preparation). With these tools we can also study the correlation between neuronal differentiation and behaviors (Stephenson et al., 2012) as well the genes underlying essential behaviors (Lahr et al., 2012). We, for the first time, generated neural stem cells from adult zebrafish brain thus illuminating the mechanism underlying hormone induced reversion of Kallmann Syndrome in humans (Cortés-Campos et al., submitted to *Nature*). Currently we have two papers submitted that resulted from new collaborations within the lines of investigation: with Dr. P. Orio (Line 1) to develop an image analysis method to quantify migration of cell populations in time lapse videos (Boric et al 2012 *PLOSOne* submitted) and with

Dr. E. Couve (Line 4) to analyze effects of ethanol exposure on cilia in migrating neural crest cells (Boric et al., submitted *Zebrafish*).

4. Systems Neuroscience. This line of research investigates the neurobiology of sensory capacity, learning, and memory during normal and pathological aging. During aging dramatic changes occur in central nervous system at the molecular level and at the level of neuronal circuit with consequences on the cognitive capacity of animals. This capacity deteriorates further under pathological conditions (e.g. Alzheimer`s disease). During aging, the rodent *Octodon degus* shows the main brain hallmarks of Alzheimer`s disease: decrease of cognitive performance and deterioration of neural plasticity mechanisms (LTP, LTD) that correlates with the increase of soluble oligomers (e.g. Ab*56) and phosphorylated tau proteins (Ardiles et al *PNAS* 2012) making *degus* a valuable model to explore alternatives for pharmacological rescue. We have shown the importance of Gs and Gq11 neuromodulators, which trough G-protein receptors exert a pull-push mechanism that determines the switch between LTP or LTD neural plasticity (Huang et al. *Neuron* 2012).

The research team is composed by the P.I. (A. Palacios) and Assoc. Inv. O. Schmachtenberg, P. Munoz, and Dr. A. Kirkwood, (J. Hopkins U). Associativity within the group is shown in several publications (e.g., *J. Neural Transmission.* 119:173-195 *Neuron.* 73:497-510, *Cold Spring Harbor Protocols*, 2013; PNAS 109:13835-40 and *Cold Spring Harbor Protocols*, 2013 in collaboration with line of research 2 & 3).

Cross-cutting: ***Molecular Simulations, and Computational Biology.*** With the objective of implementing a cross-cutting research area at CINV, we have been involved in a large number of transversal collaborations. To do so, we produce and use computer-based simulation tools by combining advanced mathematical modeling, thermodynamics, biophysics and chemistry, together with high performance computing (HPC) techniques. Our main goal is to produce tools for multi-scale simulation of biological systems: micro-scale (atomic level), meso-scale (cell level) and macro-scale (population level). At the same time, several projects are progressing through a modeling-experiment-modeling cycle, including K^+ channels, TRP channels and Ca^{+2} channels. Experimental validation of models is done through collaborations with Line 1.

The research team is composed by the P.I. (F. González-Nilo) and Assoc. Inv. T. Pérez-Acle and P Orio. This line of research has built strong ties with lines 2, 3, & 4 (see above).

Conclusions: The blend of neuroscientists, biophysicists, mathematicians, and computational biologists together with the collaboration of an extended international network of individual scientists and laboratories has been fundamental in the success of this first year of CINV as an ICM. This is reflected in the large increase in productivity that went from 19 ISI articles in 2011 to 38 in 2012. This increase in productivity was accompanied by an increase in the number of papers with authorship containing two or more members of CINV (2 in 2011 vs. 9 in 2012).

3) Scientific and technological research:

a) Current status of research lines:

Our research is organized along four broad lines and one ***cross-cutting theme***. We want to emphasize that most investigators (both principal and associate) are involved in more than one research line, and that specific research questions will be answered through efforts carried out by different research teams. This makes our Center a highly interdisciplinary and associative research enterprise.

1. Structure and Function of Molecular Sensors. Ion channels and pumps are the main mechanism responsible for keeping the appropriate electrical homeostasis of the cell. Confronted with a stimulus, ion channels dissipate the ion gradients built up by ion pumps using the energy contained in the ATP molecule, to modify the membrane potential. ***Using three different ion channels, the Ca^{2+}- and voltage-activated K^+ (BK), the voltage-dependent Ca^{2+} ($Ca_v1.2$) and the proton (H_v) channel, our research line is trying to unveil the general mechanisms through which the electrical energy contained in the electric field is transformed by the voltage sensor domain (VSD) into mechanical energy (the pore opening).*** In K^+, and $Ca_v1.2$ the subunits or domains share a common structure consisting in six transmembrane domains (S1-S6) where S1-S4 form the voltage sensor domain (VSD) and S5-S6 the pore domains. BK has the same general structure but each subunit contains an extra transmembrane domain (S0). The proton channel is just a VSD (S1-S4) with no pore domain. All the members of this line of research are involved in the search of a solution to the following questions: a) How important is the location of positively charged amino acid in the S4 transmembrane segment in determining channel voltage sensitivity? b) How BK β subunits modify the resting-active equilibrium of the voltage sensor in this channel and the channel gating kinetics? c) What is the contribution of each of the Ca_v channel domains to the overall voltage sensitivity of this channel? D) What kind of conformational changes the Hv channel undergoes during opening? We are in an advantageous position to answer these questions since we are combining the expertise in K^+ channels of Drs. C. Gonzalez, R. Latorre and D. Naranjo with the expertise in Ca_v channels of Dr. A. Neely and H_v channels of Dr. C. González. Experimentally we are tackling this problem using a combination of electrophysiology, mutagenesis and state-of-the-art fluorescence techniques. It is important to mention here that with the implementation of three different fluorescence techniques, voltage clamp fluorometry, patch clamp fluorometry and Luminescence Resonance Energy Transfer (LRET), we are at present in a unique position to correlate the electrical events (single channel, macroscopic and gating currents) with the structural changes the ion channels undergo during opening. On the other hand, using voltage clamp fluorometry we have made great advances in the understanding of the conformational changes that the voltage sensor of Ca_V(collaboration with Dr. Olcese laboratory, UCLA) and the voltage sensor of H_v (collaboration with Dr. Larsson laboratory, University of Miami) undergo during channel activation (see below). All the results obtained to date have opened new challenges that need new strategies to solve them. It is clear that in the particular case of H_v channels a crystal structure is necessary to understand fully how this protein, consisting only in a voltage sensor domain, transports protons.

***Shaker K^+ Channel*:** *Voltage gating.* Voltage gated Shaker potassium channels increases open probability (Po) by 20-fold with a ~6 mV depolarization. Such high voltage sensitivity is mostly due to the electrophoretic transmembrane relocation of four positively charged arginines residues in each of their four voltage sensing domains (VSD). These charge movements allow for channel opening upon membrane depolarization. To what extend positively charged residues in these positions determine channel expression and voltage sensitivity? We tested if stabilization was specific for positive charges as has been proposed elsewhere. To do this, we replaced each of the voltage sensing

arginines, R362, R365, R368 and R371, with aspartate residues, on a N-type inactivation removed background Shaker channel. Single mutations did not modify channel expression and, consistent with charge reversion on each individual arginine, they decreased the effective valence ~50% of that of the wild type channel. The double mutant R362D/R365D and the triple mutant R361D/R365D/R371D also expressed robust currents with similar voltage dependence as the single charge- inversion mutants. These results together with the comparable level of channel expression in oocytes are consistent with the idea that the voltage sensitive positions in S4 are not specific for basic residues. Shaker is a low conductance K-channel but the Shaker- P475D mutant conducts with a conductance 6-8 fold large (Naranjo et al. In preparation).

***Shaker K^+ Channel*:** *Modulation of $K_V1.1$ Fast Inactivation by mRNA Editing*. In the nervous system, A to I RNA editing has an important role in regulating neuronal excitability. Ligand-gated membrane receptors, synaptic proteins, as well as ion channels, are targets for recoding by RNA editing. Although scores of editing sites have been identified in the mammalian brain, little is known about the functional alterations that they cause, and even less about the mechanistic underpinnings of how they change protein function. Our experiments show that the channel's inactivation gate enters deep into the ion permeation pathway and the very tip establishes a direct hydrophobic interaction with the edited position. By converting I to V, the intimacy of the interaction is reduced, allowing the inactivation gate to unbind with much faster kinetics (González et al. *Nature Comm*. 2:436, 2011).

BK Channel: *β Subunits Modify BK Voltage Sensor Activation:* We have made progress in our understanding of the mechanisms by which the β subunits (β1- β4) modulate the gating of BK channels and we have proposed a new allosteric model in which the channel gate opens in two steps. β subunits are associated with BK channels in most tissues and dramatically modify their gating properties. We found that β1, β2 and β4 stabilize the voltage sensor in the active configuration and, in addition, that β4 decreases the apparent number per voltage sensor. The allosteric model we proposed is able to explain how β1, β2 and β4 are able to slow down both the activation and deactivation kinetics of the BK macroscopic ionic currents without affecting the kinetics of the voltage sensor movements (Contreras et al. *PNAS* 46:18991, 2012).

BK Channel: *Interaction Surfaces between α and β subunits:* In collaboration with Research line 2, we have demonstrated that the first transmembrane domain of the β2 subunit physically binds to the transmembrane S1 of the α subunit (*FEBS Lett* 586:2287-2293, 2012). We found that slow-twitch (soleus) and fast twitch (flexor digitorus brevis) express different BK splice variants with different Ca^{2+} sensitivities and pharmacological characteristics. These findings may have relevance for conditions affecting mainly postural muscles, such as prolonged bed-rest and microgravity, or conditions affecting fast-twitch muscles, such as periodic paralysis (*PLOS One* 7: e40235, 2012; work done in collaboration with Dr. Tricarico laboratory, University of Bari, Italy).

BK External Architecture Determined with LRET. LRET (a technique put into practice for the first time in Chile thanks to a collaborative effort with Dr. Bezanilla laboratory) allow us to measure intra and intermolecular distances with great accuracy (± 1Å). Using LRET we have determined the external architecture of the pore-forming α subunit of the BK channel and the location of the transmembrane domains of the modulatory β1 subunit in the α/β1 complex (Communicated at the Meeting of Biophysical Society 2013-USA)

Ca_V channel: *Contribution of Cav Channel Domain to Voltage sensing*: With respect to defining the relative contribution of each of the four putative voltage-sensors of high voltage-activated calcium channels to channel opening by voltage-clamp fluorometry, in collaborative effort with Dr. Olcese from UCLA, we were able to detect voltage-dependent changes in fluorescence (V-ΔF) from probes attached to each individual voltage-sensor. To our surprise only V-ΔF from the third domain matches the voltage-dependence of charge movement. Probes on the voltage sensor of domain I and

II reach half maximal changes in fluorescence at more positive voltages with respect to charge movement while probes on domain IV are to the left with respect to the voltage axis. We are currently modeling these (V-ΔF) to reproduce gating current and charge-movement. These results were presented to the 57th Meeting of Biophysical Society 2013-USA).

H_v channels: *Voltage-Sensing Amino Acid Residues in H_v Channels*. Voltage-dependent proton (H_v) channels are homologous to the Voltage Sensor Domain (VSD) of Kv channels, which have been shown to use the positive charges contained in the fourth TM segment, S4, as their voltage sensor (reviewed in Gonzalez et al., *Biophys. Rev*. 4:1-15, 2012). We hypothesized that arginines on the S4 are the gating charges of the voltage sensor in H_v channels, so we focused on these three arginines on the Cio-Hv S4 segment: R255, R258 and R261. Charge neutralization, cysteine scanning mutagenesis and accessibility experiments show that the three S4 charges are the main voltage-sensing residues responsible for the voltage dependence of H_v channels and that the extent of movement of these S4 charges is sufficient to explain the voltage dependence of opening in H_v channels (González et al., *J. Gen.Physiol*. 141:275-285, 2013).

H_v channels: *Conformational Changes during Activation in Hv Channels.* H_{v1} channels have two subunits. Each subunit has a permeation pathway, but opening of the two pathways is highly cooperative. Using voltage clamp fluorometry, we detected two conformational changes reported by a fluorophore attached to the S4 segment in Hv1 channels. The first conformational change is voltage-dependent and precedes channel opening. The second conformational change is less voltage dependent and closely correlates with channel opening. Modifications that alter dimerization or alter the intersubunit interface affect both the second conformational change and channel opening. Our data suggest that this second conformational change is a cooperative conformational change involving interactions between the two subunits of H_{v1} channels that are, at least partly, mediated by the S1-S1 interface (Qiu et al. *Neuron* 77:288-298, 2013).

Na^+/K^+ pump: In a collaborative effort with Drs. Bezanilla and Holmgren (NIH), we have taken advantage of the unique properties of the giant axon of the squid *Dosidicus gigas* present in the coastal water of Chile, to understand the mechanism by which the Na^+/K^+ pump translocate ions. The large size of the axons of this squid (up to 2 mm in diameter) have allowed us to characterized the electrical transitions involved in the movement of both Na^+ and K^+ mediated by the Na^+/K^+ pump (Castilllo et al. *PNAS* 108:20556-20561, 2011).

2. Cellular Signaling. ***Hemichannels.*** We continued to investigate the role of increased hemichannel (HC) activity in cellular degeneration in cultures astroglial cells subjected to proinflammatory conditions including proinflammatory cytokines and hypoxia-reoxygenation (Orellana et al., *Glia* 58:329-343, 2010). We used heterocellular cultures of microglia and astrocytes as well as conditioned culture media by activated microglia or astrocytes to treat highly enriched neuronal cultures. We also used transgenic mice that express glial fibrillary acidic protein tagged with green fluorescent protein. We demonstrated a sequential activation of HCs in cultured cells and in hippocampal slices. The sequence of events starts in microglia and propagates to astrocytes via TNF-α and IL-1β. Then, the increased HC activity in astrocytes leads to ATP and glutamate release that leads to activation of pannexin hemichannels (Panx HCs) and $P2X_7$ and NMDA receptors in neurons causing neurodegeneration. The inhibition of HCs in the cells types studied prevented neurodegeneration, indicating that HCs are targets to reduce or prevent neurodegeneration in diverse diseases characterized by neuroinflammation. These findings were published in several articles and commented in several reviews by invitation (Orellana et al., *J. Neurochem* 118:826-840; 2011; *J. Neurosci.* 31:4962-4977, 2011; Koulakoff et al., *Biochim. Biophys. Acta* 1818:2048-2057 2011;, Rovegno et al., *Med Intensiva.* 36:37-44 2012; Bennett et al., *Brain Res.* 1487:3-15, 2012; Eugenin et al., *J Neuroimmune Pharmacol.* 7:499-518, 2012). The effect of proinflammatory polyunsaturated

fatty acids as activators of connexin hemichannels (Cx HCs) was published (Figueroa et al., 2012). Additionally, we demonstrated that release of gliotransmitters via astroglial HCs play a relevant role in fear long term memory (Stehberg et al.,*Faseb J.* 26:3649-3657, 2012). Using similar approaches we are currently studying the role of mast cells as the first protagonist of the above-mentioned findings and the participation of HCs expressed by oligodendrocytes in demyelinization responses induced by inflammatory conditions.

Hemichannel Assembly: With regard to the molecular mechanism of HC formation we identified motifs involved in oligomerization of the gap junction protein Cx26. Individual transmembrane (TM) domains and the full-length protein were used. Using the TOXCAT assay for interactions of isolated TM α-helices, we found that TM1, a Cx26 pore domain, had a strong propensity to homodimerize. We also studied the effects of mutant TM1s associated to genetic deafness in Cx26 and found impairments of oligomerization and function that parallel the severity of disease that they cause (Martínez et al., *Biochem J.* 436:35-43, 2011; Jara et al., *Mol Biol Cell*. 23:3299-3311, 2012).

Neurotransmitter Release: At the level of neurotransmitter release, we studied whether the association of synaptophysin with dynamin controls the fine tuning of transmitter release. We showed that synaptophysin and dynamin interact in chromaffin granule-rich fractions and that this interaction relies on the C terminal of synaptophysin. Using amperometry we found that cortical actin organization and *de novo* formation of F-actin filaments depend on the GTPasa dynamin-2. We also demonstrated that interfering with actin function increases the quantum size and delays the expansion of the fusion pore (González-Jamett et al., *J Neurosci.* 30:10683-1091, 2010).

Collaborations**:** Drs. A. Martínez and J.C. Sáez published three articles (Schalper et al., 2012; Jara et al., 2012; Figueroa et al., 2012). Dr O. Schmachtenberg, .A. Martínez and J.C. Sáez are collaborating in the demonstration of gap junctions between oligodendrocytes formed by pannexin1. A.M. Cárdenas, A. Martínez and J.C.. Sáez are collaborating in the demonstration of Panx1 HCs in chromafin cells and their role in neurosecretion. An ongoing area of interest is the neuronal role in muscle atrophy, which is being studied by R. Latorre and J.C. Sáez. Also, A. Martínez, T. Pérez-Acle and J.C. Sáez are collaborating in the identification of subunit interaction during oligomerization (Jara et al., 2012).

Internationally, we are collaborating with: Dr. C. Naus, British Columbia, Canada (Hemichannels in brain trauma); C. Giaume, INSERM and Collège de France, France (HCs in animal model of Alzheimer disease), Dr. K. Willecke, Lymus Institute, Bonn University (HCs in neuronal control of skeletal muscles). Dr. G. Altemberg, TTUHSC, USA.

3. Genetic and Developmental Neuroscience.

We continue to use genetic tools to understand the development of the nervous system and the resulting behaviors. During this funding period Line 3 has developed collaborations with labs outside of Chile (Germany, England, USA) and these collaborations have resulted in four published papers in 2012.

Neurogenesis and Behavior in Zebrafish: Notably, we have, for the first time, successfully generated neurospheres (neural stem cells) and shown that they give rise to the gonadotropin-releasing hormone neuroendocrine cells essential for puberty where defects in the development of these cells underlie Kallmann Syndrome in humans (Cortés-Campos, Letelier, Westmiller & Whitlock, submitted to *Nature*; Letelier, Kim & Whitlock, in preparation). We identified regulatory elements controlling *six4b* transcription factor proposed to be involved in neurogenesis, and generated a fluorescent reporter line (*six4b:mCherry*) to follow potential stem cells in the nervous system *in vivo* (Harden et al., *Develop. Dynamics* 241:1143-54, 2012). We are now dissecting the genetic and developmental processes to understand how continually regenerating precursors are maintained in the adult olfactory epithelium (Torres & Whitlock, in preparation). With the

characterization of different olfactory behaviors (Stephenson et al., *Zebrafish* 9:68-73, 2012) we can now study the correlation between neuronal differentiation and behaviors in zebrafish.

Ecdysis Behavior in Drosophila: In *Drosophila*, we have shown through genetic analysis of behavior using null mutants null for genes encoding specific neuropeptides, which we have isolated, that two distinct neuropeptides interact to control essential ecdysis behaviors (Lahr et al, 2012, *J. Neurosci.* **32**: 6819-29; Krüger et al, in preparation). Our ongoing analysis of behavior, in elegant studies using genetically encoded calcium indicators *in vivo*, we express a calcium-sensitive GFP in different target neurons to determine how each neuropeptide contributes to the ecdysis sequence (Mena & Ewer, in preparation).

Circadian Clocks: We continued our analysis of how the circadian clock causes a daily rhythm of emergence of the adult fly, a rhythm that is known to depend on the activity of a circadian clock in the brain and one in the Prothoracic Gland (PG). We investigated how these clocks are coupled by interfering genetically with the brain and PG clock and their interaction (Millán, et al, in preparation). Collaborative work in this area resulted in a joint publication (Sundram et al,, (2012. *J. Biological Rhythms*, 27:183-95.). We were recently awarded a FONDEQUIP grant (CONICYT) for a total of M$106,000 (approx. US$212,000) to purchase an extremely high sensitivity camera to be able to monitor the activity of the circadian clock *in vitro*, using flies bearing a clock gene-luciferase transgene as well as a camera to detect calcium signals in larval zebrafish to correlate olfactory sensory neuron activity with developmental experience in living animals. Thus we have made great strides in developing tools and a knowledge base necessary for unraveling the link between neuronal differentiation and activity underlying the onset and modulation of behaviors essential for survival.

Collaborations: We continue to collaborate with the consortium that sequenced and annotated the genome of the mite, *Tetranychus urticae* (Grbić, *el al*. (2011. *Nature* 479: 487-492) with the aim of eventually sequencing and annotating the genome of the endemic Chilean mite, *Brevipalpus chilensis*.

- We are collaborating with Dr. A. Palacios, assisting with the identification in the Chilean rodent, *Octodon degus*, of gene homologues that have been implicated in Alzheimer disease. This rodent is a natural model for this important human disease, and we aim to correlate the occurrence of specific variants with the severity of the disease.
- We are collaborating with Dr. T. Perez Acle (Line 5) to identify regulatory elements controlling olfactory receptor expression in zebrafish the genome using bioinformatic tools. The olfactory receptors we have identified by qt-RTPCR that show environmentally induced down-regulation are consistent with our recent bioinformatic results showing dual activation of homeobox containing transcription factors results in genomic suppression (Calfun, Dominguez, Perez-Acle & Whitlock, in preparation).
- We have collaborated with Dr. P. Orio (Line 5) to develop an image analysis method to quantify migration of cell populations in time lapse videos (Boric et al 2012 *PLOS One* submitted) and with Dr. E. Couve (Line 4) to analyze effects of ethanol exposure on cilia in migrating neural crest cells (Boric et al., submitted *Zebrafish*).
- We are developing a collaboration between Dr J. Ewer, Dr K. Whitlock, and Dr B. Smith (University of Arizona, USA) to analyze the potential for neurogenesis in adult invertebrates using the honey bee as a model system
- A CONICYT MEC grant allowed Dr. X. Nelson (Canterbury U., New Zealand) so spend 5 months in our lab. Dr. X. Nelson is an expert on jumping spiders, and her visit allowed us to start a new project, investigating the circadian clock in these spiders, about which little is known.

4. System neuroscience.

***Circuitry and Neural Plasticity*:** We are studying the neural capacity (mechanism and circuits) for learning and memory during normal and pathological states.

Octodon degus as a Model of Alzheimer Disease. Aging causes dramatic changes – molecular and cellular - within the central nervous system, decreasing the cognitive – learning and memory capacity of animals. This capacity is further deteriorated by pathological states (e.g. Alzheimer disease). During aging the rodent *O. degus* shows the main brain hallmarks of Alzheimer disease and a decrease of cognition as evidenced by T-maze (spatial) and object recognition (working) memory tests. The latter performance is correlated with a decay of LTP neural plasticity mechanisms, the increase of soluble oligomers (e.g. dodecamer) and phosphorylated tau proteins, and the decrease of PSD-95, GluR2-AMPAR and NR2b-NMDAR subunits, suggesting that the failure of a postsynaptic mechanism is responsible for the cognitive deterioration (Ardiles et al., PNAS 109(34):13835-40 2012; featured in Alzheimer forum news, Pour la Science). This makes degus a valuable animal model to explore pharmacological alternatives. See the review article *Journal of Neural Transmission* (2012,119(2):173-195) and two methodological articles in Cold Spring Harbor Protocols (Ardiles et al. and Lee and Palacios 2013).

G Proteins as Neuromodulators. In collaboration with Dr. A. Kirkwood, we have shown the importance of G_s and $G_{q,11}$ neuromodulators, acting via G-protein receptors and exerting pull-push mechanisms that determine the switch between LTP and LTD in neural plasticity (Huang et al., Neuron 73(3):497-5102012).

Reactive Oxygen Species (ROS) and Neurodegenerative Diseases. The accumulation of oxidative modifications may contribute to aging itself and to the development of a wide spectrum of neurological disorders like Alzheimer's and Parkinson disease. Recently we have suggested that ROS are an additional class of small molecules that act as cellular messengers on different signalling pathways in synaptic plasticity. Consistent with this idea, ROS-generated neuronal activities are required for synaptic plasticity and hippocampus-dependent memory. We found that iron-generated ROS are involved in calcium signalling initiated by stimulation of NMDAR and amplified by the calcium- induced calcium release, allowing phosphorylation and nuclear translocation of ERK1/2 (Munoz et al., JBC. 286(15): 13382-92 2011) and iron is required for basal synaptic transmission and expression of LTP (Munoz and Humeres 2012 *Biometals* 25:825–83). We explored in an object recognition memory (RM) task in young (3 months), or aged (18 months) rats, the role of RYR / calcium channels. We found that young and aged rats spent more time exploring the novel object when tested 24 hr after the last session. By contrast, both groups displayed significant differences exploring repositioned objects. Six hours after the behavioural test brain, mRNA from rats was analyzed by qRT-PCR. We found in that mRNA levels of the RyR2/RyR3 isoforms increased in the hippocampus of young rats, but not in the perirhinal cortex, after a behavioural paradigm (in preparation).

Sensory Neuroscience*: Olfactory coding in teleost fishes*: Olfactory crypt cells are purported pheromone sensors in the teleost olfactory epithelium. To test this hypothesis and understand their role, crypt cells were isolated from juvenile and sexually mature rainbow trout and stimulated with different odorant classes under calcium imaging. Our data indicate that crypt cells have a broad tuning profile in juvenile specimen, being able to detect a vast array of odorants. In sexually mature specimen, their tuning profile sharpens, displaying a preference for reproduction related odorants and possible pheromones (Bazáes & Schmachtenberg, *J. Exp.l Biol*. 215:1740-8 2012). These results support a functional specialization of the three types of olfactory receptor neurons in teleost fishes, which is reflected in their morphology, brain projection patterns and molecular machinery (reviewed in Bazáes et al., in press).

Modulation of visual coding in the retina by nitric oxide (NO): Previous studies showed that retinal NO, liberated by certain amacrine cells, inhibits ganglion cell OFF responses, but little is known about its effects on OFF bipolar cells (reviewed in Vielma et al., *Brain Res*. 430:112-25 2012). We investigated if NO regulates glutamate responses in OFF bipolar cells through the activation of the

soluble guanylyl cyclase-cGMP pathway. To this end, OFF bipolar cells were recorded in vibratome sections of rat retina with whole-cell patch clamp and identified by morphological and electrophysiological criteria. Both NO-donors and cGMP analogues eliminated the slow component of the bipolar cell response to glutamate, while the amplitude of the fast component remained unchanged. On the other hand, perfusion with the NO synthase inhibitor L-NAME prolonged the timing of the glutamate response. Our data suggest that NO alters the glutamate response characteristics of OFF bipolar cells, and confirm that the retinal OFF pathway is modulated by NO, which has important implications for visual coding in the retina (manuscript in preparation).

Computational Neurobiology: How are physical signals encoded by the nervous system? How does the brain analyze spike trains? What are the underlying computational *coding* principles? At the current stage of scientific knowledge, answering those questions is still a challenge for biology and computational neuroscience. We have worked on a method to estimate the Gibbs distribution, under spatial and temporal constraints, of a retina ganglion cell population response. We showed that a model using high order statistics, instead of an Ising pair wise model, produces a fit better of the retinal spike train response. Furthermore, in a joint chapter with Dr. B. Cessac, we demonstrated that the retina is an excellent case to study "Spike Trains Statistics", where the rationale was to understand the relation between the statistical properties of a visual stimulus (an image) and the resulting neural response (coding) of the retina neural network.

5. Cross-cutting: ***Molecular Simulations, and Computational Biology***. The main objective of the Line 5 is to implement advanced bioinformatics and computational biology tools to produce multi-scale models to gain insight to the fundamental structure and dynamics underlying complex biological phenomena. With the objective of implementing a cross-cutting research area in CINV, the associated researchers have been involved in a large number of transverse collaborations.

Implementation of computational facilities: We use and produce computer-based simulation tools by combining advanced mathematical modeling, thermodynamics, biophysics and chemistry, together with high performance computing (HPC) techniques. To face these challenges, the implementation of one of the largest computer facilities for scientific computing in Chile was required. This installation was executed by a joint venture between Universidad Andres Bello (UNAB) and CINV. Specifically, these facilities have a SGI ICE 8200 server with 1,536 cores and 3 TB of RAM. At the same time, three new computer facilities associated to this line have been implemented; one at CINV Valparaiso (Computational Neurosciences Laboratory, headed by Dr. P. Orio) and two other in Santiago (Computational Biology Laboratory at Fundación para la Vida, headed by Dr. T. Perez-Acle, and Center for Bioinformatics and Integrative Biology at UNAB, headed by Dr. F. Gonzalez-Nilo). These facilities include a large amount of software for molecular simulation and bioinformatics, which are used to integrate data from genes to structures.

Molecular Simulations: While our main goal is to produce a tool for multi-scale simulation of biological systems, we also study processes at a single scale. At the micro-scale (atomic level), we use tools from molecular modeling and molecular simulations to understand the physicochemical properties that govern intercellular communication mediated by Connexin channels, trans-membrane transport mediated by Aquaporins and ligand-gated ion channels, and molecular recognition and signal transduction processes mediated by GPCRs. At the meso-scale (cell level) we use network topology to infer and characterize networks of cell signaling events, gene regulation, neural coordination and conductance-based modeling (analysis of neural excitability using conductance-based modeling). At the macro-scale (population level) we use rule-based modeling to study the behavioral adaptation of artificial populations (agents) due to the spread of infectious diseases and information. During the reported period, we have obtained results at both the micro and the macroscale. At the microscale, by using the crystallographic structure of the β2 Adrenergic

Receptor we were able to extend the evidence (*PLOS One* **6**, e23815, 2011) that support the existence of two ligand binding microdomains in the GPCR molecular architecture, as we previously suggested (*Bioorg Med Chem* **16**, 4378-4389, 2008). On the other hand, we provided structural evidence to support the role of the transmembrane helix 1 (TM1) of Cx26 in regulating oligomerization, function and permeability of hemichannels (*Mol. Biol. Cell* **23**, 3299-3311, 2012). At the macroscale, we reported an extended version of the Kappa language that will allow us to produce models using a pseudo-explicit space (*arXiv* [q-bio.PE], 20, 2012).

Additional projects are in progress through a theoretical-experimental cycle between this line and line 1 (Structure and Function of Molecular Sensors). To date, Dr. Gonzalez-Nilo´s team has implemented different molecular models of membrane (*J. Chem. Theory and Comp.*, **8** (5): 1765-1773, 2012) and ion channels, such as K^+ channels (Shaker and BK), TRP channels (TRPV1 and TRPM8) and Ca^{+2} channels, all of them validated experimentally. These models have been submitted to different approaches (Free Energy Perturbation calculations, External Electric Field, among others) in order to study the structural elements that govern the conductance of these channels. One of the more exciting advances in these collaborations is the prediction of key residues involved in the selectivity filter of Ca^{+2} channels (collaboration with Dr. A. Neely). Using these strategies, we have found novel residues and the most relevant structural mechanism involved in the modulation of the conductance of K^+ channels (collaboration with Dr. D. Naranjo, *Biophys. J.* **103**:1198, 2012). Additionally, we are implementing the use of sequence similarity networks to study the evolution of the many proteins superfamilies studied by the experimental groups, with the aim of using evolutionary information to inform function.

Modeling and analysis of neural excitability using conductance-based modeling: In the lab of P. Orio, the main interests are the role of different ion channels in the response of cold-sensitive neurons and nerve endings, and the effect of stochastic cannel gating in neural excitability. The main achievements of this period are: a) A numerical method for the simulation of a large number of discrete stochastic processes such a Markov Chains (Orio and Soudry, *PLOS One* **7**, e36670, 2012). This method, developed for ion channels but with applications in other areas, is faster than the explicit simulation of Markov Chains and more accurate than other algorithms published by the date. b) Analysis of the role of the HCN1 in the behavior of cold-sensitive nerve endings and the transduction of cold stimuli at the receptor level (*J Neurophysiol* **108**:3009-3023, 2012). In this work, numerical simulations were critical in the interpretation of experimental data. c) Using stochastic search algorithms, we fitted the parameters of a mathematical model that proposes a role of the TRPM8 channel in the acute response to cold in cold sensitive nerve endings (in preparation). Finally, there are three intra-Institute collaborations worth mentioning: conductance-base modeling of motion direction-selectivity circuits in the retina (collaboration with A. Palacios); we pioneered in the application of Optical Flow methods to analyze time-lapse images of migrating neural precursors in zebrafish embryos (collaboration with K. Whitlock; submitted), and, in collaboration with Perez-Acle and Ewer groups, we are starting the application of network inference methods to the analysis of the neural network responsible for the behavior of ecdysis in *Drosophila.*

b) Publications:

Summary table

Category of Publication	**MSI Center Members**	**Number of Publications coauthored by students**	**Total Number of Publications**
ISI Publications or Similar to ISI Standard	Associate Researchers	29	43
	Other Researchers	6	14
SCIELO Publications or Similar to SCIELO Standard	Associate Researchers	0	0
	Other Researchers	0	0
Scientific Books and chapters	Associate Researchers	3	6
	Other Researchers	0	0
Other Scientific Publications	Associate Researchers	2	3
	Other Researchers	0	0
Total of Publications		40	66

c) Other achievements:

- **Patents:** NONE
- **Intelectual Property:** NONE
- **Congress Presentations:**

Summary Table

Type of presentation	National Events [Number]	International Events [Number]
A. Associate Researchers		
Conferences, oral communications, poster communications, others (specify)	42	85
Invited presentations (not included in above row)	0	0
B. Other researchers (Adjunct Researchers, Senior Researchers, Young Researchers, Postdoctoral Researchers and Students)		
Conferences, oral communications, poster communications, others (specify)	28	40
Invited presentations (not included in above row)	0	0

- **Organization of Scientific Events:**

The second ***international workshop on Olfaction and Evolution (EVOLFII)*** held in Puerto Natales Chile. This initiative was lead by Dr. Peter Mombaerts (Max Planck Institute of Biophysics, Director, Department of Molecular Neurogenetics). Associated with this workshop we held a one-**day symposium in Valparaiso ".**

International Symposium: "Intercellular Comunication Via Pannexin and Connexin-based Channels in Health and Disease". It was part of the annual meeting of the Chilean Society for Cell

Biology, Puerto Varas, Chile (October 26, 2012). Four seminars were presented to an average of 80 assistants.
All the members of CINV were involved in the organization of the ***"Neuroscience meets Valparaíso" Meeting*** held at the Centro Cultural de Valparaiso on November 27-28, 2012. Through this series of symposia, CINV made an effort to reveal some of the secrets of this extremely complex but fascinating network of cells, the nervous system.

- **Scientific Editorial Boards:**

R. Latorre: Biological Research (ISI) since 1992, Proceedings of the National Academy of Sciences(ISI) since 2004, Journal of General Physiology(ISI) since 2004, Channels (ISI) since 2007 and Journal of Biological Chemistry (2008-2013).
J. Ewer: Journal of Insect Science (ISI) since 2006.
A. Palacios: Biological Research (ISI) since 2008, J.Pol.Complex Sys since 2013.
T. Perez-Acle:_ PeerJ since 2012.
J.C. Sáez. Frontiers in Neurosciences (ISI) since 2010.

- **Awards:**

New grants awarded. During this period, we have been awarded a two Scientific Ring project from PIA CONICYT. One directed by Gonzalez-Nilo (ACT-1107), dedicated to integrate structural biology knowledge to support the development of Bionanotechnology in Chile. The second one (ACT-1104) is lead by C. Gonzalez and deals the structure-function of voltage sensors in variety of ion channels: e P. Orio is participating in another Scientific Ring (PI R. Madrid from USACH) dedicated to understand the role of TRP channels in thermotransduction and synaptic plasticity at a systems level. We have been awarded 3 FONDECYT projects; Perez-Acle: Dissecting the structure function relationships that are coded into the molecular architecture of Cx26 hemichannels and gap junctions (1130652); Gonzalez-Nilo: Study of structural and dynamic properties that govern selectivity and conductance of K channels under non-equilibrium conditions (1131003), and Orio: Conductance-based modeling of the dynamic response of cold thermoreceptors (1130862). K. Whitlock received an award from Aquaneering Inc. for an E-rack for use in K-12 education. Postodoctoral awards from CONICYT: Jose Antonio Garate (3130547), Fanny Momboisse (3120221) and Alvaro Ardiles (3130759). Ph.D. students A. Vielma and G. Contreras were awarded additional funds for their thesis work.
Special Awards: R. Latorre received multiples awards during this period:."**Medalla Bicentenario"(2011)** Bicentennial medal from the municipality of Valparaíso aknowledging his 40 year of outstanding contributions to national science and his commitment to the recovery of Valparaíso..**Doctor Honoris Causa de la Facultad de Medicina de la Universidad de la República** (Uruguay) for his contribution to science and specifically to biophysics **Premio Juan Negrín de la Sociedad Española de Ciencias Fisiológicas** for his brilliant scientific career and contribution to the molecular physiology of ion channels and his commitment to the development of biomedical research in Spain and Latin America. **Profesor Honorario de la Universidad Nacional de la Plata** (Argentina). For his commitment to the development of biomedical research in Latin America.
Several of our graduate students have been awarded competitive international travels awards from the **Biophysical Society (G. Contreras 2011, I. Diaz and D. Baez 2012), IBRO-LARC (A. Gonzalez, A. Vielma), IBRO Travel Grants Award (A. Arias).**
Juan Andres Orellana was awarded **Best Ph.D. thesis by the Chilean Academy of Science** (2011).

4) Education and Capacity Building

a) Education and Capacity Building:

Members of the CINV participate in two interdisciplinary Neuroscience graduate programs of the University of Valparaíso: a Masters Program and a PhD Program.

The Masters Program in Neuroscience was founded in 1999 by CINV Members and is currently directed by Dr. Agustin Martinez (CINV, Research line 2). Last year it was reaccredited for 8 years (through 2018), becoming the longest accredited program of the University. To date it has graduated 35 students. The program is characterized by a high content of basic Neuroscience as well as for its multidisciplinarity. Its students are from various disciplines: biologists and biochemists as well as health professionals, engineers or mathematicians, eager to understand the biological basis of the functioning of the nervous system. During this period, the CINV-ICM has granted 7 fellowships for Master students.

The program of study is designed for 22h/week effort. First year activities include courses and electives, such as General Organization and Development of the Nervous System, Neural and Synaptic Transmission Physiology, Sensory Physiology, Movement and Motor Control, Neuropathology, Experimental Methods in Neuroscience, Microscopy, Biophysics, Biostatistics, Computational Neuroscience, Research rotations and Thesis Project. The second year is for the development of the thesis. The program can be completed in one year of full-time effort (44 h/per week), as long as the student meets the academic requirements for approval of courses and thesis project.

Requirements and application to enter the program: Holding a Diploma or Bachelor Degree in areas related to Neuroscience (biology, biochemistry, some areas of medicine such as speech therapy, physiotherapy, psychology, etc.). The applicant should send a Curriculum Vita, Certificate of Title and grades and a letter of intent. Candidates that are shortlisted are interviewed by the Program Committee and must demonstrate a basic understanding of a scientific article. Registration is through the university and program websites, www.uv.cl and www.magisterneurociencia.cl

The PhD Program in Neuroscience was created in 2002 by CINV members and is accredited through 2017. Its Director is currently Dr. John Ewer (CINV, Research line 3). It is designed to train researchers interested in the development, the structure, and the function of the nervous system. The strengths of our Program are in the areas of molecular physiology and biophysics, computational neuroscience, sensory neuroscience, neuronal plasticity and neuropathology, and development and neurogenetics.

The curriculum includes a one-year mandatory Core Course and Lab rotations in the first year, and one seminar course per semester thereafter. The Core course is composed of 6 modules, some of which may be skipped if they are equivalent to courses taken elsewhere. Students must pass a qualifying exam and thesis project defense in the third semester, and defend their thesis at the end of the Program. Chilean and foreign students are funded for 4 years by available scholarship programs (CONICYT, MECESUP, UV). In addition, during this period, the CINV-ICM granted 8 graduate fellowships. Guaranteed funding ensures that students can devote full time to their Ph.D. work. Agreements with other post-graduate programs allow students to take elective courses at other universities in Chile and abroad, as well to carry out research internships. Our Program has a double Ph.D. agreement with the Scuola Internazionale Superiore di Studi Avanzati (SISSA) of Trieste, Italy (www.sissa.it), which has expanded the range of thesis topics.

Requirements and application to enter the program: To apply to our Ph.D. program the student must hold the equivalent of a Bachelor' Degree in areas relevant to Neuroscience (biology, biochemistry, relevant areas of medicine, computer science, etc.). The applicant must send a Curriculum Vitae, a certificate of title and grades, and a letter of intent. Shortlisted candidates are interviewed by the

faculty and must demonstrate a basic understanding of a scientific article, and be interested in areas of research carried out by the Program's faculty. Web pages: www.uv.cl and www.dunuv.cl.

The **new Ph.D. program in Biophysics and Computational Biology** (Program Director: Dr. A.Neely, Research line 1) proposed in the original grant will start accepting students by September 2013 and train students in state-of-the-art biophysical techniques and concepts. It is intended to only accept a select group of students per year (less than 5) with diverse backgrounds. The program is organized in six week modules of intense training in diverse areas of physical chemistry, molecular biology, and mathematics. The specific set of courses that each student will take will be decided his/her tutoring committee based on the student's interests and weaknesses. Theoretical coursework will be complemented with intensive bench work. Students will be encouraged to attend specialized international workshops, and will be given credit for doing so.

International courses organized by the CINV

International Workshop: *"Structure and Function of Connexins and Pannexins Channels".* Workshop on the structure and function of channels formed by connexins and pannexins **(Line 2 and 4)**. This workshop brought together leading scientists in a national and international network as part of the BioN (our associated "Redes" grant) to discuss key aspects of the role of these proteins in central aspects of neuroscience and biology in general. Fifteen seminars were presented during this workshop, which were attended by an average of 60 people per session. We performed 4 laboratories for Ph.D and Master students: ***Laboratory I:*** Functional analysis of gap junction channels by intercellular diffusion of fluorescent tracers. ***Laboratory II:*** Functional analysis of gap junction channels by double whole cell patch clamp. ***Laboratory III:*** Functional analysis of hemichannels by uptake of fluorescent dyes. ***Laboratory IV:*** Functional analysis of hemichannels by electrophysiological techniques. The workshop in Valparaiso enrolled 19 students and 12 full scholarships were awarded for attendance. The workshop was organized by Dr. A. Martínez (UV) and Dr. J.C. Saéz (PUC). Faculty included (5) from the CINV-ICM, (4) from other Chilean Universities, (2) from US, (1) from Canada, and (2) from Europe.

The **"Small Brains, Big Ideas: Biomedical Insights from Invertebrates"** international practical course was offered for the second time October 29th - November 7th, 2012 **(Line 3)**. The primary objective of the course is to expose students from Latin America to the use of invertebrate preparations for basic and applied research in neurosciences and biomedicine. The course included lectures, laboratory exercises, research talks from Faculty, two outreach talks for high school students and teachers and the general public, and a symposium. As in 2010, some of these activities took place in Santiago (Medical School of the University of Chile) and others in Valparaíso (at the Naval Museum, Cerro Artilleria). As in 2010, the course was organized by Dr. J. Sierralta (Medical School of the University of Chile, Santiago, Chile), Dr. Y. Fuentes, (MIT Sloan School of Management, Boston, USA), and Dr. J. Ewer (CINV, University of Valparaíso, Valparaíso, Chile). Faculty included (5) from Chile, (1) from Argentina, (6) from USA, of which 5 were from the UMass Medical School, USA, and (2) from UK. This course will be offered every two years for the foreseeable future. Most of the information on the course can be viewed at the course's web site: www.smallbrains.org

b) Achievements and results:

The **Masters program** currently has 30 active students (14 female; 16 male): 24 students are doing their thesis, of which 15 have advisors within our CINV and will defend their thesis within the next couple of years. This year over 45 students applied to our program of which 22 were accepted.

In the **PhD Program**, 17 students have graduated since 2002 (5 female; 12 male), of which 6 graduated during this period (see list below). Of these 6 students (1) was associated with Line 2, (3)

with Line 3 and (1) with line 4; the additional students was in a lab at the University of Valparaiso which is outside our Center. The Program currently has 24 students (6 female, 18 male). During this year's recruitment period we received 26 applications and admitted 13 students, which represents a 2-3-fold increase in the number of applicants relative to our historical average. All current students typically attend one national or international conference in their area of study; funding is provided through their fellowship or by their advisor. (See Annexes 5.1 and 5.2).

Main achievements of our Ph.D. and Master Students during the period:

Ph.D. Thesis Project Approvals and Qualifying exams (Sep 2011-Dec 2012) :

1.- *Willy Carrasquel.* "Estudio de la topología de dos familias de proteínas (B y Y) moduladoras de la actividad del canal BK humanos mediante técnicas espectroscópicas". Tutor: R. Latorre **(Line 1)**.
2.- *Oscar Jara.* "Mechanism of differential regulation of Cx26 and Cx43 hemichannels by actin cytoskeleton". Tutor: A. Martínez **(Line 2)**.
3.- *Jorge Torres.* "Dissecting the roles of the transcription factors dlx3b/4b and six4b during the olfactory epithelium development in zebrafish". Tutor: K. Whitlock **(Line 3)**.
4.- *Isaac Garcia.* "Role of the amino-terminal domain of Cx26 in the biogenesis and functional state of hemichannels and gap junction channels". Tutor: A. Martínez **(Line 2)**.
5.- *Ignacio Diaz.* "Revealing the allosteric nature of the Shaker K+ channel". Tutor: D. Naranjo **(Line 1)**.

Graduations of Ph.D. students (Sep 2011-Dec 2012):

1.- *Vania Figueroa:* "Role of connexin26 hemichannels in the regulation of purinergic calcium signaling under physiological and pathological conditions" Tutor: J.C. Saéz **(Line 2)**.
2.- *Katica Boric.* "Ethanol exposure disrupts cranial neural crest migrations and primary cilia in developing zebrafish" Tutor: K. Whitlock **(Line 3)**.
3.- *Gonzalo Terreros.* "Efectos de la ingesta de un alimento funcional elaborado con quínoa y del ácido alfa linolénico sobre deterioro que produce el estrés crónico en el hipocampo de rata". Tutor: A. Dagnino and P. Muñoz **(Line 4)**.
4.- *Joaquin Letelier.* "Disruption of zebrafish prokineticin2 signaling causes kallmann syndrome related phenotypes". Tutor: K. Whitlock **(Line 3)**.
5.- *Wilson Mena..*"Mecanismo en activación secuencial de lared neuronal que comanda una conducta innata en drosophila". Tutor: J. Ewer **(Line 3)**.
6.- *Alvaro Ardiles*."Alteración cognitiva y sináptica en octodon degus: Un modelo natural de alzheimer". Tutor: A. Palacios **(Line 4).**
7.- *Claudio Elgueta.* "Neuromodulatory mechanims of retinal A17 amacrime cells". Tutor: A. Palacios **(Line 4)**.

Graduation of Master Students during the period:

1.-*Paula Vallejos.* "Application of Markov Chains for the characterization spatial memory in rodents in the open field model". Tutor: A. Palacios and M.S. Torres **(Line 4)**.
2.-*Mauricio Aspe.* "Diversity and possible heritability of the chronotype of the chilean rodent *Octodon degus*". Tutor: J. Ewer **(Line 3)**.

Graduated Doctors form other programs during September 2012 and March 2013

1.- Pablo Sáez, Ph.D. in Physiological Science (Pontificia Universidad Católica de Chile). "Pannexin1 hemichannels are involved in the Ca^{2+} code that mediates migration, but not death or maturation, in dendritic cells". (**Line 2**)
2.- Kenji Shoji, Ph.D. in Physiological Science (Pontificia Universidad Católica de Chile)."Expression of pannexin1 hemichannels and their involvement in death of lymphocytes". (**Line 2**)

3.- Luis Cea. Ph.D. in Physiological Science (Pontificia Universidad Católica de Chile). "Conditions that induce atrophy of skeletal muscles up-regulate the expression of $P2X_7$ receptors and hemichannels formed by connexins or pannexin1, which explains the increase in membrane permeability". (**Line 2**)

4.- *Calixto Dominguez*. Ph.D. in Biotechnology, UNAB. "Estudio de la regulación transcripcional activada por el morfógeno Dpp durante la formación del ectodermo dorsal en *Drosophila melanogaster*". (**Line 5)**

5- *Angel Gonzalez.* , Ph.D. in Biotechnology, UNAB: "Estudio de los Micro-Dominios Funcionales e Interacción con Ligandos en Receptores Acoplados a Proteínas G (GPCRs) Mediante Herramientas Bioinformáticas". **(Line 5**).

6.- Paula Vallejos. "Application of Markov Chains for the characterization spatial memory in rodents in the open field model". Tutor: A. Palacios and M.S. Torres **(Line 4)**.

Undergraduate students. Nineteen undergraduate students are either doing thesis work or laboratory rotations in the CINV-ICM. Most are students in Biological and Biochemical science, but some are also from physical science and engineering. The CINV-ICM has granted 5 fellowships for undergraduate students.

Postdoctoral fellows. A number of postsdocs have joined CINV labs, and the ICM has provided critical funding for (3) of them.

c) Destination of Students:

Master´s program: To date 35 students have graduated. Half of our graduates have gone on to doctoral studies, a quarter of them are working as teachers in national and international universities and the other quarter continues working in their professions.

PhD Program: Seventeen students have graduated since 2002. The first graduate of the Program (Dr. A. Chavez) graduated in 2007. He, as well as the majority of graduates of our Program, is currently carrying out postdoctoral work (with the exception of a few graduates from 2012 who are either looking for postdoctoral positions or are completing experiments and preparing their thesis work for publication).

Summary Table*:*

Obtained Degree	Academy	Industry and Services	Studies	Research	Other (Specify the other type of activity
Doctoral	6				
Master					
Undergraduate	1				
TOTAL	7				

5) Networking and other collaborative work

a) Networking:

With the support of MSI through the program "*Redes Formales de Colaboración*", we have created a program to nurture three independent but interconnected international collaborative networks: the **Biophysics and Computational Neuroscience**, the **Genetics and Development**, and the **Intercellular Communication**. As can be noted in the following paragraphs, all the networking and collaborative activities carried out during this period, were fully consistent with the objectives of strengthening ties with national and international collaborators. These activities allowed us to: 1) promote the activities of CINV, 2) ensure continuity of scientific collaboration along time, and, 3) promote the generation of measurable and quantifiable impacts that benefit both CINV and the national scientific community.

The main objective of the **Biophysics and Computational Neuroscience** network (**BiCoNeu**) is to establish a dialogue between these two sciences, enhancing the understanding of the molecular basis of neuronal excitability and how this may influence the behavior of neurons and neural networks. We are achieving this goal by strengthening existing ties between CINV investigators and colleagues and institutions abroad and consolidating partnership between the different *research* lines of our institute. Molecular Sensors local researchers: (C. González, R.Latorre, D. Naranjo and A. Neely), Systems Neuroscience (A. Palacios) and our cross cutting research line Computational Biology and Molecular Simulations (F. González-Nilo, T. Perez-Acle and P. Orio) are the local members of this network. **BiCoNeu** includes an international network composed by researchers of a NIH-FIRCA grant: F. Bezanilla (U. of Chicago), M. Holmgren, (NIH) and R. Latorre (CINV); a collaboration grant CONICYT / MINCYT with "Universidad De La Plata" (V. Milesi, U. De La Plata and C. Gonzalez, CINV) and European ANR grant that includes INRIA (France) via an "*equipe asociee*" and researchers C. Bruno, T. Vieville, F. Alexandre and M. J. Escobar from UTFSM, among others. This network started on 2012 with a ***Summer School on Biophysics and Computational Biology (SSBCB)*** aimed at graduate students from Chile and other countries (particularly from Latin America). This course will be offered every two years and take place at CINV. Defined as a theoretical/practical school, students face morning lectures and in-situ lab experiences during the afternoon.

We have organized the ***II LACONEU Summer School in Computational Neurosciences*** with the participation of 13 faculties from around the world, 40 students and funding from IBRO-LARC, CINV, MECESUP, INRIA, ISCV, ANR-CONICYT, UV. LACONEU main objective is to promote the advancement of computational neuroscience in Latin America. The multidisciplinary study of neural coding using neuroscience, statistical mechanics, mathematics and computational approaches will help us to understand how the nervous system makes sense of the environment in which it is immersed. In this school, we hope to create a highly participatory environment where the exchange of students and teachers, based on knowledge and skills, help build partnerships that remain over time (see details at www.laconeu.cl)

The main objective of the **Genetics and Development Network** is to strengthen knowledge in genetics and developmental biology regarding the development and function of the nervous system. We consider that it is essential to the formation of our graduate students, to expose them to senior researchers that have succeeded conducting sciences within these fields. Thus, a major goal of this network is to create opportunities for our graduate students to interact with researchers conducting high impact work in Chile and other countries. We organized for the second time, together with Dr. J. Sierralta (Medical School of the U. de Chile, Santiago) and Dr. Y. Fuentes, (MIT, Boston, USA) the international practical course, **"*Small Brains, Big Ideas*: Biomedical Insights from Invertebrates.** The primary objective of the course is to expose students from Latin America to the use of invertebrate preparations for basic and applied research in neurosciences and biomedicine.

The international participants were Dr. S. Reppert, Dr. V. Budnik, Dr. M. Alkema, Dr. M. Freeman, Dr. M. Francis, University of Massachusetts Medical School, USA; Dr. F. Ceriani, Fundación Instituto Leloir, Argentina; Dr. S. Waddell, University of Oxford, UK; Dr. B. Smith, Arizona State University, USA.

We, with Dr. P. Mombaerts, organized the second **international workshop *Evolution and Olfaction* (EVOLFII)** in Puerto Natales, Chile. Associated with this workshop we held a one-day symposium, ***Synthesis of Scents*,** in Valparaiso. The participants in these events were Dr. B. Hansson Director, Max Planck Institute for Chemical Ecology; Dr. J. Bohlmann University of British Columbia; Dr. Bart Kempenaers, Director Max Planck Institue for Ornithology Behavioural Ecology and Evolutionary; Dr. B. Key, Director, Brain Growth and Regeneration Lab, University of Queensland; P. Mombaerts, Director Molecular Neurogenetics, Max Planck Institute of Biophysics; Dr. A. Schäfer, Max Planck Institute for Medical Research; Dr. Kathleen Whitlock, CINV; Dr. D. Wicher, Max Planck Institute for Chemical Ecology.

As a result of these interactions we are organizing the International Practical Course ***Olfaction and the Environment*** (October 2013, Valparaiso) which will be funded through Milenio REDES. Finally, in 2014 we will participate in organizing the third version of an international course in Developmental Biology, in collaboration with the Universidad Andrés Bello.

The **Intercellular Communication Network** is aimed at exchanging, sharing, disseminating and teaching the latest knowledge on the role of synaptic neurotransmission at chemical synapses and gap junctions at electrical synapses. To do so, we conducted various activities during this period: One symposium at the Annual meeting of the Chilean Society for Cell Biology, one workshops at the University of Valparaiso and several national and international conferences. It is also our goal to motivate new students from pre-and post-graduate to conduct research into these areas. So far, the network has been funded by two ECOS-CONICYT projects (C08 B01 completed in March, 2012), one DAAD-CONICYT (ends in July, 2012), one IBRO project, and also funding from local universities. This network is formed by CINV researcher belonging to lines System Neuroscience (A. Palacios and O. Schmachtenberg), Cellular signaling (J.C. Sáez, A.M. Cárdenas and A. Martinez) and Computational Biology and Molecular Simulations (T. Pérez-Acle). During this period we were visited by Dr. C. Giaume, College de France, Paris, France; Dr. L. Leybaert, University of Ghent, Belgium, Dr. S. Penuela, University of Ontario Canada and Dr. R. Johnson, University of Minnesota, USA.

Four seminars were presented to an average of 80 assistants. Also we organized **an International**

We have organized the ***International Symposium: "Intercellular Communication Via Pannexin and Connexin-based Channels in Health and Disease".*** It was part of the annual meeting of the Chilean Society for Cell Biology, Puerto Varas, Chile (October 26, 2012). Four seminars were presented to an average of 80 assistants. - It is noteworthy that application to held 2015 International Gap Junction in Valparaiso on the year was recently approved. It will be the first time it takes place outside of Europe or USA. This conference is the main activity in gap junction research and is repeated every two years.

In addition to activity related to formal network, CINV as whole participated in the following networking activities:

- The first Meeting with CINV Scientific Advisory Committee (SAC) took place on November 26 and November 29, 2012. The Scientific Advisory Committee of CINV is at present composed by: K. Anderson (Developmental Biology Program, Center for Stem Cell Biology, Member of the National Academy of Science, USA), M. Bennett (Albert Einstein College of Medicine, Member of the National Academy of Science, USA), J. Diamond (NIH, NINDS, Senior Investigator Synaptic Physiology Section), E. Neher (Max Planck Institute of Biophysical Chemistry, Nobel Prize in Physiology, 1991) and K. Willecke (LIMES Institute, University of Bonn).

- All the members of CINV were involved in the organization of the ***"Neuroscience meets Valparaíso" Meeting*** held at the Centro Cultural de Valparaiso on November 27-28, 2012. Through this series of symposia, CINV made an effort to reveal some of the secrets of this extremely complex but fascinating network of cells, the nervous system. The first symposium (line 1 and 5) aimed to provide an informative and up to date account of the techniques and concepts used in understanding ion channels. C. González (CINV), F. González-Nilo (CINV, Universidad Nacional Andrés Bello and CINV), F. Bezanilla (University of Chicago, CINV), M. Holmgren (NIH, NINDS, CINV) and T. Pérez-Acle were the speakers in this symposium. Cell-to-cell conversations are mediated by gap junctions and the second symposium was dedicated to these channels that allow the electrical and metabolic communication between cells (line 2). The speakers in this Symposium were J.C. Sáez (Universidad Católica, CINV), K. Willecke (LIMES Institute, University of Bonn), O. Schmachtenberg (CINV), J. Stehberg (Universidad Nacional Andrés Bello) and M. V.L. Bennett (Albert Einstein College of Medicine). Synapsis and neurotransmission was the subject of the third symposium (line 3) and here a special emphasis is given to the mechanism of neurotransmission mediated by exocytosis. Speakers in this Symposium were: A.M. Cárdenas (CINV), E. Neher (Max Planck Institute for Biophysical Chemistry), A. Chávez (Albert Einstein College of Medicine), J. Diamond (NIH, NINDS) and A. Palacios (CINV). The fourth symposium tackled the question of how the nervous system develops and produces behavior (line 4). K. Anderson (Developmental Biology Program, Center for Stem Cell Biology), T. Bestor (Columbia University Medical Center), J. Ewer and K. Whitlock (CINV) were the speakers in this Symposium.

b) Other collaborative activities:

A collaboration with the laboratory of Dr. A. Patapoutian (The Scripps Research Institute, California Campus) was established in 2010 and is still active. Through this collaboration one of Dr. Latorre (Line 1) PhD students, H. Moldenhauer, learned the high throughput mutagenesis technique applied to the cold receptor TRPA1.

During 2012 a collaboration with Dr. R. Olcese laboratory at UCLA to perfect voltage-clamp fluorometry technique was strengthen with a visit by A. Neely and G. Contreras. An abstract describing the first fluorometric detection of voltage-sensor movement in Ca channels was submitted to the 57th meeting of the Biophysical Society.

During the last semester of 2011 we started collaborating with Dr. Feng Qin, Professor at the Department of Physiology and Biophysical Sciences, State University of New York, Buffalo. Ph.D. Student David Báez-Nieto did a 3-month research internship in Dr. Qin laboratory with the purpose of learning and building a fast temperature clamp. This equipment is able to change the temperature of the medium surrounding the cell in less than a millisecond and has proved to be very useful when working with thermoTRP channels (TRPV1 and TRPM8). Using this equipment, D. Báez was able to perform a complete analysis of the thermodynamic parameters that determine the high temperature sensitivity of TRPM8 (a cold receptor).

6) Outreach and connections with other sectors

a) Outreach:

The CINV's mission is to become reference for scientific research in the field of Neuroscience and a bridge that bring science closer to the community by actively collaborating in the development of Valparaíso. Activities that have been implemented in this end have resulted in CINV being recognized by the region's diverse community leaders, raising the awareness of the importance of science for the country's development. This process also has strengthened relationships with a wide range of local institutions along with a progressive positioning in the local media that shows a greater interest in covering CINV's activities.

General Objective: CINV's goal is to bring science closer to diverse audiences, from elementary school students to university students, professionals and the community in general. Specifically, the activities have focused on: bringing science closer to other disciplines (*Tertulias Porteñas*); promoting awareness of the scope of the field of Neuroscience and CINV's research (*Neuromantes*); bringing Neuroscience to high school students who have shown promise in the sciences (*Coloreando Neuronas*); bringing science to local schools in order to integrate it into the curriculum (*¿Qué tienes en mente?* and *Ciencia al Tiro*); and raising awareness about the life and work of a noted Chilean scientist (*La ciencia del ser*).

Criteria of the Center for Promoting Outreach Activities:

- To promote increased knowledge about science using broad and direct language, drawing comparisons with day to day experiences and using common technological tools;
- To promote practice, "learning by doing," ensuring coherence with the counterpart's level of knowledge;
- To include professionals from other disciplines in order to enrich teaching-learning processes, making an effort to ensure that the knowledge contributed is comprehensive and lasting;
- To promote discussion of issues which are of interest to our center through social networks and active participation in activities organized by CINV.

Achievements: These activities have increased CINV recognition by various community leaders. In addition, we have progressively positioned our center in the local electronic and print media, which have begun to show greater interest in and familiarity with our activities. This has allowed us to develop strong relationships with political leaders in the city and the region resulting in widespread support of our scientific endeavors.

Brief description of the key activities implemented during this period. (The remaining activities are described in the corresponding appendix.)

Launch of CINV as a Millennium Institute: The process of scientific dissemination in our region began with the launch of CINV as a Millennium Institute in the Hall of Honor of the Chilean Congressional Building. The event was used to highlight the importance of having a Millennium Institute outside of Santiago that is housed in a public university. Attendees included the Regional Governor, the Mayor of Valparaíso, and the Rector of the University of Valparaíso, several regional officials and local academics, and the Executive Director of the Millennium Scientific Initiative.

Date: October 25, 2011

Participants: 250 people

Sponsoring institutions: the Chilean House of Representatives and Senate

Tertulias Porteñas. CINV organized meetings of scientists, artists and intellectuals designed to bring Neuroscience closer to society. Cristián Warnken, a well-known Chilean TV personality, moderated conversations about topics in neuroscience creating spaces for an interdisciplinary discussion. The

target audiences included university students, professionals, and community groups. The events were held at the Regional Library of Valparaíso, a recently restored building that houses the country's first public library. A second goal of the initiative was to increase the visibility of this historical space in the community. The events were transmitted via streaming video due to the significant size of the audience.
-Dates: August 29 and December 13, 2012
-Attendees: 500 people (total)
-Sponsoring institutions: Universidad de Valparaíso, Regional Public Library, Radio Valentín Letelier
-Sponsors: local newspaper Diario El Mercurio de Valparaíso, Hotel Gervasoni

Coloreando neuronas. This workshop was directed at students in their last year of high school from the Valparaíso region and was organized in collaboration with EXPLORA CONICYT. The main purpose of "*Coloreando neuronas*" was to encourage students who have shown promise in science by providing them with the experience of working in a laboratory and giving them an opportunity to attend workshops led by CINV researchers.
-Associated institutions: Universidad de Valparaíso, EXPLORA Conicyt Program
-Duration: Second half of 2012
-Participating students: 20

¿Qué tienes en mente? CINV made an ongoing commitment to offer scientific talks in schools throughout the Valparaíso region in the context of the Year of Neuroscience organized by the program EXPLORA CONICYT as a key topic for 2012. A total of 3000 students were reached through eight lectures offered by CINV researchers in different provinces of the region. The speakers also took part in the launch of the program in the cities of Valdivia.
-Events: Eight lectures in schools and two regional inaugurations
-Total number of participating students: 3,000 in Valparaíso and 500 in Valdivia
-Associated Institutions: EXPLORA CONICYT, Universidad de Valparaíso, Universidad Austral

Homenaje a Francisco Varela. CINV and the "Instituto de Sistemas Complejos de Valparaíso" organized a series of activities commemorating the tenth anniversary of the death of renowned Chilean neurobiologist Francisco Varela in collaboration with the Universidad de Valparaíso. The event featured the launch of the book *La ciencia del ser* published by Universidad de Valparaíso; the screening and premiere of two documentaries produced by Swiss filmmaker Franz Reichle; and an exhibit on Francisco Varela life in the National Council of Culture and the Arts of Valparaíso (Consejo Nacional de la Cultura y las Artes en Valparaíso).
-Dates: November 28-December 12, 2012 in Valparaíso and Santiago
-Participants: 500 people (total)
-Sponsoring Institutions: Universidad de Valparaíso, Complex Systems Institute of Valparaíso, National Council of Culture and the Arts, National Cinema Library
-Participating Businesses: Xstrata, Codelco Andina, and Mellafe y Salas.

Ciencia al Tiro. This project, directed by Dr. Kathleen Whitlock, was initiated by funding from MSI Millennium Nucleus Center for Genomics of the Cell. The project is being continued by the CINV Millennium Center under the direction of Dr. Whitlock. In December 2011, the Center's work with Escuela Básica Pacífico de Playa Ancha de Valparaíso drew to a close with the inauguration of a solar shower system. It was the first system of its kind installed in a public school in the Valparaíso region.

During 2012, we have worked on the publication of the book *La alegría de la ciencia* as well as lectures and other activities which will once again involve direct contact with students beginning in 2013.
-Dates: Ongoing since 2009
-Participants: 400 students in 2011
-Participating Institutions: Middle schools Arabe-Siria and Basico Pacifico, Neuroscience PhD. program from the Universidad de Valparaíso

b) Connections with other sectors:

During its first year as a Millennium Institute, the CINV set a goal approaching the community and positioning itself at the local level as a key institution in the region's development, actively taking part in the recovery of the historic sector of Valparaíso. In this context, we focused our efforts on developing connections with public sector institutions. We have implemented the following actions related to this goal in collaboration with those entities.

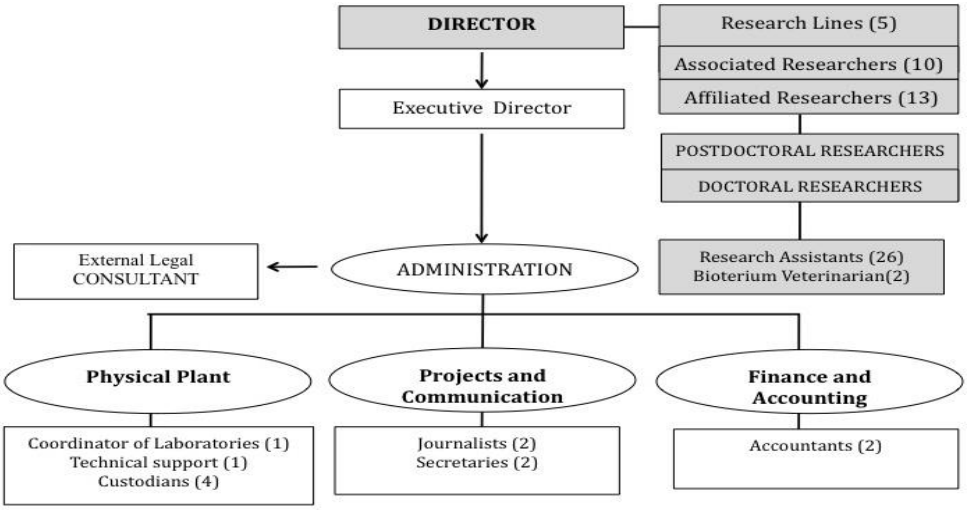
Advising Services for the Municipality of Valparaíso on the Master Plan for Heritage Management : The Manager of CINV was invited by the Municipality of Valparaíso to serve on the Consulting Advisory Committee for the Master Plan for Heritage Management during 2012. In this capacity, he provided input regarding the Port Area where the CINV building will be located.

Cooperation Agreement with the Valparaíso Cultural Park: In an effort to position Valparaíso as a "beacon of science in Chile," CINV agreed to offer symposia and scientific conferences at the Parque Cultural de Valparaíso. The Latin American Society for Biophysics and Chilean Society for Neuroscience will hold their conference there in 2013, and Valparaíso has been proposed as the headquarters for the International Gap Junctions Conference in 2015. It would be the first Latin American city to host the event.

CINV Severin Building Project: We are planning a new house for CINV at an historical area of Valparaiso to revive a sector currently very degraded. In addition to laboratories and animal facilities this project includes outreach areas such as an auditorium, meeting rooms and an exhibit hall. The Government of Valparaiso co-financed the project, and the Ministry of Public Works acts as technical assistance, ensuring top laboratories standards and helping to fulfill the requirement of UNESCO heritage sites. In 2012 the architectural design was concluded and we obtained approval from the National Monuments Council and the Ministry of Housing and Urban Development.

7) Administration and Financial Status

a) Organization and administration:



Organization of the administrative staff: An Executive Director supervises and coordinates all the administrative duties according to the need of the Director and other Investigators. He coordinates outreach and networking with the private sector and community leaders together with all effort aimed at securing funds and the construction of the new building to house the CINV. Beside all the booking and accounting, there is a team to fulfill researchers' needs for purchasing equipment and computer maintenance and another team offering support for development, submission of grants, and outreach activities. Each host institution provides office and laboratory space to individual investigators holding faculty positions. Base salaries for individual investigators are also assumed by the corresponding host institution. The Universidad de Valparaíso has set up an institutional grant to help with operational expenses.

Category	Female	Male	TOTAL
Assistant & Technicians	23	11	34
Administrative Staff	4	3	7
TOTAL	27	14	41

b) Financial Status:

At present, MSI contribute with 55% of the administrative cost while CONICYT with 23% and Universidad de Valparaíso with 18%. The remaining 4% corresponds to funds allocated for specific projects.

MSI outcome concentrate mainly in equipments (34%) to increase scientific productivity. In human resource, we have privileged students and technicians (24%) over incentive to investigators (18%). Six per cent of MSI budget was spend on supplies to complement funds from other grants that cover most of research operational cost.

Annex 1.- Institute/Nucleus Researchers

1.1 Associate Researchers

Full Name	Research Line	Nacionality	Gender	Date of birth	Profession	Academic Degree	Affiliation	Current Position	Relation with Center
Adrián Palacios	4	Chilean	M	18/03/1958	Psychologist	D	Universidad de Valparaíso	Professor	2
Alan Neely	1	Chilean	M	15/04/1956	Biologist	D	Universidad de Valparaíso	Professor	2
Ana María Cardenas	2	Chilean	F	01/04/1960	Pharmacist	D	Universidad de Valparaíso	Professor	2
David Naranjo	1	Chilean	M	17/10/1957		D	Universidad de Valparaíso	Professor	2
Fernando González	5	Chilean	M	09/12/1968	Chemist	D	Universidad Andres Bello.	Professor	2
John Ewer	3	Chilean	M	23/02/1961	Biologist	D	Universidad de Valparaíso	Professor	2
Juan Carlos Saez	2	Chilean	M	02/02/1956	Biochemist	D	Universidad Católica de Chile	Professor	2
Kathleeen Whitlock	3	American	F	27/08/1963		D	Universidad de Valparaíso	Professor	2
Ramon Latorre	1	Chilean	M	29/10/1941	Biochemist	D	Universidad de Valparaíso	Professor	1
Tomás Pérez Acle	5	Chilean	M	09/09/1970		D	Fundación Ciencia para la Vida	Assoc. Investigator	2

1.2 Young Researchers

NONE

1.3 Senior Researchers

NONE

1.4 Others

Full Name	Research Line	Nacionality	Gender	Date of birth	Profession	Academic Degree	Affiliation	Current Position	Relation with Center
Agustín Martínez	2	Chilean	M	14/08/1968		D	Universidad de Valparaíso	Assoc. Professor	2
Alfredo Kirkwood	4	Chilean	M		Biologist	D	John Hopkins University	Professor	2
Carlos Gonzalez	1	Cuban	M	13/12/1965	Biochemist	D	Universidad de Valparaíso	Assoc. Professor	2
Francisco Bezanilla	1	Chilean	M	17/05/1944	Biochemist	D	Chicago University	Professor	2
José Hurtado	4	Chilean	M	19/02/1969		D		Assoc. Investigator	2
Miguel Holmgren	1	Chilean	M			D	Porter Neuroscience Research Center	Senior Inves.	1
Oliver Schmachtenberg	4	German	M	12/12/1970		D	Universidad de Valparaíso	Assoc. Professor	2
Osvaldo Alvarez	1	Chilean	M	14/10/1942	Biochemist	D	Universidad de Chile	Professor	2
Pablo Muñoz	4	Chilean	M	19/01/1973		D	Universidad de Valparaíso	Assoc. Professor	2
Patricio Orio	5	Chilean	M	03/12/1973	Biochemist	D	Universidad de Valparaíso	Assoc. Professor	2
Ralph Greenspan	3	American	M			D	Kavli Institute for Mind and Brain	Professor	2
Verónica Milesi	1	Argentinian	F	02/12/1962		D	Universidad Nacional de La Plata	Professor	2
Daniel Aguayo	5	Chilean	M	08/08/1978		D	Universidad Andrés Bello	Professor	2

NOMENCLATURE:

[Gender]
[M] Male [F] Female

[Academic Degree]
[U] Undergraduate [M] Master [D] Doctoral

[Relation with Center]
[1] Full time [2] Part time

Annex 2.- Research Lines

Nº	Line Research	Objective	Description	Researcher	Discipline	Starting Date	Ending Date
1	STRUCTURE AND FUNCTION OF MOLECULAR SENSORS	We try to understand how ion channels and pumps can respond to a variety of stimuli.	It is a combination of molecular biology, electrophysiology, modern fluorescence techniques, simulations and molecular modeling.	D. NARANJO, R. LATORRE,A. NEELY, O. ALVAREZ, F. BEZANILLA, M HOLMGREN, V MILESI, CGONZÁLEZ.	73	08-08-2011	
2	CELL SIGNALING	Investigate how protein-protein interactions and covalent modifications of dynamin control neurosecretion and trafficking of ion channels.	Using patch clamp amperometry and total internal reflection fluorescence microscopy the handling by the cell of vesicles containing neurotransmitters is characterized.	J.C. SAEZ, A.M. CARDENAS, AMARTINEZ.	61	08-08-2011	
3	DEVELOPMENTAL GENETICS AND BEHAVIOR	Understanding how the nervous system develops and produces complex behaviors.	Using zebrafish and *Drosophila* as biological models, the development of the olfactory system and the genetic pathways controlling behavior are studied.	KWHITLOCK, JEWER, RGREENSPAN.	63 y 74	08-08-2011	
4	SENSORY AND SYSTEMS	To investigate the mechanisms of neuronal encoding the visual, olfactory and cerebral physiological and pathological conditions.	Using different animal models, including Degu, a natural model for studying AD. The molecules identified by Group 2 as regulators of neurosecretion will be tested in the context of neuronal plasticity.	APALACIOS, AKIRKWOOD, P MUÑOZ, OLIVER SCHMACHTENBERG, J HURTADO.	61 y 73	08-08-2011	
5	COMPUTATIONAL BIOLOGY AND MOLECULAR SIMULATION	Using high performance computing for molecular modeling of membrane proteins, drug design assisted by computer, and inference and dynamics of biological networks.	Interaction between theoretical and experimental biologist to create new methods, models and hypothesis suitable to be tested by the experimental groups	F. GONZALEZ, T.PEREZ ACLE, P. ORIO	6, 59 y 73	08-08-2011	

Annex 3.- Publications

Students (former and present) co-authoring a publication are underlined.

3.1.- ISI Publications or Similar to ISI Standard

3.1.1 Associate Researchers:

1. Baez-Nieto D, Castillo JP, Dragicevic C, Alvarez O, Latorre R. (2011). RThermo-TRP channels: Biophysics of polymodal receptors, Advances in Experimental Medicine and Biology, Vol.704:469-490

2. Castillo JP, De Giorgis D, Basilio D, Gadsby DC, Rosenthal JJ, Latorre R, Holmgren M, Bezanilla F. (2011) Energy landscape of the reactions governing the Na+ deeply occluded state of the Na+/K+-ATPase in the giant axon of the Humboldt squid. Proc Natl Acad Sci U S A. 108:20556-20561

3. González A, Perez-Acle T, Pardo L, Deupi X. (2011) Molecular Basis of Ligand Dissociation in b-Adrenergic Receptors. Plos One. 6(9): e23815

4. Grbić M, Van Leeuwen T, Clark RM, Rombauts S, Rouzé P, Grbić V, Osborne EJ, Dermauw W, Ngoc PC, Ortego F, Hernández-Crespo P, Diaz I, Martinez M, Navajas M, Sucena É, Magalhães S, Nagy L, Pace RM, Djuranović S, Smagghe G, Iga M, Christiaens O, Veenstra JA, Ewer J, Villalobos RM, Hutter JL, Hudson SD, Velez M, Yi SV, Zeng J, Pires-daSilva A, Roch F, Cazaux M, Navarro M, Zhurov V, Acevedo G, Bjelica A, Fawcett JA, Bonnet E, Martens C, Baele G, Wissler L, Sanchez-Rodriguez A, Tirry L, Blais C, Demeestere K, Henz SR, Gregory TR, Mathieu J, Verdon L, Farinelli L, Schmutz J, Lindquist E, Feyereisen R, Van de Peer Y. (2011) The genome of Tetranychus urticae reveals herbivorous pest adaptations. Nature. Nov 23;479(7374):487-92.

5. Latorre R, Brauchi S, Madrid R, Orio P. (2011) A cool channel in cold transduction. Physiology (Bethesda). 26(4):273-85.

6. Miranda-Laferte E, Gonzalez-Gutierrez G, Schmidt S , Zeug A, Ponimaskin EG, Neely A, Hidalgo P. (2011) Homodimerization of the Src homology 3 domain of the calcium channel beta-subunit drives dynamin-dependent endocytosis. Journal of Biological Chemistry. 286(25): 22203-10.

7. Orellana JA, Shoji KF, Abudara V, Ezan P, Amigou E, Sáez PJ, Jiang JX, Naus CC, Sáez JC, Giaume C. (2011) Amyloid β-induced death in neurons involves glial and neuronal hemichannels. Journal of Neuroscience. Mar 30; 31(13):4962-77.

8. Orellana JA, Froger N, Ezan P, Jiang JX, Bennett MV, Naus CC, Giaume C, Sáez JC. (2011) ATP and glutamate released via astroglial connexin 43 hemichannels mediate neuronal death through activation of pannexin 1 hemichannels. Journal of Neurochemistry. 118(5):826-40.

9. Orellana JA, Figueroa XF, Sanchez HA, Contreras-Duarte S, Velarde V, Saez, JC, (2011) Hemichannels in the Neurovascular Unit and White Matter Under Normal and Inflamed Conditions. CNS & Neurological Disorders-Drug Targets 10 (3): 404-414

10. Paul TA, Rovnak J, Quackenbush SL, Whitlock KE, Zhan H, Gong Z, Spitsbergen J, Paul R, Bowser PR, Casey JW. (2011). Transgenic expression of Walleye Dermal Sarcoma accessory genes in zebrafish does not result in tissue proliferation. Marine Biotechnology (NY). 13(2):142-50.

11. Stephenson JF, Whitlock K, Partridge J. (2011) Zebrafish Preference for Light or Dark Is Dependent on Ambient Light Levels and Olfactory Stimulation. Zebrafish 8 (1): 17-22

12. Acuña MA, Pérez-Nuñez R, Noriega J, Cárdenas AM, Bacigalupo J, Delgado R, Arriagada C, Segura-Aguilar J, Caviedes R, Caviedes P. (2012) Altered voltage dependent calcium currents in a neuronal cell line derived from the cerebral cortex of a trisomy 16 fetal mouse, an animal model of down syndrome. Neurotoxicity Research. 22(1):59-68.

13. Aguayo D, Gonzalez-Nilo F, Chipot C. (2012) Insight into the properties of cardiolipin containing bilayers from molecular dynamics simulations, using a hybrid all-atom/united-atom force field. Journal of Chemical Theory and Computation. 8(5):1765-1773.

14. Ardiles AO, Tapia-Rojas CC, Mandal M, Alexandre F, Kirkwood A, Inestrosa NC & Palacios AG. (2012) Post-synaptic dysfunction is associated with spatial and object recognition memory loss in a natural model of Alzheimer's disease. Proceedings of the National Academy of Science. 109(34):13835-40 .

15. Bennett MV, Garré JM, Orellana JA, Bukauskas FF, Nedergaard M, Sáez JC. (2012) Connexin and pannexin hemichannels in inflammatory responses of glia and neurons. Brain Research. 1487:3-15.

16. Braidy N, Muñoz P, Palacios AG, Castellano-Gonzalez G, Inestrosa NC, Chung RS, Sachdev P, Guillemin GJ. (2012) Recent rodent models for Alzheimer's disease: clinical implications and basic research. J Neural Transm. 119(2):173-95.

17. Cárdenas AM, Ardiles AO, Barraza N, Baéz-Matus X, Caviedes P. (2012) Role of Tau Protein in Neuronal Damage in Alzheimer's Disease and Down Syndrome. Archives of Medical Research. 43(8):645-54.

18. Cea LA, Riquelme MA, Cisterna BA, Puebla C, Vega JL, Rovegno M, Sáez JC. (2012) Connexin- and annexin-based channels in normal skeletal muscles and their possible role in muscle atrophy. Journal of Membrane Biology. 245(8):423-36.

19. Contreras GF, Neely A, Alvarez O, Gonzalez C, Latorre R. (2012) Modulation of BK channel voltage gating by different auxiliary β subunits. Proceedings of the National Academy of Sciences. 109(46):18991-6

20. Cortés C, Eugenin E, Aliaga E, Carreño LJ, Bueno SM, Gonzalez PA, Gayol S, Naranjo D, Noches V, Marassi MP, Rosenthal D, Jadue C, Ibarra P, Keitel C, Wohllk N, Court F, Kalergis AM, Riedel CA. (2012) Hypothyroidism in the adult rat causes incremental changes in brain-derived neurotrophic factor, neuronal and astrocyte apoptosis, gliosis, and deterioration of postsynaptic density. Thyroid. 22(9):951-63.

21. Dinardo MM, Camerino G, Mele A, Latorre R, Conte Camerino D, Tricarico D. (2012) Splicing of the rSlo gene affects the molecular composition and drug response of Ca2+-activated K+ channels in skeletal Muscle. Plos One, 7(7):e40235.

22. Eugenin EA, Basilio D, Sáez JC, Orellana JA, Raine CS, Bukauskas F, Bennett MV, Berman JW. (2012) The role of gap junction channels during physiologic and pathologic conditions of the human central nervous system. Journal of Neuroimmune Pharmacology. 7(3):499-518.

23. Fiori MC, Figueroa V, Zoghbi ME, Saez JC, Reuss L, Altenberg GA.(2012) Permeation of calcium through purified connexin 26 hemichannels. Journal of Biological Chemistry. 87(48): 40826-34

24. Gonzalez C, Báez-Nieto D, Valencia I, Oyarzún I, Rojas P, Naranjo D, and Latorre, R. (2012) K channels: function-structural overview. Comprehensive Physiology. 2: 2087-2149.

25. Gonzalez W, Riedelsberger J, Morales-Navarro SE, Caballero J, Alzate-Morales JH, González-Nilo FD, Dreyer I. (2012) The ph-sensor of the plant K+ uptake channel Kat1 is built of a sensory cloud rather than of single key amino acids. Biochemical Journal. 442(1):57-63.

26. Harden MV, Pereiro L, Ramialison M, Wittbrodt J, Prasad MK, McCallion AS, Whitlock KE. (2012) Close association of olfactory placode precursors and cranial neural crest cells does not predestine cell mixing. Developmental Dynamics. 241(7):1143-54.

27. Huang S, Treviño M, He K, Ardiles A, Di Pasquale R, Guo Y, Palacios A, Huganir R, Kirkwood A. (2012) Pull-push neuromodulation of ltp and ltd enables bidirectional experience-induced synaptic scaling in visual cortex. Neuron. 73(3):497-510.

28. Jara O, Acuña R, García IE, Maripillán J, Figueroa V, Sáez JC, Araya-Secchi R, Lagos CF, Pérez-Acle T, Berthoud VM, Beyer EC, Martínez AD. (2012) Critical role of the first transmembrane domain of Cx26 in regulating oligomerization and function. Molecular Biology of the Cell. 23(17):3299-3311.

29. Koulakoff A, Mei X, Orellana JA, Sáez JC, Giaume C. (2012) Glial connexin expression and function in the context of Alzheimer's disease. Biochimica et Biophysica Acta. 1818(8):2048-2057.

30. Lagos CF, Araya-Secchi R, Thomas P, Pérez-Acle T, Tapia RA, Salas CO. (2012) Molecular modeling of Trypanosoma cruzi glutamate cysteine ligase and investigation of its interactions with glutathione. Journal of Molecular Modeling. 18(5):2055-64.

31. Lahr EC, Dean D, Ewer J. (2012) Genetic analysis of ecdysis behavior in Drosophila reveals partially overlapping functions of two unrelated neuropeptides. Journal of Neuroscience. 32(20):6819-29.

32. Miranda-Laferte E, Schmidt S, Jara AC, Neely A, Hidalgo P. (2012) A short polybasic segment between the two conserved domains of the β2a-subunit modulates the rate of inactivation of R-type calcium channel. The Journal of Biological Chemistry.287(39):32588-97.

33. Morera FJ, Alioua A, Kundu P, Salazar M, Gonzalez, C, Martinez AD, Stefani E, Toro L, Latorre R. (2012). The first transmembrane domain (TM1) of β2-subunit binds to the transmembrane domain S1 of α-subunit in BK potassium channels. FEBS Letters. 586(16):2287-93.

34. Moscoso C, Vergara-Jaque A, Márquez-Miranda V, Sepúlveda R, Valencia I, Díaz-Franulic I, González-Nilo F, Naranjo D. (2012) K+ Conduction and Mg2+ Blockade in a Shaker Kv-Channel Single Point Mutant with an Unusually High Conductance. Biophysical Journal, 103(6):1198-1207.

35. Orellana JA, Sáez PJ, Cortés-Campos C, Elizondo RJ, Shoji KF, Contreras-Duarte S, Figueroa V, Velarde V, Jiang JX, Nualart F, Sáez JC, García MA. (2012) Glucose increases intracellular free Ca(2+) in tanycytes via ATP released through connexin 43 hemichannels. Glia. 60(1):53-68.

36. Orellana JA, von Bernhardi R, Giaume C, and Sáez JC. (2012) Glial hemichannels and their involvement in neurodegenerative diaseases. Reviews in Neuroscience. 23(2): 163–177.
37. Perez-Armendariz EM, Cruz-Miguel L, Coronel-Cruz C, Esparza-Aguilar M, Pinzon-Estrada E, Rancano-Camacho E, Zacarias-Climaco G, Fernandez P, Espinosa AM, Becke, I, Saez JC, Berumen J, Perez-Palacios G. (2012) Connexin 36 is expressed in beta and connexins 26 and 32 in acinar cells at the end of the secondary transition of mouse pancreatic development and increase during fetal and perinatal life. Anatomical Record-Advances In Integrative Anatomy And Evolutionary Biology. 295(6): 980-990.
38. Schalper KA, Riquelme MA, Brañes MC, Martínez AD, Vega JL, Berthoud VM, Bennett MV, Sáez JC. (2012) Modulation of gap junction channels and hemichannels by growth factors. Molecular BioSystems. 8(3): 685-698.
39. Stehberg J, Moraga-Amaro R, Salazar C, Becerra A, Echeverría C, Orellana JA, Bultynck G, Ponsaerts R, Leybaert L, Simon F, Sáez JC, Retamal MA. (2012) Release of gliotransmitters through astroglial connexin 43 hemichannels is necessary for fear memory consolidation in the basolateral amygdala. FASEB Journal. 26(9):3649-57
40. Stephenson JF, Partridge JC, Whitlock KE. (2012) Food and conspecific chemical cues modify visual behavior of zebrafish, danio rerio. Zebrafish. 9:68-73.
41. Sundram V, Fanny S, Ng FS, Roberts MA, Millán C, Ewer J, Jackson FR. (2012) Requirements for LARK in the Drosophila circadian system. Journal of Biological Rhythms. 27(3):183-95.
42. Vasquez JC, Palacios AG, Marre O, Berry II MJ, Cessac B (2012). Gibbs distribution analysis of temporal correlation structure on multicell spike trains from retina ganglions cells. Journal of Physiology Paris. 106(3-4):120-7.
43. Vergara-Jaque A, Poblete H, Lee EH, Schulten K, Gonzalez-Nilo FD, Chipot CJ. (2012) Molecular basis of drug resistance in A/H1N1 virus. Journal of Chemical Information and Modeling. 52(10):2650-6.

3.1.2 Other researchers

1. Brauchi S, Orio P (2011) Voltage Sensing in ThermoTRP Channels. Advances in Experimental Medicine and Biology 704:517-530.
2. Couve E, Schmachtenberg O. (2011) Autophagic Activity and Aging in Human Odontoblasts. Journal of Dental Research, 90(4): 523-8.
3. Delgado LM, Schmachtenberg O. (2011) Neurogenesis in the adult goldfish cerebellum. The Anatomical Record: Advances in Integrative Anatomy and Evolutionary Biology 294: 11-15.
4. Gonzalez C, Lopez Rogriguez A, Srikumar D, Rosenthal J & Holmgren M (2011) Editing of human KV1.1 channel mRNAs disrupts binding of the N-terminus tip at the intracellular cavity. Nature Communications. 2:436.
5. Gonzalez C*, Manzanares D*, Ivonnet P, Chen RS, Valencia-Gattas M, Conner G.E, Larsson HP, & Salathe M *Contributed equally.(2011) Functional apical BK channels are required for maintenance of airway surface liquid volume. Journal of Biological Chemistry Jun 3; 286(22):19830-9.

6. Martínez AD, Maripillán J, Acuña R, Minogue PJ, Berthoud VM, Beyer EC (2011) Different domains are critical for oligomerization compatibility of different connexins Biochemical Journal 436: 35-43.

7. Muñoz P, Humeres A, Elgueta C, Kirkwood A, Hidalgo C. and Núñez MT. (2011) Iron Mediates N-Methyl-D-aspartate Receptor-dependent Stimulation of Calcium-induced Pathways and Hippocampal Synaptic Plasticity. Journal of Biological Chemistry 286, (15): 13382-13392

8. Salazar M, Moldenhauer H, Baez D. (2011) Could an Allosteric Gating Model Explain the Role of TRPA1 in Cold Hypersensitivity?. (Journal Club) The Journal of Neuroscience. 31(15):5554-5556.

9. Bazáes A. and Schmachtenberg O. (2012) Odorant tuning of olfactory crypt cells from juvenile and adult rainbow trout. Journal of Experimental Biology. 215(Pt 10):1740-8.

10. Couve, E., Osorio, R. and Schmachtenberg, O. (2012) Mitochondrial autophagy and lipofuscin accumulation in aging odontoblasts. Journal of Dental Research. 91(7): 1-6.

11. Muñoz P and Humeres A. (2012) Iron deficiency on neuronal function. Biometals 25:825–835.

12. Orio P, Parra A, Madrid R, González O, Belmonte C, Vania F. (2012) Role of ih in the firing pattern of mammalian cold thermoreceptor endings. J Neurophysiol. 108(11):3009-23.

13. Orio P, Soudry D. (2012) Simple, fast and accurate implementation of the diffusion approximation algorithm for stochastic ion channels with multiple states. PLoS ONE. 7(5): e36670

14. Vielma AH, Retamal M, Schmachtenberg O. (2012) Nitric oxide signaling in the retina: What have we learned in two decades?. Brain Research. 430:112-25.

3.2.- SCIELO Publications or Similar to SCIELO Standard.

NONE

3.3.- Scientific Books and Chapters

3.3.1 Associate Researchers:

1 Orellana JA, Giaume C, Sáez JC. (2011) Neurodegenerative diseases – processes, prevention, protection and monitoring Edited by: Raymond Chuen-Chung Chang ISBN 978-953-307-485-6 Publisher: InTech

2 Palacios AG, Cosmelli D, Cohen-Varela A. (2011) Un recorrido junto a Francisco Varela. Pag. 11-22. Edit. Adrian Palacios y Amy Cohen-Varela. La ciencia del ser: Las rutas de Francisco Varela. Editorial Universidad de Valparaiso. 320 p.

3 Cessac B and Palacios AG. (2012) Spike train statistics from empirical facts to theory: the case of the retina. In Mathematical Problems, in Modeling in Computational Biology and Biomedicine. Cazals F, Kornprobst P, Springer, pag. 261-302.

4 Latorre R. and Báez-Nieto, D. (2012) Ca2+ activation of K+ channels: RCK domains. In: Encyclopedia of Biophysics. Springer Verlag. Gordon C.K. Roberts, editor. DOI 10.1007/978-3-642-16712-6.

5 Latorre R, Gonzalez C, Rojas P. (2012) Signal-Transduction-Dependent Channels. Neuroscience in the 21st Century, Eugene Martin and Donald Pfaff, eds. Springer. 81-107

6 Pertusa M, Moldenhauer H, Brauchi S, Latorre R, Madrid R, Orio P (2012). Mutagenesis and temperature-sensitive little machines. In: Mutagenesis, Mishra, R. Ed., InTech Open Access publisher. Chapter 11, pp 221-246.

3.4.- Other Publications

3.4.1 Associate Researchers:

1. Gonzalez C, Contreras G, Peysler A, Larsson P, Neely A, Latorre R. (2012) Voltage sensor of ion channels and enzymes. Biophysical Reviews. 4(1): 1-15.
2. Muñoz P (2012) Iron-mediated redox modulation in neural plasticity. Communicative & Integrative Biology 5:2,166–168.
3. Nunez F, Ravello C, Urbina H, Perez-Acle T. (2012) A Rule-based Model of a Hypothetical Zombie Outbreak: Insights on the role of emotional factors during behavioral adaptation of an artificial population. arXiv:1210.4469v1 [q-bio.PE].

3.4.2 Other researchers:

3.5.- Collaborative publications:

Category of Publication	**1 researcher**		**2 researchers**		**3 researchers**		**4 or more researchers**	
	Nº	**%**	**Nº**	**%**	**Nº**	**%**	**Nº**	**%**
ISI Publications or Similar to ISI Standard	42	64,6	9	13,9	4	6,2	1	1,5
SCIELO Publications or Similar to SCIELO Standard								
Books and chapters	5	7,7	1	1,5				
Other Publications	2	3,1			1	1,5		
Total of publications	49	75,4	10	15,4	5	7,7	1	1,5

Annex 4.- Organization of Scientific Events

Scope	Title	Type of Event	City	Country	Responsible Researcher
National	The Neuroscience in action	Symposium	Valparaiso	Chile	Ramón Latorre
International	40 years of ion channels a Marriage of Convenience	Symposium	Valparaiso	Chile	Ramón Latorre
International	Workshop in Molecular Simulation & Drug Design	Workshop	Talca	Chile	Danilo González
National	Dynamic Clamp: Playing with Models in Real Neurons	Course	Valparaíso	Chile	Patricio Orio
International	Synthesis of Scents. Olfaction: From Molecule to Mind.	Symposium	Valparaíso	Chile	Kathleen Whitlock
International	Structure and Function of Connexin and Pannexin Channels.	Workshop	Valparaíso	Chile	Juan Carlos Sáez
International	Small Brains, Big Ideas: Biomedical Insights from Invertebrates	Course	Santiago	Chile	John Ewer
International	Small Brains, Big Ideas	Symposium	Valparaíso	Chile	John Ewer
International	Neuroscience meets Valparaíso	Symposium	Valparaíso	Chile	Ramón Latorre

Annex 5.- Education and capacity building

5.1 Capacity Building inside MSI Centers

MSI RESEARCHER	NUMBER												TOTAL NUMBER PER MSI RESEARCHER		
	Undergraduate students			Graduate students						Postdoctoral researchers					
				Masters			Doctoral								
	F	M	**T**	F	M	**T**	F	M	**T**	F	M	**T**	F	M	**T**
Adrián Palacios					1	1	1		1				1	1	**2**
Agustín Martínez	2		2		4	4		2	2		1	1	2	7	**9**
Alan Neely	2		2		1	1		1	1				2	2	**4**
Ana María Cárdenas	2		2	3		3	1		1	1		1	7		**7**
Carlos González	1	1	2					1	1				1	2	**3**
David Naranjo					1	1		2	2					3	**3**
Fernando González	1		1				2		2				3		**3**
John Ewer	1		1		1	1	1	2	3				2	3	**5**
Juan Carlos Sáez	2		2	1		1	5	5	10		3	3	8	8	**16**
Kathleen Whitlock				1		1	1	3	4		1	1	2	4	**6**
Oliver Schmachtenberg		1	1		2	2		1	1		1	1		5	**5**
Pablo Muñoz	4	1	5				1	1	2				5	2	**7**
Patricio Orio								2	2					2	**2**
Ramón Latorre				1		1	2	5	7				3	5	**8**
Tomás Pérez-Acle		3	3											3	**3**
TOTAL	**15**	**6**	**21**	**6**	**10**	**16**	**14**	**25**	**39**	**1**	**6**	**7**	**36**	**47**	**83**

Annex 5.2.- Short-term Traineeships of MSI students

Student Name	Institution	Country	Advisor	Project Description	Starting Date	Ending Date
Raúl Araya Secchi	IBM Thomas J. Watson Research Center	USA	Dr. R. Zhou	The goal of this project is to study the effect that hereditary-deafness associated mutations have over the structure, function and transport properties of Cx26 hemi-channels and gap-junction channels using molecular modeling and simulation techniques.	20/04/2012	20/10/2012
Mauricio Aspé	SISSA	Italy	R. Rumiati	Training in techniques to study human behavior	00-09-2012	00-12-2012
David Baez	State University of New York at Buffalo, Buffalo, NY, USA	USA	Dr. F. Qin	Learning and building a fast temperature clamp to apply to thermoTRP channels for is Thesis.	20/09/2011	20/12/2011
Gustavo Contreras Cáceres	David Geffen School of Medicine at UCLA, Los Angeles	USA	Dr. R- Olcese	To learn voltage clamp fluorometry applied to calcium channels.	01/11/12	31-11-2012

Annex 6.- Networking and other collaborative work

6.1 Networking

Network Name	Network Scope	Network Participants [Number]				Institutions
		From the Center		External		
		Researchers	Postdocs/ Students	Researchers	Postdocs/ Students	
Network 1 **Biophysics and Computational Neuroscience** network (**BiCoNeu**) is to establish a dialogue between these two sciences, enhancing the understanding of the molecular basis of neuronal excitability and how this may influence the behavior of neurons and neural networks	I	6	24	9	10	CINV-Universidad de Valparaíso, Chile Pontificia Universidad Católica de Chile, Chile Centro de Modelamiento Matemático (CMM), FCFM - Universidad de Chile University of Minnesota, USA Department of Anatomy and Cell Biology; University of Western Ontario, Canada INSERM, Collège de France, France Universidad Del Desarrollo, Chile The University of Chicago, USA Universidad de Ghent, Bélgica.
Network 2 The main objective of the **Genetics and Development Network** is to strengthen knowledge in genetics and developmental biology regarding the development and function of the nervous system.	I	4	10	3	10	CINV-Universidad de Valparaíso, Chile UMass Medical School (USA) Welcome Trust (UK), AMSUD Pasteur, and Company of Biologists (UK) Medical School of the U.Chile, Santiago MIT, Boston, USA

Network Name	Network Scope	Network Participants [Number]				Institutions
Network 3 The **Intercellular Communication Network** is aimed at exchanging, sharing, disseminating and teaching the latest knowledge on the role of synaptic neurotransmission at chemical synapses and gap junctions at electrical synapses.	I	6	10	4	10	University of Ghent, Belgium University of Ontario Canada University of Minnesota, USA

Annex 6.2.- Other collaborative activities

Activity Name	Co-Participant Institution(s)	Participants [Number]				Products [Type & Number]
		MSI center		External		
		Researchers	Postdoc/Students	Researchers	Postdocs/Students	
High throughput mutagenesis technique applied to the cold receptor TRPA1	Scripps Research Institute, California Campus	1	1	1	4	Scientific exchange
Voltage-Clamp fluorometry applied to voltage-dependent Calcium channels	Department of Anesthesiology Cardiovascular Research Laboratory. Brain Research Institute David Geffen School of Medicine University of California Los Angeles	1	1	1	2	Scientific exchange
Fast temperature clamp to study high temperature sensitivity of TRPM8	Department of Physiology and Biophysical Sciences, State University of New York, Buffalo	1	3	1	3	Scientific exchange

Annex 7.- Outreach
7.1.- Outreach activities throughout the period

a) International events

Title of the Event	Type of Event	Date	Place Region	Target Audience
Cátedra en el área de la salud Miguel Alemán Valdés	Conference	29/10/2011	Ciudad de México, México	University Students
The amazing intelligence of bees: how tiny brains solve complicated problems	Conference	30/10/2012	Valparaíso, Chile	Secondary - Primary Students

b) National events

Title of the Event	Type of Event	Date	Place Region	Target Audience
Inauguration of CINV as a Millennium Institute	Ceremony	25/10/2011	Valparaíso, Chile	General Public
"El arte del descubrimiento, una mirada desde la ciencia"	Conference	05/11/2011	Valparaíso, Chile	General Public
"Diferencias entre la energía solar térmica y la energía solar eléctrica"	Workshop	01/11/2011	Valparaíso, Chile	Grammar school Students.
"El arte del descubrimiento, una mirada desde la ciencia"	Conference	18/11/2011	Quillota, Chile	Grammar and Middle school Students
Photographs exhibition	Exhibit	28/11/2011	Valparaíso, Chile	General Public
Tribute to R. Latorre Congreso Nacional	Ceremony	30/11/2011	Santiago, Chile	General Public
Documentary Monte Grande	Exhibit	05/12/2011	Santiago, Chile	General Public
Duchas Solares (Solar showers)	Exhibit	06/12/2011	Valparaíso, Chile	Basic school Students
Documentary Monte Grande	Exhibit	06/12/2011	Valparaíso, Chile	General Public
"La Ciencia del Ser. Las rutas de Francisco Varela"	Book Launching	07/12/2011	Valparaíso, Chile	General Public
Documentary Francisco Cisco Pancho	Exhibit	07/12/2011	Valparaíso, Chile	General Public
Documentary Francisco Cisco Pancho	Exhibit	12/12/2011	Santiago, Chile	General Public

Title of the Event	Type of Event	Date	Place Region	Target Audience
"Alteración de prok2/prokr2 en pez cebra provoca genotipo relacionados al síndrome de Kallmann" (Dr. Christian Wilson – USA)	Seminar	05/03/2012	Valparaíso, Chile	University Students and Faculty
Opening lecture for Neuroscience Year	Conference	20/03/2012	Valdivia, Chile	Secondary - Primary Students
"A single molecule approach for studying sub-nanometer effects of force on proteins and DNA" (Dr. Christian Wilson, University of California, USA)	Seminar	26/03/2012	Valparaíso, Chile	University Students and Faculty
"On the evolution and development of cells that make the vertebrate skeleton" (B.Frank Eames, Ph.D – University of Saskatchewan, Canadá)	Seminar	30/03/2012	Valparaíso, Chile	University Students and Faculty
"Examining ion cannel properties using free-energy methods" (Dr. Carmen Domene, University of Oxford South, USA.)	Seminar	02/04/2012	Valparaíso, Chile	University Students and Faculty
"From confocal microscopy to time correlated single photon counting" (Dr. Werner Zuschratter, Leibniz Institute for Neurobiology Magdeburg, Germany)	Seminar	09/04/2012	Valparaíso, Chile	University Students and Faculty
Opening lecture for Neuroscience Year	Conference	11/04/2012	Valparaíso, Chile	Grammar and Middle school Students
Seminar by Alex Vielma y Hans Moldenhauer (CINV students)	Seminar	20/04/2012	Valparaíso, Chile	University Students and Faculty
"Simulación molecular de canales de K" (Danilo González, CINV, Chile)	Seminar	27/04/2012	Valparaíso, Chile	University Students and Faculty
Seminar by Jorge Castex y Severín Lions (CINV students)	Seminar	04/05/2012	Valparaíso, Chile	University Students and Faculty
"Un mecanismo molecular para el proceso de activación-deactivación en el canal de Mg2+ CorA" (Dr. Eduardo Perozo, University of Chicago, USA)	Seminar	11/05/2012	Valparaíso, Chile	University Students and Faculty
"Entendiendo la transmisión dopaminérgica a través de interacciones proteína-	Seminar	18/05/2012	Valparaíso, Chile	University Students and Faculty

Title of the Event	Type of Event	Date	Place Region	Target Audience
proteína" (Gonzalo Torres, PhD, University of Pittsburgh, USA)				
Seminar by Christian Cortés e Ignacio Díaz (CINV)	Seminar	25/05/2012	Valparaíso, Chile	University Students and Faculty
Seminar by Angelina Palacios y Daniel Aguayo (CINV students)	Seminar	01/06/2012	Valparaíso, Chile	University Students and Faculty
Neuroscience lecture series: ¿Qué tienes en mente?	Conference	13/06/2012	Los Andes, Chile	Primary - Secondary Students
Neuroscience lecture series:¿Qué tienes en mente?	Conference	14/06/2012	Valparaíso, Chile	Primary - Secondary Students
Neuroscience lecture series: ¿Qué tienes en mente?	Conference	20/06/2012	Limache, Chile	Grammar and Middle school Students
Seminar by Alejandra Arias y Juan Pablo Castillo (CINV students)	Seminar	22/06/2012	Valparaíso, Chile	University Students and Faculty
Neuroscience lecture series:¿Qué tienes en mente?	Conference	27/06/2012	La Ligua, Chile	Primary - Secondary Students
Seminar by Nicolás Enrique y Gaspar Herrera (Argentina)	Seminar	30/06/2012	Valparaíso, Chile	University Students
Neuroscience lecture series:¿Qué tienes en mente?	Conference	04/07/2012	Viña del Mar, Chile	Grammar and Middle school Students
Seminar by Alan Astudillo (CINV students)	Seminar	06/07/2012	Valparaíso, Chile	University Students and Faculty
Seminar by Mauricio Aspe y Ximena Báez (estudiantes CINV)	Seminar	13/07/2012	Valparaíso, Chile	University Students and Faculty
¿Se murieron las células? Impacto de la actividad TRP" (Dr. Pablo Olivero, UV, Chile)	Seminar	20/07/2012	Valparaíso, Chile	University Students and Faculty
"Descifrando los códigos celulares y moleculares del tráfico del receptor 2 de apolipoproteína E (ApoER2):relevancia para la función de su ligando Reelina" (Dra. María Paz Marzolo, PUC, Chile)	Seminar	27/07/2012	Valparaíso, Chile	University Students and Faculty
Seminar by María José Guerra y Jorge Torres (CINV students)	Seminar	03/08/2012	Valparaíso, Chile	University Students and Faculty
Neuroscience lecture series:¿Qué tienes en mente?	Conference	08/08/2012	San Antonio, Chile	Grammar and Middle school Students

Title of the Event	Type of Event	Date	Place Region	Target Audience
Coloreando Neuronas·	Workshop	08/08/2012	Valparaíso, Chile	Middle school Students econdary Students
"Control epigenético de la expresión durante la diferenciación ósea" (Dr. Martín Montecino, UNAB, Chile)	Seminar	10/08/2012	Valparaíso, Chile	University Students and Faculty
UV seminar series	Seminar	10/08/2012	Valparaíso, Chile	University Students and Faculty
Neuroscience lecture series:¿Qué tienes en mente?	Conference	22/08/2012	Quintero, Chile	Grammar and Middle school Students
Internal seminar	Seminar	24/08/2012	Valparaíso, Chile	University Students and Faculty
Tertulias Porteñas: ¿Qué sabemos de los sueños?	Other (Tertulia)	29/08/2012	Valparaíso, Chile	General Public
Seminar by Catherine Estay y Carolina Estay (CINV students)	Seminar	07/09/2012	Valparaíso, Chile	University Students and Faculty
Neuroscience lecture series:¿Qué tienes en mente?	Conference	22/08/2012	Villa Alemana, Chile	Grammar and Middle school Students
"Control de la Plasticidad Cortical" (Dr. Alfredo Kirkwood, John Hopkins University, USA)	Seminar	28/09/2012	Valparaíso, Chile	University Students and Faculty
"Efectos del alcohol en el desarrollo fetal" (Dra. Katica Boric, CINV, Chile)	Conference	05/10/2012	Valparaíso, Chile	General Public
"Efectos del alcohol en el desarrollo fetal" (Dra. Katica Boric, CINV, Chile)	Conference	09/10/2012	Viña del Mar, Chile	General Public
"Odontoblasto: Función y Longevidad" (Eduardo Couve, UV, Chile)	Seminar	12/10/2012	Valparaíso, Chile	University Students and Faculty
"El complejo mundo visual de la simple araña saltarina" (Ximena Nelson, Ph.D. University of Canterbury, Nueva Zelanda.	Seminar	19/10/2012	Valparaíso, Chile	University Students and Faculty
Seminar by Valentina Aros y Jaime Maripillán (CINV students)	Seminar	09/11/2012	Valparaíso, Chile	University Students and Faculty
Visit to Valparaíso "Programa Penta – UC "	Other	10/11/2011	Valparaíso, Chile	High School Students
M. Iacoboni. "Recent development in mirror neurons research"	Seminar	12/11/2012	Valparaíso, Chile	University Students and Faculty

Title of the Event	Type of Event	Date	Place Region	Target Audience
David Báez y Jesús Olivares (CINV students)	Seminar	16/11/2012	Valparaíso, Chile	University Students and Faculty
¿Qué es un cerebro?	Workshop	10/12/2012	Valparaíso, Chile	Grammar school Students
Tertulias Porteñas: ¿Qué sabemos de la Conciencia?	Other (Tertulia)	13/12/2012	Valparaíso, Chile	General Public
"A single-molecule fluorescence system for studying adenylate kinase under force" (Christian Wilson, Ph.D. University of California. USA)	Seminar	14/12/2012	Valparaíso, Chile	University Students and Faculty
Contest "La ilusión en la pintura"	Competition	20/12/2012	Valparaíso, Chile	Grammar school Students
Coloreando Neuronas	Workshop	21/11/2011	Valparaíso, Chile	Middle school Students
"El factor de transmisión Atonal específica neuronas involucradas en la detección del movimiento en Drosophila" (Carlos Oliva, University of Leuven, Belgium)	Seminar	21/12/2012	Valparaíso, Chile	University Students and Faculty

7.2.- Products of outreach

Type of Product	Target Public	Scope	Total
TV series	General Public	International	1
Book	University students – Community in general	National	1
Album of confocal microscopy pictures	High school students	Regional	1
Website	General Public	International	1
Exhibit	General Public	National	1
Audio recording	General Public	National	1

7.3.- Articles and Interviews

Type of media and scope	Local/Regional		National		International		
	N° Interviews	N° Articles	N° Interviews	N° Articles	N° Interviews	N° Articles	**TOTAL**
Written	3	7	4	4	0	0	**18**
Internet	0	27	3	22	1	3	**56**
Audiovisual	1	1	2	0	0	0	**4**
TOTAL	**4**	**34**	**8**	**24**	**1**	**3**	**78**

Annex 8.- Connections with other sectors

Activity and Objective	Expected Impact	Obtained Results	Type of Connection [Number]	Type of Activity [Number]	Institution Name	Institution City, Region & Country	Agent Type [Number]	Economic Sector
Advising Services for the Municipality of Valparaíso on the Master Plan for Heritage.	Local development of heritage site	Management plan for heritage site developed	2	1	Municipality	Valparaíso, Chile	2	Government
Cooperation Agreement with the Parque Cultural de Valparaíso	Making international neuroscience congress in Valparaiso	Used for 2 Symposia and reserved for an international congress in 2013	2	5	Parque Cultural de Valparaíso	Valparaíso, Chile	2	Culture and Government
Severin building a new house for CINV	Constructing the new building of CINV in heritage site of Valparaiso	Architectural Design	1	2	Ministerio de Obras Públicas, Gobierno Regional de Valparaíso	Valparaíso, Chile	2	Government

9.1 Total incomes:

Funds	Accumulated incomes to last year [$]	2012 Incomes		Total incomes to 2012 [$]
		Amount [$]	Percentage of resources used by the Center [%]	
MSI (CINV, REDES, PME)	1,013,771.00	987,186,997	100	987,186,997
UV (DEPARTAMENT,CID)	320,000.00	335,000,000	100	335,000,000
CORPORATION	1,000.00	2,500,000	100	2,500,000
CONICYT (FONDECYT, ANILLO, ANR, MINCYT)	576,009.00	421,290,000	100	421,290,000
OTHER GRANTS (MECESUP,FIRCA, COPEC)	13,626.00	6,000,000	78	6,000,000
OTHER GOVERNMENT AGENCIES (FNDR, CNTV, CONGRESS)	94,961.00	2,500,000	100	2,500,000
PRIVATE CONTRIBUTIONS NATIONALS (ARQUIMED)	3,922.00	10,000,000	100	10,000,000
PRIVATE CONTRIBUTIONS FOREIGN (LAB.PASTEUR, OTROS)	000.00	7,447,473	100	7,447,473
TOTAL	2,023,289.00	1,771,924,470		1,771,924,470

9.2 Outcome structure

ITEM	Accumulated expenses to last year [$]	2012 Expenses [$]				Total expenses to 2012 [$]	%
		Operative	Networking	Outreach	**Total**		
Honoraria Researchers	50.860.000	144.689.930			144.689.930	**144.689.930**	**18**
Honoraria students and other personnel	40.387.558	189.554.720			189.554.720	**189.554.720**	**24**
Tickets and travel expenses	14.286.719	35.966.527	2.000.000		37.966.527	**37.966.527**	**5**
Materials/supplies	10.541.420	46.336.399			46.336.399	**46.336.399**	**6**
Goods and equipment	51.262.838	273.285.885			273.285.885	**273.285.885**	**34**
Infrastructure	5.980.940	5.889.742			5.889.742	**5.889.742**	**1**
Administrative expenses	17.140.120	50.187.119			50.187.119	**50.187.119**	**6**
Publications and subscriptions	1.520.096	3.004.803			3.004.803	**3.004.803**	**0**
Consultancies	3.180.002	23.062.061			23.062.061	**23.062.061**	**3**
Overhead	0	21.600.000			21.600.000	**21.600.000**	**3**
Insurance costs	0	882.213			882.213	**882.213**	**0**
Legal expenses	0	0			0	**0**	**0**
Others	0	986.208			986.208	**986.208**	**0**
Total Expenses ($)	**195.159.693**	**795.445.607**	**2.000.000**		**797.445.607**	**797.445.607**	**100**

9.3 Financial accounting

ITEM	2012 [$]			
	Operative	Networking	Outreach	**Total [$]**
Income	918,765,000	28,030,220	40,391,777	**987,186,997**
Outcome	795,445,607	2,000,000	0	**797,445,607**
Annual balance	123,319,393	26,030,220	40,391,490	**189,741,390**