

Evaluation of FONDAP Center for Cell Regulation and Pathology "Joaquin V. Luco" (CRCP)

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Center progress and achievements

The Center has seven principal investigators, and one young investigator. The young investigator and two of the principal investigators were recruited during the first 5 year funding period of the Center. All of the investigators have active and productive research programs of a quality that would be deemed suitable at top research institutions internationally. The Center scientists are actively engaged in disseminating the knowledge obtained from their research, by publication in international journals, by presenting research seminars in Chile and internationally, and by oral and poster presentations at international scientific meetings.

The investigators are consistently publishing in international peer-reviewed journals, including a significant number of publications in highly competitive top-tier journals such as Nature Cell Biology and Journal of Biological Chemistry. The rate of generation of published research papers by investigators in the Center has remained more or less constant during the years the Center has existed. 20 publications resulted from Center-funded activities in 2006. A few of the recent publications seem to have only a tenuous relationship to the central themes of the Center. It does not seem likely that this will be a continuing problem, however, particularly because Dr. Inestrosa recently has markedly narrowed the focus of his investigations, eliminating several aims that were peripheral to his primary interests. Considering the number and quality of the publications as a whole, in relationship to the overall level of funding of the Center, the level of productivity of the Center has been excellent. However, as the senior investigators were similarly productive before the conception of the Center, it is not clear that the existence of the Center has notably increased the productivity of its members.

Of particular importance, the three recently recruited more junior investigators have been successful in establishing productive independent research programs. Each has published in 2006 – indeed several of the most important recent publications from the Center derive from the work of these recently recruited members. This engenders optimism for the continued productivity of the Center. The Center wisely recruited new investigators with research programs that integrate nicely into the Center's thematic focus, and these investigators potentially provide bridges that may foster collaborative interactions among Center members.

A recent annual review of the Center noted some concern that Dr. Inestrosa's growing emphasis on studies of function of neuronal synapses had an element of risk because of his lack of previous experience in this area but his recent progress shows that this is no longer an issue. The Center has a growing emphasis in the area of developmental biology. This represents the major thrust of Dr. Larrain's research but several other

investigators have projects with developmental themes also. The Center's role in establishing a facility for *Caenorabditis elegans* research also is significant in this context, as *C. elegans* is a premier model organism in developmental biology.

The CRCP has been very successful in having national and international collaborations, they have several manuscripts with international collaborators in topics such as:

- a) morphogens, new passengers on lipoprotein particles,
- b) endosomal transport of neurotrophins,
- c) wnt7, and nicotinic acetylcholine receptors,
- d) APB1 recycling endosomes, and APB1 controls biosynthetic,
- e) PPPSP motif determines megalin's phosphorylation and,
- f) e) p75 neurotrophin.

Examples of these international collaborators are Dr. Hengartner, in Zurich Switzerland, Dr. Rebeca Kohn from the Ursinus College in PA will join the Center to participate in a course about regulation of Nervous System Function using *C. elegans* as a model. There is also collaborations with Dr. Roberto Mayor, from University College London, Dr. Bu in Washington University, Dr. Taylor in University of La Jolla, Dr. Post from University of Holland, Dr. Stojilkovic from NIH and Dr. Ventura from Barcelona.

The Center is organizing also several international courses and meetings that are a reflect of their international and national collaborations. The investigators at the Center are very well known in the country as well as in other countries and this has been very positive for the students who have opportunities to visit other laboratories.

The Center contributes in several important ways to developing the research infrastructure of Chile. The value of providing advanced research training opportunities for students is obvious. Importantly, the Center provides the opportunity for interaction and networking of students and established investigators with scientists in other countries, both by promoting travel of Chilean scientists to international meetings, and by promoting travel to Chile of leading scientists from other countries. Students and investigators in the Center place a high value on such opportunities. Finally, the publications and the presentations at international meetings by investigators from the Center increases the international visibility and reputation of Chilean science.

The students in general are very well integrated in the academic life of the Center since it has provided them with an adequate environment to do science. They have very good collaboration interchanging space in the laboratory, reagents, protocols as well as discussion for their projects. The CRCP has opened the opportunity for them to attend foreign laboratories in different countries; also the students and postdocs are able to attend national and international scientific meetings. The fact that the students from CRCP have an adequate economical support for their projects has made a difference when compared with students from the same program that are not in the Center.

The students expressed the desire to have more and better forums, either as seminars or meetings, in which to discuss their research in a more formal way. Also a common space like a coffee room would provide the students a better environment to discuss their science informally.

It will be positive for the PhD students to be more involved in the writing of the papers since this is a very important part of their formation.

The outreach activities of the Center for this period of time were:

- 1) Chilean International Symposium on Lipoprotein receptors: from Cell biology to disease,
- 2) Theoretical and practical course on confocal microscopy,
- 3) Academia program for outstanding students from public schools, (PENTA) from the University and teacher actualization in concert with the Educational Reform,
- 4) Millenium conferences,
- 5) Workshop of young scientists of the Millenium Institute Initiative,
- 6) a book on Alzheimer's for the lay people (in Spanish),
- 7) brochure of the Center,
- 8) Seminars.

These activities have been very important for the contact of the Center with outside, not only in the scientific environment but also with younger people not involved in science or not specialists, an example of this is the book on Alzheimer disease in Spanish being authored by Dr. Inestrosa. We encourage the PIs to be more involved in these activities since is part of the initial goal of the Center.

The annual meeting of the Center should be kept as one of the most important activities of the Center. This provides an informal environment to share experiences and discuss the projects among the members of the Center.

Value added by the CENTER

Although one of the major impacts of the Center is in the students and the collaboration that exists among them, this has not been reflected in joint publications among the investigators. It is convenient to notice that the goal of the Center is to add value to the individuals as a group.

Some of the present collaborations the programs have are:

- a) Heparin, wnt signaling and Ab neurotoxicity (Dr. Inestrosa, Dr. Brandan, Dr. Marzolo),
- b) Nucleotide P2Y1 receptor regulates EGF (Dr. Huidobro, Dr. Gonzalez)
- c) Basolateral sorting of LRP in the recycling pathways (Dr. Marzolo, Dr. Gonzalez)
- d) PPAR induces expression of the Bcl-2 (Dr. Brandan, Dr. Inestrosa)
- e) Neurotrophin, cholinergic neuron (Dr. Brandan, Dr. Inestrosa)

These collaborations are reflected in papers that will be submitted in the near future. It is expected that these manuscripts will be published, and this is going to be a very good element to evaluate collaboration among the programs. It is noticeable that the researchers are making an effort to increase the collaboration among the programs, and this is evident with the environment with the students, as well as the sharing of reagents, interchange of ideas, equipment, etc. However, after seven years of the creation of the Center the collaboration among the programs should be more tangible. Several investigators, notably A. Gonzalez, do not attach great intrinsic value to collaborative research. They reasonably assert that the environment of the Center provides an important mechanism for cross-fertilization of ideas among investigators that is difficult to quantify and does not necessarily lead to publications coauthored by several investigators. Senior investigators also note that it is important for the career development of more junior members that they can demonstrate their intellectual independence by publishing papers that do not include senior investigators as coauthors.

The youngest researchers, Drs. Marzolo, Larrain, and Bronfman are making a very good effort in sharing experiences and having collaborations with the seniors PIs. This should be reflected in the future in publications, thesis, joint projects, etc.

Management

Dr. Inestrosa has provided strong leadership. The investigators indicate satisfaction with their ability to contribute to the decision-making process. A previous review noted insufficient opportunity for junior members to participate in the decision-making process but this problem appears to have been rectified. Dr. Inestrosa, and the deputy Director, Dr. Brandon, lead by example as the most prolifically publishing investigators in the Center.

Recruitment

The recruitment of post docs and students has been very successfully. All the researchers have students and most of them have post docs, and the numbers are very close to which was in the original plans. The involvement of the students in the Center is outstanding, this is probable the most tangible intra-pro-am collaboration activity that results from the Center. The postdocs and Ph D students have the same opinion about the role of the Center in their performance. With the increase in the number of students, the center members need to make an effort to regulate the number of students per investigator according to their space resources as well as the amount of quality time they have for the students. Also the Ph D students need to participate more actively in the preparation of the manuscripts since some investigators do not have this discipline. In order to have a larger number of postdocs they also should make an effort in advertise the positions outside Chile.

Institutional commitments

The support of the University could be significantly improved in several important ways. This is a point that needs future attention. The Center significantly enhances the status of the University. The University should be made aware of the needs of the Center for a common space for interaction of the students and researchers. This present lack was found to be a serious impediment to effective interactions among Center members, as reported by students as well as senior investigators. Also it is essential for the future development of Center and its members to have the laboratories of Dr. Francisca Bronfman and Dr. Juan Larrain closer to the Center to allow more effective interactions of the Center members.

Advisory Committee

The scientific advisory board presently is composed of four highly respected senior investigators – Dr. David Carey, Director of the Geisinger Clinic, USA; Dr. Francisco Barrantes, Director of the Institute of Biochemistry, Argentina; Dr. Jose Boyer, Senior Director of Molecular Pharmacology, Inspire pharmaceuticals, USA, and Dr. Enrique Rodriguez-Boulan, Director of the Dyson Vision Research Institute, Weill Medical college of Cornell University, USA. They all have strong scientific expertise in the particular fields emphasized in the Center. They have contributed energetically to the guidance of the Center, providing strongly worded recommendations that have significantly impacted decisions made within the Center. Unfortunately logistical problems forced rescheduling to January 2007 of an advisory board meeting originally scheduled for October 2006. Thus, the review committee did not have the benefit of a recent report from the advisory board but their January meeting will certainly be an important event for the Center.

The investigators of the Center have been very receptive in accommodate the concerns of the international advisory committee. They incorporated Drs. Marzolo and Larrain as a senior investigators and Dr. Bronfman as a junior investigator, and allowed them to participate formally in the decisions of the Center in response to the recommendations of the committee. Also, because of the suggestions of the advisory committee, Dr. Francisca Bronfman has been working on finishing the publications that will be published next year, she has now one paper in press, another submitted, and one more in preparation. The research line of Dr. Inestrosa has been focused now in the Project program of the Center, leaving topics such is the prion research that make his program broad and diffuse. The recommendations of the advisory committee concerning the importance of improving collaborations in the Center and crystallizing them in publications have not yet been sufficiently implemented, however.

Evaluation of Program Projects

PP1 Intracellular protein traffic: molecular mechanisms, functional implications and disease (Dr. Alfonso González:

This has been one of the more creative and successful programs in the Center. The program continues to do excellent work, but has not been particularly successful in

bringing work to the stage of publication in the present funding period. In years 2005 and 2006, Dr. González has contributed to two published review articles and has 4 manuscripts in preparation. Several original research papers have been submitted, but none have been published at present, but research progress has been described at several national and international scientific meetings. The major focus of this program is characterizing mechanisms responsible for determining sorting to basolateral versus apical surfaces of polarized epithelial cells. One major emphasis concerns characterizing the role of AP1B and similar cytoplasmic adapter proteins in determining the sorting behavior of basolateral proteins in epithelial cells. In this work, Dr. González frequently collaborates with Dr. Rodriguez-Boulan, a recognized world leader in this subject. This is an important area of research and Dr. González has made good progress. Another project characterizes cytoplasmic sorting signals of the scavenger receptor SR-B1. Other projects characterize mechanisms governing sorting of GPI-linked membrane proteins, and of the EGF receptor. These are all important questions and the González lab attacks them with great expertise. In the realm of publication, it is the style of this laboratory to publish infrequently, but to publish work of the highest quality, and in the best journals. Dr. González has been very successful in attracting graduate students to his research program and appears to be giving them excellent research training. However, it was our perception that his perfectionism leads him to involve his students insufficiently in the process of writing papers, depriving them of this important training opportunity. Dr. González does not appear to have fully embraced the notion that collaborative research is a major goal of the center – thus, although he has engaged in several collaborations with other center investigators, his activity in this realm is rather limited.

PP2 Function of proteoglycans in myogenesis and fibrosis (Dr. Enrique Brandan:

The recent productivity of Dr. Brandan has always been excellent; he has published 6 papers in 2006, including several in top-tier journals and has several other manuscripts under review. He also has been quite active in collaboration with other members of the Center. He is a leading investigator studying the role of TGF-beta in regulating myogenesis and fibrosis and the role of proteoglycans in modulating TGF-beta activity in this process. He has engaged in collaborative work with Dr. Marzolo, leading to publication of a very important paper concerning the function of the low density lipoprotein receptor-related protein as an endocytic receptor for decorin. He also has collaborated with Dr. Larrain leading to an important publication concerning the function of Biglycan is a component of the Chordin-BMP4 signaling pathway. Thus, his activities have been exemplary in scientific importance and in regard to the goals of the center. Dr. Brandam besides his own research is the vice-president of the Center and a very important support for the rest of the members.

PP3 Role of plasma membrane nucleotide receptors (DR. Juan Pablo Huidobro-Toro:)

Dr. Huidobro-Toro's work on purinergic receptors continues to be extremely productive. He has published 8 papers in international journals in the last 2 years. A major emphasis has been the role of regulation ATP released from sympathetic nerve terminals on

activation of purinergic receptors on vascular cells. Another interesting line of research concerns transactivation of EGF receptors by purinergic receptors. This work represents a collaboration with Dr. González. He has several PhD students.

PP4 Role of PPARs in neural function. Identity of PPARs physiological ligands (Dr. Miguel Bronfman):

The limited productivity of Dr. Bronfman in recent years has been a subject of concern. Although this concern was reduced somewhat by Dr. Bronfman's publication of two important papers at the beginning of 2005, the productivity of his research program continues to be somewhat slow. One major goal of his program, the identification of physiological ligands for PPAR receptors, has had only limited success. A major goal presently concerns examination of the role of PPARs in oligodendrocyte differentiation. These studies are potentially important and some progress has been made. A serious limitation of the research approach is that it relies heavily on use of the C6 glioma cell line. Use of more physiologically normal cultured glia systems will be required to allow publication in top-tier journals. A collaboration with Dr. Inestrosa has led to an important publication concerning the involvement of PPARs in limiting neurotoxic action of amyloid beta peptide. Another collaboration, with Dr. F. Bronfman, concerns the possible functional significance of association of PPARgamma with neurofilaments. This project, which is at an early stage of development, is potentially ground-breaking, but risky.

PP5 Role of the Wnt signaling pathway in synaptic function and neurodegenerative diseases (Dr. Nibaldo C. Inestrosa):

Dr. Inestrosa continues to direct a very productive research program, in addition to acting as an effective leader of the Center. We also were impressed with his success in mentoring graduate students. Dr. Inestrosa's usual outstanding pace of publication has diminished slightly in the last year or two, but is still substantial, and he has a number of research projects at an advanced stage of progress. His greatest productivity of publications recently has concerned the relationship of copper to prion disease, a subject that is not directly related to his research goals within the center.

His work on the synaptic function of Wnt proteins, and their relevance for mechanisms of Alzheimer's disease, is of high quality and great importance. Although Dr. Inestrosa does not have a substantial background in the area of study of synaptic function, he has demonstrated that he can work very productively in this area.

PP6 Early development of the Xenopus embryo (Dr. Juan Larraín):

Dr. Larraín is a junior investigator with exceptional promise. He has quickly established himself as a leading investigator of the role of proteoglycans in gastrulation. His research interests overlap in a very synergistic way with the interests of Dr. Brandan concerning proteoglycan function, and the interests of Dr. Inestrosa concerning Wnt function. Dr. Larraín's recent work led to a very important publication in a premier journal (Nature Cell Biology) concerning the role of syndecan-4 in modulating Wnt

function during gastrulation. Dr. Larrain will undoubtedly be a strong positive force in the development of the Center.

PP7 Trafficking, function and regulation of the low-density lipoprotein receptor-family members (Dr. María Paz Marzolo):

Dr. Marzolo, as a junior investigator, has had excellent success in establishing her independent research program while engaging productively in collaborative work with other members of the Center including Dr. Brandon, and Dr. Inestrosa, with whom she recently has published important papers. Her research interests concern the trafficking, processing, and signaling functions of members of the LDL receptor family. This subject provides excellent opportunities for collaborative interaction with Dr. Inestrosa concerning the role of LDLRs such as LRP in Alzheimer's disease, with Dr. Brandon concerning the interaction of LRPO with decorin in modulation of TGF-beta signaling, and with Dr. Gonzalez concerning mechanisms regulating receptor trafficking. Her continued success is key to the future of the Center and the resources provided to her by the center should reflect this.

PP8 Neurotrophin signaling: cell biology, proteolytic processing, and role in central nervous system repair and disease (Dr. Francisca Bronfman):

As the most junior member of the Center, Dr. F. Bronfman's research program is just beginning to become productive. She has authored one review paper which presently is in press. She has made substantial progress toward characterizing the mechanism of proteolytic processing of the p75 neurotrophin receptor, leading to a manuscript recently submitted for publication, and this line of investigation will continue to be a major emphasis. Two more recently initiated lines of investigation, characterizing the effects of axotomy on cholinergic phenotype of basal forebrain cholinergic neurons and assessing the effect of Niemann-Pick disease on neurotrophin signaling, are less well developed. The scope of the progress report and Dr. Bronfman's short presentation during the site visit did not allow the rationale for these studies to be fully explained, so this reviewer does not have a firm opinion about the likelihood for success. These projects do present good opportunities for fertile collaborative interactions with other Center investigators, however.