UNIVERSITY OF PENNSYLVANIA MEDICAL CENTER

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Biocomplexity Faculty Search Committee
c/o Prof. Rob de Ruyter van Steveninck
Biocomplexity Institute
Indiana University
Swain Hall West 117
Bloomington IN, 47405-

Dear Prof. van Steveninck;

It is a pleasure for me to write to recommend Dr. Martin Muschol for an Interdisciplinary Systems Biology faculty position in the Biocomplexity Institute at Indiana University. I have known Martin for seven years, since he joined my laboratory in the Department of Neuroscience at the University of Pennsylvania. Martin's initial appointment was as a Postdoctoral Fellow, but, in June of 2000 I strongly urged his promotion to Research Assistant Professor. In 1996, I selected Dr. Muschol from among a very strong pool of applicants because of his excellent background in optical physics, his obvious intellectual gifts, and recommendations from his previous mentors. He, in turn, was attracted by the prospect of applying his skills in optical physics to neurobiology in a laboratory that had pioneered the use of voltage-sensitive dyes and light scattering measurements from nerve terminals. Muschol rapidly acquired and mastered both the experimental techniques and theoretical concepts involved in this completely new field. His accomplishments over the last few years have been outstanding and indicate a clear and focused mind.

I supervise a very small laboratory with individual members each dedicated to a specific research project. It is therefore easy for me to assert that Martin has assumed complete responsibility for structuring and carrying out his research during his time in my lab; he is single-minded in his pursuit of intellectually satisfying experimental results and, increasingly over the last few years, I value his scientific judgment concerning our optical experiments above that of virtually any other colleague! Muschol's efforts have resulted in ten abstracts presented at national meetings and in three highly significant first author publications in the Biophysical Journal and the Journal of Neuroscience. (I can also assert that Dr. Muschol's first author status was entirely justified by his contribution, and in no way reflects any "courtesy".) While this published output is relatively modest, I believe that the quality and importance of the papers is exceptional. (Martin Muschol did, after all, arrive in my laboratory with only the most rudimentary knowledge of Neuroscience, and no formal training in Cell Biology. It seems fair to note that his learning curve was spectacular!)

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Dr. Muschol began by working on the rapid intrinsic optical changes that occur during stimulated release of peptide hormones in the intact neurohypophysis. He became convinced, correctly, I believe, that an optical analysis was unlikely to reveal the physical origin of the optical changes because of their domination by multiple scattering. Although we agreed that this argument does not bear on efforts to identify the physiological origin of the signals, Martin decided that he would try to approach the problem by studying the dynamics of intraterminal calcium, which is closely related to the intrinsic optical signal. Martin Muschol was the first in my laboratory to make millisecond time-resolved (or, for that matter any) measurements of calcium indicator dye fluorescence, and his data are among the very best I have seen anywhere. Strong effects of caffeine on the calcium transients were consistent with a role for calcium-induced calcium release within secretory terminals (suggested by other data from the lab). However, Martin was concerned with inconsistencies in the fluorescent indicator dye calcium data, and, as a result of a series of very elegant experiments, he showed that the bulk of the caffeine effect on fluorescence intensity had its origin in a *direct hydrophobic interaction* of caffeine with several of the most commonly used dye fluorophores (Biophys. J, 1999). This important paper has potentially very troubling implications: many reports of caffeine-induced depletion of calcium-stores in other systems need to be reexamined, and other pharmacological agents used at high concentration must have their direct effects on indicator dye fluorophores controlled.

It has long been recognized that peptide release from the neurohypophysis is highly sensitive to the pattern of action potential stimulation. Martin showed that stimulus-induced calcium transients themselves were strongly modulated during repetitive stimulation. These data provided, for the first time, a convincing basis for explaining both increases *and* decreases in release, depending on the pattern of stimulation (J. Neurosci, 2000). Recording the transmembrane voltage with fast potentiometric dyes and a high-speed camera system, we found that regulation of tissue excitability itself plays a critical role in shaping the frequency-dependence of calcium transient. During the course of these experiments, Martin suggested an elegant model that was able to resolve several confounding features of the observed action-potential modulation (J. Neurosci, 2003). In this model, which we termed "stuttering conduction", the nerve terminals and numerous in-line varicosities become refractory to stimulation while the axons themselves continue to support action-potential propagation. This model implies a novel mechanism for generating presynaptic depression and has potentially significant implications for our understanding of presynaptic plasticity.

In 2000 Martin received an NIH funded research training award (K25). His proposal received the highest score during his review cycle and was the only application funded. As a result, I persuaded the Department of Neuroscience here at the University of Pennsylvania to promote Dr. Muschol to the Research Faculty as an Assistant Professor.. Martin has utilized his training grant to widen the range of experimental techniques at his disposal, to help him develop an independent research project, and to participate in national meetings and Gordon conferences on synaptic transmission. He arranged a visit at the laboratory of my colleague Edward Stuenkel

at the University of Michigan, in order to learn how to prepare isolated nerve terminals from the neurohypophysis. He has attended the electrophysiology course at Cold Spring Harbor in order to acquire patch-clamping techniques. He is currently involved in experiments to collect preliminary data on near-membrane measurements of calcium and hormone release in preparation for an RO1 grant application. For this purpose he has purchased significant equipment for patch-clamping, flash photolysis and a research-grade solid state laser system, all of which should follow him to a new position.

I have had the opportunity to observe Martin during oral presentations at several national meetings and at a departmental seminar in the Physiology Department at the University of Pennsylvania. Martin is an excellent speaker who gives clear and well structured presentations. Based on his presentation at the 2003 Biophysical Society meeting, he was invited to give a keynote lecture about pituitary physiology at the 2004 meeting of the Society for Endocrinology, Neurobiology and Physiology in Holland. I am, therefore, confident that Martin is an effective and skillful lecturer and instructor.

On a personal note, I found Martin a pleasant and knowledgeable discussion partner. He has consistently provided critical and constructive feedback on all aspect of our research on optical signals. He interacted well with the students under his supervision and has taken on the task of training our new research associate - another physicist without background in neuroscience. Martin is well liked by members of the faculty and has been diligent about attending departmental seminars and faculty meetings.

In summary, I would rate Dr. Muschol's intellectual and research abilities among the top 2-3 of all the junior researchers with whom I have collaborated over the years. Martin has already proven to be an imaginative, productive and enthusiastic scientist with great promise for the future. He also possesses many of the intangibles that I regard as predictors of success in science. He takes great pride in the quality of his data. He is unflinching in his willingness to try new techniques if these might advance the research program. He is critical without being negative. Finally, he has established that he can secure funding for his research ideas and he has shown that he interacts well with his colleagues and students in the laboratory. I am convinced he would make an excellent addition to any department and I recommend him to you for an Interdisciplinary Systems Biology faculty position in the Biocomplexity Institute at IU, with my highest enthusiasm.

If I can be of any further assistance, please do not hesitate to call me.

Sincerely yours,

Brian M. Salzberg, Ph.D.
Professor of Neuroscience
& Physiology