



September 30, 2005

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Dr. Yves Brun
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Dear Dr. Brun:

I am happy to write in support of the application of David R. Shook for the position in your Department. I have been aware of Dave's research for a number of years, and we presently have a collaboration on gastrulation in a direct developing frog. Dave is one of the finest scholars of embryonic cell movements, and his ability to capture cellular details of these movements is remarkable. Check out his movies posted on the Web.

I am particularly happy that Dave wants to apply his talents to examine movements comparatively. He is well versed in the cellular activities during gastrulation and early development of *Xenopus laevis*. Much of this information comes from Keller's lab, where Dave has been working, and Dave has contributed to that body of knowledge. He has worked with embryos of other amphibians, such as *Ambystoma mexicanum*, so he occupies a unique niche – a scientist interested in comparative analysis who has a refined eye for cell behavior. He is the only one that I know with this expertise and interest. There are many people considering genes and gene expression from an evolutionary and developmental perspective. The consideration of cell behaviors is far more difficult, but Dave's research shows that it should be possible.

Dave does not publish frequently, but the longer length of his papers indicates a desire to present a complete story. In this way, Dave is more similar to the investigators of previous eras, and I expect that Dave would love to talk to Holtfreter. Actually, Dave's mentor, Ray Keller, published very few papers, early in his career, so perhaps Dave is emulating that model. He always has plenty of data to talk about.

I recommend Dave strongly to you. He provides a divergent perspective that has the potential to bear novel fruit.

Sincerely,

Richard P. Elinson
Professor



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September 21, 2005

Yves Brun
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RE: Reference letter for David Shook

Dear Dr. Brun,

I write in support of David Shook's application for a faculty position in your Department. David Shook was an excellent student. He was in my laboratory for only a little over two years and completed a project that has been highly cited. However, first let me describe David's previous work, done before entering my lab.

David's Master's thesis work was on the role of intermediate filaments during gastrulation in the frog *Xenopus laevis*. This work was done in the Department of Molecular, Cellular and Developmental Biology, at the University of Colorado, Boulder, with Dr. Michael Klymkowsky. This work involved testing the effect of anti-intermediate filament antibodies when injected into *Xenopus* zygotes; embryos were allowed to develop to the late gastrula stage and assayed for defects in gastrulation. This work was published as Klymkowsky, M.W., D.R. Shook, and L.A. Maynell, 1992, *PNAS* 89: 8736-8740. It provided evidence that the deep keratin filament systems of the *Xenopus* embryo act to ensure successful gastrulation.

Having become interested in evolutionary questions (David was the chairman of the organizational committee of 1992 MCD Biology Graduate Student Symposium, *Evolution from the Inside: The Influence of Cellular and Developmental Processes on Evolution*, Keynote Speaker: Stephen Jay Gould. This was a three-day symposium organized and funded entirely by graduate students), he chose a project of a more evolutionary nature for his Ph.D. thesis work. David joined my lab here in the Institute for Behavioral Genetics in 1996.

His Ph.D. thesis work took place in my lab and was on the evolution and quantitative genetics of life history traits in the nematode worm, *Caenorhabditis elegans*. His Ph.D. work was performed under the auspices of the Department of Evolutionary, Population and Organismic Biology, at U.C. Boulder. He was identifying quantitative trait loci (QTL's) specifying life traits, using recombinant inbred (RI) strains. The initial data was collected by an earlier student and David collected additional data to fill in the missing data for some RI genotypes. David completed a statistical analysis of this data, including distribution statistics, analysis of components of variance due to replicates, sets, and strains (heritability), a single marker QTL mapping analysis, an examination of the resulting putative QTLs for pleiotropies, and a thorough analysis of interactions among markers to detect putative epistatic QTLs, which are not detectable by single marker analysis. These results were then written up and are in *Genetics* (Shook, Brooks and Johnson, Mapping quantitative trait loci affecting life history traits in the nematode *Caenorhabditis elegans*. *Genetics* 142:801-817). This paper considers the quantitative genetic

implications of the data as well. One of the reviewers commented, "This will be a landmark paper in the analysis of life history traits."

The second part of David's thesis utilized phenotypic data collected himself, occasionally with the help of an undergraduate worker. This work was based on the premise that *C. elegans* are likely to be a highly r-selected species, in which case life history traits such as age at onset of sexual maturity, early fertility, and intrinsic rate of increase should be strongly selected, and might therefore be likely to show a trade off with life span. An example of such a trade off would support a popular current theory for the evolution of aging. David designed an assay that could accurately assess these early life history traits, which involved determining the mean number of progeny produced by each strain. Further analysis of this data included the calculation of several demographic parameters, including, α , the age of sexual maturity and r , the intrinsic rate of population increase. He also designed and carried out survival experiments on agar plates; previous experiments had been done in liquid media, and other results had suggested that there were strong genetic by environment interactions in *C. elegans*, especially between these two environments. In addition, David achieved better coverage of the genetic map by genotyping the RIs for more Tc1-based molecular markers. All this data has been combined in a further QTL analysis of previous and new life history traits, including the calculated demographic parameters, based on the expanded genotypic data for the RIs. This latest work was also published in the premier journal in genetics (*Genetics* 1999, **153**: 1233-1243).

David had completely retrained himself in evolutionary theory and became an expert statistician; he did this on his own and is very good at working by himself with almost no supervision. He loves to design new experiments and is a very careful worker. He is among the best of ten Ph.D. students I have had in my lab. I would certainly hire David in the future and did offer him a postdoctoral spot, which he wisely declined to pursue his own interests. David is certainly deserving of an interview and I am sure that you will find him a lively, intelligent member of your faculty and an excellent colleague and teacher.

Sincerely,



Thomas E. Johnson
Professor of Molecular and Behavioral Genetics

September 26, 2005

Yves Brun
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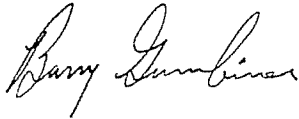
Dear Dr. Brun:

I am pleased to write a letter of reference for Dr. David Shook, who is applying for a position of Assistant Professor in the Department of Biology at Indiana University. I have known Dr. Shook for only the past year or so since I moved to the University of Virginia, mostly through periodic joint group meetings with Ray Keller's laboratory. I have not worked closely enough to him to be able to comment on his experimental skills or work habits, but I can give general comments of my impressions of him.

Dr. Shook is a bright young scientist who has developed a very strong understanding of cell behavioral and mechanical aspects of morphogenetic movements of gastrulation. He has taken a very broad approach, relying on comparisons of cell behaviors in different, but related species. This has provided insights into the generalities and variations underlying morphogenetic movements that would not be apparent from the more typical emphasis on *Xenopus laevis* or, more recently, Zebrafish. It also provides a potential approach to gaining insights into the evolutionary basis of morphogenetic processes. Dr. Shook has not yet developed molecular or genetic approaches to studying the mechanistic basis for these observed differences in cell behaviors. This is due, at least in part, to a trade-off between his broader approach to understanding the cellular basis of morphogenetic movements and a more focused approach to get at mechanisms. If he is able to develop tools and strategies for studying molecular mechanisms in these experimental models, he will have a unique handle on understanding morphogenesis. On the other hand, his approach to comparative analysis should provide important information about the range of ways in which different cell behaviors can produce different tissue outcomes.

I should also add that Dr. Shook is a highly interactive young scientist who contributes significantly to the intellectual environment. He is very curious about all aspects of problems that we discuss in our joint meetings, asks lots of questions, and provides many suggestions for experiments. Furthermore, he is the person who has committed much of the energy and effort required to initiate and organize these joint group meetings. I think that he would be a very good colleague to have on one's faculty.

Sincerely,

A handwritten signature in cursive script that reads "Barry Gumbiner".

Barry M. Gumbiner, Ph.D.
Chair and Professor
Department of Cell Biology

September 27, 2005

Yves Brun
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Dear Yves Brun,

It is a pleasure to write this letter of recommendation on behalf of **Dr. David Shook**, who has applied for a junior faculty position in your department. I have known Dr. Shook since 1996 when he came to the University of Virginia to join the Keller lab. My research interests parallel closely those of Dr. Keller's and, thus, we maintain close ties. It is primarily through regular group meetings with the Keller laboratory that I have become familiar with Dr. Shook and his work.

I will leave a more detailed description of Dr. Shook's recent research accomplishments to Dr. Keller but in brief, he has taken an interesting approach to analyzing the fundamental cell behaviors that drive early morphogenetic processes in amphibia. His primary approach is a comparative one, and involves an in depth examination of mesendodermal movements across phylogenetic boundaries. Among his more recent contributions has been the identification of "subduction" processes that drive superficial mesoderm inward at gastrulation in amphibia. His work has considerable practical importance to many investigators wishing to understand differences in morphogenic cell behaviors among *Xenopus* species (i.e., *laevis* and *tropicalis*). It also has important implications for identifying conserved biomechanical movements that underlie morphogenesis in chordates.

Dave has a varied background with experience in cell biology, developmental genetics and, more recently, "traditional" embryology. He has been a member of the Keller lab for some time now, and while one would ideally like to see greater evidence of productivity (in terms of published data – although recently he has improved this as well), I am quick to add that the kind of work that Dave has been doing is difficult, time consuming but also very important. What I admire most about his work, however, is the level of scholarship required to address such fundamental questions, in addition to the melding of his now prodigious embryologic skills with more molecular and biomechanical approaches. This is an important combination of strengths that will serve him well as he sets up his own laboratory. Dave is likely to follow "his nose" and study the biological problems of greatest interest to him even though this might not always be the best strategy for attracting maximal grant dollars. Frankly, I admire this in him but realize that this combination of talents and interests would likely not serve him well in the most

fast-paced competitive environments where NIH funded biomedical research is the single most important metric of success (e.g., most med schools). I think his particular combination of interests, talents and skills would be put to best use at an institution where quality undergraduate education is stressed. I hasten to add, however, that Dave is in an excellent position to build an independent, extramurally-funded research program. I certainly am not ruling out that he will attract NIH funding, to the contrary, but I think some of his best opportunities for funding in the immediate future will also be through the NSF. Given his interest in comparative embryology and evolution, however, I see tremendous opportunities unfolding for his work as the “post-genomic” era progresses (i.e., he is *ahead of his time* in many ways).

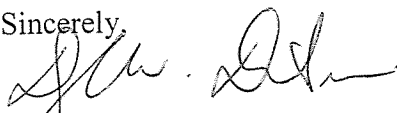
I have every confidence that Dave will be an outstanding colleague and teacher. Although the Keller and DeSimone labs have considerable overlapping interests, I daresay we would not have the rewarding level of interactions we now enjoy were it not for Dave. He has been the key player that organizes - and occasionally cajoles - us to make the effort to participate fully in joint lab meetings and other more formal venues. Most significantly, Dave took it upon himself a couple of years ago to organize the “Morphogenesis Supergroup” at the University of Virginia that brings together strengths (>10 laboratories) from across grounds for what has become an exciting monthly series of evening research talks. He stressed the importance of “chalk talk” style presentations and this format has proven highly successful and very enjoyable. At a time when e-mail and large group sizes have tended to insulate investigators from interacting outside of their laboratories, the Morph. Supergroup has been a welcome reminder to many of us why we became academic researchers in the first place. It is his recognition of the need and importance of quality scholarly interaction that I find among his most admirable traits.

In this context, it is worth mentioning that Ray also chose Dave as his co-author on a chapter that appears in Claudio Stern’s hefty tome on Gastrulation (CSH Press, 2004). This is clearly one of the “flagship” chapters in this definitive book and I view this as an important indicator of the high regard that Ray has for Dave, his contributions at the bench, and his scholarship.

I have had many opportunities to hear Dave present his research over the years but have not had first hand exposure to his teaching. Based on his research presentations and what I’ve learned from others, however, his presentations/lectures are detailed, clear and well organized. He has sought out many opportunities to teach undergraduate students and I believe this, together with his lecturing abilities, bodes well for his future as a teacher. I think he is most comfortable with developmental biology but clearly courses in cell biology, comparative embryology and evolutionary biology are well within his grasp.

In conclusion, I believe Dr. Shook will prove an outstanding colleague, teacher and scholar. In the right academic research environment his scholarly contributions are likely to be quite substantial. I recommend him to you with very high enthusiasm.

Sincerely,



Douglas W. DeSimone, Ph.D.
Professor

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