Impact of the *Xenopus* system on the mission of the NINDS

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The mission of the NINDS is to reduce the burden of neurological disease. This mission is supported by a robust portfolio of basic research efforts aimed at understanding the structure and activities of the brain, knowledge essential for diagnosing and treating human brain disease. Some important areas of NINDS basic research include: biology of the cells of the nervous system, brain and nervous system development, genetics of the brain, cognition and behavior, neurodegeneration, brain plasticity and repair, neural signaling, learning and memory, motor control and integration, sensory function, and neural channels, synapses, and circuits.

As an overview, because of the ease with which its developing and adult nervous system can be studied, *Xenopus* has been an important model system for understanding brain function. Among the most prominent early examples of general biological insights were Gurdon's studies demonstrating that a cell nucleus from embryonic intestine could drive development of an entire embryo, using nuclear transplantation at the one-cell stage[1]. The use of informative (e.g. animal cap) assays for early tissues interactions (induction) demonstrated that this model system is an engine for gene discovery in neural development[2]. Important insights into synapse formation and refinement came from studies of neuromuscular junctions, especially those in very early development[3] and from the establishment of topographic maps in the retinotectal system[4]. Our understanding of ion channel function in the nervous system has been heavily dependent on expression in *Xenopus* oocytes[5]. Neuroendocrine discoveries included the identification and isolation of melanocyte stimulating hormone[6]. These historical strengths have been followed by a series of very important new discoveries, falling within the NINDS mission, whose insights would have been much more difficult, or impossible, to obtain with other systems. Selected examples from the recent literature (2006 – 2009) are given below.

The fundamental contributions of research in *Xenopus* is documented in the recent papers listed below that were selected to illustrate the facilitation of NINDS mission objectives through use of this model system. The topics range widely and the contributions are substantive and highly visible.

Cell biology of neurons

Agathocleous Michalis; Iordanova Ilina; Willardsen Minde I; Xue Xiao Yan; Vetter Monica L; Harris William A; Moore Kathryn B **A directional Wnt/beta-catenin-Sox2-proneural pathway regulates the transition from proliferation to differentiation in the Xenopus retina**. Development (Cambridge, England) (2009), 136(19), 3289-99.

Bollmann Johann H; Engert Florian **Subcellular topography of visually driven dendritic activity in the vertebrate visual system**. Neuron (2009), 61(6), 895-905.

W. Shen, J.S. Da Silva, H. He and Hollis T. Cline **Type A GABA-receptor-dependent synaptic transmission sculpts dendritic arbor structure in *Xenopus* tadpoles *in vivo*.** (2009) J Neurosci. 29:5032-43.

Bertolesi GE, Michaiel G, McFarlane S. **Two heparanase splicing variants with distinct properties are necessary in early Xenopus development.** J Biol Chem. (2008) 283:16004-16

Carlos Carmona-Fontaine, Helen K. Matthews, Sei Kuriyama, Mauricio Moreno, Graham A. Dunn[2], Maddy Parsons[2], Claudio D. Stern[1] and Roberto Mayor **Contact**

inhibition of locomotion *in vivo* controls neural crest directional migration. Nature (2008) 456:957-961.

Ly Alice; Nikolaev Anatoly; Suresh Geetha; Zheng Yufang; Tessier-Lavigne Marc; Stein Elke **DSCAM is a netrin receptor that collaborates with DCC in mediating turning responses to netrin - 1**. Cell (2008) 133: 1241-54.

Green Jeremy B A; Davidson Lance A **Convergent extension and the hexahedral cell**. Nature cell biology (2007) 9: 1010-5.

Mitchell Brian; Jacobs Richard; Li Julie; Chien Shu; Kintner Chris **A positive feedback mechanism governs the polarity and motion of motile cilia**. Nature (2007), 447: 97-101.

Nervous system development

Strate Ina; Min Tan H; Iliev Dobromir; Pera Edgar M **Retinol dehydrogenase 10 is a feedback regulator of retinoic acid signalling during axis formation and patterning of the central nervous system**. Development (2009) 136: 461-72.

Linda W. Chang and Nicholas C. Spitzer **Spontaneous calcium spike activity in embryonic spinal neurons is regulated by developmental expression of the Na^+, K^+-ATPase β3 subunit.** (2009) J Neurosci. 29:7877-85.

N. Bardine, C. Donow, B. Korte, A.J. Durston, W. Knöchel and S.A. Wacker **Two Hoxc6 transcripts are differentially expressed and regulate primary neurogenesis in *Xenopus laevis*.** (2009) Dev Dyn. 238:755-65.

Denver, Robert J.; Hu, Fang; Scanlan, Thomas S.; Furlow, J. David. **Thyroid hormone receptor subtype specificity for hormone-dependent neurogenesis in Xenopus laevis.** Developmental Biology (2009) 326: 155-168.

Hassenklöver T, Schwartz P, Schild D, Manzini I. **Purinergic signaling regulates cell proliferation of olfactory epithelium progenitors.** Stem Cells. (2009) 27:2022-31.

CNS genetics; *Xenopus* models of neurological disease

Tam Beatrice M; Moritz Orson L **Dark rearing rescues P23H rhodopsin -induced retinal degeneration in a transgenic Xenopus laevis model of retinitis pigmentosa: a chromophore-dependent mechanism characterized by production of N-terminally truncated mutant rhodopsin**. The Journal of Neuroscience (2007), 27(34), 9043-53

Barela Arthur J; Waddy Salina P; Lickfett Jay G; Hunter Jessica; Anido Aimee; Helmers Sandra L; Goldin Alan L; Escayg Andrew **An epilepsy mutation in the sodium channel SCN1A that decreases channel excitability**. The Journal of Neuroscience (2006), 26(10), 2714-23.

Cognition and behavior; neuroendocrine regulation

Davide Dulcis and Nicholas C. Spitzer **Illumination controls differentiation of dopamine neurons regulating behaviour.** Neuron (2009) 64, 240-250.

Hu, F., Crespi, E.J. and Denver, R.J. (2008) **Programming neuroendocrine stress axis activity by exposure to glucocorticoids during postembryonic development of the frog Xenopus laevis.** Endocrinology 149:5470-5481.

Elliott Taffeta M; Kelley Darcy B **Male discrimination of receptive and unreceptive female calls by temporal features**. The Journal of experimental biology (2007) 210: 2836-42.

Vignal C, Kelley D. **Significance of temporal and spectral acoustic cues for sexual recognition in *Xenopus laevis.*** Proc Biol Sci. (2007) 274:479-88.

Yang Eun-Jin; Nasipak Brian T; Kelley Darcy B **Direct action of gonadotropin in brain integrates behavioral and reproductive functions**. Proceedings of the

National Academy of Sciences of the United States of America (2007) 104: 2477-82.
Crespi, E.J. and Denver, R.J. **Leptin (ob gene) of the South African clawed frog *Xenopus laevis***. Proceedings of the National Academy of Sciences, USA (2006) 103:10092-10097.

Neurodegeneration

Mazabrand, A. and Pollet N. **Reduced levels of survival motor neuron protein leads to aberrant motoneuron growth in a *Xenopus* model of muscular atrophy.** Neurogenetics. (2009) Jun 11. DOI 10.1007/s10048-009-0200-6
Waters Michael F; Minassian Natali A; Stevanin Giovanni; Figueroa Karla P; Bannister John P A; Nolte Dagmar; Mock Allan F; Evidente Virgilio Gerald H; Fee Dominic B; Muller Ulrich; Durr Alexandra; Brice Alexis; Papazian Diane M; Pulst Stefan M **Mutations in voltage-gated potassium channel KCNC3 cause degenerative and developmental central nervous system phenotypes**. Nature genetics (2006), 38(4), 447-51.
Boorse, G.C., Kholdani, C.A., Seasholtz, A.F. and Denver, R.J. **Corticotropin-releasing factor is cytoprotective in *Xenopus* tadpole tail: Integration of ligand, receptor and binding protein in tail muscle cell survival**. Endocrinology (2006) 147:1498-1507.

Plasticity and repair

Bonett, R.M., Hu, F., Bagamasbad, P. and Denver, R.J. **Stressor and glucocorticoid-dependent induction of the immediate early gene Krüppel-like factor 9: implications for neural development and plasticity**. Endocrinology (2009) 150:1757–1765.
Chiu Shu-Ling; Chen Chih-Ming; Cline Hollis T **Insulin receptor signaling regulates synapse number, dendritic plasticity, and circuit function in vivo**. Neuron (2008), 58(5), 708-19.
Pineda Ricardo H; Ribera Angeles B **Dorsal - ventral gradient for neuronal plasticity in the embryonic spinal cord**. The Journal of neuroscience (2008), 28(14), 3824-34.
Kumar Anoop; Godwin James W; Gates Phillip B; Garza-Garcia A Acely; Brockes Jeremy P **Molecular basis for the nerve dependence of limb regeneration in an adult vertebrate**. Science (New York, N.Y.) (2007), 318(5851), 772-7.

Neural signalling

X.F. Liu, P.K. Tari and K Haas K. **PKM zeta restricts dendritic arbor growth by filopodial and branch stabilization within the intact and awake developing brain.** (2009) J Neurosci. 29:12229-35.
N. Schwartz**,** A. Schohl and Edward S. Ruthazer **Neural activity regulates synaptic properties and dendritic structure in vivo through calcineurin/NFAT signaling**. (2009) Neuron 62:655-69.
Nishiyama Makoto; von Schimmelmann Melanie J; Togashi Kazunobu; Findley William M; Hong Kyonsoo **Membrane potential shifts caused by diffusible guidance signals direct growth - cone turning**. Nature neuroscience (2008) 11: 762-71
Sharon B. Sann, Lin Xu, Hiroshi Nishimune, Joshua R. Sanes and Nicholas C. Spitzer **Neurite outgrowth and *in vivo* sensory innervation mediated by a $Ca_V2.2$ – laminin β2 stop signal.** (2008) J. Neurosci. 28: 2366-2374.
Nicol Xavier; Voyatzis Sylvie; Muzerelle Aude; Narboux-Neme Nicolas; Sudhof Thomas C; Miles Richard; Gaspar Patricia **cAMP oscillations and retinal activity are permissive for ephrin signaling during the establishment of the retinotopic map**. Nature neuroscience (2007), 10(3), 340-7.

Learning and memory

Du JL, Wei HP, Wang ZR, Wong ST, Poo MM. **Long-range retrograde spread of LTP and LTD from optic tectum to retina** Proc Natl Acad Sci U S A. (2009) Nov 3. [Epub ahead of print]PMID: 19887635

R.C. Ewald and Hollis T Cline **Cloning and phylogenetic analysis of NMDA receptor subunits NR1, NR2A and NR2B in *Xenopus laevis* tadpoles**. (2009) Front Mol Neurosci. 2:4

Maguschak, Kimberly A.; Ressler, Kerry J. **β -catenin is required for memory consolidation.** Nature Neuroscience (2008) 11: 1319-1326

Motor control and integration

Dong W, Lee RH, Xu H, Yang S, Pratt KG, Cao V, Song YK, Nurmikko A, Aizenman CD. **Visual avoidance in Xenopus tadpoles is correlated with the maturation of visual responses in the optic tectum**. J Neurophysiol. (2009)10:803-15.

Soffe Stephen R; Roberts Alan; Li Wen-Chang **Defining the excitatory neurons that drive the locomotor rhythm in a simple vertebrate: insights into the origin of reticulospinal control**. The Journal of physiology (2009), 587(Pt 20), 4829-44.

Zornik Erik; Kelley Darcy B **Regulation of respiratory and vocal motor pools in the isolated brain of Xenopus laevis**. Journal of Neuroscience (2008), 28(3), 612-21

Rhodes, Heather J.; Yu, Heather J.; Yamaguchi, Ayako. **Xenopus vocalizations are controlled by a sexually differentiated hindbrain central pattern generator.** Journal of Neuroscience (2007), 27(6), 1485-1497.

Sensory function

Chen TW, Lin BJ, Schild D. **Odor coding by modules of coherent mitral/tufted cells in the vertebrate olfactory bulb**. Proc Natl Acad Sci U S A. (2009)106:2401-6.

Garcia-Morales Carla; Liu Chiung-Hui; Abu-Elmagd Muhammad; Hajihosseini Mohammad K; Wheeler Grant N **Frizzled - 10 promotes sensory neuron development in Xenopus embryos**. Developmental biology (2009), 335(1), 143-55

Rossi, Christy Cortez; Hernandez-Lagunas, Laura; Zhang, Chi; Choi, Irene F.; Kwok, Letitia; Klymkowsky, Michael; Bruk Artinger, Kristin. **Rohon - Beard sensory neurons are induced by BMP4 expressing non-neural ectoderm in Xenopus laevis.** Developmental Biology (2008), 314(2), 351-361.

Elliott Taffeta M; Christensen-Dalsgaard Jakob; Kelley Darcy B **Tone and call responses of units in the auditory nerve and dorsal medullary nucleus of Xenopus laevis**. Journal of comparative physiology. A, Neuroethology, sensory, neural, and behavioral physiology (2007), 193(12), 1243-57.

Channels

Bosmans Frank; Martin-Eauclaire Marie-France; Swartz Kenton J **Deconstructing voltage sensor function and pharmacology in sodium channels**. Nature (2008), 456(7219), 202-8.

Keith, Ryan K.; Poage, Robert E.; Yokoyama, Charles T.; Catterall, William A.; Meriney, Stephen D.. **Bidirectional modulation of transmitter release by calcium channel/syntaxin interactions in vivo.** Journal of Neuroscience (2007), 27(2), 265-269.

Ben-Chaim Yair; Chanda Baron; Dascal Nathan; Bezanilla Francisco; Parnas Itzchak; Parnas Hanna **Movement of 'gating charge' is coupled to ligand binding in a G-protein-coupled receptor**. Nature (2006), 444(7115), 106-9.

Synapses

Berghuis Paul; Rajnicek Ann M; Morozov Yury M; Ross Ruth A; Mulder Jan; Urban Gabriella M; Monory Krisztina; Marsicano Giovanni; Matteoli Michela; Canty Alison; Irving Andrew J; Katona Istvan; Yanagawa Yuchio; Rakic Pasko; Lutz Beat; Mackie Ken; Harkany Tibor **Hardwiring the brain: endocannabinoids shape neuronal connectivity**. Science (2007), 316(5828),

Xu, Chun; Zhao, Man-xia; Poo, Mu-ming; Zhang, Xiao-hui. **GABAB receptor activation mediates frequency-dependent plasticity of developing GABAergic synapses.** Nature Neuroscience (2008), 11(12), 1410-1418

Borodinsky, Laura N.; Spitzer, Nicholas C.. **Activity-depending neurotransmitter-receptor matching at the neuromuscular junction.** Proceedings of the National Academy of Sciences of the United States of America (2007), 104(1), 335-340.

Circuits

Dunfield Derek; Haas Kurt **Metaplasticity governs natural experience-driven plasticity of nascent embryonic brain circuits**. Neuron (2009), 64(2), 240-50

Ramdya P, Engert F. **Emergence of binocular functional properties in a monocular neural circuit.** Nat Neurosci. (2008) 11:1083-90.

Pratt Kara G; Dong Wei; Aizenman Carlos D **Development and spike timing-dependent plasticity of recurrent excitation in the Xenopus optic tectum**. Nature neuroscience (2008) 11: 467-75.

Technical innovation

Berndt, Andre; Yizhar, Ofer; Gunaydin, Lisa A.; Hegemann, Peter; Deisseroth, Karl. **Bi - stable neural state switches.** Nature Neuroscience (2009), 12(2), 229-234

Viczian Andrea S; Solessio Eduardo C; Lyou Yung; Zuber Michael E **Generation of functional eyes from pluripotent cells**. PLoS biology (2009), 7(8); PubMed ID 19688031

Butts, Christopher A.; Xi, Jin; Brannigan, Grace; Saad, Abdalla A.; Venkatachalan, Srinivasan P.; Pearce, Robert A.; Klein, Michael L.; Eckenhoff, Roderic G.; Dmochowski, Ivan J. **Identification of a fluorescent general anesthetic, 1-aminoanthracene.** Proceedings of the National Academy of Sciences of the United States of America (2009), 106(16), 6501-6506.

Junek Stephan; Chen Tsai-Wen; Alevra Mihai; Schild Detlev **Activity correlation imaging : visualizing function and structure of neuronal populations**. Biophysical journal (2009), 96(9), 3801-9

Xenopus grants funded by the Institute:

According to NIH RePORTER Search Tool, in the fiscal year of 2011, the National Institute of Neurological Disorders & Strokes (NINDS) funded fifteen grants for projects involving Xenopus. These grants total to $4,118,899.

2011 *Xenopus* White Paper - Community Needs:

Executive Summary

***Xenopus*: An essential vertebrate model system for biomedical research:**

Model animals are crucial to advancing biomedical research. Basic studies in vertebrate animals rapidly accelerate our understanding of human health and disease. Among the commonly used model animals, the frog, *Xenopus,* has great impact because of its close evolutionary relationship with mammals. Moreover, the remarkable experimental repertoire of the *Xenopus* system has made it a cornerstone of neurobiology, physiology, molecular biology, cell biology, and developmental biology.

Current NIH investment in research using *Xenopus*:

Consistent with its broad utility, the NIH has made a large and continuing investment in *Xenopus* research. Indeed, a search of the NIH rePORT database for R01 or equivalent grants using the search term "*Xenopus*" returned **678 grants for a total of over $217,000,000** for FY09-10. The NIH has also recently demonstrated its commitment to *Xenopus* community resources by approving $2.5 million to establish the National *Xenopus* Resource in Woods Hole, MA and a similar amount to maintain and expand Xenbase, the *Xenopus* Community's online database.

***Xenopus* as a model system for human disease gene function**

Given the tremendous power of the *Xenopus* system, the pace of new biological discovery by the *Xenopus* Community is vigorous. Using *Xenopus*, we have significantly improved our understanding of human disease genes and their mechanisms of action, justifying the NIH's investment. For example:

Xenopus embryos are used for *in vivo* analysis of gene expression and function:

Congenital Heart Disease – *PNAS* 2011. 108, 2915-2920
CHARGE Syndrome – *Nature* 2010. 463, 958-962.
Bardet-Biedl and Meckel-Gruber Syndromes – *Science* 2010. 329, 1337-1340.
Hereditary hypotrichosis simplex – *Nature* 2010. 464, 1043-1047.
Hutchison-Gilford Progeria – *Dev. Cell* 2010. 19, 413-25.
Cutis laxa – *Nat Genet.* 2009. 41, 1016-21.
Colorectal cancer – *Genome Res.* 2009. 19, 987-93.
Nephronophthisis – *Hum Mol Genet.* 2008. 17, 3655-62; *Nat Genet.* 2005. 37, 537-43.

Xenopus egg extracts are used for *in vitro* biochemical studies:

Fanconi Anemia – *Mol. Cell.* 2009. 35, 704-15; *Science.* 2009, 326, 1698-701.
C-myc oncogene – *Nature*. 2007. 448, 445-51.
BRCA1 – *Cell*. 2006. 127, 539-552

Xenopus oocytes are used to study gene expression and channel activity:

Rapid-onset dystonia-parkinsonsim – *Nature* 2010. 467, 99-102.
Trypanosome transmission – *Nature* 2009. 459, 213-217.
Epilepsy, ataxia, sensorineural deafness – *N Engl J Med.* 2009. 360, 1960-70.
Catastrophic cardiac arrhythmia (Long-QT syndrome) – *PNAS* 2009. 106,13082-7.
Megalencephalic leukoencephalopathy – *Hum Mol Genet.* 2008. 17, 3728-39.

***Xenopus* as a model system for understanding basic biological processes:**

Xenopus also plays a crucial role in elucidating the basic cellular and biochemical mechanisms underlying the entire spectrum of human pathologies. Just a small fraction of the many recent discoveries are highlighted here:

Xenopus contributes to our understanding of vertebrate genome organization.
(*Science.* 2010*. 328, 633-636*).

Xenopus egg extracts reveal fundamental aspects of cell division.
(*Cell.* 2010. 140, 349-359; *Nature*. 2008. 453, 1132-6; *Science*. 2008. 319, 469-72).
Xenopus reveals new aspects of eukaryotic nuclear structure and function.
(*Cell*. 2010. 143, 288-98; *Science. 2010.* 318, 640-643).
Xenopus embryos are used for studies of Wnt and TGF-β signal transduction.
(*Science*. 2010. 327, 459-463; *Cell*. 2009. 136,123-35).
Xenopus embryos are used for studying mucociliary epithelia.
(*Nat Cell. Biol.* 2009 11 1225-32; *Nature*. 2007. 447, 97-101).
Xenopus embryos are used for studying development of the vasculature.
(*Cell*. 2008.135, 1053-64).
Xenopus egg extracts provide key insights into DNA damage responses.
(*Mol Cell.* 2009. 35,704-15; *Cell.* 2008.134, 969-80).
Xenopus embryos link telomerase to Wnt signaling.
(*Nature*. 2009. 460, 66-72).
Xenopus are used for small molecule screens to develop therapeutics.
(*Nat Chem Biol.* 2010. 6, 829-836; *Blood*. 2009. 114, 1110-22; *Nat Chem Biol.* 2008. 4, 119-25).

Despite its demonstrated utility and despite the recent investments by the NIH, *Xenopus* still lacks many resources that are considered entirely essential for other model systems. It is the consensus of the *Xenopus* community that their biomedical research could be greatly accelerated by the development of key resources of use to the entire *Xenopus* research community.

At the 2010 International *Xenopus* Conference, developmental, cell, and molecular biologists gathered to discuss the resources needed and the priority that should be assigned to each. There was broad community-wide consensus that eleven resources are currently needed, and these were prioritized into two categories: Immediate Needs and Essential Resources:

The Immediate Needs of the *Xenopus* research community:

1. **Generation of the *Xenopus* ORFeome:**
   -Will enable genome-wide *in vivo* analyses of gene function.
   -Will enable genome-wide *in vivo* analyses of protein localization.
   -Will enable, when combined with transgenesis, the first large-scale biochemical determination of protein-protein interactions in specific tissues and at specific embryonic stages.
   -Will facilitate more-rapid functional characterization of specific proteins.

2. **Improvement of the *Xenopus* genome sequence:**
   -Will accelerate molecular studies by providing a complete catalogue of *Xenopus* genes.
   -Will enable completion of the *Xenopus* ORFeomes.
   -Will enable genomic analyses & systems biology approaches for novel gene discovery.
   -Will facilitate proteomics approaches and peptide analysis.

Essential Resources for *Xenopus* research community:

In addition to these most-pressing needs, the community has identified nine other Essential Resources that should be developed as soon as possible, so that *Xenopus* biologists can more effectively fulfill the missions of the NIH. The *Xenopus* community considers all of these additional resources to be essential, but understands that priorities must be set, and therefore ranks these as indicated below:

3. **Improvement of long-range contiguity in the *Xenopus laevis* genome**

4. **Improvement of *Xenopus* antibody resources**
5. **Loss of function: Zinc Finger Nucleases/TILLING**
6. **Loss of function: Small inhibitory hairpin RNAs**
7. **Novel loss of function/knockdown/knockout technologies**
8. **Intergenic annotation of the *Xenopus* genome**
9. **Improvements of the *X. tropicalis* genome – long range contiguity**
10. **Additions and improvements to Xenbase: the *Xenopus* Model Organism Database**
11. **Frogbook: A comprehensive resource for methods in *Xenopus* biology**

Community Recommendations for Attaining Resources:

The *Xenopus* Community feels that in order to attain these much needed resources it will be imperative to renew the PAR-09-240/1: "Genetic and Genomic Analyses of *Xenopus*". This mechanism can help to direct funding to the establishment of resources that will accelerate research by the entire community. Development of research resources is essential to the NIH mission, but because such work is not hypothesis-driven, these proposals fare poorly in standard CSR study sections. Moreover, the standard study sections typically lack the depth of expertise that is needed to properly evaluate these proposals. The "Genetics and Genomic Analyses of *Xenopus*" PAR allows for a focused and expert review of resource development proposals, and its renewal will help to ensure a continuing return on the current NIH investment in biomedical research using *Xenopus*.

The *Xenopus* Community also feels that, given the ease with which massive amounts of biological samples can be obtained using this organism, a new PAR to support systems biology using *Xenopus* is warranted. A new PAR in this area would allow all biomedical researchers to exploit the emerging genomic resources for *Xenopus* to perform systems-level analyses *in vivo*, in a vertebrate, and in a cost-effective manner. Such research would generate significant advances into the "New Biology" described below.

Anticipated Gains for Biomedical Research:

Xenopus as an animal model continues to have a broad impact for biomedical research. Given its already long history of large-scale screens of gene function and its broad use in molecular, cell, and developmental biology, the establishment of additional community-wide resources will greatly facilitate the impact of *Xenopus* as a premier vertebrate model for systems-level analyses.

The National Research Council and the National Academy of Sciences have recently called on the United States "to launch a new multiagency, multiyear, and multidisciplinary initiative to capitalize on the extraordinary advances recently made in biology". This report (http://www.nap.edu/catalog.php?record_id=12764) recommends the term "New Biology" to describe an approach to research where "physicists, chemists, computer scientists, engineers, mathematicians, and other scientists are integrated into the field of biology." The promise of systems-level analysis in *Xenopus*, combined with its already proven strengths, make *Xenopus* the ideal model organism for pursuing "New Biology."

Specifically, genome improvements will provide *Xenopus* researchers with the ability to perform genome-wide screens for biological activities that will in turn allow the rapid assembly and analysis of gene regulatory networks and their relationship to phenotypes. The ORFeome will greatly facilitate such genome-wide screening by allowing all ORFs to be rapidly analyzed or large numbers of proteins to be tagged for analysis of protein-protein interaction or for *in vivo* visualization. Using extracts and biochemical purification coupled with mass-spectrometry and genomic sequence, protein interactomes can be

rapidly identified and validated. *Xenopus* offers a unique resource because it is the only *in vivo* vertebrate animal model that couples vast amounts of biological material and a sequenced genome, thus cell-type specific interactomes can also be identified. Large-scale genetic screens will identify important novel genes in developmental pathways, especially given the relatively simple genome of *X. tropicalis* compared to zebrafish. Finally, the flexibility of both *Xenopus* extracts and embryos make this system ideal for chemical biology screens.

Identifying gene-regulatory networks, interactomes, and novel genes will be only the first steps. The well-established power of *Xenopus* for rapid analysis of gene function will then allow deeply mechanistic analyses to complement the systems-level approaches described above. It is the combination of these characteristics that distinguishes *Xenopus* from other vertebrate model systems such as mouse and zebrafish and allows for a systems-level approach to understanding biological mechanisms. The tremendous impact of the *Xenopus* model cannot be realized, however, without the immediate development of community-wide research resources. This White Paper presents the needed resources, and we look to the NIH for guidance in how to best achieve these goals.

For complete details of the 2011 *Xenopus* White Paper, please visit
http://www.xenbase.org/community/xenopuswhitepaper.do

Appendix

Project Number	Project Title	Activity	Principal Investigator	Organization Name	Total Cost
5R21NS065153-02	DEVELOPMENT OF A ZEBRAFISH ASSAY FOR THE IDENTIFICATION OF ALS DRUG TARGETS	R21	BEATTIE, CHRISTINE E	OHIO STATE UNIVERSITY	$187,782
3R01NS034727-13S1	STRUCTURE OF THE GABA A RECEPTOR BINDING SITES	R01	CZAJKOWSKI, CYNTHIA M	UNIVERSITY OF WISCONSIN MADISON	$359,313
2P01NS049134-06	PROJECT 4	P01	EISENBERG, DAVID	UNIVERSITY OF CALIFORNIA LOS ANGELES	$108,724
1R01NS069714-01	DENDRITIC INTEGRATION IN THE ENTORHINAL CORTEX	R01	GASPARINI, SONIA	LOUISIANA STATE UNIV HSC NEW ORLEANS	$297,763
1ZIANS003016-04	STRUCTURE OF INTEGRAL MEMBRANE PROTEINS	ZIA	GRISSHAMMER, REINHARD	NATIONAL INSTITUTE OF NEUROLOGICAL DISORDERS AND STROKE	$755,350
1R01NS061809-01A1	GENE THERAPY FOR ALPHA-MANNOSIDOSIS	R01	HASKINS, MARK	UNIVERSITY OF PENNSYLVANIA	$199,375
5K08NS052454-05	GABA-A RECEPTOR FUNCTION IN PEDIATRIC FOCAL CORTICAL DYSPLASIA	K08	JANSEN, LAURA	SEATTLE CHILDREN'S HOSPITAL	$168,291
7K08NS045077-06	CONTROL OF GENOMIC STABILITY BY EMI1 AND SECURIN	K08	LEHMAN, NORMAN	HENRY FORD HEALTH SYSTEM	$161,329
5R01NS029967-17	NEURAL CALCIUM CHANNELS-REGULATION AND FUNCTION	R01	LIPSCOMBE, DIANE	BROWN UNIVERSITY	$292,226
3R01NS056415-02S1	NEUROMUSCULAR JUNCTION FORMATION	R01	MEI, LIN	MEDICAL COLLEGE OF GEORGIA (MCG)	$224,522
7R01NS049417-04	THE NIPA 1 PROTEIN IN SPASTIC	R01	NICHOLLS, ROBERT D	UNIVERSITY OF	$278,677

	PARAPLEGIA AND DEVELOPMENT			PITTSBURGH AT PITTSBURGH	
5R01NS056281-04	AXON REGENRATION: SYNERGISTIC ACTIONS OF THE MAPK AND CYCLIC AMP PATHWAYS	R01	PEARSE, DAMIEN	UNIVERSITY OF MIAMI SCHOOL OF MEDICINE	$331,341
1R15NS067566-01	NEUROTRANSMITTER FATE SPECIFICATION AND THE ROLE OF VOLTAGE-GATED CALCIUM CHANNEL	R15	SAHA, MARGARET S	COLLEGE OF WILLIAM AND MARY	$213,017
5R21NS062204-02	IDENTIFICATION OF LIGANDS THAT BIND TO THE ORPHAN DELTA2 RECEPTOR	R21	TRAYNELIS, STEPHEN F.	EMORY UNIVERSITY	$169,531
5R01NS035909-13	GENES ESSENTIAL TO MOTOR AXON GUIDANCE IN DROSOPHILA	R01	VAN VACTOR, DAVID L	HARVARD UNIVERSITY (MEDICAL SCHOOL)	$371,658
				Total	$4,118,899