1 U54 GM072967-01 GLAZIER, J

RESUME AND SUMMARY OF DISCUSSION:
With this new U54 proposal, Dr. James Glazier and his colleagues propose the establishment of a National Center of Excellence called The Tissue Simulation Toolkit. Their objective is developing the computational capabilities support comprehensive multi-scale models of cells and cell communities, with particular emphasis on the subcellular, multicellular (million cells), and tissue levels. The Center will involve collaborations among investigators at Indiana University, Bloomington, IN; Indiana University- Purdue University, Indianapolis (IUPUI), IN; University of Notre Dame, South Bend, IN; Purdue University, West Lafayette, IN; and the University of Kansas Medical Center, Kansas City, KS. This research team is strong in grid computing know-how and toolkit development, but may be deficient in expertise related to modeling and model development. For example, Core 1 proposed the development of a simulation toolkit for developmental biology, which is an important endeavor, particularly if connected with the Virtual Cell project, the Physiome project, and other ongoing efforts in the community, as is proposed. However, the project places nearly exclusive emphasis on the Cellular Potts Model with little attention given to other levels such as the tissue level; only vague plans are presented for ensuring seamless integration with other sites; and critical computer science issues in grid applications are not addressed. A similar theme applies to the work proposed in Core 2 on modeling and model integration, where conflicting statements and a disjointed presentation are viewed as indications that additional input would be beneficial. The three Driving Biological Projects (DBPs) in Core 3 address classic problems in developmental biology (heart development, vascular development, and limb regeneration), are headed by leaders in the field, and are likely to produce interesting results. However, concern exists about the synergy between these efforts and the computational efforts in Cores 1 and 2, and the value of a national Center in facilitating either the computational or the biological goals. The infrastructure component in Core 4 is somewhat ambitious, and the administrative components in Cores 5 - 7 are mostly appropriate and adequate. Overall, despite high levels of enthusiasm for the importance of developing computational tools to facilitate modeling of cells, cell interactions, and morphogenesis, enthusiasm is tempered by a disjointed presentation that draws into question the viability of these efforts if they were to be united into a national Center.

DESCRIPTION (provided by applicant):
The National Center for Tissue Modeling (CTM) links four major Indiana universities: Bloomington, IUPUI, Notre Dame, and Purdue. By developing simple and adaptable open source tools and toolkits for cell-level simulations of tissue and organ development, the CTM will realize the twenty-year vision of modern systems biology - development of reliable multiscale simulations reaching from genome to organism. CTM tools will link seamlessly with both larger scale continuum models and microscopic models. The CTM includes with this proposal signed letters of agreement with Physiome, Virtual Cell, SciRun and BioSpice to support co-development, model integration and platform sharing. CTM simulations will be a hierarchical, organized set of biologically motivated levels where parameters and properties pass from coarser to finer scale models. The CTM will develop a simulation environment via the Tissue Simulation Toolkit (TST), enabling simulations that cover the mesoscale from single structured cells to aggregates of millions of cells, a scale neglected by other major Computational Biology Centers. Its Cellular Potts Model (CPM) tool combines multiscale algorithms with scalable parallelism from PCs to supercomputers. This tool will be presented as a Grid service with a portal interface. Visualization tools will be based on SCIRun and the simulation framework on the CCA to facilitate model interconnection. The CTM is comprised of three major scientific components; computer science, model development and experiment, organized from a unified, interdisciplinary point of view. Experimental activity is devoted to three tractable problems in development and structural disease: vascular development, heart development, and limb regeneration, delivering parameters and test data for simulations and motivate modeling. The computer science activity delivers tools for modeling in conjunction with experiments. CTM member institutions will develop a set of shared interdisciplinary

curricula, expanding successful outreach and training programs to educate and train a new generation of computational biologists.

OVERALL CRITIQUE (primary reviewer):
This is a strong group of both senior and junior researchers addressing the critically important area of developing, validating and making available predictive tools to model three scales related to tissue behavior, namely sub-cellular, cellular, and continuum. The bulk of the effort is at the single to million cell level where the investigators utilize the Cellular Potts Model (CPM) (cell automata) developed for biological applications by senior members of this team. The team plans to implement methods on a range of relevant computer architectures and networks (e.g., parallel clusters, grids) with which they have considerable collective experience. Additional strengths include the careful adherence to emerging standards, and the participation in furthering of the standards process, as well as the desire to seamlessly integrate with large computational research programs below (Virtual Cell) and above (SCIRun) the CPM scale. It was also thought that the successful delivery of validated predictive tools that were fairly seamless in their integration across scales would be a significant national resource. The integration of modeling and experimentation as part of the discovery and validation was deemed a strength. The environment, investigative team, and administrative structure were considered first-rate, as were the plans for outreach. The proposed infrastructure and support will provide the kind of experience for users appropriate to a national center.

However, there were some significant concerns that tempered enthusiasm. With regard to the computer science activities (Core 1) related to the toolkit, the novelty of scaling up a cellular automata approach to grid computing and/or large scale parallel environments is fairly straightforward. Thus the panel questioned whether the advances in computer science were of appropriate strength and significance. On the other hand, issues associated with scale linking, and integration with projects like Virtual Cell and SCIRun were not well articulated, and are considered significant challenges. Finally, there was concern that too much emphasis is placed on methods relevant to CPM at the expense of the scales adjacent to CPM.

With regard to Core 2, there were concerns that CPM was the only method suggested to treat the chosen level, and that in the end, it would not produce a truly predictive tool. More significant was the lack of theoretical details on the scale-bridging methods. In addition, the theoretical and computational activities at non-CPM scales are not appropriately represented (in investigators or effort) or described, and they do not appear to link well with the CPM level. In some of the descriptions, the link between scales is not at all evident. Because there was a sense that considerable work remains on CPM before it is ready for national delivery, the panel also considered the delivery of the simple user interface and training materials mentioned in Core 4 to be overly ambitious (but significant for the future).

CORE 1:

CRITIQUE 1:

SIGNIFICANCE: Core 1 describes the tissue simulation toolkit (TST) which represents the core Computer Science components of the project. The overarching goal of this toolkit is to support the execution of multiscale tissue simulations along different time and space scales. The toolkit will enable the integration of 'best-of-breed' tissue simulation models and tools and high-end visualization, and will make them accessible to a wide community of end-users using Web portal GUI interfaces from the desktop. The problem area is significant and intellectually challenging from both the biomedical/systems biology and computing standpoints.

APPROACH: The TST will be realized by the adoption of Grid technologies. The Grid has been proposed as a model for cyberinfrastructure and is being successfully deployed in other physical science domains, most notably high-energy physics. The appeal of Grid technologies lies in their promise of harnessing large amounts of computational resources and in recent strong efforts towards integration via Web/Grid services. In both respects, the adoption of Grid technologies is logical and justifiable. The proposed infrastructure heavily leverages standards (e.g. SBML, CCA, Web Services) and widely used tools (e.g. SCiRun) and this greatly increases the potential interest and impact of the project. The portal approach for desktop access, the use of web/grid services, and the reliance on standards make a sound approach.

INVESTIGATORS: The Computer Science investigators (Gannon, Fox, and Lumsdaine) have solid track records in Grid computing research, development and adherence to standards, and in producing useful software artifacts (Webflow, XCAT, MPI, etc). Several have also been involved in interdisciplinary research projects with application collaborators.

INNOVATION: There are several parts to the Core 1 proposal that attempt to address the multiscale nature of the simulations, particularly in scaling up from 2 cells to 10^6 cells. The TST begins with the adoption of CPM and SBML as its core technologies. One of the proposed research challenges is to parallelize CPM for large-scale Grid execution. This is a useful thing to do and makes good use of Grid technology, however since this is a Monte-Carlo code it is not clear that this is particularly challenging. For example, Folding home runs Monte-Carlo simulations over 1000s of nodes. CPM will also be modified to utilize mathematica primarily for its visualization capability. Visualization components appear later in the proposal, so perhaps this work is redundant. Other optimizations include message blocking to reduce message latency penalty. It is not clear why this is particularly challenging within the CPM context. It is also claimed that parallelizing the multi-scale techniques within the toolkit (I assume this applies to CPM) will require dynamic selection of components based on the current Grid conditions as has been done in ATLAS and LAPACK. This is an interesting area, but the proposal did not give adequate details as to how this would be done?

My next set of comments pertains to the Grid discussion. It is suggested that workflow will be used to express multiscale model integration, and that the semantic web ideas will be useful. Greater motivation and examples of TST-relevant workflows would have been helpful. What are the technical challenges inherent in the workflow approach?

It also seems intuitive to run different models at different scales onto different Grid resources as is mentioned. But it is not clear how this will be done. What information is needed from the models/applications/tools to do this? How will the author handle multiple job requests in the environment, i.e. resource management?

The Core also talks about the TSTGrid (presumably referring to the specific resources upon which the TST will be deployed), the TSTPSE, TST Data Grid, and middleware. The TSTPSE, data grid, and middleware are not well described and their precise roles are unclear, as well as the research challenges. It is also not clear how the CPM core tool will relate to the other tools that are to be included in the TST; one presumes this would be part of the workflow specification, but it is not clear.

Several aspects of this Core are noteworthy:

1. Ensuring that the TST will allow the user to select the appropriate scale and abstraction criteria seems useful.

2. Leveraging OGSA-DAI (Grid DB interface) is responsive to the biocommunity. I would move this up to year 1 in the schedule.

3. The requirement for CCA compliance to support tool integration is a good idea, but since many/most of the TST services will be Web/Grid services which also provide integration, might there be some overlap here?

ENVIRONMENT: In terms of computing environment, Purdue and IU are already part of the NSF's TeraGrid and IU has a local Grid (IP) infrastructure. IU also has excellent visualization resources. The amount of resources that are available to CTM as opposed to TeraGrid and local (non-CTM) users is not clear. The adoption of standards for tool wrapping (CCA), tool access (Web/Grid services), and the current Grid infrastructure experience (IP and TeraGrid) increase the likelihood that the CTM could be extended nationally in the future.

OVERALL EVALUATION: In general, the Core 1 proposal strengths are its clear standards-focus, leveraging widely-used computing and biomedical tools, and consideration of integration. However, the proposal was somewhat weak is highlighting the key issues on putting TST onto the Grid. What are the real research problems? Tool re-engineering to fit the TSTGrid framework is a good idea, but this strikes the reviewer as mostly engineering and retrofitting. The reviewer was also concerned about the amount of effort for the CS faculty. A stronger proposal would identify more specific computing challenges relating to tissue modeling (as opposed to the somewhat generic term multiscale), and develop IT/CS research around these challenges.

CRITIQUE 2:

Core 1 is directed mainly toward the computer science tools that are needed by the Center for the Tissue Simulation Toolkit (TST). The background and description of the theories and approaches that comprise their proposed analyses are given mainly in Core 2. Computer science issues include general algorithmic, hardware, and networking (e.g. cluster computing, grid computing), standards, interoperability and interfaces. Thus the comments below relate primarily, although not exclusively, to the computer science issues.

SIGNIFICANCE: Core 1 addresses issues that are critical to the success of a national biocomputing center as envisioned by the NIH RFA, and in this regard, the significance of the proposal is high. The toolkit will subscribe to accepted standards, and as a national center will be one of the contributors in setting such standards. One key will be interfaces with the evolving 'markup languages', and it appears the Center will make an effort through planned use of SBML and interactions with the Virtual Cell (UCMC), Physiome (Australia) and SCIRun (Utah) efforts. Of significance also is the experience with, use and testing of, and plans for implementing a variety of parallel computational environments including clusters and grids. Successfully providing a toolkit which can be run seamlessly on any of the high performance platforms (present and future) without unusual user intervention is of critical importance.

There are two areas of concern. First, there is significant emphasis in this proposal on the Cellular Potts Model (CPM) which has been developed by members of the research team. While not given exclusive attention in the toolkit, there is a possibility that CPM-related high performance computing issues will so dominate the developments that important issues at the scales above and below CPM will not be adequately covered. The second concern relates to usability by the community. It is somewhat unclear how the non-specialist user outside of the Center participants will interface with the toolkit and/or the level of expertise required for that interaction.

APPROACH: The overarching approach of the Center is to evaluate existing solutions and approaches, adopt or adapt as appropriate, and develop only when needed.

In additional to the fundamentally sound overarching approach, there are many strengths of the proposed Center toolkit activity. The adherence to standards that are emerging in markup languages

and the promotion of additions to these standards is a critically emerging area. Contacts have been made with several of the key major efforts in this area (Physiome, Virtual Cell, BioSpice and SCIRun), and in some cases, interactions with these Centers have already taken place. Subscription to the Common Component Architecture (CCA) is also planned, including a somewhat lukewarm commitment to change current codes to CCA. A related issue is to allow users of the TST access to the toolkits and methods of the other programs mentioned above.

Another strength is the experience with and plans for implementation on a wide range of computing platforms, particularly at the parallel, cluster and grid levels. The team has extensive experience with such systems and members participate on influential committees planning future directions of such systems.

The planned Center activity relative to tissue developmental biology carries the overwhelming weight and is also a strength. The group has extensive experience with the method, is well aware of the computational issues that need to be addressed, and has articulated a solid plan for achieving goals relative to more widespread access to the CPM approach and significantly larger, more complex, and presumably more representative analyses based on CPM. There is a desire to make the use of the tools a relatively transparent process.

There are also some weaknesses that in some cases could be related to a lack of detail in the description. There is clear advantage to having a national center with the capabilities of simulation tools of tissue developmental biology in the range of one to millions of cells in a 3-D space. However, for a national center, it is equally critical that the computer hardware / software issues for the links to (at least) the scales below and above CPM be adequately addressed. The plan to interact with appropriate centers / activities is in place, but the articulation of the issues is less clear.

A related potential weakness is the user interface and the level of expertise needed to use the TST. It will be helpful to more explicitly define the range of users, outside of the Center, and their anticipated expertise. Details of a very simple (perhaps overly simple) user interface, and methods to ease the training of users are given in Core 4, so this comment is directed toward a lack of detail on how to achieve this extremely ambitious goal.

Particularly at the level above CPM, but increasingly at and below the CPM level, bioengineers are making fundamental discoveries about tissue development and properties, and they would almost certainly be interested in many of the TST tools under consideration. A potential weakness, therefore, is lack of input and participation from that area of expertise. This issue is discussed in more detail under Core 2, where the weakness is more apparent.

INNOVATION: The toolkit concept and the computational hardware and software associated with the toolkit are not radically new ideas. However, there is strong innovation in the plan to broadly enhance the capabilities for simulation at the CPM level, and to establish relatively transparent, user-friendly links to the scales above and below. This will provide innovative capabilities currently out of reach to most users.

INVESTIGATORS: The team assembled for the purposes of Core 1 is outstanding. There is no question of the hardware and software expertise and extensive experience to successfully implement the TST concept. The investigators are also well placed in the community, planning future advances in networks and parallel computing infrastructure. With regard to articulating new aspects of the CPM approach as they relate to Core 1, the team has leading experts in the field. The collection of experts in math and science (computer, biology and chemistry) is extensive and outstanding. The input from the team will be critical to developing the TST in a fashion useful to the worldwide science community.

ENVIRONMENT: The environment for developing and delivering the TST is outstanding. Much of the computational testbed is already in place and the support for the environment appears to be strong.

CRITIQUE 3:

SIGNIFICANCE: This proposal identifies the Cellular Potts Model (CPM) as the central concept, formalism, and algorithm to integrate the diverse work of the many researchers associated with this proposal. This reviewer's experience as a theoretical and computational physicist suggests that the use of any kind of cellular automata model should not be the foundation for an expensive, multi-year, interdisciplinary scientific effort.

APPROACH: Cellular automata (including Potts models whose origin lie in understanding the physics of equilibrium phase transitions) are models in which space, time, and field values take on discrete values. Except in rare idealized cases, the discrete values and the rules that determine how the site values evolve over time can be quite difficult to relate to experiment since the fundamental physical description of nature (quantum mechanics, the Maxwell equations, molecular dynamics via Newton's equations of motion, equations of stress and strain, the Navier-Stokes equations) involve continuously evolving space, time, and field values. Many scientific cellular automata have been useful to demonstrate that certain ingredients are capable in principle of explaining some phenomenon of interest (say a spiral cardiac wave in an excitable medium, formation of stripes or hexagons in the skin of an animal, the formation of dendrites in a snow flake), but quantitative prediction, especially as physical parameters are varied, is almost never attained.

In the original context of phase transitions, it was not so important to relate the properties of a Potts model to experimental properties because experiment and theory showed that the properties of second-order phase transitions depend only weakly on details of the medium, specifically only on the spatial dimension and symmetry of the lattice but not on chemical properties. For phenomena like clumping and evolution of biological cells, no such universal details are expected and then it is necessary to find ways to relate features of the CPM to experimental quantities if quantitative predictions are to be made and tested.

There is not a well-defined mechanism to connect the many parameters of the CPM to fundamental physical and chemical quantities other than by fitting to data. This means that the CPM becomes a sophisticated statistical, rather than physical, tool for describing data. There is little doubt that it is a potentially valuable statistical tool that can explain subtle properties of cell aggregations and how they evolve and Professor Glazier and his collaborators are to be congratulated on discovering that such a model can be so useful. But the model is unsatisfying and may ultimately be limited in utility because there is not a constructive connection between its parameters and more fundamental chemistry and physics.

OVERALL EVALUATION: The computer science side of this proposal is on more solid ground since the computer science issues are more technical than conceptual; there is little doubt various algorithms and visualizations can be carried out as proposed and will lead to useful insights and software. The heart of this proposal is the conceptual issue of how to model interacting cells each of which has great complexity (genetics, internal cytoskeletal structure, surface chemistry and mechanics, etc.) The proposal fails to make clear how the CPM will be capable of dealing with this kind of complexity specifically because a connection between the parameters of pages 244-246 and 306-315 and experiment (other than statistical fitting) is missing.

CRITIQUE 4:

This is an outstanding proposal. The assembled PIs and key personnel are well qualified and definitely capable of carrying out the mission of the proposed Center. The scope and the mission of the Center is clearly identified and its computational (Core 1), modeling (Core 2) and experimental (Core 3) activities,

as well as its supporting activities (Core 4 through Core 7) are well formulated and well coordinated, and show every indication that the Center would succeed. Architecture of the system is well defined and supporting infrastructure is available. There are no human subjects involved, issues related to experiments on vertebrate animals are explicitly addressed. Educational and outreach activities, including involvement of underrepresented groups, are appropriate and adequate. There is very close and effective coupling and synergy between Core 1 and Core 2 activities. These research efforts involve cutting edge computer science, computational science, and biological modeling. Cross-verification and validation with real-world experimental efforts (Core 3) are well formulated and appropriate. Literature is adequate.

This proposal inspires confidence that the CTM team will succeed.

CORE 2:

CRITIQUE 1:

SIGNIFICANCE: Core 2 describes the modeling and integration methodologies for the CTM. The resulting models and tools will be realized within the TST from Core 1. This Core consists of various mathematical models and techniques for intra- and inter-cellular processes that simulate biological phenomena at various scales. The problem is significant and important.

APPROACH: The 'core' of this Core is the Cellular Potts Model (CPM) and there is a focus on interfacing CPM to single-cell and reaction kinetic models (RK) at a lower level and also to higher-level tissue and organ models (e.g. FEM methods). Clearly this model is in widespread use, but were there other alternatives to explore? Why or why not? Three motivating projects will drive the research - heart development, vascular development, and limb regeneration. Part of the research will be to develop specific models for these projects within the CPM framework. These DBPs will also provide experimental parameters needed by CPM.

INNOVATION: The work in RK will be in part to determine parameters to large-scale models. Existing RK approaches will be evaluated for 'best'. The reviewer was uncertain how 'best' was determined. It was also not clear what 'develop additional RK related material' meant exactly. Evidently, there are a large variety of data sources to consider. Will there will some kind of common representation for these within TST? This was not discussed.

The FEM approach for continuum modeling will be extended and incorporated into CPM and TST, and will be validated using real data. The immersed boundary method also deals with cellular geometries and so it was not clear if this is fully complementary with the FEM approach or is it addressing a different aspect of the problem?

The section on multiscale modeling seemed rather thin relative to the other sections. Evidently, it will utilize CPM at lower levels, and FEM at tissue scale. What are the challenges, difficulties, here? Some things seemed to be pushed to the TST -- averaging tools for interpolating variables between levels. Isn't this a modeling issue which needs some description and explanation - while TST is simply where it will be implemented? The Core also claims that CMP scales well to the tissue/organ level - this seems to contradict a bit with Core 2 which describes this as a research challenge? Core 1 also says it is "easy" to connect CPM to models at other scales; however this reviewer thought the multiscale aspect of this work was one of the more challenging aspects.

The Core also indicates that there are existing toolkits that attempt to link models. How does this overall effort compare with those?

The need for distributed computing, Grids, large-scale resources, etc. advocated in Core 1 was not so clear from this section. After reading Core 1, it is not precisely clear what the computing challenges really are.

ENVIRONMENT: The environment for the Core 1 seems appropriate for the scale and scope of the proposed work. Much of the work involves model development and integration and there appear to be sufficient resources at the constituent Universities to support this work. There is excellent physical proximity to the IU Grid resources. There is also sufficient support from the experimental projects to provide needed data to help parameterize the simulations.

INVESTIGATORS: The PIs in this Core seem well qualified to carry out the modeling research as they have done prior work in the area and appear to know the related work very well.

OVERALL EVALUATION: The approach is sound combining best practice models and tools, and will include development of specific solutions where needed for their driving DBPs. Hopefully, existing tools and models will be highly leveraged.

Using experimental data to validate CPM and provide parameters to it, and using large-scale models to validate CPM, and CPM to validate lower level models is a logical and sound approach.

There is a great deal of proposed work. It is not clear to the reviewer what the incremental steps will be. The reviewer also felt that one of the key ideas, multiscale modeling, was not deeply explored and lacked details. Finally, the computing challenges
inherent in this Core were not clear to the reviewer.

CRITIQUE 2:

Core 2 deals extensively with the details of the Cellular Potts Model (CPM) as well as work below and above the CPM scale (from one to millions of cells). Note that the CPM approach is a stochastic cellular automata approach. While extensive coverage is given to the CPM approach, there is discussion of previous work and plans at all three scales, as well as plans to link the scales. In general, the activities within the CPM level should be considered a considerable strength, but weaknesses are noted, particularly at scales above CPM, and in the linking of scales.

SIGNIFICANCE: The ability to have a validated, predictive simulation tool applicable at a scale of one to millions of cells will be of major value to researchers in tissue developmental biology. As is nicely articulated by the authors, simulation will ultimately become an integral partner to basic biology research. Of equal significance is the ability to carry the analysis to levels above and below the CPM level, activities also planned by the Center. Finally, the subscription to and advancement of standards emerging to describe relevant details of the many scales associated with biomedical research is of major importance.

APPROACH: As mentioned with Core 1, the overarching approach of the Center is to evaluate existing solutions and approaches, adopt or adapt as appropriate, and develop only when needed. This approach is a clear strength.

There are a number of clear strengths of the proposed Center. Activities based around the CPM approach are extensive, well organized and well articulated. In general, through a series of experiments, simulations, modifications / additions to the components of CPM, and further experiments, the investigators hope to produce a validated simulation toolkit that is predictive with respect to tissue activity at the scale of one to millions of cells. Achieving this goal will be a major strength of this Center. Each of the broadly grouped projects has an appropriate and apparently reasonable set of goals and aims, that in general contribute to the overarching goals of the Center. Because of the wide range of

activities within the CPM scale, it is difficult to assess whether the levels of effort (faculty, post-doc and graduate assistant) are appropriate for a given project. The chief concern within the CPM scale activities is the degree to which the tissue developmental biology community believes in the approach.

In addition to the CPM level work, which dominates the effort and budget, activities are planned at the hierachical scales above (e.g., continuum tissue models) and below (e.g., cell reaction kinetics models). Aspects of these plans involve working with similar toolkit projects (Virtual Cell and SCIRun, in particular). Efforts are planned to link these three scales, and throughout, to maintain compatibility with standard programming architecture (CCA) and standards in markup languages. Successful completion of all these plans will be a strength of the Center activity.

There are some notable weaknesses, primarily related to the non-CPM scales and to the linking of scales. While there is overwhelming expertise at the CPM scale, there is much less involvement at the other scales, and in the linking of scales. First and foremost, this appears to be a matter of the number of participants with appropriate expertise and the level of their activity. This is most striking at the continuum tissue level where it appears as a lack of coordination and integration with the overall project. There are examples from each of the CPM to continuum activities, but let me cite one for example. The bone tissue modeling at Notre Dame (led by Dr. Neibur) is an outstanding project in its own right. However the integration in the project is only vaguely referenced in one sentence that indicates the future possibility of linking to bone growth, CPM modeling (and design of prosthetics). It is also unclear why a bone failure analysis is of highest priority for the Center. In what further illustrates the more obviously disjointed writing of Core 2 (figures missing and mis-referenced, sentences incomplete, some conflicting plans), is a section discussing the bone growth project (following the description of bone modeling), with no mention of the Notre Dame bone modeling, but rather a reference to using ANSYS to do an equivalent viscoelastic model of bone.

A second example is the fluid flow modeling (Immersed Boundary Method) where a fairly broad description is given, with virtually no mention of how this relates to the CPM activities or to the links to plans to use toolkits like SCIRun and Physiome. While it is reasonable to tap the expertise and the opportunities of the CPM approach, activities above and below that level should be given a more significant and coordinated role in order for the goals of the Center to be realized.

There are similar concerns about the scale linking efforts. Plans to develop the averaging tools to move from lower to higher scale, and the lifting tools to move from higher to lower scale are appropriate and essential to success. However, there is relatively little discussion of plans in this area, and there appears to be far too little effort devoted to this aspect of the project.

A final concern on the CPM approach related to a more diverse collection of expertise and input. At the scale of a cell and above, we know that biomechanical signals, for example, influence bone development, and biofluid forces influence cells near flows. The inclusion of such influences, outside of the force that derives from energy gradient, is minimal. While at subcellular levels, activity is ultimately biochemical, it seems that a greater level of sophistication and inclusion is required when talking about multiple cells.

INNOVATION: The toolkit concept and the computational hardware and software associated with the toolkit are not radically new ideas. However, there is considerable potential for innovation in having a predictive computational tool at the one-to-many cell level, in successfully linking to predictive analyses at levels above and below, and in providing a seamless interface to similar efforts at other institutions.

INVESTIGATORS: The investigators in Core 2 are outstanding. They include senior faculty who are internationally known for developments at the cellular and subcellular levels, and outstanding young faculty who are engaged in innovative research. There is no doubt that they have the expertise for successful completion of projects in their area. The weakness of the investigative team is not related to the present participants, but to the absence of more balanced expertise. Most of the weaknesses

mentioned above can be attributed to a lack of participants in the areas discussed. Increasingly the successful projects in tissue development or tissue engineering, are primarily oriented to bioengineering.

ENVIRONMENT: The environment for developing and delivering the modeling and methods is outstanding. There are no perceived environmental weaknesses that would affect successful completion of the plan.

CRITIQUE 3:

SIGNIFICANCE: This is an unconvincing proposal, in the opinion of this reviewer. The investigators plan to model nothing less than cells, tissues, and organs -- their development, functions, and interactions -- all on the basis of an assortment of existing modeling methodologies which they hope to incorporate into a common computational environment. A central role in their proposal is played by the cellular Potts model (CPM), which is itself unconvincing because what it really models is the physics of interaction of immiscible liquids, a process which may capture certain features of cell sorting in certain situations, but which seems inherently too simple to model correctly the many complexities of cell-cell interactions.

APPROACH: In this connection, it is telling that the investigators define a biological cell (within the context of the CPM) as a connected set of computational elements that are all in the same state. Similarly, they define the boundary between two biological cells as a boundary across which the state of the computational elements is different on the two sides of the boundary. An obvious consequence of these definitions is that there can never be a set of contiguous but distinct biological cells in the same state (if there were, they would be considered as a single biological cell according to the above definitions). This makes sense for liquids, for which the only boundaries that matter are those between different liquids, but hardly for biological cells, which have membranes that separate one cell from the other regardless of whether those cells are of the same type or of a different type.

More fundamentally, the CPM relies too strongly on concepts drawn from physics such as the 'energy' of interaction between two cell types, or the 'temperature' associated with the seemingly random motions of cells. These terms are here put in quotation marks to emphasize that their use in CPM does not correspond to actual energy or temperature
of the system. The 'temperature', for example, is related to the supposedly random motions of a cell produced by the cell's locomotive machinery, at the expense of metabolic energy. There is little reason to think that that the statistics of these motions resemble those of thermal motions, and therefore little reason to think that the statistical mechanical notion of temperature is actually relevant. Similar remarks could be made about the supposed 'energy' of interaction between cells of different types, and even more so about the supposed Hamiltonian dynamics derived from this 'energy'. These
concepts of the CPM are at best metaphors, and most likely misleading ones, since there is no a priori reason why the biological system should obey the 'laws' that are normally associated with these words. Living systems are driven by metabolic processes with their accompanying energy fluxes that have no counterpart in a passive system like two liquids in contact. It would be strange indeed if the former could be reliably modeled by the latter.

Although it is clear that the investigators have a lot of experience with the CPM, the same cannot be said of the other modeling strategies they propose. Large parts of the proposal read like a laundry list of modeling methodologies (developed by other people) that the investigators say they intend to utilize. The proposal does not inspire confidence that these methodologies will be used effectively, or that they will work well together.

OVERALL EVALUATION: Individual parts of the proposal describe interesting research, but the attempt to put all this under the umbrella of a large project is not successful. Sometimes the whole is worth less than the parts, and that seems to be the case here.

CORE 3: SUMMARY

The overall project proposal - NIH National Center for of Excellence: Tissue Simulation Toolkit (or National Center for Tissue Modeling - CTM) is concerned with development of a nationally available (over the network) reliable multiscale simulation set reaching from genome to organism. Central to the effort is the Tissue Simulation Toolkit (TST) that enables simulations which can range from single structured cells to aggregates of millions of cells. Its core is a suite of scalable Cellular Potts Model based algorithms. The plan is to present the toolset as a grid service. The offering will also include visualization tools. System development will be based on sound software and system engineering principles. Experiments are tied with the use and/or development of the TST associated algorithms. In addition CTM member institutions will develop interdisciplinary curricula, and expand outreach and training. The proposed work is clearly within the scope of the RFA.

There are three experimental efforts in the plan. All three Driving Biological Projects (DBP) are concerned with morphogenesis. In one case it is the morphogenesis of heart development, in another of vascular development, and in the third of limb regeneration. Personnel associated with the experiments are appropriately qualified to conduct them. The science behind the experiments is sound, and experiments are well formulated and scoped.

Common goals and objectives of the Core 3 projects are clearly identified and described (get data, predict effects by simulation, validate simulations, refine simulations). Technical and scientific commonalities among the projects (all are focused on morphogenesis, mix experiments and simulations, involve genetic perturbation and analysis) are clearly stated, explained and explicitly related to the proposed simulation environment, processes and feedback loops. It is easy for a reader to see not only why certain experiments were chosen, but what the benefits of the experiments are, how experiments benefit from the proposed CTM modeling, visualization and computational environment, and how the results of the experiments verify, validate and feed back into the furthering of the CTM tools. The timelines of all three proposed experimental DBP efforts are explicitly stated and interrelated with the in silico studies and tool use. Plans are presented to compete for independent funding for continuation of the work. Plans are also presented to recruit and select additional DBPs after collaborations with the initial 'founding' DBPs under the NIH NCBC have been completed.

The Core 3 proposal inspires confidence that the science described in Core 3 would provide a new way to access dynamically and with new methods in molecular biology, descriptive data that has been available for many years in the literature.

All the Core 3 experiments are interesting, doable, and valuable and should generate much interesting data, e.g., about the morphogenesis of the developing mammalian 4-chambered heart or limb regeneration. (Note: It is somewhat unclear how the proposed research differs from ongoing NIH-funded projects in the same laboratories).

One reviewer stated that in terms of the overall proposal whose purpose is to create a Tissue Simulation Toolkit that will aid the understanding and interpretation of experiments, Core 3 is unfortunately weak. The critical discussions of how experiments will couple to theory and modeling and vice versa (see pages 366-367, pages 385, 498-409) are much too short and vague for such an important aspect of this proposal. This reviewer felt that the experimentalists do not have a strong need for the Toolkit to help with their experiments, that they do not understand what quantities to measure as input into the Toolkit, nor what kinds of predictions the Toolkit will make that will influence the ongoing

experiments. Others on the panel strongly disagreed with this suggestion but there was concern whether this part of the research could be done.

On page 408, the researchers say honestly that 'Such data is not yet available in sufficient quantity to do computer simulation involving growth factor production and cell signaling in limb regeneration.' and then go on to say that they will work on some other issues in the meantime. This confirmed the concern mentioned in the above paragraph. The researchers on pages 385 give a list of parameters that is a direct list from an earlier part of the proposal which is fine except that the parameters are treated as of equal relevance; there seems to be no prior thinking about which parameters matter or which should be determined first. Section III.5.v.a.9 on page 385 is extremely brief, vague, and optimistic about how the experimental data will be connected to the CPM. The discussion of Section III.4.vii on pages 366-367 is the most detailed and promising but still strikes one as vague as to what specific hypotheses will be tested and how the CPM or experiment will have to be modified if disagreement arises.

In summary, Core 3 is solid and interesting experimentally but weak in terms of how the proposed experiments will aid the refinement of the CPM, which in turn will couple back to computer simulation/in silico experiments.

CORE 3:

CRITIQUE 1:

SIGNIFICANCE: This work is important because it addresses fundamental issues about how molecular and cellular events shape cardiovascular development. Congenital heart disease and cardiovascular diseases arising later in life ultimately claim more lives than any other cause of death. By enhancing our understanding of the basic process of cardiovascular development, new insights into the prevention and treatment of cardiovascular disease are likely to emerge.

The inclusion of components to specifically examine (i) heart, (ii) vascular, and (iii) wound healing development is ambitious and risky in the sense that it dilutes the focus of the application, but these concerns are diminished by the potential to integrate commonalities between these different areas of research. Heart development is intimately connected with vascular development, and the wound-healing efforts have the potential to provide new mechanistic approaches to the important clinical problem of how to regenerate cardiac muscle and vasculature.

Although much progress has been made in the area of cardiovascular development, there are presently only limited computer models available to facilitate these research efforts. It would be highly advantageous to the field if an integrated molecular, cellular, and morphological model were developed, especially if the model were adaptable and could be used as a foundation for future experimental endeavors of this kind (e.g., to evaluate the impact of the loss or overexpression of a new gene or genes, growth factor/hormone treatments, interactions with novel biomaterial matrices, etc.). If successful, researchers could perform simulated experiments to test their hypotheses in silico before expending precious time and resources to do the actual biological experiments. This would also have the added benefit of reducing the number of laboratory animals that would otherwise be needed. The major benefit of this approach would not be to ultimately eliminate the biological experimennts, but rather to develop a useful tool or set of tools that would help guide researchers efficiently towards the most productive and informative experiments. The Driving Biological Projects described in this proposal have the capacity to deliver this laudable goal.

APPROACH: As alluded to above, the approach is an ambitious one that encompasses three broad areas: (i) Heart development, (ii) Vascular development, and (iii) Wound healing. Each of these project areas has multiple sub-projects, each of which is substantial in its own right. A drawback of this

approach is that the application must successfully integrate complex networks of information from these seemingly dissimilar areas of research. An argument could be made, for example, that the application would benefit from focusing solely on heart development. Certainly, such an approach would fall under the scope of the RFA in that it would focus on a particular organ. It would also satisfy the criterion that "an investigator will propose 2-4 collaborations with NIH-funded biomedical researchers" (4 such collaborations are proposed within the heart sub-project alone: Field, Conway, Firulli, and Shou). Moreover, the heart sub-project appears to be the strongest component of this application. It is clearly written, specific, highly goal-oriented, feasible, and likely to succeed. Each project addresses different issues (role of myocyte proliferation on ventricular wall development, transcriptional control of right/left ventricular development, valve development/repair, and the role of BMPs and other growth factors on ventricular compaction and trabeculation). The data generated from each of these projects is expected to be similar in nature (identification of cell position and migration in 3D using fluorescence-tagging and 2-photon fluorescence microscopy) even though it will be gathered using distinct mouse model systems. This is viewed as a major advantage since it should facilitate integration of the data from the various heart development sub-projects for the proposed computational analyses.

Although the heart development sub-projects are largely harmonious and complementary, the choice of genetic markers for the BMP-10 sub-project (III.4.vi) was somewhat puzzling. The strategy itself appears sound, but it requires the procurement of an endothelial cell marker mouse (Tie2-GFP) and generation of a new transgenic mouse model (NCX1-GFP) to obtain a cardiomyocyte-specific marker. The investigator cites a paper by Motoike et al. (2000) indicating that the Tie2-GFP mice are "freely available". No letter or other documentation indicating that these mice are indeed available was provided. This is likely a minor point. Of greater concern is the proposed construction of an NCX1-GFP mouse. While this is certainly feasible and should not derail the study, why not use the MHC-GFP mouse to mark cardiomyocytes? The MHC-GFP mouse is being used for precisely that purpose in the first heart development sub-project headed by Dr. Field. It would seem that using the same mouse to address these two different issues (proliferative and growth factor effects) would not only be a more efficient use of resources, but it should also help to facilitate integration of the biological data sets.

The vascular development sub-projects, though less well-defined than the heart development sub-projects, are also expected to be informative. Vascular development is clearly a logical extension of cardiac development and increased understanding of this process could have major clinical impacts. The investigators have developed or otherwise have access to appropriate ADEP and EDEP cells that they can isolate and study following transplantation into either quail embryos or mouse allantois. The choice of quail as a model poses a minor concern that the applicants readily acknowledge. They cite studies, however, that indicate that heterologous (i.e., mouse) EDEPs appear to appropriately migrate and differentiate in quail embryos. Even so, there will always be an underlying concern that species-specific cues may be misinterpreted or missed altogether in such a heterologous system. This concern is dampened by the fact that the quail embryo is a well-established model for these types of studies, and it is further ameliorated by the fact that all of the studies for this part of the proposal will be done in mouse allantois as well as quail. By comparing the data from the two different recipient "hosts", the investigators should be able to use the differences between these model systems to their advantage.

Although it was at first difficult to decipher what data exactly would be generated from the vascular studies (e.g., a list of "parameters" finally appears on pg. 385 - nearly 14 pages after the beginning of the vascular studies section. - would have been helpful to have this info up-front), the data presented appear convincing and the proposed experiments are straightforward. It is anticipated that a large amount of data will be generated for the computational analyses. The in vitro capillary formation studies should complement the transplantation experiments nicely and be of general interest to the field since this is a widely-used and powerful bioassay.

The last sub-project group in this core focuses on wound-healing. By the applicants' own admission, this is the least-developed section of Core III. Yet, in many ways it is the most interesting, particularly in the context of this application. Two basic models, amphibian limb regeneration and Zebrafish fin

regeneration, are proposed. No cell-specific GFP markers or appropriate transgenic frog or fish models appear to be available for the studies proposed, so the information obtained is expected to be of a much more basic (but no less important.) nature. The investigators will initially evaluate gene expression patterns in regeneration-competent and regeneration-deficient tissues. Selected candidates from these screens will then be employed to test their potential influence on the regeneration process by either blocking their expression or overexpressing them. These studies will be performed in concert with morphological evaluation of the regenerating limbs (or fins).

The experiments are logically presented, straightforward technically, and therefore have a high chance for success. The real trick will be to integrate this information (data) with that from the heart and vascular development sub-projects so that it can be applied to specific problems associated with these processes.

INNOVATION: The application is highly innovative. Its strength (and weakness) is its diversity and bold embrace of wound-healing as a complement to core cardiovascular developmental issues. The applicants have developed a number of innovative models and are using cutting-edge technology (e.g., 2-photon laser-scanning fluorescence microscopy) wherever possible. Conventional approaches are also widely used throughout the application, as appropriate.

INVESTIGATORS: The investigators are well-trained and most of them are independently funded. They appear to be fully competent to perform the proposed studies.

ENVIRONMENT: The research environment at the respective institutions involved (mostly in Indiana) are excellent and more-than-adequate to carry out the proposed work. It is particularly pleasing to see several prominent universities in a common geographical area come together to form this type of partnership.

ADDITIONAL CRITERIA: The work proposed in this application is expected to establish an integrated national biomedical computing environment that will be useful and available to other researchers and clinicians with interests in cardiovascular development and regeneration. The proposed work, in its entirety, is not essential to establishing this environment, but a concentrated effort to computationally model cardiovascular development using a multi-tiered system such as this is viewed as vital for moving this critical area of research forward sooner rather than later. All of the projects described in this application have an "exit strategy" that involves spinning the findings generated from these studies into individual R01 applications and other similar types of independent grant awards within ~ 3 yrs after the initiation of funding.

VERTEBRATE ANIMALS: No major issues, but two minor ones were identified. First, it is noted that IU accreditation is "pending". Does this mean that it is not currently accredited and if so, what impact does this have on the ability of the applicants to perform the proposed animal studies at IU? Second, cervical dislocation is cited as the method of euthanasia for some of the studies. This procedure has been discontinued at a number of institutions, though this reviewer is not sure about the NIH guidelines regarding this matter. Perhaps swift decapitation could substitute as a reasonable alternative?

CRITIQUE 2:

III.4ii.a. Experimental Platform 1: Modeling the effects of cardiomyocyte proliferation on the developing ventricular wall:

Earlier studies using retroviral tagging in chicks suggest that proliferation of individual progenitor cells within the tubular heart gives rise to transmural, cone-shaped growth units which ultimately assemble with one another, resulting in the formation of three-dimensional ovoid structures, which contribute to the formation of the ventricular wall. The proposed studies in this Experimental Platform 1 (Project 1) will provide baseline-imaging data to model the growth of individual transmural growth units within the

normal embryonic heart. Data will include spatio-temporal information of cardiomyocyte position, number, and division rates during normal heart development. They will, in parallel, conduct imaging studies wherein the division rate of individual progenitor cells are altered via genetic manipulation. Real-time imaging data from the genetically altered progenitor cells will be compared to normal controls. The investigators will use this information to generate a computer simulation of the events of cardiac organogenesis. They will then initiate in silico computer-generated experiments whereby the division rate of individual progenitor cell is altered. It is anticipated that computer simulations allow the separate representations and control of individual mechanisms. This will be essential if they are ever to disentangle the complex interacting phenomena of embryonic pattern formation (the spatiotemporal control of cell differentiation) and morphogenesis (the generation of the appropriate three-dimensional relationships among tissues). Such simulation allows integration of these data under different conditions and with other experimental results. Ultimately the simulations should be multiscale and quantitatively predict phenomenology.

III.4.iii Experimental Platform 1: Modeling the effect of cardiomyocyte proliferation on the developing ventricular wall (Loren Field, coordinator):

The investigators point out that although the morphogenetic transformation of the primitive heart into a four-chambered structure is well characterized from an anatomical standpoint, issues pertaining to the origin of the heart forming cells, regulation of cardiomyogenic induction and proliferation, and the molecular regulation of morphogenesis during early embryonic life remain largely unanswered.

III.4.iii.a Specific Aims

Proper development of the ventricular musculature depends in part upon well-coordinated regulation of cardiomyocyte cell cycle activity and cell/tissue migrations in the embryonic heart. Retroviral tagging experiments in chicks using b-galactosidase-expressing viruses revealed that upon myogenic differentiation, proliferation of a single infected myocyte in the tubular heart gives rise to transmural, cone-shaped growth units (Mikawa et al., 1992a; Mikawa et al., 1992b; Fischman and Mikawa, 1997). Mikawa et al., have hypothesized that the assembly of multiple transmural growth units contributes to the three-dimensional ovoid structure of the ventricular walls. More recent studies with neonatal and adult transgenic mice carrying chimerically expressed reporter genes further supported this conclusion (Meihac et al., 2003). These studies have provided important insights into the relationships among the progenitor cells in the primitive heart and the subsequent three-dimensional structure of the ventricular wall; the analyses were static and largely two-dimensional in nature. Consequently, the extent to which cell cycle activity contributes to the formation of a three-dimensional structure has not been quantitatively determined. The studies proposed in this experimental platform (Project 1) should generate baseline data to facilitate the development of a computer simulation of the formation of transmural growth units. In addition, the investigators propose to do experiments in silico and in situ to model and experimentally determine, respectively, the consequences of altered cardiomyocyte cell cycle activity on the morphogenesis of the transmural growth units.

The aims of Experimental Platform One are:
1. Specific Aim 1: Image analysis in cultured explanted hearts of transmural growth units derived from cardiomyocyte progenitors with normal cell cycle activity;
2. Specific Aim 2: Image analysis in cultured explanted hearts of transmural growth units derived from cardiomyocyte progenitors with genetic modifications results in enhanced cell cycle activity;
3. Specific Aim 3: Computer simulation of transmural growth units derived from cardiomyocyte progenitors with normal or genetically enhanced cell cycle activity.

These proposed aims should provide key information describing the real-time formation of the cardiac growth units in terms of cell number, division rates and anatomical position as well as the interrelationships among these parameters. These data will permit the generation of in silico models providing three-dimensional morphogenetic information on normal or genetically-altered cardiac

development, and additionally can apply directly to therapies which rely on enhancing cardiomyocyte proliferation to effect myocardial repair in diseased hearts.

SIGNIFICANCE: This is a well-written proposal by a well-qualified coordinator, Dr. James Glazier of IUPUI. The work follows a logical sequence from an excellent base of background data that has accumulated. In particular, the earlier work of Mikawa et al., 1992a, 1992b; and Fischman and Mikawa, 1997, have set the stage for the kind of research that will be carried out with computer simulation approaches, in particular, the work of Mikawa and others that suggest the logic of formulation of cone-shaped patterns of growth in formation of the heart wall. In addition, cell death (apoptosis) does notcontribute to this process as previously suggested, an important step forward in our knowledge. The proposed experiments will build on existing data to further examine the relationships among the cardiac cell cycle, the formation of growth cone lineages and the overall formation of the ventricular wall.

APPROACH: The preliminary studies are compelling and impressive. The investigators have succeeded in developing a transgenic reporter gene with chimeric expression in a mouse model (Rubart et al., 2003) that uses the a-cardiac myosin heavy chain promoter to target expression of enhanced green fluorescent protein (EGFP) in cardiomyocytes.
Several mouse lines (strains) show a high penetrance of transgenic expression with virtually all of the cardiomyocytes exhibiting EGFP staining. Other mouse lines exhibit mosaic patterns of transgenic expression varying from < 1-20% exhibiting EGFP fluorescence in the adult heart. It is interesting that different genetic backgrounds in the mice influenced the level of mosaic transgene expression.

Aims 1 and 2 will depend upon this method and the preliminary data predicts success with a good deal to be learned from the actual data collection as well as the computer simulations.

For Aim 2, analysis of the explanted hearts with enhanced cell cycle activity, the investigators also have generated several transgenic mouse models with enhanced cardiomyocyte cell cycle activity in the developing ventricle. They have selected two of these models, the MHC-cycD1 mice which have 3-fold more cardiomyocytes than their non-transgenic littermates. They will also use the MHC-TAG which shows a marked proliferative response in the developing ventricle due to encoding of oncoproteins. These two transgene models will be used in Aim 2 to compare with each other (i.e., slight versus extensive phypoplasia) and to controls and compare the effects of cell cycle on ventricular wall development.

The first two Aims are logical and very doable as demonstrated by the high quality preliminary data. As mentioned earlier, Aim 1 will involve the image analysis in cultured explanted hearts of transmurol growth units derived from cardiomyoctye progenitors with normal cell cycle activity. They will use the MHC-EGFP trangene labelled cardiomyocytes using the methods shown in their very compelling preliminary data. The other method of collecting the embryos, culturing them and imaging them with confocal and two-photon microscopy, including after counter staining with d-4-anepps which stains t-tubules, is well established in the laboratory at Indiana University Medical School and is well within the expertise of the group. While the proposed method should work very well in the proposed experiments, two back-up protocols are proposed - the Buckingham approach is certain to work if it becomes necessary.

Specific Aim 2 is designed to basically do the same studies as Aim 1 on cultured explanted hearts of transmural growth units derived from cardiomyocyte progenitors with genetic modifications resulting in enhanced cell cycle activity. The basic approach will be the same as in Aim 1 with respect to confocal microscopy and image analysis. In addition, the investigator will examine the penetrance in the different strains by intercrossing double transgenic mice with non-transgenics and also create chimeric embryos from 4-cell embryos and reimplant them into foster mothers. This aim has much promise, in that the investigators expect to be able to increase the rate of cardiomyocyte mitosis by intermingling growth units. While the aim has significant promise, it is not as well developed as it might be and the preliminary data to take the embryos through all of the steps to a final product that is consistent,

reproducible and, for that matter, doable in every way has not been demonstrated in the Preliminary Data section. The complication of adding experiments to answer questions about background expression differences for the genes is a distraction from the main emphasis of evaluating the role of cell cycle in formation of the ventricular wall. This "side issue" should be addressed in a separate Aim since it complicates the main question to be addressed. This is not to say that the strain issue is not important to pursue - it is just that it seemed hastily put together at the expense of a carefully written cell cycle set of experiments. Never the less, the idea is a good one and the preliminary data does show that at least the first phase of the Aim is straight forward in the sense that it will be possible to compare the effects of enhanced cell cycle activity in the formation of the ventricular wall. This is a logical next step. The third Aim will be to pass this data on to the computational platform for analysis. This should overall be a most interesting parameter of heart development to evaluate by this method and should contribute much to our understanding of the ventricular wall formation in developing embryos with respect to the parameters being examined.

INVESTIGATORS: Dr. Loren Field will serve as the coordinator of Experimental Platform One, Core 3. Dr. Field received his Ph.D. degree at SUNY, Buffalo and continued as a postdoctoral fellow there and at the Roswell Park. He has an excellent record of achievement in the field and is well qualified to serve as a coordinator on this project.

BUDGET: The budget appears reasonable.

CORE 4:

CRITIQUE 1:

Core 4 efforts focus on the establishment and appropriate maintenance of the environment in which open source CTM and other computational toolsets will be developed and will operate. There is ample computational, storage, networking and other infrastructure. Software engineering, scientific workflow, and scientific computational support are adequately addressed, and explicitly related to the goals of the project and the needs of the end-users.

A timeline for a set of deliverables is outlined. It is clear that the Principal Investigators are not only aware of the latest trends in the scientific workflow and modeling support technologies, but are capable of implementing them and intend to make them part of the solution.

The Core 4 proposal inspires confidence that the Core 4 team, in collaboration with the whole CTM team, will succeed.

CRITIQUE 2:

Core 4 details support and infrastructure for user tool development and user access to the tools to run applications (including external, non-Center users). The Center institutions have a strong history of providing the type of support needed by the Center, including Knowledge Base applications, support for software bugs, and software engineering support. There is every indication, therefore, that the technical expertise and experience exist to accomplish the stated goals of the Center with regard to support and infrastructure.

Concerns in this area relate to the underlying materials. Developing high quality educational materials for distance learning is an enormous task. The training tasks in this case will include both use of the analysis software (step-by-step use of software) as well as an appropriate level of understanding of the methods required to run independent analyses. The later will require significant content-expert input, and it is unclear that adequate time has been devoted to this activity. The authors recognize the

challenge of offering this at levels below and above the CPM approach, but offer only guiding principles. The achievement of the goals of this section may be unrealistic given the state of the underlying tools and the timetable for getting those ready for prime time.

CORE 5:

CRITIQUE 1:

Core 5 efforts are in line with NIH Roadmap and are concerned with the education of scientists, clinicians, and technologists to combine and intertwine knowledge of biology with mathematical, computational and physical knowledge in order to produce a workforce that is capable of tackling the challenges of post-Human Genome Project biology. Educational programs planned as part of CTM are outlined, and clear goals and objectives are given. An appropriate and adequate description is provided of individual graduate training programs by participating institution. This is followed by an overview of postdoctoral and technician training programs, and a description of the administrative infrastructure, and distance learning plans.

A timeline for educational deliverables is given. It is clear that the Principal Investigators are not only aware of the latest trends and solutions in that space, but are capable of implementing them and intend to make them part of the solution.

The Core 5 proposal inspires confidence that the Core 5 team, in collaboration with the whole CTM team, will succeed

CRITIQUE 2:

The essence of Core 5 is educational programs for graduate students and additional learning opportunities for post-docs. This includes new interdisciplinary degrees that span departments and institutions, and new courses that integrate specialties. Much of the groundwork and some of the courses are in place. Curricular changes, interdepartmental programs, and collaborations between the institutions, which although not entirely directed toward the CTM, have been put in place and suggest the likely success of the Centers proposed activities.

CORE 6:

CRITIQUE 1:

Core 6 efforts are in line with the Roadmap and are concerned with outreach activities. This section describes: a) web-based (portal) repository of software and related activities; b) dissemination of the CTM outputs via delivery of short-courses, tutorials and participation in conferences and meetings; c) newsletter and seminar activities, d) annual workshops and all hands meetings; e) visitors program and industry outreach; f) outreach to traditionally underserved groups and individuals. All of the described activities are appropriate and well defined and support CTM goals. A timeline for deliverables is given. It is clear that the Principal Investigators are not only aware of the outreach needs, but are capable of implementing them and intend to make them part of the solution.

The Core 6 proposal inspires confidence that the Core 6 team, in collaboration with the whole CTM team, will succeed.

CRITIQUE 2:

The Center proposes a wide range of outreach activities that include web resources, help desk, short courses, newsletter, seminars, annual scientific workshops, Center meetings, visitors program, and outreach to industry and traditionally underserved groups. These activities should more than meet the needs of the Center participants as well as members of the scientific community planning to use Center tools.

CORE 7:

CRITIQUE 1:

Core 7 efforts are concerned with administrative activities related to CTM. This section describes CTM organizational structure, administrative functions, and the chain of responsibility. It confirms that CTM will be well coordinated and capable of sustaining strong research and development enhancing integration and feedback across projects and participating institutions, in line with the NIH center needs. A timeline for deliverables is given. It is clear that the Principal Investigators are capable of running CTM in a way that is consistent with its mission and with RFA.
The Core 7 proposal inspires confidence that the Core 7 team, in collaboration with the whole CTM team, will succeed.

CRITIQUE 2:

The administrative structure for the Center appears to be appropriate to the challenge of coordinating four institutions. The major strength in this area is that the directors have experience with similar multi-institutional centers, some among the four institutions involved in the CTM. There is a mechanism in place for soliciting proposals for new Biologically Motivating Problems (BMPs), and coordinators will be appointed to each of the BMPs. In principle, this structure can provide the necessary coordination, but there is always concern in projects of this magnitude that the coordination of research projects within a BMP will be a more demanding task than anticipated. It would be good to see details of the mechanism within a BMP for starting new projects (which are not anticipated when preparing the proposal), helping existing projects stay on course, and eliminating projects which are either unproductive or no longer retaining relevance to overall Center goals. That said, the personnel involved in the Center clearly have the experience to handle this challenge.

THE FOLLOWING RESUME SECTIONS WERE PREPARED BY THE SCIENTIFIC REVIEW ADMINISTRATOR TO SUMMARIZE THE OUTCOME OF DISCUSSIONS OF THE REVIEW COMMITTEE ON THE FOLLOWING ISSUES:

VERTEBRATE ANIMALS: ACCEPTABLE
The studies proposed in Core 3, Project require the use of axolotls and zebrafish as model organisms, due to their ability for limb regeneration and wound healing. These processes are not ones shared by humans. Axolotls will be housed in the IUPUI School of Science animal facility with professional veterinary care, and zebrafish will be purchased from local pet stores and maintained in temperature-controlled water tanks. Appropriate and detailed procedures are explained for their maintenance, minimizing discomfort during the experimental procedures, and euthanasia.

COMMITTEE BUDGET RECOMMENDATIONS: The budget was recommended as requested.

NOTICE: The NIH has modified its policy regarding the receipt of amended applications. Detailed information can be found by accessing the following URL address: http://grants.nih.gov/grants/policy/amendedapps.htm

NIH announced implementation of Modular Research Grants in the December 18, 1998 issue of the NIH Guide to Grants and Contracts. The main feature of this concept is that grant applications (R01, R03, R21, R15) will request direct costs in $25,000 modules, without budget detail for individual categories. Further information can be obtained from the Modular Grants Web site at http://grants.nih.gov/grants/funding/modular/modular.htm