INVESTIGATIVE MEDICINE PROGRAM

Ph.D. Program for Physician Scientists

Graduate Student Handbook 2023-2024

INTRODUCTION

The Investigative Medicine Program is a freestanding, interdisciplinary Ph.D. granting program of the Yale Graduate School. The program provides special training in clinical investigation for highly selected physicians in clinical departments who are interested in careers in biomedical research. The overall goal of the program is to provide the training required to develop a broad knowledge base, analytical skill, creative thinking and the hands-on experience demanded of clinical researchers devoted to either laboratory-based or clinically-based patient-oriented research.
The student handbook is intended to be a source of information for graduate students studying for the Ph.D. degree in the Investigative Medicine Program at Yale. In this handbook, students and faculty will find the specific program requirements set forth by the Investigative Medicine Program. The Graduate School of Arts and Sciences Programs and Policies Bulletin is the definitive source of information about academic rules and regulations as well as general policies that apply to all graduate programs.

GRADUATE PROGRAM OFFICE

Campus Address:

Investigative Medicine Program
Yale Center for Clinical Investigation
Investigative Medicine Program
2 Church Street South, Suite 112
New Haven, CT 06519

Mailing Address: (USPS only, for courier service use above address):
Investigative Medicine Program
Yale Center for Clinical Investigation
Investigative Medicine Program
2 Church Street South, Suite 112
New Haven, CT 06519

Website: http://medicine.yale.edu/investigativemedicine

STRUCTURE OF THE INVESTIGATIVE MEDICINE PROGRAM

Program Staff

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Director and Head of Laboratory-Based Patient-Oriented Research
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Professors

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Joseph Craft, M.D., Professor of Medicine and Immunobiology
James Dziura, Ph.D., MPH, Associate Professor of Emergency Medicine and of Medicine
David Fiellin, M.D., Professor of Medicine, Public Health, and of Investigative Medicine
Thomas Gill, M.D., Professor of Medicine, Epidemiology, and of Investigative Medicine
Fred Gorelick, M.D., Professor of Medicine and Cell Biology
Jeffrey Gruen, M.D., Professor of Pediatrics, Genetics and of Investigative Medicine
Harlan Krumholz, M.D., M.Sc., Professor of Medicine, Public Health, and of Investigative Medicine
Chirag R Parikh M.D., Ph.D., F.A.C.P., Associate Professor of Medicine and of Investigative Medicine
Eugene Shapiro, M.D., Professor of Pediatrics, Epidemiology, and of Investigative Medicine
George Tellides, M.D., Professor of Surgery and of Investigative Medicine
Mary Tinetti, M.D., Professor of Medicine and of Investigative Medicine

Graduate School of Arts and Sciences

Lynn Cooley, Ph.D.
Dean, Graduate School of Arts and Sciences

Jasmina Besirevic Regan, Ph.D.
Associate Dean, Graduate Education

Robert Harper-Mangels, Ph.D.
Associate Dean for Admissions and Financial Support

Allegra di Bonaventura
Associate Dean for Academic Support

Michelle Nearon Ph.D.
Senior Associate Dean and Director of Office for Graduate Student Development and Diversity

Current Graduate Students

Christine Miller, Matriculated 2022
Lab Track, Department of Pediatrics

Nadia Solomon, Matriculated 2022
Clinical Track, Department of Radiology and Biomedical Imaging

Enock Teefe, Matriculated 2022
Lab Track, Department of Psychiatry

Natalia Festa, Matriculated 2022
Clinical Track, Department of Geriatrics

Clancy Mullan, Matriculated 2021
Lab Track, Department of Surgery

Michael Mensah, Matriculated 2021
Clinical Track, Department of Psychiatry

Tara Thompson-Felix, Matriculated 2021
Lab Track, Child Study Center

Prajwal Boddu, Matriculated 2020
Lab Track, Department of Oncology

Benjamin Lu, Matriculated 2020
Lab Track, Department of Oncology

Holly Blackburn, Matriculated 2019
Lab Track, Department of Surgery

Vadim Kurbatov, Matriculated 2019
Lab Track, Department of Surgery

Alumni

Avesta, Arman, 2023
Developing Capsule Networks for Brain Image Segmentation
Tu, Long, 2023
Strategies to Optimize Neuroimaging in the Emergency Department
Foster, Gena, 2023

Curtis, Susanna, 2020
Cannabinoids for the Reduction of Pain and Inflammation in Sickle Cell Disease
Liu, Elise, 2022
The Role and Production of Gut Food- Specific Immunoglobulin A
Shung, Dennis , 2022
Deep Risk Prediction of Embedding Patient Data: Application to Acute Gastrointestinal bleeding
Gruenbaum, Benjamin, 2020
Neuronal Mechanisms of Impaired Consciousness in an Awake Rodent Absence Seizure Model
Zhang, Yu, 2021
Modulation of T Cell Response Hierarchy by Coinhibition and Costimulation With Cancer Immunotherapy
Siebel, Stephan, 2021
Minimizing Tracer Interference to Assess in vivo Hepatic Metabolism with Glutamine-generated Mass Isotopomers
O'Neill, Kathleen, 2021
Improving Care for Survivors of Gun Violence
Mori, Makoto, 2021
Characterization and Prediction of Postoperative Recovery After Cardiac Surgery
Curtis, Susanna, 2020
Cannabinoids for the Reduction of Pain and Inflammation in Sickle Cell Disease
Gruenbaum, Benjamin, 2020
Neuronal Mechanisms of Impaired Consciousness in an Awake Rodent Absence Seizure Model
Oliveira, Carlos, 2019
Estimating the Effectiveness of Human Papillomavirus Vaccine: A Case-Control Study with Bayesian Model Averaging
Ashima Gulati, 2019
Identification of genetic variants contributing to intracranial aneurysm formation in autosomal dominant polycystic kidney disease
Branagan, Andrew, 2019
A Novel Influenza Vaccination Strategy for Patients with Plasma Cell Disorders
Gristotti, Gabriella, 2018
Decreased lymphangiogenesis alters angiogenesis and dermal remodeling in wound healing
Lee, Christopher, 2018
Timing of Therapeutic Hypothermia Initiation in Cardiac Arrest Survivors
Merola, Jonathan, 2018
Generating Human Endothelial Cells that Evade Alloimmunity
Moledina, Dennis, 2018
Targeted Cytokines as Biomarkers for Human Acute Tubulo-interstitial Nephritis
Gruenbaum, Shaun, 2017
The Role of Branched-Chain Amino Acids in Glutamate Metabolism and Seizure Modulation in a Rat Model of Mesial Temporal Lobe Epilepsy
Hammond, Christopher, 2016
Stress, Neurophysiology, Affective States, and Co-occurring Cannabis and Tobacco use in Adolescents
Ryder, Alex, 2016
Vesicular stomatitis virus vectored chimeric hemagglutinin constructs as broadly cross-reactive influenza vaccines
Belcher, Justin, 2015
Acute Kidney Injury in the Setting of Cirrhosis

Lemaire, Mathieu, 2015
Discovery of novel genes that cause rare pediatric renal diseases using exome sequencing
Marion, Chad, 2015
Chitinase 3-Like-1 (Chil1) as an Immune Modulator in Gram-negative Pneumonia
Williams, Kyle, 2015
Characterization and Treatment of Inflammation and Autoimmunity in Pediatric Obsessive Compulsive Disorder
Hoffman, Ellen, 2014
Functional Analysis of Genes Associated with Autism Spectrum Disorders in a Zebrafish Model System
Hsieh, Evelyn, 2014
Osteoporosis Among HIV-Infected Individuals in China
Kim, Sang, 2014
Characterization of CD4 T Cells That Help Memory B Cells and Their Clinical Relevance
Koraishy, Farrukh, 2014
The crosstalk of Hgf and canonical Wnt signaling in kidney repair
Popov, Violeta, 2014
Role of β -catenin in Regulating Hepatic Lipid Metabolism in NAFLD
Protack, Clinton, 2014
The role of EphB4 during arteriovenous fistula maturation
Huen, Sarah, 2013
Regulation and Function of Macrophage Polarization in Ischemic Injury
Westphal, Alexander, 2013
Clinical Characterization and Amygdala Hypothesis of Childhood Disintegrative Disorder
Das, Rita, 2012
Role of Macrophage Migration Inhibitory Factor (MIF) in the Pathogenesis of Tuberculosis
Jou, Roger, 2012
The Structural Neural Phenotype of Autism Spectrum Disorder: Heterogeneous and Distributed Abnormalities in the Social Brain and its Long-Range Connectivity
Panda, Alexander, 2012
Age Associated Alterations in Toll-like Receptor Function of Human Dendritic Cells and Correlation With Influenza Vaccine Response.
Jastreboff, Ania, 2011
Neural Response to Stress and Food Cues in Obese Individuals
Kim, Agnes, 2011
AMPK Regulation and Function in the Ischemic Heart
Nygaard, Haakon, 2011
The Role of Cellular Prion Protein in β-Amyloid Induced Neuronal Network Dysfunction in Alzheimer's Disease
Sherr, Jennifer, 2011
β before α: The importance of residual β-cell function for α-cell secretion of glucagon in response to hypoglycemia
VonKohorn, Isabelle, 2011
Predicting Which Mothers Will Go Back to Smoking After Pregnancy: An Application of the Theory of Planned Behavior
Provost, Karin, 2010
Regulation of the Airway Epithelial Responses in Asthma
Moriarty, Katie, 2009
Estrogen Receptor-Mediated Rapid Signaling in Endothelial Progenitor Cell Contributions to Ischemia
Kim, Nancy, 2008
Persistence with Oral Hypoglycemic Therapy One Year After Initiation
Mehra, Vishal, 2008
Free Fatty Acids on VEGH Signaling Abnormalities
Miller, Edward J., 2008
The Cardioprotective Effects of AMP-Activated Protein Kinase

Paglino, Justin, 2008
Understanding and Enhancing the Oncoselectivity of Minute Virus of Mice
Tang, Paul, 2008
Role of MyD88-Dependent Inflammation in Flow-Mediated Remodeling
Hassan, Hatim, 2007
Mechanisms of Regulation of Anion Exchanger SLC26A6
Liao, John, 2007
Studies in Therapeutic Vaccination and Immune Evasion by PHV Oncoproteins
VanDuin, David, 2007
Toll-Like Receptors in Older Adults and Response to Vaccination
Dorsey, Karen, 2006
Measurement of Physical Activity in Obese and Non-Obese Children
Lee, Warren, 2006
Characterization of Candidate Susceptibility Genes in Human Temporal Lobe Epilepsy with Mesial Temporal Sclerosis
Herzog, Erica, 2005
Bone Marrow Derived Stem Cells as Progenitors of Alveolar Epithelium
Hardy, Susan, 2004
Patterns, Probability and Predictors of Recovery from Disability in Activities of Daily Living among Community-Dwelling Older Persons
Lee, Samuel, 2004
Trafficking and Secretion of Virulence-associated Proteins in Candida albicans
Samuel, Varman, 2004
Mechanisms of Fat Induced Hepatic Insulin Resistance
Weiss, Ram, 2004
Metabolic Characteristics of Obese Children with Normal vs Impaired Glucose Tolerance
Ariyan, Charlotte, 2003
Strategies to Improve Islet Cell Transportation

TIMELINE FOR STUDENT PROGRESS

YEAR ONE

Required Course Work July - May
Qualifying Committee Meeting Before January 31
Comprehensive Qualifying Examination & Admission to Candidacy Before December 31 of 2nd year

YEARS TWO THROUGH COMPLETION

Minimum of two electives
Meeting of Thesis Committee at least every 6 months
Dissertation

COURSE WORK

The minimum overall course requirements for the doctorate program are nine courses. Full-time course work will extend for 12 months, starting in July. Students must enroll in a minimum of two courses in each of the first three terms: summer, fall, and spring. The majority of the course requirements are to be completed by the end of the first year of study. Elective courses are often taken in the second year, with the expectation that they be completed by the end of the second year. Electives are chosen in consultation with the student's advisor. The Director of Graduate Studies provides approval of the course selections by the students. To be eligible to take the comprehensive qualifying examination, students must achieve the grade of Honors in two courses (one course if a full-year course), have a minimum grade average of High Pass, and have completed a minimum of six courses.

When requirements are met (typically by December 31 of the second year), students submit their thesis prospectus and undertake the comprehensive qualifying examination. In order to be admitted to candidacy, students must pass both the written and oral examinations and submit a thesis prospectus which has been approved by their qualifying committee. The remaining degree requirements include completion of dissertation project, the writing of the dissertation, and its oral defense.

Course Requirements for Laboratory-Based Patient-Oriented Research

1. IMED 625 Principles of Clinical Research (summer – year 1)
2. IMED 645 Introduction to Biostatistics in Clinical Investigation (summer – year 1)
3. CBIO 601 The Molecular and Cellular Basis of Human Disease (fall & spring – year 1)
4. IMED 630 Ethical and Practical Issues in Clinical Investigation (fall – year 1)
5. CB&B 740 Clinical and Translational Informatics (fall – year 1)
6. IMED 635 Directed Reading in Investigative Medicine (spring – year 1)
7. IMED 680 Topics in Human Investigation (spring – year 1)
8. IMED 655 Writing Your First Grant Proposal (spring – year 2)
9. Elective

Course Requirements for Clinically-Based Patient-Oriented Research

1. IMED 660 Methods in Clinical Research – Part I (summer – year 1)
2. IMED 661 Methods in Clinical Research – Part II (fall – year 1)
3. IMED 630 Ethical and Practical Issues in Clinical Investigation (fall – year 1)
4. IMED 662 Methods in Clinical Research – Part III (spring – year 1)
5. IMED 680 Topics in Human Investigation (spring – year 1)
6. IMED 635 Directed Reading in Investigative Medicine (spring – year 1)
7. IMED 655 Writing Your First Grant Proposal (spring – year 2)
8. Elective
9. Elective

Grades

Course grades for regular term courses in the Graduate School are recorded as Honors, High Pass, Pass, and Fail.

Honors Requirement

To meet the minimum Graduate School quality requirement, students must maintain an overall grade average of High Pass and achieve the grade of Honors in at least one quarter of the courses (standard rules for rounding apply) taken in each of the first two years after matriculation in the Graduate School during the academic year.

The Honors requirement must be met in courses other than those concerned exclusively with dissertation research and preparation. A grade of Honors awarded at the conclusion of a full-year course in which no grade is awarded at the end of the first term would be counted twice towards this requirement. A student who has not met the Honors requirement at the end of the second and fourth terms of full-time study will not be permitted to register. In exceptional circumstances, the director of graduate studies may petition the degree committee, through the appropriate dean, that a student who has not met the Honors requirement be permitted to continue study. Such a petition should be made before the end of the second or fourth term of study in time to be considered by the degree committee at its meeting that term.

In order to achieve the minimum average of High Pass, each grade of Pass on the student's transcript must be balanced by one grade of Honors. Each grade of Fail must be balanced by two grades of Honors. If a student retakes a course in which he or she has received a failing grade, only the newer grade will be considered in calculating this average. The initial grade of Fail, however, will remain on the student's transcript. A grade awarded at the conclusion of a full-year course in which no grade is awarded at the end of the first term would be counted twice in calculating this average.

Incompletes

Arrangements between students and instructors concerning incomplete work are constrained by deadlines set by Graduate School regulations: if a student and instructor have agreed that an extension is appropriate, the student must submit a request for the Temporary Incomplete (TI) with the intended completion date signed by the instructor and the Director of Graduate Studies. The instructor will indicate the mark of TI on the grade sheet, which is submitted to the Office of the Registrar by the appropriate deadline. In a single term, only one TI is permitted. Temporary Incompletes received in an academic year must be converted to final grades by October 1 of the following academic year. If a grade is not received by the Graduate Registrar by this date, a TI will be converted to a permanent Incomplete (I) on the student's record.

Course Waivers

The program may, with Graduate School approval, waive a portion of the Ph.D. course requirement in recognition of previous graduate-level work done at Yale or elsewhere. To request a course waiver, the student must submit to the Director of Graduate Studies an outline of the course(s) taken at Yale or elsewhere for which the waiver is requested. Note that a waiver only means that a required course need not be taken, but course credit towards the degree is not granted for a waived course.

Auditing

A student who wishes to audit a course must receive permission from the instructor before enrolling as an auditor, as not all faculty permit auditors in their classes. The minimum general requirement for auditing is attendance in two-thirds of the class sessions; instructors may set additional requirements for auditing their classes.

QUALIFYING/THESIS COMMITTEE

Thesis Advisor

For new students with a defined research area and a thesis advisor already identified, the committee is selected immediately on entering the program. For undifferentiated students, the Director of Graduate Studies serves as the academic advisor until the thesis advisor is chosen. The thesis advisor must have a graduate school appointment.

Students may go to the Program Registrar or the Director of Graduate Studies for help with questions about requirements or procedures. The Program Registrar maintains records on the academic progress of the students.

Responsibility of the Qualifying/Thesis Committee

The Qualifying/Thesis Committee plays a number of roles:

1. To provide a source of scientific expertise. A major role of the Qualifying/Thesis Committee is to provide advice to the student from a variety of perspectives beyond those available from the Thesis Advisor.
2. To advise the student (and Thesis Advisor) as to the general direction of the thesis. It is not unusual, for instance, for the Qualifying/Thesis Committee to recommend a more focused and less ambitious project than that which was originally outlined.
3. To determine when the student is ready to present a prospectus or to defend the thesis.
4. To evaluate the thesis prospectus for the dissertation.
5. To provide a critical audience in front of which a student can hone skills in presenting his/her work.
6. To evaluate the progress of the thesis, and to provide feedback to the Director of Graduate Studies on a semi-annual basis.
7. To identify the topics for Directed Reading Investigative Medicine (IMED 635).
8. To conduct a comprehensive written and oral examination of the student's broad knowledge of areas related to the proposed thesis work.
9. To write a brief summary of each meeting and to send this report to the Registrar for review by the Director or Deputy Director.

Guidelines for Composition of the Committee
The student and Thesis Advisor will decide together with the Program Director or Deputy Director on the composition of the Qualifying/Thesis Committee.

The committee will consist of the Thesis Advisor and 3-4 other interdisciplinary faculty members.

- A minimum of 2 members must be from clinical departments.
- At least one member must have a primary appointment in the Graduate School.
- Two members of the committee must have a Graduate School appointment.
- No more than two members may have a primary appointment in the same section of a department.

Schedule for Qualifying and for Thesis Committee Meetings
New students must meet at least twice in the first year with their committee. Additional meetings may be needed at the discretion of the committee. It is expected that by January 31 of the first-year students are to hold their first Qualifying Committee meeting at which time the student will present his/her preliminary prospectus. Students who have been admitted to candidacy must meet with his/her Thesis Committee at 6-month intervals (at a minimum) throughout the duration of his/her training. The meetings will be scheduled in advance. The Thesis Advisor serves as chair of the committee and all committee meetings, except for the Prospectus meeting. The Thesis Advisor will report to the Program Registrar and the Director of Graduate Studies about the outcome of each meeting, including progress of the student and any recommendations by the committee.

PREPROSPECTUS MEETING

The Preprospectus Meeting is typically held by January 31 of the first year. In advance of the meeting, the student should prepare a preliminary prospectus (aims, background and studies/plans to date) of the proposed project and distribute to the committee members. The format for the preliminary prospectus should follow the format of the written prospectus described under the Prospectus section.

The student must prepare a written preprospectus, to be submitted to the IMP office and members of the thesis committee at least one week prior to the preprospectus meeting, after review by the primary mentor.

The preprospectus proposal should be one-page (single-spaced, 1-inch margins, 11-point Arial, or Helvetica font) in length, and include the following:

- Title, and student and mentor's name
- Introduction (1 or 2 paragraphs) outlining the general problem to be studied and relevant background.
- Specific aims
- Brief description (1 or 2 paragraphs) of methods of approach to be undertaken.
- 3-4 pertinent references

Format of Preprospectus Meeting
The Preprospectus meeting should begin with a brief meeting, approximately 10 minutes, of the Qualifying Committee, in the absence of the student. The thesis advisor serves as chair of the committee. The student will then begin with a 20–30-minute presentation of the proposed research. Following the presentation, the committee then has an opportunity to discuss the project, addressing issues such as feasibility, scope, suitability and scientific importance.

At this meeting, the reading topics for the Directed Reading Course (IMED 635) are assigned to the student by the committee. The topics are chosen by the committee, advisor and student. Typically, one topic is assigned to each committee member (except the advisor) and the student will read about 10-20 primary and appropriate review articles with that member. The student picks the articles and sends to the committee member for his/her approval

before embarking on the reading; the student then meets twice with the committee member between the Preprospectus and the Prospectus meeting to discuss articles.

A rough timetable for the Prospectus Meeting, which is also the qualifying exam for candidacy, should be determined. The Prospectus meeting should be about 3-4 months after this Preprospectus meeting and after the student has had time to read.

The student should write a brief report summarizing the Preprospectus Meeting and forward to the advisor for approval. The approved report should then be emailed to the Program Registrar as a record of the meeting. The report should include the committee's opinion of the thesis project, the topics on which the student will examined at the Prospectus Meeting and the timetable for the Prospectus meeting.

FORMAT/STRUCTURE FOR DIRECTED READING COURSE

The following is the required format/structure of the reading period for Investigative Medicine students.

- A minimum of three sessions is required, to be completed before the qualifying exam.
- The topics, all of which should have relevance to the proposed thesis work, are initially chosen by the student in consultation with the thesis advisor and approved by the qualifying committee at time of the preliminary prospectus meeting.
- One topic is to be assigned to each qualifying committee member, excluding the thesis advisor. The sessions are divided approximately equally among the members of the qualifying committee.
- The student is to compile a list of 10 – 20 papers on each of the topic areas assigned. The list of suggested papers is to be sent to the committee member with whom the student will review that topic. The committee member can then approve the list, as well as make substitutions or deletions. Once the papers are approved, a time to meet can be decided upon.
- Students will keep a record of session dates and papers reviewed. The suggested general format is available from the program registrar. The completed reading list will be submitted by the end of the spring term to the Investigative Medicine Program office.

ADMISSION TO CANDIDACY

Admission to Candidacy indicates that the student is prepared to conduct original and independent research. It is expected that students be admitted to candidacy by December 31 of the second year. The student will be admitted to candidacy when he/she has fulfilled the requirements below. The Qualifying Committee will vote to admit the student to candidacy during the comprehensive oral examination. Once the student has been admitted to candidacy, the Qualifying Committee will then become the Thesis Committee.

- Course Requirements
- Honors Requirement
- Comprehensive Qualifying Examination
- Thesis Prospectus

COMPREHENSIVE QUALIFYING EXAMINATION

By December 31 of the second year, students are expected to take their qualifying examination. The qualifying examination is comprised of two components: 1) written examination and 2) oral examination and defense of thesis prospectus. Typically, students should complete the oral examination first and the written examination second. The goal of the written examination is to evaluate the student's ability to investigate a topic using the available knowledge base. For the oral examination, students must present the preliminary hypothesis and direction of their proposed

research project. The goal of the oral examination is to assess knowledge specific to the proposed thesis work through the defense of the prospectus.

The grade for the qualifying examination will be a "Pass" or "Fail". For a truly outstanding performance, a pass with "Distinction" shall be given. The grade will be decided based upon the quality of the written examination and the performance on the oral examination. Students who receive a grade of "Fail" will be given one further opportunity to retake the qualifying examination. Unacceptable performance on the re-examination will result in a review of the student's candidacy.

Students may receive a conditional pass if the Qualifying Committee feels that there is one particular area from the examination that needs additional work although the student demonstrates competency in all other areas. Correction of a deficient area will be accomplished, for example, by a written paper from the student or repeat oral questions by the committee chair within 2 weeks following the examination. Upon successful completion, the student will be granted a pass.

Comprehensive Written Examination

The student's Qualifying Committee will evaluate the student's general knowledge. Students will be given several questions in their area of research written by members of their committee. They will select two questions and will have 20 working days to prepare a written response. The questions will require the student to formulate a hypothesis, and then write a 6-page proposal in the format of an NIH application, including Specific Aims, Background and Significance, Preliminary Data, Research Design and Methods, and Bibliography (the latter will not count towards the 6-page limit). The response will be reviewed by the writer of the question and by the Director or Deputy Director and will be graded either pass/fail.

Comprehensive Oral Examination and Defense of Thesis Prospectus

The oral exam and defense are completed in one session. The Qualifying Committee has the responsibility of evaluating the thesis prospectus for the dissertation and of assessing the adequacy of preparation of the student in the more specific knowledge necessary to conduct the research for the thesis.

The oral exam begins with the student's defense of the prospectus. After the presentation, the student is questioned on topics related to the prospectus and related to the directed readings with a focus on how the reading has been integrated in the prospectus. The Thesis Advisor must be present for the Comprehensive Oral Examination. The role of the Thesis Advisor will be that of an observer rather than active participant. One member of the Qualifying Committee will serve as chair; he/she will be responsible for communicating the results of the oral examination to the Director of Graduate Studies and Program Registrar. After the defense and oral examination, the student leaves the room for the Qualifying Committee's discussion and vote about candidacy.

Format for Thesis Prospectus

The student must prepare a written prospectus using the format of a grant proposal. It should not cumulatively exceed 13 pages (single-spaced, 1- inch margins, 11-point Arial or Helvetica font) in length. The prospectus is to be submitted to the IMP office at least one week before the date of the oral examination and simultaneously distributed to all members of the thesis committee; it should be reviewed with the primary mentor before submission. The written proposal should have the following sections:

1. Title Page
2. Specific Aims (1 page or less): A concise statement of the general problem under study and the explicit goals of the project.
3. Background and Significance (no more than 3 pages): This section should place the proposal in context and describe the system in a manner intelligible to a non-specialist. This should include a brief, but critical, evaluation of the relevant literature and a description of how your research project will advance knowledge in the field.
4. Progress to date (2-3 pages): Description of the preliminary data and your interpretation of the data generated.

5. Proposed Research Plan (3-4 pages): Outline the research envisioned at this time and indicate how they will help you attain the overall goals of the project. Acknowledge pitfalls and limitations of your experimental approach, and if possible, suggest alternative strategies.
6. References: Should be included at the end (not counted in the page limit). If you wish, you can also include up to two additional pages of diagrams, figures or tables, with appropriate brief legends, as long as these are referred to in the text.

DISSERTATION

The most important part of the Ph.D. program is research training, culminating in the writing of a dissertation. The dissertation should demonstrate the student's mastery of relevant resources and methods and should make an original contribution to knowledge in the field.

It is the responsibility of each qualified student to petition the Graduate School for the Ph.D. degree at the appropriate times. Dissertations must be submitted to the Graduate School by March 16 to ensure consideration for the May degree and by October 1 for the December degree. Students should consult with the Director of Graduate Studies and Program Registrar to make sure they have satisfied all requirements.

Preparing and Submitting the Dissertation

Before beginning to write the dissertation, students should download the "Dissertation Submission Checklist" from the Graduate School website. This checklist provides step-by-step instructions for preparing and submitting the dissertation. In addition, students should notify the Program Registrar of their intent to submit the dissertation so that the program can initiate the assignment of dissertation readers (see Readers of the Dissertation below).

Format of Dissertation

The dissertation must describe original research making significant new contributions to knowledge. Its form and content should be of the quality expected of papers submitted to major scientific journals. The booklet entitled "Preparation and Submission of the Doctoral Dissertation" addresses the aspects of the dissertation format that are required by the Graduate School. It may take several months to write the dissertation, or less time for students who have already published papers that will form the core of the dissertation. Published papers can be reformatted to form the core of the dissertation (including changing all the "we's" to "I's"), but the student must be the principal writer of the papers. Remember that the dissertation must be a scholarly work of your own creation. If another person obtained a result shown in the dissertation, then the contribution of this person must be explicitly acknowledged, and the amount of the work of others that constitutes the meat of the dissertation should be limited. If you wish to copyright your dissertation, you must obtain permission to reproduce any published materials (even if your own) in your dissertation. You should draw your own illustrative diagrams rather than using published ones with minor modification.

Defense of Dissertation

The student must give an oral defense of the dissertation when the student's Thesis Committee is satisfied that the work is complete, and the student has a complete draft of the dissertation ready to submit to the Graduate School. The complete draft must be provided to the Thesis Committee two weeks before the defense takes place.

The defense consists of a presentation of the results followed by a question and discussion period, which is open to anyone in the Program and guests.

Submission of Dissertation

The dissertation should be submitted to the Graduate School as soon as the defense has been passed and any final corrections to the dissertation have been made. This must be completed within one month of passing the defense. A copy of the final dissertation must also be given to the Program Registrar. After the dissertation has been submitted, copies are sent to the readers (see below), who read the dissertation and complete a Reader's Report

form. When all the Readers' Reports are in, they are given to the Director of Graduate Studies to recommend the degree. The recommendation is then forwarded, along with the reader's reports, to the Graduate School's Degree Committee. The Degree Committee reviews and votes on the conferral of the degree.

Readers of Dissertation

The Graduate School requires that the Dissertation be read by at least 3 readers, with at least 2 having ladder or ladder-track positions at Yale. An outside reader is not required by the Graduate School. Investigative Medicine requires that the Dissertation be read by the Thesis Committee, excluding the thesis advisor. After all reader evaluation forms have been returned to the Graduate School and all requested changes to the dissertation have been made, the DGS will sign the form recommending award of the Ph.D. degree. Then the Graduate School Degree Committee and finally the Yale Corporation will vote to approve conferral of the degree.

REGISTRATION

General Information

Graduate School registration is required of all students, whether engaged in course work, preparing for qualifying examinations, or dissertation research in residence or in absentia. Students who fail to register for any term, in which the student has not been granted a leave of absence, will be considered to have withdrawn from the Graduate Program. Privileges associated with registered status will be withdrawn.

No student may register for any term unless he or she is making satisfactory progress toward the degree. Satisfactory progress means that the student has met the program requirements set forth in the timeline for student progress and has met the requirements set by the Graduate School.

Students who fail to meet the program or Graduate School requirements by the designated deadlines, and students who have been admitted to candidacy who fail to submit the annual Dissertation Progress Report, will be administratively withdrawn.

Tuition and Continuous Registration Fee

All students are charged four years (eight terms) of full tuition. Students who have met the full-tuition obligation are charged a Continuous Registration Fee (CRF) of $737 per term.

Leave of Absence

Students in good standing who wish to interrupt their study temporarily for personal reasons may, with approval of the director of graduate studies and the associate dean, be granted a leave of absence (see Graduate School *Programs and Policies*).

MISCELLANEOUS INFORMATION

Dissertation Progress Report
Students must maintain a satisfactory rate of progress toward the Ph.D. to remain in good standing in the Program. The Graduate School requires that students who have been admitted to candidacy complete an annual dissertation progress report (DPR). The report maps out their achievements in the past year, goals and timeframe for the upcoming year.

Each year in April, students who have been admitted to candidacy but not yet petitioned for the Ph.D., will receive an email from the Graduate School notifying them to complete the online DPR. Once completed, the thesis advisor and the DGS will be notified to review and approve the report. A student who fails to complete the DPR at the end of the spring term will not be allowed by the Graduate School to register in the following fall term.

Policy on K Awards
Students enrolled in the Investigative Medicine Program are permitted to apply and hold K awards from the National Institute of Health by waiving the institutional requirement that K Award recipients have faculty appointments. This waiver applies only to students in the Investigative Medicine Program and only for the period during which they are enrolled as full-time graduate student.

Policy on Faculty Titles
It is a Graduate School regulation that a graduate student is not permitted to hold a faculty position/title and be a student at the same time.

Policy on Part-Time Employment
Study toward the Ph.D. degree is expected to be a full-time activity. For the Investigative Medicine Program, it is recommended that clinical duties be restricted to ½ day per week, this counts as part of your educational activities. In addition, it is a Graduate School policy that part-time employment be restricted to an average of ten hours per week (additional clinical duties beyond ½ day clinic, such as moonlighting).

Language Requirement
There is no foreign language requirement for students.

Departmental Responsibilities
Students are expected to participate fully in all academic activities of their chosen graduate school discipline, including journal clubs, lab meetings, research-in-progress sessions, and departmental retreats.

Training Activities

Students are expected to attend and participate in the following training activities for the entire time they are in the Program.

1. Research in Progress: On a monthly basis, the program's faculty and students will meet. One student will be selected each month to present research in progress to the program faculty and students. Each student is expected to present yearly in this forum. The presentation should be in the form of a PowerPoint presentation. All students are expected to attend.

2. The dissertation defense of fellow students in the program.

Appendix

Appendix A.

Academic Calendar 2023-2024 / Schedule of Academic Dates and Deadlines

Graduate School Academic Calendar Home Page

Appendix B.
Timetable Prospectus and Qualifying Exam

1. Upon matriculation student and mentor to receive Preprospectus Prospectus Procedures
2. Mid-Semester, term 1 following matriculation student and mentor to determine committee composition.
3. Once Committee is composed, committee members, student and mentor will receive Preprospectus Prospectus Procedures
4. Two weeks prior to exam all committee members will receive Preprospectus Prospectus Procedures and Qualifying Examination Overview.
5. Following meeting committee chair will submit qualifying examination report

Appendix C.
Preprospectus Prospectus Procedures

Guidelines for Composition of the Committee

The committee will consist of the Thesis Advisor and 3-4 other interdisciplinary faculty members.

- A minimum of 2 members must be from clinical departments.
- At least one member must have a primary appointment in the Graduate School.
- Two members of the committee must have a Graduate School appointment.
- No more than two members may have a primary appointment in the same section of a department.

Schedule for Qualifying and for Thesis Committee Meetings

New students must meet at least twice in the first year with their committee. Additional meetings may be needed at the discretion of the committee. It is expected that by January 31 students are to hold their first Qualifying Committee meeting at which time the student will present his/her preliminary prospectus.

Students who have been admitted to candidacy must meet with his/her Thesis Committee at 6-month intervals (at a minimum) throughout the duration of his/her training. The meetings will be scheduled in advance. The Thesis Advisor serves as chair of the committee and all committee meetings, except for the Prospectus meeting. The Thesis Advisor will report to the Program Registrar and the Director of Graduate Studies about the outcome of each meeting, including progress of the student and any recommendations by the committee.

Preprospectus Meeting

The Preprospectus Meeting is typically held by January 31 of the first year. The student must prepare a written preprospectus, to be submitted to the IMP office and members of the thesis committee at least one week prior to the preprospectus meeting, after review by the primary mentor.

The preprospectus proposal should be one-page (single-spaced, 1-inch margins, 11-point Arial, or Helvetica font) in length, and include the following:

- Title, and student and mentor's name
- Introduction (1 or 2 paragraphs) outlining the general problem to be studied and relevant background.
- Specific aims
- Brief description (1 or 2 paragraphs) of methods of approach to be undertaken.
- 3-4 pertinent references

Meeting Format

The Preprospectus meeting should begin with a brief meeting, approximately 10 minutes, of the Qualifying Committee, in the absence of the student. The thesis advisor serves as chair of the committee. The student will then begin with a 20–30-minute presentation of the proposed research. Following the presentation, the committee then has an opportunity to discuss the project, addressing issues such as feasibility, scope, suitability, and scientific importance.

At this meeting, the reading topics for the Directed Reading Course (IMED) are assigned to the student by the committee. The topics are chosen by the committee, advisor, and student. Typically one topic is assigned to each committee member (except the advisor) and the student will read about 10-12 primary

and appropriate review articles with that member. The student picks the articles and sends to the committee member for his/her approval before embarking on the reading; the student then meets once or twice with the committee member between the Preprospectus and the Prospectus meeting to discuss articles.

A rough timetable for the Prospectus Meeting, which is also the qualifying exam for candidacy, should be determined. The Prospectus meeting should be about 2-4 months after this Preprospectus meeting and after the student has had time to read.

The thesis advisor should write a brief report summarizing the Preprospectus Meeting and email to the program registrar as a record of the meeting. The report should include the committee's opinion of the thesis project, the topics on which the student will be examined at the Prospectus Meeting and the timetable for the Prospectus meeting.

Format/Structure for Directed Reading Course

The following is the required format/structure of the reading period for Investigative Medicine students.

- A minimum of three sessions is required, to be completed before the qualifying exam.
- The topics, all of which should have relevance to the proposed thesis work, are initially chosen by the student in consultation with the thesis advisor and approved by the qualifying committee members at time of the preliminary prospectus meeting.
- One topic is to be assigned to each qualifying committee member, excluding the thesis advisor. The sessions are divided approximately equally among the members of the qualifying committee.
- The student is to compile a list of 10 – 20 papers on each of the topic areas assigned. The list of suggested papers is to be sent to the committee member with whom the student will review that topic. The committee member can then approve the list, as well as make substitutions or deletions. Once the papers are approved, a time to meet can be decided upon.
- Students will keep a record of session dates and papers reviewed. The suggested general format is available from the program registrar. The completed reading list will be submitted by the end of the spring term to the Investigative Medicine Program office.

Admission to Candidacy

Admission to Candidacy indicates that the student is prepared to conduct original and independent research. It is expected that students be admitted to candidacy by December 31 of the second year. The student will be admitted to candidacy when he/she has fulfilled the requirements below. The Qualifying Committee will vote to admit the student to candidacy during the comprehensive oral examination. Once the student has been admitted to candidacy, the Qualifying Committee will then become the Thesis Committee.

- Course Requirements
- Honors Requirement
- Comprehensive Qualifying Examination
- Thesis Prospectus
- Training in the Responsible Conduct of Research (IMED 630)

Comprehensive Qualifying Examination

By December 31 of the second year at the latest, students are expected to take their qualifying examination. The qualifying examination is comprised of two components: 1) written examination and 2) oral examination and defense of thesis prospectus. The goal of the written examination is to evaluate

the student's ability to investigate a topic using the available knowledge base. For the oral examination, students must present the preliminary hypothesis and direction of their proposed research project. The goal of the oral examination is to assess knowledge specific to the proposed thesis work through the defense of the prospectus.

The grade for the qualifying examination will be a "Pass" or "Fail". For a truly outstanding performance, a pass with "Distinction" shall be given. The grade will be decided based upon the quality of the written examination and the performance on the oral examination. Students who receive a grade of "Fail" will be given one further opportunity to retake the qualifying examination. Unacceptable performance on the re-examination will result in a review of the student's candidacy.

Students may receive a conditional pass if the Qualifying Committee feels that there is one particular area from the examination that needs additional work although the student demonstrates competency in all other areas. Correction of a deficient area will be accomplished, for example, by a written paper from the student or repeat oral questions by the committee chair within 2 weeks following the examination. Upon successful completion, the student will be granted a pass.

Comprehensive Written Examination

The student's Qualifying Committee will evaluate the student's general knowledge. Students will be given several questions in their area of research. They will select two questions and will have 10 working days to prepare a written response. The questions will require the student to formulate a hypothesis, and then write a 6-page proposal in the format of an NIH application, including Specific Aims, Background and Significance, Preliminary Data, Research Design and Methods, and Bibliography (the latter will not count towards the 6-page limit). The response to each question is to be reviewed by the committee member that wrote the question and will be either pass/fail.

Comprehensive Oral Examination and Defense of Thesis Prospectus

The oral exam and defense are completed in one session. The Qualifying Committee has the responsibility of evaluating the thesis prospectus for the dissertation and of assessing the adequacy of preparation of the student in the more specific knowledge necessary to conduct the research for the thesis.

The oral exam begins with the student's defense of the prospectus. After the presentation, the student is questioned on topics related to the prospectus and related to the directed readings with a focus on how the reading has been integrated in the prospectus. The Thesis Advisor must be present for the Comprehensive Oral Examination. The role of the Thesis Advisor will be that of an observer rather than active participant. One member of the Qualifying Committee will serve as chair; he/she will be responsible for communicating the results of the oral examination to the Director of Graduate Studies and Program Registrar. After the defense and oral examination, the student leaves the room for the Qualifying Committee's discussion and vote about candidacy.

Format for Thesis Prospectus

The student must prepare a written prospectus using the format of a grant proposal. It should not cumulatively exceed 13 pages (single-spaced, 1- inch margins, 11-point Arial or Helvetica font) in length. The prospectus is to be submitted to the IMP office at least one week before the date of the oral examination and simultaneously distributed to all members of the thesis committee; it should be reviewed with the primary mentor before submission. The written proposal should have the following sections:

1. Title Page (also include student's and mentor's names)
2. Specific Aims (1 page or less): A concise statement of the general problem under study and the explicit goals of the project.
3. Significance (3-4 pages): This section should place the proposal in context, highlighting its significance and describing the system in a manner intelligible, including to a non-specialist. This should include a brief, but critical, evaluation of the relevant literature and a description of how your research project will advance knowledge in the field.
4. Progress to date (3-4 pages): Description of the preliminary data and your interpretation of the data generated.
5. Proposed Research Plan (3-4 pages): Outline the research envisioned at this time and indicate how they will help you attain the overall goals of the project. Acknowledge pitfalls and limitations of your experimental approach, and if possible, suggest alternative strategies.
6. References: Should be included at the end (not counted in the page limit). Figures, tables, and diagrams should be included in the text as needed for explication; figure legends can be 10-point font.

Appendix D.
Prospectus Chair Letter Sample

Dear Dr. [Committee Chair]:
As a reminder, please send a written summary stating the results of XXXX's oral qualifying exam.

The summary should state that the student has taken the oral qualifying examination, the date, the committee members present, the topic, and the outcome of the examination.

Please copy the committee members, the student, Dr. Craft, and me. A sample summary is attached outlining the required information.

Summary sample of committee chair's report on qualifying examination, to be sent to committee chair.

Program Registrar
Investigative Medicine Program
imp@yale.edu

Dear Ms./Mr. Registrar:

The oral exam for *Name* was held on *Date*. Committee members present included me, and list names of members present. (Also list any members absent.)

Name presented plans for *his/her* dissertation research on *Dissertation Title*. *They* successfully answered questions from the committee, as well as questions that assessed knowledge specific to the proposed thesis work.

Student Name passed the oral qualifying exam unconditionally.

Signed,

Committee Chair

cc: *Each committee member*
Student Name
Joseph Craft, Director and DGS, IMP
Eugene Shapiro, Associate Director, IMP

Appendix E
Qualifying Examination Overview

RE: Student
Qualifying Examination
Investigative Medicine Program

Dear Committee Members' names:

I'm writing to provide a brief overview of the Investigative Medicine Program's process and procedures of the qualifying exam and admission to candidacy for Student. A detailed copy of the process and procedures in its entirety is attached.

The prospectus meeting (oral exam) follows the same format as the pre-prospectus meeting, except Student will hand out his/her prospectus plans; hypothesis, aims, background, preliminary data if any, research plan and references - about 13 pages in all.

The exam begins with Student's defense of his/her dissertation prospectus (the slides and talk should be limited to around 30 minutes, to leave ample time for questions and discussion). Following the presentation, the committee should ask questions on areas and topics pertaining to the dissertation prospectus and readings. Questions and discussion throughout the presentation are also appropriate. After the question and discussion period, Student will leave the room and the committee will hold a discussion to determine if she/he is prepared to proceed with his/her dissertation research.

One member of the committee (not the thesis advisor) should be elected as chair prior to the meeting and is responsible for conducting the meeting and keeping any minutes. This is the only committee meeting the advisor does not chair. The Thesis Advisor is responsible for communicating the results of the oral exam to Dr. Joseph Craft, IMP Director and DGS, Eugene Shapiro, IMP Associate Director and myself (I will send you a template for this letter).

At the oral exam, the role of the thesis advisor is that of an observer rather than an active participant. The advisor can provide clarifications where appropriate but should not answer questions unless and until the student is given a chance to answer them first.

In addition to the oral exam, students in Investigative Medicine have a written exam. The goal of the written examination is to evaluate the student's ability to investigate a topic using the available knowledge base.

Each committee member, less advisor, is requested to write two questions that are hypothesis driven and based upon the readings. Student will then select two questions to write on — a six or so page answer, in the form of a grant proposal. She/he will have 20 business days to prepare a written response. The response to each question is be read and approved by the committee member that wrote the question. Example student exam questions previously distributed are attached.

The grade for the qualifying examination (oral and written) is Pass or Fail and will be decided based upon the quality of the written examination and the performance on the oral examination. For a truly outstanding performance, a pass with Distinction shall be given.

Students who receive a grade of Fail will be given one further opportunity to retake the qualifying examination. Unacceptable performance on the re-examination will result in a review of the student's candidacy.

If you have any questions, please let me know.

Thank you,

Program Registrar
Investigative Medicine Program
imp@yale.edu

cc: Student
Joseph Craft, Director and DGS, IMP
Eugene Shapiro, Associate Director, IMP

Appendix F
Defense Procedures

Dear *Thesis Advisor*:

Student's name defense is scheduled for *date, time*. It will be held in *Place*. Outlined below is the general format for the defense and process following.

Prior to the Defense

· The thesis committee should approve the request for the student to defend.
· The student should provide a written version (or .pdf document if preferred by the committee) of the thesis to each committee member 2 weeks prior to the defense date.
· Each committee member should read the thesis draft, be prepared to ask questions of the student at the defense and offer suggested corrections so that the student can submit the final, edited version of the thesis to the graduate school after the formal defense, assuming committee approval (see below: Process Following Defense).

Defense Format

· The thesis advisor welcomes the audience, announces the event (thesis defense of *Student's name*), provides a brief background of the student and his/her accomplishments during thesis work, and describes that the defense will proceed according to the following format
· Student presents a summary of the thesis work (45–50 minutes).
· Questions are asked by the general audience and questions of a broad nature may be asked by members of the Thesis Committee (10–15 minutes).
· After the presentation and questions, the general audience will be asked to leave.
· The student will then meet with members of the thesis committee. Questions may be asked beyond those asked at the presentation. The student may be asked to leave the room while the thesis committee holds a discussion and vote.

Mentor, during the introduction you may want to include that a celebration of cake and soft drinks will be held after the defense. (This is assuming committee approval. The department sponsoring the student typically provides refreshments; please specify the place and approximate time of the celebration.)

Process Following Defense

After the defense has been passed, the mentor will send a brief note to me, with ccs to Drs. Craft and Shapiro, that the defense of *Student's name* was successful. *Student's name* will then submit his/her dissertation to the Graduate School. This must be completed within one month of passing the defense.

Each reader will receive an email invitation from the Dissertation Submission Office with a web link to provide you access to the Online Reader's Report (ORR). As part of the dissertation evaluation, readers are asked whether the dissertation contains significant errors in typing, grammar, spelling, reference citations or other textual matters.

Readers are expected to submit their reports within 30 days of receipt of the email.

If you have any questions regarding the process, please let me know.

Thank you,

Registrar
Investigative Medicine Program

cc: *PLEASE CC EACH COMMITTEE MEMBER And STUDENT*
Dr. Joseph Craft
Dr. Eugene Shapiro

Appendix G.
Sample Laboratory Based Written Exam Questions

Student 1
Project: Studies in Therapeutic Vaccination and Immune Evasion by HPV Oncoproteins

Question 1: A novel isoform of the human EGF receptor has been identified, i.e., sEGFR p110. This protein product arises via alternative splicing of a transcript arising from the EGFR gene. The protein product encoded by this transcript contains all of the ligand-binding sequence of this receptor, and a unique 78 amino acid carboxy-terminal sequence. Antibodies directed toward this unique carboxy-terminal sequence do not cross react with any other known proteins, including the full-length EGF receptor. Using tissue/tumor microarrays and immunocytochemistry a large number (> 400) cervical carcinomas have been analyzed for p110 sEGFR expression. Kaplan-Meier curves based on these expression data indicate that on average, women with higher levels of p110 sEGFR expression (than seen in normal cervical tissue) live, six months longer than women with the same or lower levels of p110 sEGFR expression. Cervical tissues/tumors are also known to express full-length EGF receptors, but there is discordance in the literature regarding the prognostic significance of this marker. What is known about the role of the EGFR/ErbB family in the etiology/progression of cervical cancer? Is there a relationship between EGFR and HPV-induced tumorigenicity? If so, how might p110 sEGFR be implicated in this process, and how might you test this hypothesis? Is there a need for improved methods of stratifying cervical cancer patients for treatment? Is there a need for the development of new treatment targets and/or prognostic markers in the management of this disease? Why might there be discordance in the current literature regarding the utility of EGFR expression as a prognostic marker in these patients? How might the discovery of this new receptor isoform be used to approach one (or more) of those important clinical questions?

Question 2: What is the clinical/immunological evidence that immunotherapy of human papillomavirus diseases may be effective? What are the limitations to the currently available animal models for immunotherapy of papillomavirus-induced disease, and how can they be improved?

Question 3: It is known that many people in areas endemic for Malaria do not develop long-term acquired immunity and will experience multiple malaria infections every year. You have developed an animal model to study acquired immunity to malaria. You find that upon the first infection of animals with a mouse adapted strain of plasmodium yalie ~ 50% control the infection and survive, but the other half succumb to the infection. You find that the group that survived the first infection, when infected a second time, again only 50% survive the infection. Upon a third infection you find that less than 30% survive the infection and that with a fourth infection all the animals die. Please provide two hypotheses that may explain this series of results and at least two experiments per hypothesis that you would use to test your ideas.

Student 2
Project: Genetic Signatures in Human Temporal Lobe Epilepsy

Question 1: Discuss methodological and conceptual issues on the application of expression microarray technology to research on the human brain. The following is a suggested outline:

1. The nature of human brain tissue:
   a. Sources of tissue.
   b. Cellular heterogeneity.
   c. Complexity of gene expression (e.g., some have estimated that 60% of the genes in the genome are expressed at some point in the brain).

2. RNA-related issues:
   a. What procedures are necessary for evaluating RNA quality and quantity?
   b. What strategies will maximize RNA yield and quality?
   c. Discuss advantages and disadvantages in different approaches to preparation of complimentary DNA and RNA. Example: poly-T vs. random primer-based reverse transcription.
3. Sources of variability in micro-array data:
   a. Hybridization-related issues: do you need to optimize stringency? If so, how. Are there other sources of variability related to the hybridization step?
   b. What impact, if any, does variation in amount of RNA (or derivative molecules) applied to the array have on the relative strengths of signals?
   c. Is chip-to-chip variability an issue?
4. Signal-to-noise issues:
   a. How does one identify "true" differences in mRNA levels?
   b. What statistical approaches can be applied to help evaluate the significance of possible differences?
   c. How does one minimize type I and type II errors? (In other words, how sensitive and specific are microarrays for detecting true changes in gene expression)?
5. Interpretation and biological importance:
   a. How can the problem of cell-type specificity be addressed when the material derives from a heterogeneous tissue such as brain?
   b. What procedures are available to confirm findings made with arrays?
      - Message level
      - Protein level
      - Cell level

Question 2: Using microarray results to generate hypotheses: Consider the following vignette:
In their recent full-length Nature article, Lee and colleagues (2005) compared mRNA expression in cingulotomy specimens removed from patients with intractable epilepsy. Half of their sample set came from patients with clear anatomic and histological evidence of mesial temporal sclerosis, and the other half had histologically normal-appearing tissue. Remarkably, the comparison groups were perfectly matched for age, sex, race, duration of illness, and medication history, and all had experienced their last seizure precisely 6 days prior to surgery. Both the surgical and laboratory dissections were extremely precise. Using a micro-array approach, the investigators noticed a 10-fold change in levels of message encoding RINP-I (really interesting neuronal protein-I, a member of the potentially Nobel-winning superfamily). The direction of the change will soon be reported by the team (ie, Warren—you choose the direction). Further, they confirmed the difference using RNase protection assays, and other quantitative methods. In-situ hybridization and immunocytochemistry established neuronal specificity, and localized the change to CP (coolest possible) neurons, which have long been hypothesized to function differently in MTE (+) and MTE(-) epileptic brains.

In the discussion section, they elaborated 3 hypotheses regarding biological mechanisms or implications of their results. They proposed one hypothesis each at the three levels of the central dogma of molecular biology (i.e., DNA□RNA□Protein).

Formulate and defend one such hypothesis for each level (ie., inheritance of DNA, transcription of RNA, and translation/function of protein), and suggest experiments (in humans or experimental models) to test each one. The hypotheses may address either mechanisms mediating the observed differences, or functional implications of the results (make any necessary elaborations to the data necessary to answer the question).

Student 3
Project: Mechanisms of Regulation of Anion Exchanger SLC26A6

Question 1: By genetic means, you have identified the gene for a new transporter thought to be involved in HH re-absorption in the proximal tubule. The gene is predicted to have 12 trans-membrane domains. The 23 residue N-terminus (KLYPFLRRGSTSDYSHPKKIMDE) is predicted to be in the cytosol and the 12 residue C-terminus (DESGPRKIGKKGK) is predicted to be in the lumen. There is a 87 residue stretch predicted to form an extracellular loop, between TMD 4 and 5, and a 56 residue stretch predicted to form a cytoplasmic loop between TMD 7 and 8. All the other loops connecting TMD are 14 residues or less.

You hypothesize that the HH transporter is localized to a pool of vesicles which are stored in the apical domain of proximal tubular epithelial cells. You also propose that vesicles are stimulated to fuse with the apical plasma membrane when the small molecule IMP03 is added to the apical surface at >10 micromolar.

You also propose that the HH transporter is internalized from the apical plasma membrane back to the internal pool of receptors (e.g., it is not degraded). The process occurs in constitutive fashion, assuming that you as yet have no reagents to this transporter (but will develop them).

Choose one of the following projects and write an NIH style proposal to:

1. Investigate the localization of the transporter, the kinetics of exocytosis in response to IMP03, the kinetics and mechanism of constitutive internalization, and the fate of the internalized receptor. Assume that you are testing the hypotheses outlined above.
2. Investigate the interaction of HH with other proteins in the epithelial cell, to specifically test the hypothesis that protein-protein interactions with one of the HH cytoplasmic domains is required for vesicle exocytosis. Provide three methods for identifying interacting proteins, and for testing the involvement of these proteins in exocytosis.
3. Investigate the role of HH in normal physiology, by using at least two different techniques for knock out/silencing of the HH gene in mice.

Student 4
Project: The Cardioprotective Effects of AMP-Activated Protein

Question 1: Positron emission tomographic studies have demonstrated that myocardial glucose uptake in increased in patients with congestive heart failure compared to healthy individuals. Work from your lab suggests that AMP-activated protein kinase expression is downregulated in failing hearts. Propose a mechanism by which the decrease in expression of AMPK increases glucose uptake in the failing heart and describe experiments and animal models that will be used to test this hypothesis and that will determine if modulation of AMPK activity may be a useful therapeutic target for the treatment of heart failure. Write a 6-8-page proposal in the format of an NIH application.

Question 2 (Mechanism of action): You have been asked to serve as a consultant for a small biotech company whose name is Metab Inc. They are interested in developing some novel compounds to enhance insulin sensitivity in type 2 diabetic patients. They have several potential targets in mind. Give your opinion on each one:

1. Adiponectin Analogues
2. AMP Kinase Activators

3. PPAR 〈 Activators
4. PPAR ???Activators

Please discuss all of the below points (Points 1-3 below can form the background section of your proposal):

1. Theoretical mechanisms by which each agent would improve insulin sensitivity in type 2 diabetic patients and specify which organs would be involved.
2. The potential long-term salutary effects of each compound.
3. Potential Side effects of each agent.
4. Out of the list of the four, select your favorite candidate and justify why. Provide a detailed protocol for examining the mechanisms of action for this compound. Please write a 6–8-page proposal in the format of an NIH application addressing this issue, according to the attached format.

Question 3: A major pharmaceutical company has developed an agent Cblxxx that reportedly activates the Cbl signaling pathway, which may be involved in insulin-stimulated glucose uptake in fat cells. They are proposing that it might be used to promote heart glucose uptake and protect the heart against ischemic injury. They are prepared to make this agent available for your experiments. The proposal should provide strategies to understand the mechanisms of action of this agent in the heart, the physiologic role of the Cbl pathway in the heart and the potential cardioprotective role of this agent.

Question 4: A technician at Jackson Labs accidentally discovers a new strain of mutant mouse that exhibits impaired glucose tolerance. Initial characterization of this strain reveals that skeletal muscle cells in these animals do not translocate Glut4 from cytoplasmic vesicles to the plasma membrane in response to insulin stimulation. Genetic analysis suggests that this phenotype is inherited in an autosomal recessive fashion and is associated with the mutation of a single gene. Consider the cellular processes in which the product of this gene might be likely to participate. Construct a NIH style proposal in which you propose studies designed to identify the mutant gene and characterize its role in insulin stimulated Glut4 translocation.

Student 5

Project: Understanding and Enhancing the Oncoselectivity of Minute Virus of Mice

Question 1: Recently, a new single stranded RNA virus, named HF2005, has been isolated from a human fibroblast cell line that is routinely used in the laboratory. Design experiments to further characterize the virus in order to: 1. Determine the coding potential of the viral genome. 2. Classify the virus; 3. Identify the cellular compartment where the virus replicates. 4. Identify the cellular receptor(s) required for viral entry.

Question 2: The Fong group has generated a G207 herpes simplex virus which autonomously expresses AFP-driven UL39. You are asked to write a proposal which will first propose appropriate preclinical testing for this new construct and will then include a clinical trial designed to address the hypothesis that this new agent will be significantly better at control of hepatocellular carcinoma compared to unmodified G207.

Question 3: By a subtractive hybridization and/or cDNA microarray analysis of cDNA derived from highly purified hematopoietic stem cells, OR alternatively from primitive lineage-specific precursor cells, you have identified a previously unknown cDNA sequence, which when translated appears to encode a transcription factor containing zinc finger motifs. In the form of an NIH grant proposal, describe the

steps you would take to support your hypothesis that this newly discovered transcription factor plays an important functional role:

1. In hematopoietic stem cell biology
   OR
2. In lineage-specific hematopoietic cell differentiation along either the erythroid, megakaryocytic or granulocytic pathway (select one of these three pathways of your choice)

Answer either #1 or #2, not both.

Question 4: You are studying the growth of an autonomously replicating parvovirus that is defective in the expression of the small regulatory protein NS2. Both this mutant and the wildtype are unable to proliferate in the primary human embryonic kidney cell line HK, or a derivative culture, HK-TERT, that has been immortalized by a retrovirus vector expressing the catalytic subunit of human telomerase. You find that both mutant and wildtype can grow productively in a HK-TERT derivative, HK-ELR, that has been fully transformed, in a stepwise fashion, using two further retrovirus vectors expressing the SV40 early region and activated H-*ras*, respectively. You then find that the wildtype, but not the NS2 mutant, can also grow in a HK-TERT derivative, HK-E67ras, that was transformed with a retrovirus vector expressing both HPV16 E6 and E7 proteins, followed by the vector expressing activated *ras*. What might be the reason for the failure of the NS2 mutant to grow in the HK-E67ras cell line? How would you go about confirming your hypothesis?

Student 6
Project: Regulation of the Airway Epithelial Responses in Asthma

Question 1: Asthma is presumed to be a Th2 cell-mediated disease. In your thesis you outline three pathways by which Th1 cells, through effects of IFN-, inhibit Th2-induced mucus, chitinase and eosinophilia.

Many respiratory viral infections lead to exacerbations of pre-existing asthma, symptoms of increased coughing, wheezing and sputum production. Interferon gamma is typically observed in such infections and is increased in the BAL of virally infected patients with asthma exacerbations. For this proposal, assume that IFN- has been shown to be the cause of these acute asthma exacerbations.
Propose three distinct pathways, which are initiated by known effects IFN-, that could lead to asthma exacerbation; pathways stimulated by IFN- that would increase allergic airway inflammation, airway pathology or airway hyperresponsiveness.

Test your hypotheses *in vivo* using the following reagents to create your model:1) Wild-type BALB/c mice 2) house dust mite (HDM) antigen 3) recombinant murine IFN- (as much as you need). To induce allergic asthma in your mice you will use HDM administered over 3 weeks as described by Johnson et. al. (Johnson, J.R., R.E. Wiley, R. Fattouh, F.K. Swirski, B.U. Gajewska, A.J. Coyle, J.C. Gutierrez-Ramos, R. Ellis, M.D. Inman, and M. Jordana. 2004. Continuous exposure to house dust mite elicits chronic airway inflammation and structural remodeling. *Am J Respir Crit Care Med* 169:378-385.)

Question 2: Epicutaneous exposure to allergens has been thought to either sensitize or exacerbate subsequent allergic asthmatic responses. You have established a murine model in which epicutaneous sensitization to ovalbumin leads to Th2 priming and Th2-mediated pulmonary inflammation upon inhaled ovalbumin challenge. Further analysis using this model indicates that no pulmonary inflammation is observed in MyD88 deficient mice. However, TLR4 deficient mice respond normally. You are interested in the role of MyD88 in this model and decide you have enough preliminary data to write a KO8 proposal.

1. Formulate a hypothesis to explain the role of MyD88 in the ovalbumin-mediated lung inflammation. (*There is no right hypothesis here; take a stand and test below)*
2. Write specific aims that definitively test your hypothesis. (*State your aims in the form of a question).*
3. Outline experimental approaches for each of your specific aims that test your hypothesis. Indicate possible outcomes and alternative approaches. Point out pitfalls and weaknesses in the experiments proposed as appropriate.

This proposal should include: An analysis of the role of MyD88 in the priming and/or challenge phase; an analysis of the responding cell types and the importance of these cells during the in vivo response; an analysis of specificity in the response; a proof of concept set of experiments to definitively test your ideas in vivo. All reagents and assays are available to you; you can create any type of genetically manipulated mouse.

Question 3: The plasmacytoid dendritic cells are potent secretors of type I interferons in response to viruses. Recent studies implicate plasmacytoid dendritic cells to be an important regulator of Th2 responses in the lungs. Depletion of plasmacytoid dendritic cells in vivo during antigen inhalation leads to exacerbated IgE, airway eosinophilia and goblet cell hyperplasia.

1. Based on these observations, formulate a hypothesis for the role of plasmacytoid dendritic cells in the generation of adaptive immunity in the lung.
2. Outline experimental approaches to test this hypothesis. Consider the role of innate immune receptors expressed by the plasmacytoid dendritic cells and how the function of these cells might be controlled at the level of pattern recognition.

Question 4: An international health colleague of yours recently returned from a trip to the orient. She tells you that she saw rural physicians treat asthma with an herbal medicine that was remarkably effective. She also tells you that she was able to procure a stock of the herbal extract and that it is soluble and can be used in animal and in vitro systems. Please describe the approach you will use to determine if the herbal extract has appropriate "antiasthma effects" in preclinical investigations.

Question 5: Airway remodeling is commonly proposed as an anatomic basis of airway dysfunction in asthmatics. In order to explain variations in airway remodeling among patients with relatively similar degrees of inflammation, it has been proposed that those patients with more severe remodeling have fibroblasts that are more responsive to tissue remodeling signals. One major signaling pathway driving fibroblast proliferation is the gp130/stat3 pathway induced by interleukin 6 and interleukin-11. Transgenic overexpression of either IL-6 and IL-11 in the airways are known to lead to excess matrix accumulation around airways. Depending on the strain background of the mouse in which the transgene is expressed, the amount of collagen accumulation is vastly different. Suggest approaches from both cell biology and genetics to determine the mechanism by which this occurs.

Student 7

Project: Mechanisms of anti-CD45RB mAb Induced Tolerance

Question 1: Immunization with ovalbumin, a soluble protein antigen, without adjuvants can induce ovalbumin-specific CD4 Th2 cell and surprisingly CD8 T cell responses. Formulate a hypothesis to explain the mechanism by which 1) Th2 but not Th1 cells are primed and 2) CD8 T cells are primed. Use this hypothesis as a basis of a new grant proposal to test definitively the feasibility of your ideas.

Question 2: There has been a dramatic increase during the past 15 years in the incidence of asthma in more developed countries. Formulate a hypothesis to account for this observation. Use this hypothesis as a basis of a new grant proposal to test your ideas in a murine model.

Student 8

Project: Bone Marrow Derived Cells as Progenitors of Alveolar Epithelium

Question 1: You have designed a new type of vaccine using antigen loaded dendritic cells (DC). This approach involves the use of DC precursors isolated from peripheral blood mononuclear cells. The cells are cultured with infectious agent(s) and factors that lead to dendritic cell maturation. These mature dendritic cells are reintroduced into the individual to induce immunity to the infectious agent(s) with the hope that both T and B cell immunity will be observed.
An initial test of the DC vaccine approach in mice showed the following results.

TABLE: Induction of listeria monocytogenes specific immunity.

DC Treatment in vitro	Listeria Specific T Cell Proliferation	Listeria Specific Antibodies
No antigen	-	-
Irrelevant antigen pulsed DC	-	-
Listeria pulsed DC	+++	-

Legend: DC were isolated from peripheral blood mononuclear cells, matured for 2 weeks in the presence of GM-CSF, and exposed to either no antigen, irrelevant antigen, or listeria in vitro. After 24 hours, the cells are injected into mice intravenously. After 2 weeks, splenic T cells were isolated from some mice and analyzed for their ability to respond by proliferation to listeria in vitro. After 3 weeks, listeria specific antibody responses were measured by Elisa.

1. Formulate a hypothesis to explain the results observed. Test your hypothesis experimentally in a murine model using Listeria monocytogenes as your infectious agent.
2. What types of immune response are needed ideally for effective immunity to infections agents? How would you improve upon your vaccine design above to generate the appropriate immune response?
3. How would you determine the effectiveness of the vaccine over time? For example, how would you determine if vaccinated individuals are immune 2 years after the vaccination?

Question 2: Respiratory syncytial virus (RSV) can cause upper or lower tract infection, with the latter correlating with the development of asthma.

1. Formulate a hypothesis for the specific relationship between RSV lower tract infection and asthma.
2. Outline experimental approaches to test this hypothesis. Include a description of various strategies for monitoring gene expression in the relevant target tissues, and approaches to relate gene expression profiles to subsequent development of asthma.

Question 3: A colleague in your lab has identified a new protein secreted by endothelial cells that appears to be a member of the fibroblast growth factor family and has named it FGF 25. You determine by western analysis that FGF 25 is expressed by endothelial cells in the embryonic lung. Construct a hypothesis for a role (or lack of a role) for FGF 25 in early lung development and branching morphogenesis, and outline a series of experiments to test your hypothesis.

Student 9

Project: Processing and Secretion of Virulence Proteins

Question 1: You have used the URA-blaster protocol to generate heterozygote and homozygous disruption mutants of the *YFG-1* gene in *C. albicans* in order to try to understand the function of the gene product. Your competitors have used the PCR-based gene disruption protocol of Mitchell et al. (J. Bacteriol.) to construct heterozygote and homozygous disruption mutants for testing in an oral inoculation model. As you stand side-by-side at adjoining posters, you compare your results. Both groups used the same murine species and the same inoculating dose.

Animal Model	YFG-1 URA blaster			YFG-1 PCR mutation		
	WT	het	hom	WT	het	hom
Tail-Vein Model of Fungemia (CFU/gm liver)	10^5	10^4	10^2			
GI Colonization Model (CFU/gm cecum)				10^3	10^3	10^3

Develop a grant proposal that outlines how the protein encoded by *YFG-1* might be working and what novel therapies might be developed. Be sure to discuss why the results obtained by the two methods are different, and what experiments and controls you would perform in vitro and in vivo in order to test your hypothesis.

Question 2: You have just cloned and sequenced the *BIG-1* gene from *C. albicans* on the basis of a promoter-trapping experiment that pointed to this gene as highly expressed in fungemic but not colonized animals. In a grant proposal, outline one genetically-based and one protein-based method by which you could translate this discovery into a reproducible test for rapid diagnosis of Candida infections. Discuss the pros and cons of each method. Be sure to address issues of sensitivity, specificity, and positive/negative predictive value.

1. PCR-based diagnosis – caveat: need to establish whether *BIG-1* is specific for *C. albicans* or for *Candida* spp. in general
2. ELISA-based diagnosis – caveat: need to generate two different antibodies, time required to develop ELISA's.
3. Latex agglutination (coat beads with antibody to Big1p) – caveat: level of sensitivity

Question 3: A monoclonal antibody (IgG1) made by injecting mice with emulsified gel containing the C. albicans protein TUF-1 readily identifies the expressed protein on Western blot and binds actively to the outer cell wall of C. albicans blastospores. However, the antibody fails to bind to C. albicans hyphae, even though a Tuf1p-GFP fusion shows abundant surface fluorescence of hyphae by every detection technique (flow cytometry, confocal microscopy, devolution microscopy). Provide at least two explanations for this observation and discuss how you might use this apparent piece of "bad luck" to develop a novel therapeutic agent.

1. Explanation #1: conformational change of the detected epitope when *TUF-1* is expressed on hyphae. Make a Mab to the hyphal form of the *TUF-1* gene product.
2. Explanation #2: *C. albicans* hyphae express an Fc receptor. Purify the protein or use a consensus sequence to clone the gene encoding the Fc receptor, then develop polyclonal or monoclonal antibodies to block its function.

Question 4: A polyclonal rabbit antibody (Ab) to a known *Candida albicans* (Ca) cell surface protein is to be tested for efficacy in preventing Ca gastrointestinal invasion in an animal model. The rationale for the

animal experiments is preliminary data obtained from in vitro experiments looking at attachment and invasion of Ca through a polarized epithelial cell monolayer, shown in the table below. Construct a proposal to test your hypothesis for how the antibody is working, what additional in vitro data you would like to generate, and how you will test the antibody in an in vivo model.

% Inhibition by Ab (100µg/ml) in comparison to control (no Ab)	Ab and Ca added to apical side of monolayer	Ab and Ca added to basolateral side of onolayer
Attachment	88%	75%
Invasion	85%	2%

Student 10

Project: The Impact of Fat on Liver Glucose Metabolism: Defining the link between non-alcoholic fatty liver and hepatic insulin resistance

Question 1: Nobartis Pharmaceuticals Intl is proposing to develop a novel pharmacologic agent to treat poorly controlled type 2 diabetics by inhibiting hepatic gluconeogenesis at the level of fructose 1,6 biphosphatase. Through robotic screening they have identified a compound NP 10036 that appears to inhibit fructose 1,6 biphosphatase with a very high affinity. Preliminary in vivo pharmacokinetic data suggests that a single dose of 1mg/kg body weight per day should be effective in inhibiting this enzyme for ~24 hours in a rat. Before proceeding further they would like to perform some additional mechanistic studies in rats to prove: 1) the drug is working at the proposed step, 2) that it does inhibit gluconeogenesis in vivo and 3) that it might be an effective drug to treat type 2 diabetes, Construct a proposal to examine these questions.

Question 2: A Nobartis competitor, Samuel Pharmaceuticals, is proposing to develop a novel pharmacologic agent to treat poorly controlled type II diabetes, by augmenting translocation of GLUT4 vesicles to the plasma membrane. Preliminary data indicates that treatment of myocytes in vitro with SP001 results is a three fold increase of GLUT4 expression on the plasma membrane, but does not augment the Km, Vmax or initial rate for glucose transport in vitro. Nonetheless, the drug shows good efficacy in an animal model of diabetes, substantially lowering plasma glucose levels and decreasing insulin requirements. Construct a hypothesis, and a proposal to test this hypothesis, to explain the discrepancy between the in vitro and in vivo results. SP001 is Samuel Pharmaceuticals only promising drug, so they are very anxious to resolve this issue as quickly as possible, especially since the FDA commissioner, Dr Kestle, is skeptical about their results. Please be sure to outline the time frame for conducting and completing the studies.

Student 11

Project: Free Fatty Acids and VEGF Signaling Abnormalities

Question 1: eNOS is an enzyme that may be coupled to produce NO or uncoupled to produce superoxide. Since eNOS is part of a multiprotein complex devise several strategies to isolate the complex, identify the components and test the functional significance of the components in the coupled or uncoupled state.

Question 2: Impaired NO bioactivity is a hallmark of several cardiovascular diseases. Making an assumption that the decreased bioactivity is not due to changes in eNOS function, describe a series of hypothetical experiments to identify a molecular mechanism.

Question 3: You have been asked to serve as a consultant for a small biotech company whose name is Metab Inc. They are interested in developing some novel compounds to enhance insulin sensitivity in type 2 diabetic patients. They have several potential targets in mind. Give your opinion on each one:

1. Adiponectin Analogues
2. AMP Kinase Activators
3. PPAR alpha Activators
4. PPAR delta Activators

Please discuss all of the following points (Points 1-3 below can form the background section of your proposal):

1. Theoretical mechanisms by which each agent would improve insulin sensitivity in type 2 diabetic patients and specify which organs would be involved.
2. The potential long-term salutary effects of each compound.
3. Potential Side effects of each agent.
4. Out of the list of the four, select your favorite candidate and justify why. Provide a detailed protocol for examining the mechanisms of action for this compound.

Question 4: You are head of R&D at Morck, a major pharmaceutical company interested in metabolic diseases.

A. Please describe and provide the rationale for a novel drug target for treatment of insulin resistance in type 2 diabetes.
B. The Morck preclinical division has come up with a drug that works on your target and it has proved to shows some promise to reverse insulin resistance in rodents and obese monkey models of type 2 diabetes. It also looks to be safe in humans as reflected by initial Phase I studies. Describe in detail the key Proof of Concept protocol (s) in humans that you would implement to test out whether Morck should move forward with this novel drug.

Question 5: Peripheral arterial disease is common and tends to be more severe in patients with diabetes mellitus, and is associated with a poor clinical outcome. Define a mechanistic hypothesis related to this clinical phenomenon. How might you employ targeted molecular imaging to explore this hypothesis? What existing experimental models and/or transgenic mice could be employed to address your mechanistic hypotheses?

Question 6: Diabetics with peripheral arterial disease have impaired wound healing. What is the role of angiogenesis in this process? How important is VEGF in wound healing among diabetics? How would diabetic control, metabolic therapies, and the accumulation of advanced glycosylation end-products affect the angiogenic response? Define the role of metabolic alterations associated with diabetes, in the angiogenic response to ischemia injury associated with peripheral arterial disease?

Student 12 Project: Estrogen Receptor-Mediated Rapid Signaling in Endothelial Progenitor Cell Responses to Ischemia

Question 1: The past few years have brought a revolution in gene therapy and drug delivery so that many of the previous limitations have been largely bypassed with novel systems. A viral vector that can deliver sustained expression of up to 3 gene products under inducible control by 3 different non-toxic drugs has been developed and is commercially available under the trade name “Novogene”. The NIH has issued a Request for Application (RFA) entitled “Use of Novogene Delivery Systems for Induction and Sustained Function of Angiogenesis in Ischemic Vascular Disease”. You are very excited to submit an application since your lab has made fundamental discoveries regarding factors that control the complex process of growth and maintenance of functional blood vessels in an animal model of ischemic vascular

disease and you believe you are in a perfect position to obtain funding through this mechanism. Write an NIH format proposal describing which genes you propose to be used in this new system. The proposal should provide information about the rationale for each choice including which phases of angiogenesis are to be manipulated and why. Don't forget that since the Novogene vector allows you to use up to 3 separate inducible genes, you will be able to control the timing and degree of gene expression in your animal model.

Question 2: You are having a coffee with one of your colleagues and discussing common research interests. She tells you one of her mouse strains has been dying off at an alarming rate. The animal care facility at your institution has been plagued by a variety of infections despite rigorous control of potential sources. Recently, the room that houses your colleague's animals has been found to be contaminated with agent 101. The veterinarian in the animal facility has assured your colleague that this infectious agent has never been shown to cause adverse health effects in mice. However, your colleague remains convinced that the infection is responsible for the deaths of her mice. She has performed autopsies on several of the dead animals and found an alarming degree of inflammation in multiple vascular beds. The animals appear to have died from massive apoptosis of endothelial cells and resultant hemorrhage and ischemia in the brain and other crucial organs. She looked back over the pedigrees of the animals that have died and discovered that the susceptibility to the agent is inherited in an autosomal recessive fashion. She also found that these animals develop a similar syndrome in response to a wide variety of infectious agents as well as after administration of TNF alpha or lipopolysaccharide. She asks if you will help her to determine which gene is responsible for the susceptibility to inflammatory-induced endothelial apoptosis since this is far outside her area of expertise and your lab is actively involved in studies of oxidant stress and endothelial function. You are intrigued by this problem and decide to write an NIH style proposal outlining studies that might help you to discover the mechanism for this observation. You have definite ideas about candidate genes for this response and also how you might identify the gene if it is novel. You hope that the gene responsible for this susceptibility might be a therapeutic target for many types of inflammatory vascular disease.

Question 3: Determine the contribution of endothelial progenitor cells to the process of arteriogenesis and angiogenesis in critical limb ischemia.
In peripheral vascular disease (PVD), a reduction in blood flow and tissue perfusion leads to hypoxia. Excision of the femoral artery leading to ischemia in the lower limb can serve as a model of PVD. In this model, changes in arteriogenesis and angiogenesis have been shown to contribute to the recovery of blood flow to almost normal levels after four weeks. Recent studies have suggested that ischemia induces the mobilization of endothelial progenitor cells (EPC) that might contribute to the ability to recover flow.

Your project is aimed at elucidating the contribution of EPCs to the process of angiogenesis and arteriogenesis following ischemia in the mouse model. It is imperative that you design experiments that distinguish possible effects on angiogenesis and arteriogenesis.

Question 4: Examine the role of endothelial cell vacuole formation in the process of angiogenesis.
In vitro Matrigel 3-D angiogenesis assays indicate that vacuole formation in endothelial cells might contributes to blood vessel lumen formation. The exact signaling events that are activated in the formation of vacuoles have not been elucidated, but a role for the small GTPases cdc42, Rac, and Rho has been proposed. A recent study in zebrafish provided the first in vivo demonstration of the vacuole-to-lumen transition and implicated the activation of Rac.

Your task is to design in vivo experiments in mice utilizing Matrigel to investigate whether the vacuole-to-lumen transition occurs in angiogenic ECs. In parallel, your experiments should be designed to allow you to investigate the requirement for the activation of the small GTPases, especially Rac, in the formation of vacuoles and lumens.

Question 5: There is controversy in the literature regarding whether BM derived cells can differentiate into cardiac myocytes. How can this issue be resolved? What is your hypothesis regarding the seemingly disparate data?

Question 6: How could one best test whether endothelial cells can be used therapeutically to treat ischemic injury?

Student 13
Project: Role of Innate Immunity in Flow-Mediated Vascular Remodeling

Question 1: What would constitute proof of the direct role of JNK vs ERK vs p38 to vascular remodeling in your system? What approaches would define the important cell type involved?

Question 2: How would you assess the contribution of a specific MMP to the matrix remodeling in your system? How would you address the molecular mechanism?

Question 3: The incidence of atherosclerosis increases dramatically in women after menopause. However, it is unclear whether there are changes in hemodynamics that accompany this hormonal shift, and if there are such changes, whether they affect the incidence of atherosclerosis. Form a hypothesis to test whether hemodynamics affects estrogen-mediated cell signaling. Use this hypothesis to test your ideas in a murine model.

Question 4: Arterial disease, both occlusive and aneurysmal, is clinically noticed to be accompanied by inflammation. Form a hypothesis to test the significance of inflammation in both occlusive AND aneurysmal disease. Use this hypothesis to test your ideas in an animal model.

Question 5: There is increasing evidence that genetic factors determine the emryonic organization of the vascular system into arteries and veins. Devise a series of experiments to test the relationship between genetic factors and hemodynamics in regulating the remodeling of blood vessels.

Question 6: Flow dependent signaling is important for endothelial cell modulation of vascular function. However, it has difficult to define mechanosensing mechanisms compared to the abundance of data identifying flow-activated downstream pathways. Devise a series of experiments to identify mechanosensors in endothelial cells.

Question 7: About 5% of adult cases of polycystic kidney disease (dominant PKD types I and II) are associated with cerebral aneurysms. Propose a mechanism for development of these aneurysms in PKD and discuss approaches that might be taken to test the mechanism you propose. These approaches might involve specific mechanisms directly related to PKD or mechanisms indirectly related to PKD. If possible, extend this mechanism to the development of sporadic cerebral aneurysms.
Question 8: Somebody sites are devoid of vasculature, most notably the cornea. What is the mechanism by which this tissue is maintained in an avascular state? How does this finding conflict with the results of classical experiments in angiogenesis? Propose a resolution for this conflict and an experiment to test that hypothetical resolution. Can you think of ways the information you find could be used therapeutically?

Student 14
Project: Role of Palladin in Neocortical Development

Question 1: You are analyzing the phenotype of Fibroblast Growth Factor Receptor (Fgfr) knockout mice, which you generated by deleting floxed Fgfr2 alleles via the GFAPCre line. You have discovered that the Fgfr2 KO mice have lower numbers of pyramidal neurons in the cerebral cortex. BrdU birthdating experiments allowed you to observe, after BrdU incorporation at E14, a decreased number of BrdU-labeled neurons in the E16 cortical plate with a relative increase in the intermediate zone. You hypothesize that there is a decrease in neuronal migration in these knockout mice.
In the form of a grant proposal, describe experiments that will demonstrate whether or not Fgfr2 is responsible for regulating neuronal migration in the cortical plate, considering that this receptor may also regulate cell proliferation in the developing cortical wall.

Question 2: You are analyzing the phenotype of Fibroblast Growth Factor Receptor (Fgfr) knockout mice, which you generated by deleting floxed Fgfr2 alleles via the GFAPCre line. You have discovered that the Fgfr2 KO mice have lower numbers of pyramidal neurons in the cerebral cortex. BrdU birthdating experiments allowed you to observe, after BrdU incorporation at E14, a decreased number of BrdU-labeled neurons in the E16 cortical plate with a relative increase in the intermediate zone. You hypothesize that there is a decrease in neuronal migration in these knockout mice.
Fgfr2 is a tyrosine kinase transmembrane receptor, which you found to be expressed mostly by radial glial cells. In the form of a grant proposal, describe experiments that will demonstrate whether the hypothesized cell migration defect is due to a cell-intrinsic lack of Fgfr2 in migrating neurons or whether cell-cell interactions or paracrine phenomena may be responsible.

Question 3: Your advisor just returned from a meeting and told you that someone identified a novel intracellular signaling molecule, named Alma. Alma has an actin-binding domain and has high similarity to ARF-GEF (G-protein exchange factors). You started doing pilot experiments and propose that Alma is part of the Semaphorin/Neuropilin signaling pathway. Describe a serious of experiment in which you test (biochemical and functional) if Alma contributes to Sema signaling. Alma is an imaginary protein.

Question 4: You conducted an interactive screen to identify proteins binding to the intracellular domain of Neuropilin. You identified several interesting proteins, among them OLAC. Genetic evidence in invertebrates previously implicated that OLAC is involved in cell polarity. You confirmed the interaction using co-IP experiments and you found that it is expressed in cortical dendrites. You propose that OLAC is essential for the formation of cortical dendrites. Describe a serious of experiments in which you test your hypothesis. All reagent, including mice deficient in OLAC are available to you. OLAC is an imaginary protein.

Question 5: There are two main types of projection (pyramidal) neurons in the cerebral cortex. Those that project to subcortical regions and those that send axons within the cortex. Birth dating of the two types of neurons showed that subcortical projection neurons are generated first and populate deep-layers (V-VI) of the cerebral cortex, while the cortically projecting neurons are born later and populate primarily the upper-layers (II-IV) of the cortex. What is not clear is whether the two types of neurons are generated by the same progenitor cells in a temporal manner, or by two different lineages of progenitor cells in the embryonic brain. A recent study by Sally Temple's lab indicates that at least within dissociated cell cultures, individual early cortical progenitors can generate neurons with molecular characteristics of both deep and upper layers.
In the form of a grant proposal, design an experimental study that would distinguish between the two possibilities (one lineage vs. two lineages) of how different cortical layers are generated in vivo.

Question 6: Cortical projection neurons are generated and migrate into the cortical plate in an inside-first, outside-last pattern. Studies in the Reeler mutant mice have shown that it is not laminar position but the timing of neuron generation that determines the axonal target of cortical projection neurons (i.e., subcortical vs cortical). This, however, does not exclude the possibility that other factors, such as the local cellular environment of a cortical layer contribute to some of the differences in the molecular and cellular phenotype between deep-layer subcortically projecting neurons and upper-layer cortically projecting neurons.
Based on your readings, describe a series of experiments in which you test the contribution of the local environment to the determination of layer-specific dendritic, axonal and molecular phenotypes of projection neurons.

Student 15
Project: AMPK Regulation and Function in the Ischemic Heart

Question 1: While attending a meeting on medicinal therapies at the University of Hunan, you hear about a compound that dramatically reduces myocardial injury following an MI. The investigators have isolated an active component and state that they have some preliminary data that it may affect a protein kinase. The investigators know of your work on protein kinases, and offer to collaborate with you to identify the target of this drug if you can provide them with a good experimental plan. They offer you the full assistance of their medicinal chemists in your studies. How would you propose to study this compound?

Question 2: During studies in a cell-free system, you find evidence that AMPK can phosphorylate a critical myocardial plasma membrane ion transporter. What are the limitations of extrapolating this finding to an intact cell and how would you study the relevance of this finding to an intact system?

Question 3: A pharmaceutical company approaches you to evaluate a new compound they feel may have cardioprotective properties. The agent, BR-549, is a cardioselective proton ionophore that induces mitochondrial uncoupling of citric acid cycle flux from ATP synthesis. In addition to decreasing reactive oxygen species generation during ischemia reperfusion, the agent appears to activate AMP-activated protein kinase (AMPK). Please write a 6-8 page research proposal, using the NIH format, in which you propose a mechanism by which the compound activates AMPK and also determine whether the activation of AMPK is responsible for the cardioprotection rather than inhibition of reactive oxygen species generation.

Question 4: Insulin stimulation has been shown to cause redistribution of hexokinase II between a soluble cytosolic fraction and an organelle fraction consisting predominantly of mitochondria. This is thought to enhance the activity of the mitochondrial adenine nucleotide translocase by decreasing the ATP concentration and increasing the ADP concentration in the cytosolic microenvironment, thereby increasing the exchange of mitochondrially derived ATP for cytosolic ADP. You hypothesize that ischemia may have similar effects. Please write a 6-8 page research proposal, using the NIH format, in which you propose a specific mechanism by which this occurs as well as the experiments you will perform to test this hypothesis.

Question 5: You have discovered that the cells you have been studying for the past 6 months respond to hormone zzz by an increase in intracellular calcium. Outline the experiments you would do to determine where the calcium is coming from and what pathway it uses. When you examine your cells with better spatial resolution you notice that the calcium rises are oscillations. How could this occur? Why would the cell use this form of signaling?

Question 6: Describe at least two properties (other than their location) that differ between voltage-gated calcium channels and intracellular calcium channels and explain why they are important for cell function. How would you use these properties to develop specific pharmacological agents?

Student 16
Project: Toll-like Receptor 7, B Cells, and Plasmacytoid Dendritic Cells at a Nexus in Systemic Autoimmunity

Question 1: How would you test the hypothesis that TLR7 represents a first line of defense against single-stranded polynucleotide genomes, such as the single-stranded DNA of parvoviruses, as well as single-stranded RNA molecules, such as those elaborated by either positive or negative RNA viruses?

Question 2: Design an experiment to test the hypothesis that plasmacytoid DCs are both necessary and sufficient for an experimental autoimmunity model.

Question 3: There is conflicting data in the field regarding the role of HMGB1 as an endogenous activator in autoimmunity models. Design experiments to clarify the confusion.

Question 4: A company wants to develop an interferon blocking agent to improve SLE disease manifestations. They consult you to determine if such an approach is justifiable, is it?

Question 5: How could a dynamic computational model be developed and applied to investigate the role of feedback loops (e.g., through IFN-alpha and ICs)?

Question 6: How could you integrate data from existing studies to investigate whether there are disease subtypes, possibly associated with distinct underlying causes?

Student 17
Project: Characterization of Abnormal Neural Connectivity in Autism Spectrum Disorders using Multimodal Magnetic Resonance Imaging Techniques

Question 1: The issue of disordered brain connectivity in autism is interesting in light of various theoretical/psychological models of CNS functioning in autism. Would you pick one of the several overarching theoretical schemes (central coherence, theory of mind, enactive mind, or executive function) and provide a short (4 page) summary of how this theory might translate into problems of brain connectivity.

Question 2: One of the most interesting aspects of autism over the years has been the observation of individuals with islets of unusual ability (autistic savants). A large literature, mostly consisting of case reports but some more controlled studies exists (going back to a classical paper on this topic by Kurt Goldstein). Would you provide us with a summary (4 to 6 pages) of how problems in brain connectivity might be understood as contributing to this phenomenon and the even more common issue of marked scatter in developmental attainments/functioning.

Question 3: Design a hypothesis driven study to examine the question of altered connectivity in the brain in the development of autism and its relationship to cognitive impairment using diffusion MR imaging. With a focus on a particular network, describe the subject population you would examine, the data you would collect, the analysis methods you would perform and the statistical comparisons you would make.

Question 4: Please describe in detail the processing techniques used for the DTI group analysis using FSL. This should include a description of the data mapping to common space and the quantification.

Question 5: Please describe in more detail the registration techniques you are proposing to use towards completing the aims of your proposal.

Student 18
Project: Toll-Like Receptors in Older Adults and Response to Vaccination

Question 1: A newly developed avian influenza vaccine was given intramuscularly to healthy adults and age-matched patients with diabetes. The serologic efficacy of the vaccine was determined by measuring avian influenza virus antibody titer before and at 1 month after the vaccination. The results of the study showed that the increase in avian influenza virus antibody titer after vaccination was lower in patients with diabetes compared to healthy individuals.
Based on observations above, develop a strategy to determine the potential mechanisms for impaired antibody production in patients with diabetes in response to the newly developed avian influenza vaccine.

Question 2: Through your research you have identified two groups of patients, one of which had significant increase in the incidence of viral infections (HSV and CMV), whereas the other had increased incidence in gram-positive bacterial infections. Your goal is to identify the molecular basis for these immuno-deficiencies. How would you go about it, what would be the rationale for each step of your analysis and how would you test your hypothesis?

Question 3: You identify a cluster of Icelandic families in which life-threatening infections with S. aureus, L. monocytogenes and M. tuberculosis are observed in multiple family members in several generations. You suspect that mutations in TLR2 may be responsible for the observed clustering of infectious diseases, and you find that TLR2-mediated signaling responses (e.g. upregulation of co-stimulatory molecules, cytokine production) are markedly reduced in affected individuals. Write a proposal to test the hypothesis that mutation(s) in the TLR2 locus are responsible for these clinical observations, and describe your approach to identifying such mutation(s).

Question 4: You are head of R&D at Morck, a major pharmaceutical company interested in metabolic diseases:

A. Please describe and provide the rationale for a novel drug target for treatment of insulin resistance in type 2 diabetes.
B. The Morck preclinical division has come up with a drug that works on your target and it has proved to shows some promise to reverse insulin resistance in rodents and obese monkey models of type 2 diabetes. It also looks to be safe in humans as reflected by initial Phase I studies. Describe in detail the key Proof of Concept protocol (s) in humans that you would implement to test out whether Morck should move forward with this novel drug.

Question 5: The federal government has issued an RFA for vaccine development for aquatic flu in light of concern for a possible future pandemic. A Science Park biotech firm has investigated gateway receptors that are believed to modulate response to the virus responsible for aquatic flu. There is evidence that an existing vaccine to another virus has putative, but weak, protective activity toward aquatic flu and gateway receptors are known to modulate response to the other virus and the vaccine. The Science Park firm has developed a compound that enhances the activity of gateway receptors and believes that using this approach in conjunction with an existing vaccine would be both quicker and

more cost effective than developing an entirely new vaccine. As they are not primarily a vaccine company, they believe this approach, the time and cost savings, and their expertise in gateway receptors and the new compound would be to their competitive advantage in this application. The caveat is that the vaccine has been associated with the rare occurrence of a debilitating neurodegenerative disorder. The only known risk factor for who gets the presumed side effect is age (younger at greater risk). Gateway receptor activity, on the other hand, decreases with age. The concern is that by giving the compound that enhances gateway activity the risk for the dreaded side effect may increase and the potential liability would be phenomenal given the relatively younger age of those affected. As proposed federal liability protections for vaccine development have not yet been approved, and it's not clear they would apply in this case anyway given the novel approach chosen, the firm is obviously concerned that this potentially lucrative grant may result in liability exposure that would bankrupt the small company. Having heard about your work with similar Toll-like receptors and your medical and clinical epidemiology background, they would like for you to collaborate with them in designing a substudy that would assess the risk factors for developing the neurodegenerative disorder and ideally, demonstrating that it was not the enhancement of gateway activity that was responsible should any increased incidence occur. While this is neither your primary area of interest nor expertise, there is synergy with your own work and this company's, as well as the fact that they are a well-established and cutting edge firm, so the prospect of future collaboration on projects closer to your areas of interest is intriguing. Write a 6-8 page proposal for this substudy in the format of an NIH application, according to the attached format.

Student 19
Project: Age Associated Alterations in Toll-like Receptor Function of Human Dendritic Cells and Correlation with Influenza Vaccine Response

Question 1: Describe all the potential mechanisms for the transcriptional and post-transcriptional control of the cell surface expression of a gene product. In addition, describe how each mechanism may be experimentally approached.

Question 2: If the aim of a research project is to establish the effect of age on the expression of PRAT4A an endoplasmic reticulum chaperone, how should the study be designed? How do you decide who to screen and sample? How many time points would you sample the participants? What is an age effect? How would you establish an age effect? What are the factors that you would need to know to address these design and sampling questions? If a cross-sectional design were chosen then there is an assumption that PRAT4A is time and environmental stable. How would someone know if this were true? Why does the design of the experiment influence whether you might find differences that truly exist?

Question 3: Using the design you selected, what would be the analytic methods you would use to test whether there is an age effect on the expression of PRAT4A? How would you account for heterogeneity among the human participants? How would you account for correlated measures on participants? How would you address collinearity and multiplicity? Discuss the model assumption and its strengths and limitations.

Question 4: A colleague has preliminary data that suggests that deficiency in the PRAT4A in dendritic cells in older adults contributes to reduced antigen presentation function. A second colleague has data that suggests PRAT4A over expression in DC has no functional consequences regarding antigen presentation and cell expression of TLRs. The first researcher hypothesizes that deficiency of TLRs in DC in older adults was substantially due to either reduced or aberrant proteolytic processing of TLRs. How would you assess this experimentally?

Question 5: Write a position statement on the clinical use of Pneumococcal vaccine in older patients. Address the clinical practice of repeated vaccinations. This statement should be supported by the current basic science and human research data. Address gaps in the available literature which both support or refute your position on the use of the vaccine. Describe how you could address the gaps in current knowledge with both basic and clinical study designs, include research methods, feasibility and limitations.

Student 20

Project: The Role of Cellular Prion Protein in β-Amyloid Induced Neuronal Network Dysfunction in Alzheimer's Disease

Question 1: There are many signaling pathways identified that contribute to the development of long term potentiation and long term depression (LTP and LTD). Outline the roles of trafficking and function of individual glutamate receptors and kinase cascades in the development of LTP. How might deficits in LTP and intracellular signaling be perturbed in Alzheimer's disease? Where in these pathways would you expect A¾ to have an effect?

Question 2: The Kandel group has suggested that a prion-like oligomerization process might underlie the strengthening of individual synapses. In addition, a current important hypothesis on the pathological mechanisms underlying Alzheimer's disease is that there is a direct effect of A¾ on the function and strength of individual synapses. Outline the evidence that A¾ oligomers function as neurotoxins because they disrupt synaptic function and plasticity. Provide a model based on the evidence in the literature how the interaction between A¾ oligomers and prion proteins might alter synaptic strength.

Question 3: While performing chronic EEG recordings you have found that a high percentage of transgenic mice overexpressing the human amyloid precursor protein die in their cage. This does not occur in mice without prion protein. You discover that death is prevented by oral treatment with an SSRI or by dietary supplementation with L-tryptophan.
You perform additional experiments that indicate death is due to postictal respiratory depression.
Design experiments that will:

a) Determine how seizures cause respiratory depression.
b) Define how serotonin is involved.
c) Define the role of prion protein in predisposing to this cause of death.

Explain what could be done to relate your findings in mice to sudden unexpected death in epilepsy (SUDEP) in human AD patients.

Question 4: You establish that there is a large increase in frequency of spontaneous seizures in transgenic mice over-expressing the human amyloid precursor protein with, but not without, prion protein. You find evidence of mossy fiber sprouting on histological examination of the hippocampus. EPSPs induced by stimulation of the stratum radiatum are increased. On EEG recording you discover that seizures occur exclusively in sleep, primarily during REM. Design *in vivo* and *in vitro* electrophysiological experiments that:

1. Define the cellular mechanisms that lead to an increase in seizures during sleep.
2. Identify how neuromodulators released during wakefulness (e.g. monoamines and acetylcholine) interact with prion proteins to change excitability of neurons in the hippocampus.

Test the hypothesis that seizure threshold is influenced by a change in neuromodulatory tone in the hippocampus

Question 5: Please propose a study specifically testing the ER-Ca2+ hypothesis of Alzheimer's disease. The proposal should include a brief review of the ER-Ca2+ hypothesis (e.g., what is the hypothesis and how does the ER Stress Response and the Unfolded Protein Response contribute to neurodegeneration). The study should identify and address some of the weaknesses of previous studies we've discussed suggesting that Ca2+ dysregulation is a primary determinant of AD pathology.

Question 6: Assuming that you've shown that ER Ca2+ dysregulation is a primary determinant of Alzheimer's disease-associated neuropathology, propose a study examining how deviations from normal neural activity might trigger or contribute to AD pathology. More specifically, consider neural activity implicated in AD in which there is likely to be a lot of Ca2+ flux into the ER (e.g., during seizures or some phases of sleep).

Student 21

Role of Macrophage Migration Inhibitory Factor (MIF) in the Pathogenesis of Tuberculosis

Question 1: On the basis of genetic studies in the human population, IFN-□ and its receptor-mediated signaling cascade represents a major nexus of control for host immune protection against mycobacterial infections. Using this information together with evidence from cellular studies and experimental animal models, design an approach to identify the likely downstream effectors of this response. Moreover, outline how this information can be assembled together with genetic studies of patient cohorts to validate findings from the experimental setting.

Question 2: Recent evidence has highlighted the emerging roles of C-lectin type receptors in recognizing and mobilizing the innate immune response to TB infection. Chief among these is the Dectin family of receptors that may operate via Syk-CARD9 to stimulate effector functions and immune cytokine release.
Describe in detail an experimental strategy to test how the human family of Dectin receptors may operate to restrict *Mycobacterum tuberculosis* (*Mtb*) growth at the level of the infected cell (eg. macrophages and dendritic cells). Place specific emphasis on each stage of that process: (i) recognition of *Mtb* by each member, (ii) signaling events required, and (iii) target genes elicited that may be directly involved in bacillary containment.

Discuss the experimental limitations of each approach, what appropriate controls have been chosen and why, and how predicted outcomes may be interpreted. Provide alternative strategies for experiments that may have potential pitfalls. Lastly, postulate how the information derived from these studies may be useful for or applicable to vaccine design against TB infection in the human population.

Question 3: Identify and describe three characteristics of experimental and control populations necessary to pursue your observations in humans.

Question 4: Describe a single population in which longitudinal studies of might provide insight into MIF role in pathogenesis of infection in humans.

Question 5: Mechanisms of granuloma formation in tuberculosis: There is a growing body of literature on pulmonary granuloma formation, but relatively little is known about the degree to which this response represents a fundamental host defense against infection, or a pathogen driven phenomenon aimed at creating a protective environment within the host. In order to address this question, please do the following:

1. Formulate a hypothesis on the role of granuloma formation in host-pathogen interaction
2. Detail a series of experiments aimed at characterizing host and pathogen requirements for granuloma formation in vivo.
3. Detail experiments aimed at probing the role of granuloma formation in both infection and disease (two distinct phenomena) in response to M. tuberculosis

Student 22

Functional Analysis of Rare Genetic Variants in Neuropsychiatric Disorders in a Zebrafish Model System

Question 1: Autistic spectrum disorders (ASDs) are associated with persistent deficits in social communication and interactions, as well as restricted, repetitive patterns of behavior, interests, and activities. Discuss promising phenotypes of zebrafish relevant to the human behavioral phenotypes seen in ASDs. How could such phenotyping procedures add to our understanding of the molecular pathways by which rare genetic variations contribute to the emergence of ASDs. Formulate and defend one such hypothesis and suggest experiments using one or more zebrafish phenotypes. The hypothesis may address either mechanisms mediating the observed differences, or functional implications of the results.

Question 2: Induced pluripotent stem cells (iPSCs), developed from mature somatic cells, have allowed the development of patient-specific neural cultures to be observed in real-time. They have allowed some neuronal-specific abnormalities to be corrected with pharmacological intervention in tissue culture. What would be the advantages and disadvantages of such an approach in the study of cells derived from individuals in family where there is known to be a rare functional mutation segregating with a childhood onset neuropsychiatric disorder? Formulate and defend one such hypothesis and suggest experiments using one of the rare genetic variants that you are currently incorporating into a zebrafish model system.

Question 3: There are two main types of projection (pyramidal) neurons in the cerebral cortex. Those that project to subcortical regions and those that send axons within the cortex. Birth dating of the two types of neurons showed that subcortical projection neurons are generated first and populate deep-layers (L6-L5) of the cortex, while the cortically projecting neurons are born later and populate primarily the upper-layers (L4-L2). What is not clear is whether the two types of neurons are generated by the same progenitor cells in a temporal manner or by two different lineages of progenitor cells in the embryonic cortical ventricular/subventricular zones. Design an experimental study that would distinguish between the two possibilities.

Question 4: Cortical projection (pyramidal) neurons are generated and migrate into the cortical plate in an inside-first, outside-last pattern. Studies in the Reeler mutant mice have shown that it is not laminar position but the timing of neuron generation that determines the axonal target of cortical projection

neurons (i.e., subcortical vs cortical). This, however, does not exclude the possibility that other factors such as the local cellular environment contribute to some of the differences in the molecular and cellular phenotype between deep-layer subcortically projecting neurons and upper-layer cortically projecting neurons. Based on your readings, design an experimental study that would test the contribution of the local environment to the determination of dendritic, axonal and molecular phenotype of different types of projection neurons.

Question 5: The behavioral phenotypic analysis of mutant zebrafish to analyze the function of a gene with a putative neuronal function.

Question 6: The morphological analysis of mutant zebrafish to analyze the function of a gene with a putative neuronal function.

Student 23
Targeted Nanoparticles for HDAC Inhibitor Delivery to Advanced Prostate Cancer.

Question 1: The development of castrate resistant prostate cancer (CRPC) is a complicated process which involves the androgen receptor. Novel therapies such as abiraterone can decrease residual androgens in the castrate state along with tumor regression and improve survival in patients. However, all these castrate resistant tumors eventually relapse. Please describe the alternative escape routes for these patients and describe possible intervention for patients that have progressed not only on androgen ablation therapy but on these lysase inhibitors such as abiraterone.

Question 2: Histone deacetylase inhibitors are novel compounds that have shown a wide range of clinical activity in pre-clinical models yet there clinical benefit has been limited to a few indications. Please identify some of the limitations of histone deacetylase inhibitors in patients and describe some alternative solutions which may improve therapeutic ratio of this class of agents.

Question 3: Thrombospondins (TSPs) belong to the group of matricellular proteins, which are non-structural extracellular matrix proteins that modulate cell-matrix interactions and cell function in injured tissues or tumors. They interact with different matrix and membrane-bound proteins due to their diverse functional domains. Recently, antibody array profiling reveals serum TSP-1 as a marker to distinguish benign from malignant prostatic disease. Describe experiments that could allow you to determine the significance of TSP-1 in tumor progression.

Question 4: Recent studies have shown that microRNA (miRNA) inhibitory activity can be quantified by examining their target mRNA expression levels. In a recent study, 8 mRNA microarray datasets were examined to infer and compare the miRNA activities between prostate cancers (PCs) and normal tissues (NTs). Gene expression analyses showed that miRNA activity is stronger in PCs. This conclusion was consolidated by target protein expression and also showed that miRNA activity is more reproducible than miRNA expression across different datasets, which suggests that miRNA activity is a good feature for the classification of cancer subtypes. However, no attempts have been made to develop miRNA-based therapies for prostate cancers. Describe experiments that will allow you to identify target miRNAs with the potential to treat prostate cancers. Describe experimental in vivo delivery models to examine the efficacy or treating cancers by targeting miRNA.

Question 5: Histone Deacetylase inhibitors (HDACi) have a long history in drug development yet their application to human tumors is currently limited to the treatment of Cutaneous T Cell Lymphoma. What have been barriers to the development of HDACi for solid tumors? Is there any evidence that HDACi will

have therapeutic efficacy in prostate cancer, or other solid tumors at this stage of development? How do newer strategies for development of HDACi hope to overcome these barriers?

Question 6: While HDACi appear to be selective against cancer cells, the mechanism of action of these agents is complex. Which of the proposed mechanisms appear to be most relevant in targeting solid tumors? Are there specific biomarkers that may help select which patients could respond to this therapy? How would a clinician-scientist develop a biomarker development strategy to better define which patients could benefit?

Question 7: What is the clinical evidence that PLGA nanoparticles may be effective? With respect to clinical development, what are the advantages of these nanoparticles, and what are the potential limitations of this approach?

Question 8: You have been asked to serve as a consultant to a Yale-sponsored biotech company that is attempting to develop siRNA's as therapeutic molecules for the treatment of cancer. One of the main challenges that this company is facing is how to optimally formulate these siRNA's. Please cite several types of delivery systems that could be used and discuss all of the below points:

1. Theoretical mechanisms by which each system would selectively improve delivery of siRNA's into tumor cells.
2. Potential long-term antitumor effects of each system.
3. Potential side effects of each system.

Student 24

Regulation and Function of Macrophage Polarization in Ischemic Renal Injury

Question 1: Constitutive activation of the kinase LKB1 leads to the formation of polarized domains in the plasma membranes of epithelial cells, even in the absence of cell-cell contact. Under normal circumstances, intercellular adhesive and occluding junctions are thought to be required to establish and maintain the compositional integrity of polarized domains. How can the activation of a single kinase lead to the formation of spatially and compositionally differentiated membrane domains? Design experiments to investigate how those domains might be formed, and what prevents them from intermixing in the absence of tight junctions.

Question 2: Energy depletion leads to activation of AMPK in a variety of tissues, which acts through downstream effectors to turn on pathways that enhance energy generation and turn off pathways that utilize energy. Thus, AMPK is viewed as an "energy sensing" kinase that helps to mitigate potential damage to cells from energy deprivation. In the renal proximal tubule, energy depletion leads to loss of epithelial polarity (see for example: Fish EM, Molitoris BA. N Engl J Med. 1994 330:1580-1588) and to endocytic internalization of a substantial fraction of the normally cell surface Na,K-ATPase. This makes sense, in that down-regulation of sodium pump function will reduce proximal tubule cell energy utilization and thus help to protect the individual proximal tubule cells from damage due to energy depletion. In the Drosophila system, however, energy depletion and AMPK activation appear to promote epithelial polarization. How does this all fit together? Why might it make sense for an organism to activate mechanisms to maintain epithelial polarity and cell surface pump distribution in the proximal tubule, even if doing so puts the proximal tubule cells themselves in greater jeopardy of damage due to energy depletion? Formulate a testable hypothesis that connects the role of AMPK in responding to energy depletion to its possible role in generating or preserving polarity in epithelia in general and in the proximal tubule in particular. Design experiments to determine whether AMPK activation in the proximal tubule preserves epithelial polarity in the face of energy depletion and, if so, whether this is a beneficial response to ischemic injury.

Question 3: Connective tissue matrix proteins traditionally have been considered to have a primary role in imparting structural integrity to tissues. Accordingly, these proteins are critical anatomic determinants of organ function. Research over the last 20 years nevertheless has established key roles for different matrix proteins or matrix protein domains in the regulation of different cellular functions, such as in the areas of cell lineage specification, cell differentiation and survival signaling, angiogenesis, and immunity.

Provide examples of two matrix proteins, or specific matrix protein domains, that subserve such cellular functions and discuss how these functions are integrated into the homeostatic and repair properties of a particular tissue or organ.

Question 4: Antenatal tissue injury (or surgical intervention) is noteworthy in that healing proceeds in the absence of scarring.

- What may be the physiologic basis and possible regulatory mechanisms responsible for this remarkable observation? Why may this property be lost post-natally?
- How may this observation be exploited for therapeutic benefit in adults?
- Thoughtful speculation and discussion are welcome.

Question 5: Macrophages are prominent components of atherosclerotic plaques. The populations of macrophages within plaques are morphologically heterogeneous- some contain large lipid droplets (foam cells) and others do not. Activated T cells are also present within plaques. Please design a research plan to answer the following questions: What effects might T cells have upon plaque macrophage phenotypes and functions? What T cell-derived molecules are likely to mediate such effects? Macrophages are thought to participate in complications of atherosclerosis such as plaque rupture and thrombosis. How might these complications relate to specific patterns of macrophage activation?

Question 6: A modified version of IL-4 that binds to its receptor(s) but does not signal has been reported to benefit patients with asthma. Please outline an investigation that you might perform in patients to discover how this agent affects the lungs of asthmatic patients? (Hint: consider obtaining samples by airway biopsies and by bronchial alveolar lavage of airspace cell populations.)

Student 25

Novel insights into proximal tubule function: finding novel disease-causing genes in a large cohort of patients with Dent's disease without mutation in CLCN5 or OCRL1 using whole exome sequencing.

Question 1: Mutations in the proximal tubule phosphate transporter NaPi-IIa have been associated not only with phosphaturia, but with Fanconi's Syndrome and multiple proximal tubule transport defects.

a. Formulate at least 2-3 cellular and molecular hypotheses to explain this observation.
b. Design experiments in cell lines and/or animal models to test these hypotheses

Question 2: ClC-5, the protein product of the mutant gene causing Dent's disease, is a chloride-proton exchanger.

a. Formulate at least 2-3 cellular and molecular hypotheses to explain how mutations in ClC-5 could lead to the pattern of proximal tubule transport defects seen in Dent's disease.
b. Design experiments in cell lines and/or animal models to test these hypotheses

Question 3: There is a great interest of exploring the role of rare variants in complex diseases. A commonly adopted and often successful protocol is to sequence diseased patients and identify "interesting" variants unique to these individuals with reference to a database of variants created from

a sample of "control" individuals, possibly in combination with some public databases. Write an NIH style proposal to request funding to collect and sequence additional control individuals to complement your current database of 200 people. You need to justify the need for additional samples, discuss the strategy to select the control individuals, and describe the biostatistics and bioinformatics tools to be implemented to facilitate data analysis.

Question 4: More than 70 chromosomal regions have been implicated in Crohn's disease. Write a proposal with a budget of $2 million for genotyping/sequencing to identify more regions associated with Crohn's disease. Assume that there is no need to budget cost for sample collection. You need to justify why more resources are still needed for this disease and why your proposed study will have sufficient statistical power to identify additional disease associated regions.

Student 26

Regulation of VEGFR Receptor 2 Expression by FGF Signaling and Cholesterol

Question 1: Statins lower LDL cholesterol and reduce CV morbidity and mortality. However, there is ample evidence that statins exert cardioprotective actions independently of lowering LDL cholesterol. Propose a series of experiments to identify such mechanisms to test this idea and develop a novel strategy how to manipulate this pathway to test its relative importance in vitro and in vivo.

Question 2: You have discovered a novel gene (cholestrin) that regulates cellular cholesterol homeostasis. The loss of function of this gene increases free cholesterol levels, and the expression of LDL-R. Explain how this may occur and design a series of experiments to dissect the molecular pathways leading to the phenotype.

Question 3: A novel protein kinase is identified that promotes angiogenesis when activated by compound E. Design a proposal including a series of experiments to determine the mechanism of action of the kinase and whether it has a physiologic or therapeutic roles in angiogenesis.

Question 4: Compound S is identified in a large chemical screen to inhibit smooth muscle cell proliferation. Design a proposal to assess its mechanism of action and to test its potential efficacy for clinical application.

Appendix H.
Sample Clinically Based Written Exam Questions

Project: Metabolic Characteristics of Obese Children with Normal Glucose Tolerance vs. Impaired Glucose Tolerance

Question 1: Design a study to identify risk factors for development of diabetes in obese adolescents. Based on these risk factors, develop and validate a predictive model for diabetes in obese adolescents. Be sure to include how risk factors will be identified and narrowed for inclusion in the model, and detail the methods by which the model will be validated. Please write a 6-8 page proposal in the format of an NIH application, according to the attached format.

Question 2: Pfyzer Pharmaceuticals has developed a new compound (RW2003) for treatment of glucose intolerance that appears to work through inhibiting intramyocellular muscle lipid deposits. Phase I and Phase II studies have been completed, which have revealed that oral doses of 500 mgs BID thought to be in the therapeutic range, achieve good peak serum levels (21 mcg/ml +/- 7.2 (SD), n=55 subjects). Toxicity is seen mainly at doses greater than 2.5 gms/day, and consists of nausea, diarrhea, and muscle aching (n=75 subjects). One subject developed significant CPK elevations at low dosage, necessitating discontinuation of the drug. Pfyzer has now approached you to design a Phase III trial for testing the efficacy of RW2003. They want your guidance in designing the clinical trial, including (but not be limited to) enrollment and exclusion criteria, sample size, randomization and blinding methodology, monitoring process/adverse effects, duration of follow-up, study endpoints, analyses and handling of missings/drop outs. Please write a 6-8 page proposal in the format of an NIH application, according to the attached format.

Project: Patterns, Probability, and Predictors of Recovery from Disability in Activities of Daily Living

Question 1: Design a study to identify risk factors for functional decline during the first 6 months of hemodialysis in older persons. Based on these risk factors, develop and validate a predictive model for functional decline during the first 6 months of hemodialysis in older persons. Be sure to include how risk factors will be identified and narrowed for inclusion in the model, and detail the methods by which the model will be validated. Please write a 6-8 page proposal in the format of an NIH application, according to the attached format.

Question 2: Review of previous literature and your preliminary research work suggest that an intervention consisting of mild exercise and cognitively stimulating therapeutic activities may help to prevent functional decline during the first 6 months of hemodialysis in older persons. Design an intervention trial to test the efficacy of this intervention strategy. Your description should include (but not be limited to) enrollment and exclusion criteria, sample size, randomization and blinding methodology (if applicable), monitoring process, duration of follow-up, study endpoints, analyses and handling of missings/drop outs. Please write a 6-8 page proposal in the format of an NIH application, according to the attached format.

Project: Persistence with Oral Hypoglycemic Therapy One Year after Initiation

Question 1: Design a study to identify social psychological risk factors for medication non-adherence in older persons. Use a theoretical framework for developing and validating a predictive model that includes these risk factors. Include a societal level risk factor (e.g., cultural norms) and a social risk factor (e.g. that involves social dynamics between at least two people). Detail the method by which the

model will be validated. Please write a 6-8 page proposal in the format of NIH application, according to the attached format.

Question 2: Design a study to identify risk factors for medication non-adherence in the first 6 months after a new prescription for a stating for the treatment of hyperlipidemia. Based on these risk factors, develop and validate a predictive model for non-adherence. Be sure to include how risk factors will be identified and narrowed for inclusion in the model, and detail the methods by which the model will be validated. Please write a 6-8 page proposal in the format of an NIH application.

Question 3: Review of the literature suggests that a telephone-linked computer system, whereby patients receive automated telephone calls reminding them about the importance of the medication and providing strategies for self-monitoring of pill taking may increase adherence during the first 6 months after a prescription for a statin. Design an intervention trial to test the efficacy of this intervention strategy. Your description should include (but not be limited to) enrollment and exclusion criteria, sample size, randomization and blinding methodology (if applicable), monitoring process, duration of follow-up, study endpoints, analyses and handling of missings/drop outs. Please write a 6-8 page proposal in the format of an NIH application, according to the attached format.

Question 4: Self and family management of illness requires various levels of intervention depending on the nature of the problem and the state of the science specific to the problem. Identify a self and/or family management problem that needs to be examined. Describe and substantiate an approach to developing a self or family management intervention for the problem you have identified based on very little being known about the problem. In your response consider the phases of intervention development, survey, experimental and quasi-experimental approaches, and qualitative and quantitative methods. Use a specific illness management problem to provide context for your response. Please write a 6-8 page proposal for a study in the format of NIH application based on these concepts according to the attached format.

Question 5: Self and family management in the context of chronic illness is a dynamic and complex phenomenon with multiple related factors that affect response and outcomes. Focusing on either self or family management, discuss how the concept has been defined and the extent to which there are shared or competing views of the concept in the literature. What outcomes (individual or family) have been linked to self or family management and what variables have been shown to mediate and or moderate such outcomes? Please write a 6-8 page proposal for a study in the format of NIH application based on these concepts according to the attached format.

Project: ® before ⟨: The importance of residual ®-cell function for ⟨-cell secretion of glucagon in response to hypoglycemia

Question 1: Defective glucose counter regulation with associated loss of “warning symptoms” of impending severe hypoglycemia (AKA hypoglycemia unawareness) remains one of the major obstacles to successful intensive treatment of type 1 diabetes mellitus (T1DM). Design a randomized clinical trial to determine whether use of continuous glucose monitoring to reduce exposure to biochemical hypoglycemia can improve or normalize counter regulatory hormone responses to hypoglycemia. Your description should include (but not be limited to) background, enrollment and exclusion criteria, sample size, randomization, monitoring process, duration of follow-up, primary and secondary study endpoints, analyses and handling of missing data and drop outs. Please write a 6-8 page proposal in the format of an NIH application, according to the attached format.

Question 2: Assume that the recent phase 3 studies from Macrogenics have demonstrated that its anti-CD3 monoclonal antibody is effective in preserving residual beta cell function in T1DM and this drug has

been approved by the FDA. Your new biotech company has a new drug in this class that may have important advantages over the approved monoclonal antibody. Design a pivotal phase 3 study that is designed to be used to obtain FDA approval for your agent and to shown its advantages over the currently approved compound. Your description should include (but not be limited to) background, enrollment and exclusion criteria, sample size, randomization, monitoring process, duration of follow-up, primary and secondary study endpoints, analyses and handling of missing data and drop outs. Please write a 6-8 page proposal in the format of an NIH application, according to the attached format.

Question 3: Can a closed loop mechanical pancreas maintain endogenous insulin secretion longer in patients with new onset type 1 diabetes receiving immunotherapy in attempts to reverse the disease?

Question 4: Can a closed-loop mechanical pancreas restore hormonal responses and awareness to hypoglycemia in patients with long-standing T1DM with hypoglycemia unawareness?

Project: Toll-Like Receptors in Older Adults and Response to Vaccination

Question 1: A newly developed avian influenza vaccine was given intramuscularly to healthy adults and age-matched patients with diabetes. The serologic efficacy of the vaccine was determined by measuring avian influenza virus antibody titer before and at 1 month after the vaccination. The results of the study showed that the increase in avian influenza virus antibody titer after vaccination was lower in patients with diabetes compared to healthy individuals.

Based on observations above, develop a strategy to determine the potential mechanisms for impaired antibody production in patients with diabetes in response to the newly developed avian influenza vaccine.

Question 2: Through your research you have identified two groups of patients, one of which had significant increase in the incidence of viral infections (HSV and CMV), whereas the other had increased incidence in gram-positive bacterial infections. Your goal is to identify the molecular basis for these immuno-deficiencies. How would you go about it, what would be the rationale for each step of your analysis and how would you test your hypothesis?

Question 3: You identify a cluster of Icelandic families in which life-threatening infections with S. aureus, L. monocytogenes and M. tuberculosis are observed in multiple family members in several generations. You suspect that mutations in TLR2 may be responsible for the observed clustering of infectious diseases, and you find that TLR2-mediated signaling responses (e.g. up regulation of co-stimulatory molecules, cytokine production) are markedly reduced in affected individuals. Write a proposal to test the hypothesis that mutation(s) in the TLR2 locus are responsible for these clinical observations, and describe your approach to identifying such mutation(s).

Question 4: You are head of R&D at Morck, a major pharmaceutical company interested in metabolic diseases:

a) Please describe and provide the rationale for a novel drug target for treatment of insulin resistance in type 2 diabetes.
b) The Morck preclinical division has come up with a drug that works on your target and it has proved to shows some promise to reverse insulin resistance in rodents and obese monkey models of type 2 diabetes. It also looks to be safe in humans as reflected by initial Phase I studies. Describe in detail the key Proof of Concept protocol

(s) in humans that you would implement to test out whether Morck should move forward with this novel drug.

Question 5: The federal government has issued an RFA for vaccine development for aquatic flu in light of concern for a possible future pandemic. A Science Park biotech firm has investigated gateway receptors that are believed to modulate response to the virus responsible for aquatic flu. There is evidence that an existing vaccine to another virus has putative, but weak, protective activity toward aquatic flu and gateway receptors are known to modulate response to the other virus and the vaccine. The Science Park firm has developed a compound that enhances the activity of gateway receptors and believes that using this approach in conjunction with an existing vaccine would be both quicker and more cost effective than developing an entirely new vaccine. As they are not primarily a vaccine company, they believe this approach, the time and cost savings, and their expertise in gateway receptors and the new compound would be to their competitive advantage in this application. The caveat is that the vaccine has been associated with the rare occurrence of a debilitating neurodegenerative disorder. The only known risk factor for who gets the presumed side effect is age (younger at greater risk). Gateway receptor activity, on the other hand, decreases with age. The concern is that by giving the compound that enhances gateway activity the risk for the dreaded side effect may increase and the potential liability would be phenomenal given the relatively younger age of those affected. As proposed federal liability protections for vaccine development have not yet been approved, and it's not clear they would apply in this case anyway given the novel approach chosen, the firm is obviously concerned that this potentially lucrative grant may result in liability exposure that would bankrupt the small company. Having heard about your work with similar Toll-like receptors and your medical and clinical epidemiology background, they would like for you to collaborate with them in designing a substudy that would assess the risk factors for developing the neurodegenerative disorder and ideally, demonstrating that it was not the enhancement of gateway activity that was responsible should any increased incidence occur. While this is neither your primary area of interest nor expertise, there is synergy with your own work and this company's, as well as the fact that they are a well-established and cutting edge firm, so the prospect of future collaboration on projects closer to your areas of interest is intriguing. Write a 6-8 page proposal for this substudy in the format of an NIH application, according to the attached format.

Project: Measurement of Physical Activity in Obese and Non-Obese Child

Question 1: Considerable attention is now focused on the prevention of childhood obesity, but little is known about exactly what is to be prevented and the priorities for intervention at either the individual or the social level. Most basic is the issue of whether diet, activity, or both must be targeted, but beyond this specific questions also remain about the importance of education, of changing a child's relationship with food, of modification of television time, of broader interventions that would create healthier environments in schools, of decreasing exposure to advertising, etc. Design a study or a series of studies that would identify the key individual and/or environmental risk factors that would offer the most fruitful opportunities for the prevention of childhood obesity.

Question 2: You have been approached by a colleague at Dartmouth Medical School about a recent apparent increase in the incidence of temporal lobe epilepsy in the Connecticut river valley in upstate Vermont and New Hampshire. Shown below are the numbers of new cases presenting to Dartmouth-Hitchcock Medical Center for the past year, in comparison to the preceding 5 years. The state of residence of the patients is provided.

	Total New Cases TLE	From VT	From NH	From Mass

1998	5	2	2	1
1999	6	3	2	1
2000	9	3	4	2
2001	4	4	0	0
2002	6	4	1	1
2003	39	28	8	3

The populations from which these patients are drawn are as follows: VT: 750,000; NH: 625,000, MA: 400,000.

Your colleague has done a preliminary calculation suggesting that this increase is highly statistically significant. She would like you to verify her calculation. She also has done some preliminary investigation and believes that the most likely risk factor is consumption of venison by hunters. The deer population in VT has recently been shown to be infected with the dreaded IMP2004 virus. This virus is neurotropic for the temporal lobe of deer, and causes the animals to be attracted to bright orange hats and vests. However, she cannot exclude the role of two other potential confounders in the results: 1) Consumption of locally prepared moonshine, previously shown by her to cause TLE; 2) Excessive reading in the Dartmouth Hitchcock library, previously shown by her to protect against TLE. She would like you to design a study to determine if venison for the increase in TLE. She wants you to specify study design, enrollment and exclusion criteria, sample size, monitoring process, duration of study, and appropriate end points.

Question 3: Design a study that examines the effect of a community-based intervention to increase physical activity on adoption and maintenance of this behavior. However, the study should also include as specific aims identification of predictors, mediators, and modifiers of adoption and maintenance of an increased amount physical activity. The study should be conducted within a diverse community (i.e., mix of SES, ethnicity, ages, body composition, and comorbidities). Be sure to include your approach to recruitment , eligibility criteria, sample size, randomization, duration of follow-up, data collection methods, how your variables (i.e., predictors, mediators, and modifiers) will be identified and narrowed for inclusion in the study, design of the intervention, handling of compliance and adherence, and data analyses.

Question 4: Design a study for a multi-level intervention to prevent obesity in children. Based on the risk factors and knowledge of the role of health care providers, suggest an intervention that can target individual, family, and provider behaviors. Design a clinical trial to examine the efficacy of this intervention.

Question 5: Design a school-based exercise intervention for obese children recently diagnosed with insulin resistance. In designing this intervention, please consider the following factors:

- The children are from a low-income Hispanic community;
- Caloric restriction may or may not be included as part of the intervention;
- The mode, frequency, intensity, and duration of exercise best suited to lipid metabolism;
- The duration of the intervention best suited for retention and adherence, but sufficiently long in duration to observe changes in the outcome variables of interest;
- What outcome variables will be measured and how often?
- What effect modifiers will be considered?

Question 6: Please summarize the evidence that physical activity (or lack thereof) is a major factor in the recent apparent increase in the prevalence of obesity. Design a study to prove (or disprove) this hypothesis.

Project: Characterization of Abnormal Neural Connectivity in Autism Spectrum Disorders using Multimodal Magnetic Resonance Imaging Techniques

Question 1: The issue of disordered brain connectivity in autism is interesting in light of various theoretical/psychological models of CNS functioning in autism. Would you pick one of the several overarching theoretical schemes (central coherence, theory of mind, enactive mind, or executive function) and provide a short (4-page) summary of how this theory might translate into problems of brain connectivity.

Question 2: One of the most interesting aspects of autism over the years has been the observation of individuals with islets of unusual ability (autistic savants). A large literature, mostly consisting of case reports but some more controlled studies exists (going back to a classical paper on this topic by Kurt Goldstein). Would you provide us with a summary (4 to 6 pages) of how problems in brain connectivity might be understood as contributing to this phenomenon and the even more common issue of marked scatter in developmental attainments/functioning.

Question 3: Design a hypothesis driven study to examine the question of altered connectivity in the brain in the development of autism and its relationship to cognitive impairment using diffusion MR imaging. With a focus on a particular network, describe the subject population you would examine, the data you would collect, the analysis methods you would perform and the statistical comparisons you would make.

Question 4: Please describe in detail the processing techniques used for the DTI group analysis using FSL. This should include a description of the data mapping to common space and the quantification.

Question 5: Please describe in more detail the registration techniques you are proposing to use towards completing the aims of your proposal.

Project: Characterization of Clinical Characterization and Amygdala Hypothesis of Childhood Disintegrative Disorder

Question 1: Previous studies of head circumference and brain size in individuals with autism have provided evidence for developmental differences in the first two years of life, indicating an early phase of excessive brain growth, although these findings are not entirely consistent. Briefly review the literature on volumetric measurements of overall brain size as well as individual cortical structures in individuals with autism, focusing on the amygdale, and describe in detail how you would characterize and test for differences in amygdala volume in individuals with ASD and individuals with CDD. A short discussion of any practical and methodological difficulties you envisage should be included. Based on your thesis proposal, formulate a hypothesis about potential differences in growth trajectories for specific structural characteristics of the amygdala that may differentiate individuals with CDD from individuals with ASD or typically developing controls. Describe what measurements you would make, and what statistical tests you would implement, to evaluate your hypothesis. Conclude how your results may shed light on differences or similarities in brain development and amygdala function for CDD vs. ASD, relative to the thesis in your proposal regarding the etiology of these disorders.

Question 2: Your clinical topic is very much concerned with the issue of regression. This is an interesting and complicated topic in the more general area of autism and I wonder if you could address the

following: a) in what ways is the regression in CDD different than that (relatively commonly) reported in autism, b) is there some evidence that these are similar or distinctive phenomena?, c) is regression observed in normal development or children with other developmental problems and if so how does this differ from that observed in CDD and d) is it possible that the regressive process in CDD is one that is more generally observed in autism and related disorders and our current (CDD) approach 'reifies' what might be an artificial distinction?

Question 3: As with many conditions, both analyses and diagnoses of Childhood Developmental Disorder (CDD) are often conducted in terms of sets of discrete characteristics. These are often treated as lists of characteristics or symptoms -- such as those listed in Table 1 of your Thesis Proposal. However, cognitive psychologists have emphasized that we represent causal knowledge in terms of theories and structured models rather than via lists of unconnected features. In fact, this is true even for practicing clinicians, whose underlying knowledge of the mental disorders that they treat is far more structured than the lists upon which their diagnoses are (supposed to be) based. (For examples of this work from Yale, see Woo-kyoung Ahn's recent research -- for example: Kim & Ahn, 2002, Journal of Experimental Psychology: General, "Clinical psychologists' theory-based representations of mental disorders predict their diagnostic reasoning and memory.") What causal models might underlie the lists of characteristics that are typically associated with CDD? Might some of the characteristics depend on others? (Even from your initial evaluation of the 27 charts, you could in principle look at the *conditional* probabilities that relate these characteristics: if you have one characteristic, which others are you more or less likely to have?) Which possible causal models would be most theoretically interesting? Does the 'amygdala hypothesis' make any predictions about which features should be causally central? In addition, in general, how could the actual structure of characteristics in CDD be empirically determined?

Question 4: A key element in this dissertation proposal is that a key element in the development of Childhood Disintegrative Disorder is the prodromal development of abnormal levels of anxiety and fear. There is an extensive literature linking the neurobiological substrates of affiliative behaviors and stress response. Briefly, the neural circuitry of both systems intimately involves the hypothalamus particularly the paraventricular nucleus (PVN) and the amygdala. Indeed the PVN contains adjacent neurons that produce oxytocin (OT) and corticotrophin-releasing factor. This observation suggests that an examination of peripheral levels of OT and cortisol before and after a stressor (such as participating in an MRI scan) might be of value.

Question 5: What testable paradigms might you be able to adapt to your study population in order to better understand the underlying neurobiology of the amygdala that is at the core of your proposal? Specifically, how could the vast literature on amygdala function, pediatric anxiety disorders and neuroimaging be adapted to your study population? Importantly, what kind of testable hypotheses might you be able to develop based on some of these paradigms, and applied to your study subjects as well as to (much more easily recruited) control groups, which could include unaffected siblings, age- and IQ-matched subjects with autism, or with mental retardation without autism? Please address feasibility of recruiting such individuals, as well as challenges to performing some of these paradigms among subjects with profound intellectual disabilities. Select references are provided below, and especially from the work from Daniel Pine's lab in NIMH. (references are not included in this document).

Project: Predicting Which Mothers Will Go Back to Smoking After Pregnancy: An Application of the Theory of Planned Behavior

Question 1: Consider the role of individual differences in stress reactivity in the severity of nicotine dependence and conversely in the likelihood of achieving abstinence. That is, propose a study to assess

how nicotine dependence might serve to moderate the physiological and psychological consequences of chronic stress.

Question 2: Consider the role of parental investment in a new infant as a moderating factor in the severity of nicotine dependence and, conversely, in the likelihood of achieving sustained abstinence. Propose a study that uses models of reward or attachment salience as these are related to parenting to better understand parent's efforts to achieve abstinence?

Question 3: You find that cumulative stress predicts return to smoking in your dissertation. What is the next study that you would like to do and how would you apply the findings to real world benefit. Provide the design, rationale and hypotheses for such a study.

Question 4: What are the potential psychobiological mechanisms that could explain the relationship between cumulative stress and planned behaviors? Select at least 1-2 mechanisms for testing in a randomized clinical trial to treat high cumulative stress in infants and children.

Question 5: An educational intervention.
Based on what you have learned so far from your qualitative interviews, design and educational intervention during either the prenatal or postpartum period (or both) that would decrease the number of women who return to smoking after pregnancy. Use a theoretical framework for your intervention. Write a six-page proposal including specific aims, background and significance, preliminary data, research design and methods, and bibliography (the latter will not count towards the 6-page limit).

Question 6: Your hypotheses about the roles of various factors in predicting relapse to smoking after abstinence during pregnancy has been confirmed in your study. By drawing on what you learned, describe the aims and methods (including outcomes) as well as the potential impact of a randomized clinical trial that tests the efficacy of an intervention to prevent relapse to smoking after pregnancy.

Question 7: Stress as a predicator of smoking relapse after pregnancy.
Design a qualitative study to explore the phenomenon of stress in postpartum relapse to smoking. Be sure to specify the sample and the reason for choosing this sample, your qualitative methodology and the details for both the design and the analysis of your research. Choose a theoretical framework for thinking about your research. Justify your choices. Highlight how you will ensure the reliability and validity of your study. Write a six-page proposal including specific aims, background and significance, preliminary data, research design and methods, and bibliography (the latter will not count towards the 6-page limit).
It is recommended that you read select sections of Karen Glanz's book Health Behavior and Health Education. Select references are provided below.

- Curry LA, Nembhard IM, Bradley EH. Qualitative and Mixed Methods Provide Unique Contributions to Outcomes Research. Circulation 2009;119: 1442-1452.
- Côté L, Turgeon J. Teacher, Appraising qualitative research articles in medicine and medical education. Medical Teacher 2005; 27(1):71–75.
- Malterud K. Qualitative research: standards, challenges, and guidelines. Lancet 2001; 358: 483–88.

Project: Neural Response to Stress and Food Cues in Obese Individuals

Question 1: Describe a study to determine reproducibility or fMRI techniques and the methods that you are using in your study.

Question 2: What is the link between the insulin status and dopaminergic brain function and the implications for human health and disease?

Question 3: Your data suggest that the level of glucose in the fasted condition has an effect on food=reward motivation. Design a study that will test this possibility independent of the level insulin and gut hormones. How would you determine if the effect of glucose is dependent on the presence of insulin resistance? Please write a 6-8 page proposal in the format of a NIH application to address these issues.

Question 4: Design a study to test the hypothesis that insulin per-se acts on food-reward motivation independent of changes in glucose and that this effect is altered in patients with insulin resistance. Are there other factors (fuels and hormones) influenced by insulin that might directly mediate insulin's effects? Please write a 6-8 page proposal in the format of a NIH application to address these issues and point out the limitations of such a study.

Question 5: The NIH has just released a request for applications to identify neural risk factors for the development of obesity. Identify an overarching goal for a new application and three specific aims that would address this goal. Each aim should have a specific prediction and a few sentences describing how you will test the prediction. In the next section you will outline the background and significance of the area of research you propose. Make sure to highlight specific aspects of your proposal that are innovative and promise to yield original insights beyond what is currently known. You should also include a section of potential pitfalls and make sure to justify the method you will use to test your hypothesis. Place your emphasis on framing the importance of your question and upon the justification for you particular method. The proposal should be 6-8 pages in the format of an NIH application.

Question 6: Recent data from fMRI studies shows that the insular cortex is hyper-responsive n overweight compared to lean individuals. This is true in a variety of tasks. For example, the insula responds more when subjects are in a hungry state (but just resting during the scan). It responds more to the sight of food images and the taste of palatable foods. Based on what is known about insular cortex make a prediction about what this ubiquitous hyperactive insular response might mean. Please write a 6-8 page proposal in the format of an NIH application in which you outline an experiment designed to test your prediction. Discuss how your preliminary data might support your prediction. Make sure to include a section on potential pitfalls and alternative interpretations of potential findings.

Question 7: In general, reward processing can be dissected into component processes. In a 6-page NIH-style application, describe how you might investigate brain- and behavior-based aspects of reward evaluation or responsiveness that are related to food and those that are independent of food, and how you might investigate aspects of reward processing that relevant to obesity.
Question 8: Individual differences, for example in genetic and environmental factors related to stress, may influence brain structure and function that is found in individuals with obesity. In a 6-page NIH-style application, describe how you might investigate such stress-related factors with respect to brain function in obesity.

Project: Cognitive Function, Symptom Severity & β2*nAChR availability in the Psychosis Prodrome

Question 1**:** A number of studies have suggested that there is a significant effect of maternal smoking on sensory attentional function. What are the animal studies that have attempted to identify the neuronal mechanisms responsible for this effect of developmental smoke exposure? How have the animal studies helped clarify the human data and vice versa?

Question 2: A number of molecular mechanisms have been proposed for how nicotinic receptors might alter function of cortical and thalamic neurons over the long term. What do you think are the most important molecular mechanisms involved in the ability of nAChRs to modulate activity in this circuit?

Question 3: Describe a study that could investigate whether nicotine exposure increases or decreases the risk that a prodromal patient will develop frankly psychotic illness. Consider alternative designs and describe the strengths and limitations of the selected design and alternatives.

Question 4: Describe a study that could investigate whether specific behavioral effects of nicotine could make prodromal patients vulnerable to nicotine exposure and subsequent addition. Include discussion of anxiolytic, stimulant, and precognitive effects and how to determine their relative importance.

Question 5: Adolescence is marked by rapid developmental changes in executive control functions including inhibitory control. Further nicotine use increases in adolescence. Discuss how to design a study of the impact of nicotine use on the developmental trajectory of adolescent EF changes.
Question 6: Clinically there are often reported larger numbers of adolescents with mild to moderate hallucinatory experiences that appear 'normative' or never develop into a disorder per se. How might this group relate clinically to those who are in a prodromal phase and how might you think about the effect of puberty and nicotine exposure on these kinds of 'epiphenomenon' in adolescent development.

Question 7: The post mortem data suggest that smokers with schizophrenia fail to upregulate β2*nAChR receptors. Preliminary findings at Yale using the nicotinic agonist [123I]5-IA in SPECT imaging suggest differences in availability of β2*nAChR's between non-psychiatric smokers and smokers with schizophrenia. While the in vivo data suggest lower β2*nAChR availability in schizophrenic smokers, one cannot determine from the current in vivo data whether this reflects a failure to upregulate β2*nAChRs. It is not clear whether the abnormalities in β2*nAChR availability in schizophrenia are due to illness, medications, smoking or interactions between these factors. As you have stated in your prospectus, the study of prodromes will help "partially parse medication and illness effects."
Question: Explain how you will parse this out?

Project: Osteoporosis among HIV-infected individuals in China

Question 1: Consider 3 major concepts you have identified in your social science reading this semester that are applicable to patient choices among selected heath behaviors, including adherence to medical regimens. Name and describe the 3 concepts and their sources. Then, integrate the concepts into the relevant hypotheses underlying the relevant to the aims of your dissertation, and describe how you might test those hypotheses.

Question 2: Assuming your work shows a particularly advantageous medical regimen for the patient population being studied and you wanted to scale up that set of interventions to be used nationwide in China, describe some critical steps to orchestrate that scale up, based on your readings of the diffusion literature.

Question 3: Design a community-based behavioral intervention study to predict the adoption of an osteoporosis preventive behavior in China.

Question 4: Given data showing that patients with a serious candidate disease (ex. HIV, Cancer, RA) are under-treated for co-morbid conditions, design an intervention study to improve quality of care delivered