The final statement for a Ford/Knight project should include the following components, in this order:

1. Name and title of faculty director

Kalani J. Seu, Assistant Professor of Chemistry

2. Title of project

Techniques for Investigating the Interaction of EGCG with Lipid Vesicles

3. What is the research to be conducted? What are the anticipated learning goals and outcomes for yourself, the students, and the community? How does the project relate to the mission of the college? Please be specific. Note that items #1, 2 and 3 could be included in reports describing F/K projects, and posted on the F/K web page.

Lipids are a major component of cell membranes and the chemical and physical properties of lipids, as well as their role in many cellular processes, are not completely understood. The lipid research field is constantly growing and work in this field has provided insights into some of the physiological roles of lipids as well as some of their alternate uses (e.g. drug delivery systems). Of recent interest in the field of lipid research is the effect of antioxidants on membrane stability. Epigallocatechin gallate (EGCG), an antioxidant extracted from green tea, has recently shown promise as a therapeutic for HIV, cancer, and many other disorders. EGCG has also recently been shown to disrupt the integrity of lipid membrane vesicles. The mechanism and details of the interaction between EGCG and lipid membrane vesicles is still unclear.

For the past 3 summers and past 3 semesters, I have worked with a total of 6 students investigating the effects of membrane composition on EGCG-induced vesicle leakage with the hope of learning more about the fundamental chemical and physical properties of lipids. We have been using a known fluorescence vesicle leakage assay to quantify the amount of membrane leakage induced by EGCG. As the lipid composition of the vesicle is altered, we observe changes in the amount of membrane leakage caused by the EGCG. Our most recent results have provided some insights into the mechanism of EGCG-induced membrane leakage. The data suggest that incorporation of some common membrane lipids (i.e. PG, PE, and cholesterol) results in a reduction in vesicle leakage. We suspect that membrane charge, viscosity, and rigidity play an important role in the mechanism of EGCG-induced LUV destabilization. Investigation into the effects of other lipids on EGCG-induced leakage is currently underway and my students and I hope that charge density calculations and lipid diffusion data from the literature will provide some additional information.

This project has shifted slightly from learning about the fundamental chemical and physical properties of lipids to becoming more focused on the mechanism of EGCG interaction with lipid membranes. Though we have evidence that membrane charge and membrane viscosity play a significant role in EGCG induced membrane leakage, we do not have direct chemical evidence of this interaction; in particular, spectroscopic evidence (e.g. NMR, FTIR, Raman, etc.).

I would like to work with a team of 3-4 students investigating the use of spectroscopic techniques to provide direct evidence of EGCG-lipid interactions. That is, students would identify available techniques, explore the techniques for their viability, and then design and conduct the initial experiments to observe EGCG-lipid interactions. This data would be useful in that it might confirm/deny or support/refute the EGCG-membrane interactions currently being observed using the vesicle leakage assays. One favorable technique might be the use of Fourier transform infrared spectroscopy (FTIR), however, I am not convinced that this technique will work. We have a variety of other spectroscopic techniques available and I would like to work with students to investigate which technique(s) will serve our purposes best and how to design and develop a series of experiments to test this.

As part of its mission statement, Earlham College emphasizes collaborative student/faculty research and the importance of providing students with opportunities to work with other students and faculty in a cooperative manner. This Ford/Knight research project is one such

opportunity. This project also emphasizes the recently adopted ten-year mindset. As students think more about their future directions, it is my hope that this Ford/Knight project will provide them with a glimpse of what a researcher or doctoral student would be doing on a regular basis. Students will have the opportunity to collaborate with each other and me in a manner that allows them to apply the skills that they have gained in their course work, develop new skill, and actively participate in the reflective learning process. Additionally, both the Chemistry and Biology/Chemistry Interdisciplinary Majors require research for graduation. This Ford/Knight project would be an additional research opportunity for students to fulfill this requirement.

Student goals:

- To provide students with an independent student-faculty research experience that:
  - is truly collaborative where the faculty member is learning along with the student and students have the opportunity to apply the chemistry knowledge that they obtained in their course work.
  - expands on their existing knowledge; for example, investigating new applications for instruments that they have used previously for other purposes.
  - gives them first hand experience tackling a scientific problem including researching the topic, preparing an organized experimental plan, carrying out the experiments, and evaluating and adjusting appropriately throughout the process.

Faculty goals:

- To provide an environment in which I can explore and learn along with the students as a small team.
- Incorporation of the products of this work into the courses I teach, particularly in labs and Special Topics courses.
- To develop a new direction for the current research work that is being done on this project. New data will help to confirm/deny or support/refute what we are currently seeing using the vesicle leakage assay experiments. Hopefully this will provide additional data that, when combined with the vesicle leakage assay data, could be sufficient for a publication.

Outcomes.

Students should be able to:

1. systematically approach, design, and tackle a problem as a team.
2. apply and expand on existing chemistry knowledge to solve a problem.
3. use the reflective learning process which involves constant evaluation and adjustment(s) in order to achieve a goal.

4. Exactly how will the work be collaborative? Specifically, what research and analytical work will the students do? How will the students' work be evaluated? Please be specific, especially in relating assessment to student learning.

I will collaborate directly with students and students will collaborate with each other as we determine the appropriate technique and experimental protocol for detecting the direct interaction of EGCG with lipid vesicles. My expertise lies in lipids and I am familiar with a variety of different instrumental techniques, however, I do not have a clear vision as to what technique will work best for the next step of this project. I will be investigating and learning, along side the students to formulate a plan and to test our options.

Students will do background research on the available techniques and instrumentation. I will work with the students to search the literature to find other techniques/instrumentation that have been used for purposes similar to ours. We will then design, prepare, and carryout the appropriate control experiments and measurements to test each viable possibility. Students will then process and analyze all of the data that is produced.

Throughout this project, we will use a reflective learning process to direct us toward accomplishing our goal/solving our problem. This will involve constant evaluation of our experimental design and making the appropriate adjustments based on what does and does not

work. As research is an active process in which students learn by doing, students will be evaluated largely on participation. Students will also be evaluated on their brainstorming and problem solving contributions; as this project is collaborative in nature I will be looking for students to contribute ideas and suggest both logical and creative solutions to problems. Students will be asked to informally present information, and propose possible applications of said information, from literature articles. Students will also keep laboratory notebooks. Lastly, at the end of the semester, students will prepare a poster summarizing the findings of their work and present it at either the Earlham Annual Research Conference (ARC) or the Natural Science Division Undergraduate Research Poster Conference.

5. What academic preparation should students have?

Students participating in this project will likely be Chemistry or Biology/Chemistry Interdepartmental Majors having completed Equilibrium and Analysis (CHEM331) with a 'B-' or better.

6. How many students will be involved? Four students is the norm, but past projects have had as few as three and as many as eight students.

3-4 students will be involved in this project.

7. What is the schedule for the project? Please indicate

- ***semester and year during which the project will occur***
- ***how the work will be accomplished during a semester, and perhaps***
- ***how time before and/or after the research term is to be used.***

This project will be conducted in the spring of 2015. As each student participating in this project will be receiving 3 credits, they will be required to work 3 hrs for each credit hour they receive (total of 9 hrs per week). This includes both scheduled working/lab/meeting times as well as unscheduled time spent doing literature searches and processing data.

8. How will your released time be provided? If you are to be replaced, how will this be arranged?

The ES1 FTE replacement allotted to the Chemistry Dept. has allowed for the incorporation of this F/K project into the departmental unit plan. No replacement costs will be necessary, as this F/K research project will be a part of my regular teaching load.

9. What is the budget for the entire project?

No replacement costs will be necessary as this research project will be a part of my regular teaching load (see question 8). Based on preliminary work conducted by both my students and myself, some of the necessary chemicals and equipment will include EGCG (5 x \$63/50mg), D_2O (4 x \$97/100g), IR windows (\$156/pair), lipids (\$1200), extruders (2 x \$425/kit), and column material.

Release:	\$0
Chemicals/Supplies	\$3,000
TOTAL:	**\$3,000**

10. How will the project impact your teaching generally?

As my biochemistry research has direct connections to many of the courses I teach, this project may provide opportunities for the incorporation of experiments/techniques into my classes, particularly in labs and Special Topics courses. Additionally, this work will help me keep current in my research field and thus, up-to-date with course material.

11. What are your plans for sharing your research results with the Earlham community? Publications, exhibits, departmental colloquia, presentations at the Earlham Annual Research Conference, and readers' theater performances are just a few of the vehicles used in the past.

The findings of this work will be presented at either the Earlham Annual Research Conference (ARC) or the Natural Science Division Undergraduate Research Poster Conference. Depending on the progress made and the nature of the results, this work may also be presented at the Annual Indiana Local Section American Chemical Society Poster Session.