Project Summary

Cell division is a costly activity for a cell, requiring many structural and metabolic resources within the cell to complete. Likewise, fusion of haploid cells, a part of the reproductive cycle for many organisms, is also resource intensive. Preliminary data in our labs indicates that the presence of the glucose sensor, Gpr1p, in yeast is necessary for induction of the mating pathway in MAT**a** cells. However, there is no evidence that the pathways for Gpr1p and Ste2p (both G protein coupled receptors, GPCRs) converge. We propose to examine the hypothesis that glucose sensing via Gpr1p and yeast mating are interdependent. Our first aim is to further examine the interdependence of Gpr1p signaling and yeast mating through direct measurement of Gpr1 activity upon pheromone stimulation and Ste3p activity in *GPA1/GPA2* deletion mutants. The possibility that receptor-associated proteins are involved will be addressed by screening mutants for these proteins from a deletion library. Our second aim is to examine interactions between these GPCRs. Direct protein-protein interactions will be explored using a variety of techniques including immunoprecipitation, phage display, and screening of available mutants of Ste2p. Interaction through membrane colocalization will be examined by isolation of yeast lipid raft equivalents in wild type and mutants for lipid biosynthesis. Our final aim is to determine if other components of these seemingly divergent pathways interact through screening of a yeast gene deletion library and through microarray analysis. The ability of the cell to coordinate glucose sensing and mating is of tremendous physiological importance. However, very little is known about how nutrient availability plays a role in yeast mating.

Intellectual Merit

This project will fill a large gap in the understanding of the relationship between nutrient sensing and mating in yeast. Identification of interactions between Gpr1p and a mating receptor either through protein-protein interaction or through membrane colocalization has the potential to produce an excellent model for the study of GPCR interactions in a haploid eukaryote with only 2 GPCRs. This proposal includes development of novel approaches to study GPCR oligomers that have the potential to impact the broader field.

Broader Impacts

This proposed project will significantly impact undergraduate students, faculty and science departments at three small colleges. Undergraduate students will be exposed to current research during an intensive 10 week summer project with the opportunity to continue the research during the academic year. This proposed research will give undergraduate students the opportunity to design, carry out, interpret, and disseminate the results of their own experiments in a community of peers and faculty from primarily undergraduate institutions (PUIs). The students involved in this project will be recruited from rural Appalachian student populations at participating institutions, with an emphasis on recruiting typically underserved, minority students. This project utilizes established methods that are appropriate for undergraduates, most of which have been utilized by undergraduates under the supervision of the PIs. This project represents an opportunity to improve upon the skills of the PIs who will translate the research into their teaching labs. Subsequently, the science education of hundreds of students at the three participating PUIs will be enhanced.