

PROTEOMICS AND MS FACILITY
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Personnel
Indiana University

Postdoctoral Reference for Flaubert Mbeunkui, Ph.D.

Applicant for: Postdoctoral Position in Experimental Molecular Biochemistry, Immunobiology,
Bioanalytical Chemistry and Biochemical Surface Treatment

Dr. Flaubert Mbeunkui is currently on a postdoctoral appointment in my research group in the Cancer Research Institute at the University of South Alabama. He did his undergraduate work in his home country, and came to me after completing his Ph.D. in Germany. He started in January 2004 on a two year appointment developing proteomics methods for studying changes in cancer. His appointment was been extended to 31 March 2006 to allow his current research projects to be completed. The lack of funding has limited any further extension. While Flaubert's first language is French, he has a good command of English and has taken additional classes at USA. I do not have any problems communicating with him.

While Flaubert has worked on several research projects, his main effort has involved the development of methods for examining secreted proteomes from cancer cells lines and contrasting those from cell lines that have similar origins but different metastatic characteristics. Obtaining clean secreted proteomes is difficult and the best in the literature from mammalian cells was only 50% enriched at best. Flaubert did some careful examination of the sources of the other proteins and can now routinely obtain secreted proteomes that are much better than 90% enriched, often containing no sign of the most abundant contaminating proteins. Thus he has established ways to wash the cells so as to remove all traces of the serum they were originally grown in, and determined the conditions in which almost no cell lysing occurs. This required searching the literature and running trial analyses, using the results of both to feedback into the method to refine the approach. Once he had the "clean" secreted proteomes he thoroughly checked the internet and predictive engines to establish the validity of the proteins identified. The method is the subject of a paper he wrote which has just been published on-line. He is now working on two other sets of cell lines related to breast cancer and melanoma. This is the work that we plan to have completed by the end of March, and submitted for publication soon after.

Flaubert has also been worked on two other associated projects. One involved the isolation of membrane proteins from cells with the aim of performing the same differential proteomic analyses on these across cell lines similar to what is done with the secreted proteomes. He also had a personal interest in MMP and other enzymes that are involved in changing the extra-cellular environment. Both of these efforts gave way to the secreted proteome work when he had developed this into a routine procedure. He has also actively participated in discussion on other research projects in my group, especially that involving glycosylation and other posttranslational modifications. He actively participates in discussions within the facility and presents internal CRI seminars on his research.

During his time with me he has advanced his knowledge on many methods. Our emphasis is on the use of proteomic methods based on mass spectrometry to study cancer. He is quite aware of the things to avoid in method development for MS, and established a procedure for the concentration of proteins from dilute solutions (e.g. media) and the partial fractionation of these on a t-C2 column. We now use this approach routinely in several of our projects. Flaubert has used many analytical approaches such as ultracentrifugation and density gradient separations in conjunction with chromatographic approaches. He has used gel analyses (and in-gel digestion) to identify proteins and complemented this with the selection of antibodies followed by Western Blot analyses. He is extremely familiar with the preparation and digestion of proteins for MS analyses and the interpretation of MS-based sequence data for protein ID. He makes routine use of the Mascot search engine we have in-house to analyze his data, but hand interprets data as needed.

While I hope I have covered the main areas for the advertised position, I will be more than happy to answer any other questions you may have. Please contact me by email or phone.

Sincerely:

A handwritten signature in black ink, appearing to read "L. Pannell", is written over a vertical red line.

Lewis K. Pannell, Ph.D.

Head, Proteomics and Mass Spectrometry Research Facility,
and Professor of Biochemistry and Molecular Biology