Fiscal Year:	FY 2009	**Task Last Updated:**	FY 11/05/2008
PI Name:	Tai, Yu-Chong Ph.D.		
Project Title:	In-flight Blood Analysis Technology for Astronaut Health Monitoring		
Division Name:	Human Research		
Program/Discipline:	NSBRI		
Program/Discipline--Element/Subdiscipline:	NSBRI--Smart Medical Systems and Technology Team		
Joint Agency Name:		**TechPort:**	Yes
Human Research Program Elements:	(1) **ExMC**:Exploration Medical Capabilities		
Human Research Program Risks:	(1) **Medical Conditions**:Risk of Adverse Health Outcomes and Decrements in Performance Due to Medical Conditions that occur in Mission, as well as Long Term Health Outcomes Due to Mission Exposures		
Space Biology Element:	None		
Space Biology Cross-Element Discipline:	None		
Space Biology Special Category:	None		
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Zip Code:	91125	**Congressional District:**	29
Comments:			
Project Type:	GROUND	**Solicitation / Funding Source:**	2007 NSBRI-RFA-07-01 Human Health in Space
Start Date:	10/01/2007	**End Date:**	09/30/2011
No. of Post Docs:	1	**No. of PhD Degrees:**	2
No. of PhD Candidates:	3	**No. of Master' Degrees:**	0
No. of Master's Candidates:	0	**No. of Bachelor's Degrees:**	0
No. of Bachelor's Candidates:	0	**Monitoring Center:**	NSBRI
Contact Monitor:		**Contact Phone:**	
Contact Email:			
Flight Program:			
Flight Assignment:			
Key Personnel Changes/Previous PI:			
COI Name (Institution):	Ho, Chih-Ming (University of California, Los Angeles) Kasdan, Harvey (IRIS International) Adams, Thomas (IRIS International)		
Grant/Contract No.:	NCC 9-58-TD01301		
Performance Goal No.:			
Performance Goal Text:			

Task Description:	The specific aims of the project include (a) 5-part WBC differential, (b) analysis of WBC subtypes (e.g., CD4+ T helper and natural killer cells), and (c) serum/plasma protein biomarker analysis (e.g., for infection, radiation and bone loss monitoring). This project is a continuation of a related project entitled "Handheld Body-Fluid Analysis System for Astronaut Health Monitoring," in which we explored electrical impedance sensing, fluorescence optical sensing, and flow separation of blood cells in microfluidic devices and portable platforms. We successfully demonstrated fluorescent sensing and counting for WBC count and 2-part differential with a portable prototype micro flowcytometer. For the current project, a major effort is proposed to extend the 2-part WBC differential to a 5-part WBC differential, add cell surface marker detection and analysis capability to the platform repertoire, and add plasma protein detection and analysis capability to the platform repertoire. Our approach to achieve the objectives is to extend the capability of the micro flowcytometer to enable a more comprehensive WBC differential, and allow detection of fluorescent labels attached to ligands used for cell surface marker and plasma protein detection. The second component necessary for extending the platform capability is the offline data analysis software. This software is being developed in Matlab to facilitate both quantitative assessment of fluorescence detection and cell and analyte recognition and quantitation. A total of four demonstration units will be available at the end of the first funding year. Two units are complete and operational. The remaining two units are awaiting power supply components that are scheduled to arrive shortly. Of the completed units one is in use at Caltech, and the second is in use at IRIS International. It will serve as a baseline comparison for the modifications planned to the current 2-color design. Both the excitation and detection capability will be enhanced for the modified units. At the excitation end we will be investigating more powerful sources to provide increased emission intensity. At the detection end we will be investigating methods of increasing the number of colors that can be detected simultaneously. Ability to discriminate among multiple color emissions, even those that do not differ significantly in wavelength will provide the capability to detect multiple ligands simultaneously, and may help in performing a 5-part WBC differential with a single stain such as acridine orange. For the demonstration unit at Caltech, we are working with scientists from Wyle and NASA Johnson Space Center to test this prototype on one of their zero-G flights. We have been investigating the operation of the unit to suit the zero-G flight environment on ground, including mechanical robustness of the system, electrical safety of the system, long-term reliability of the testing, the actual testing protocol, and the data interpretation. We have also been providing instrument documentation, safety documentation, and experimental design to enable the proposed zero-G flight testing. We believe successful completion of the zero-G flight testing will be a major milestone for this project. To analyze WBC subtypes, we propose to use continuous flow separation at upstream and dielectrophoretic (DEP) enabled Coulter counting at downstream. For the flow separation part, we improved from previous design and achieved a very compact design capable of continuous cell separation. Pillars are placed within the microchannel to alter the fluid flow pathway, allowing particles of a certain size to be diverted toward a specified route. In order to accurately calibrate the dielectric parameters (i.e. membrane capacitance, cytoplasm resistance) of different types of white blood cells, a microelectrode-array (MA) was designed and fabricated to simultaneously carry out the impedance measurement for a large amount of cells under a wide range of frequency. The challenge of this task is to immobilize single cells onto this MA with precise position controllability. During the last year, we developed a novel cell immobilization method to accurately control the white-blood cells adhesive and repellent molecules functionality with high spatial resolution. Photolithographically patterned hexamethyldisilazane (HMDS) micron-sized patterns present hydrophobic terminal that were used to physically adsorb the cell capturing antibodies. The non-specific antibody binding was prevented by passivating the other surface without HMDS micropatterns by poly(ethylene glycol) (PEG). Specific biotin-streptavidin complexation was explored to immobilized cell-specific antibodies. High patterning selectivity was achieved and the immobilized antibodies retained their bioactivities to a great extent. By controlling the size of the antibody micropatches, single-cell patterning resolution was achieved using cultured DG75 B lymphocytes as model cells. We believe that using microfluidic networks to accurately control the shear stress imparted on the immobilized cells can further improve the patterning qualities. For the coming year, with the improvement of excitation with laser diode and detection with PMT array on the micro flowcytometer platform, we will explore WBC counting and differential with fluorophore conjugated antibodies. This can greatly expand the capability of the platform. Also a cocktail of fluorescent dyes including acridine orange will be investigated to stain blood for five part WBC differential. For WBC subtype separation and counting, with the successful optimization of the continuous flow separation device, integration of micromixer and deionier as well as DEP focusing devices and Coulter counters will be investigated. The fluorescent particle immunoassay (FPIA) will be investigated for on-chip plasma protein detection.
Rationale for HRP Directed Research:	
Research Impact/Earth Benefits:	The devices under development can be used for earth-based applications. The proposed device use cartridge and hand held system. The cartridge will be cheap and disposable. The results will be available almost immediately to the patients without going through central lab facilities. The device can be used in emergency room, on ambulance as well as at home. As the senior population continuous to grow, this kind of device will find more and more appealing in point-of-care applications.
Task Progress:	According to the original proposal, for the first funding year, we propose to optimize acridine orange staining and testing procedure for 5-part WBC differential, optimize hydrodynamic separator for WBC subtype separation, and plasma preparation for plasma biomarker detection. We are on the schedule. 1. Acridine orange staining and testing procedure optimization: We are in the process of improving the previous two color micro flowcytometer. Excitation with laser diode and detection with diffraction grating and multi-channel PMT will improve the system sensitivity and capability. We also made progress in system software to enable more comprehensive and accurate data processing. 2. Fluorophore conjugated antibody staining of whole blood: As an alternative to chemical staining, we are investigating using fluorophore conjugated antibody to staining whole blood. Antibody staining has been proposed recently as a potential new standard way for five part differential. Antibody staining followed by micro flow cytometer detection might provide a more accurate and specific way to count some subtypes of WBCs. 3. Preparation for zero-G flight test: As a crucial step toward in space use, we are preparing the portable micro

	flowcytometer prototype for zero-G flight test. We are in the process of reinforcing the mechanical structure, performing the reliability test, and customizing the system to fit the zero-G flight test scenario. We plan to run the testing continuously for the whole duration of the flight and record data for both low-G and high-G periods. The data processing will be performed after the flight. Intervention and troubleshooting by crew members are not necessary. 4. Optimization of hydrodynamic separator for WBC subtype separation: Based on our previous devices for continuous size based particle separation in microfluidic devices, a new design offers two orders of magnitude reduction in the separation region, while still achieving the same functional purposes for particle separation. 5. Plasma preparation for plasma biomarker detection: The same continuous size based particle separation device can be used to separate plasma from whole blood as demonstrate in the previous funding period.
Bibliography Type:	Description: (Last Updated: 08/30/2018)
Articles in Peer-reviewed Journals	Zheng S, Lin JC- H, Kasdan HL, Tai YC. "Fluorescent labeling, sensing, and differentiation of leukocytes from undiluted whole blood samples." Sensors and Actuators B: Chemical 2008 Jun 16;132(2):558-67. http://dx.doi.org/10.1016/j.snb.2007.11.031 , Jun-2008
Articles in Peer-reviewed Journals	Zheng S, Liu JQ, Tai YC. "Streamline-based microfluidic devices for erythrocytes and leukocytes separation." Journal of Microelectromechanical Systems. 2008 Aug;17(4):1029-38. http://dx.doi.org/10.1109/JMEMS.2008.924274 , Aug-2008
Papers from Meeting Proceedings	Lillehoj P, Li N, Tsutsui H, Ho CM. "A compact microfluidic continuous flow separator for particle and cell sorting." IEEE 21st International Conference on Micro Electro Mechanical Systems (MEMS '08), Tucson, Arizona, January 13-17, 2008. IEEE 21st International Conference on Micro Electro Mechanical Systems 2008. MEMS 2008. p. 292-295, 2008. http://dx.doi.org/10.1109/MEMSYS.2008.4443650 , Jan-2008
Papers from Meeting Proceedings	Zheng S, Tai YC. "Dual frequency resonance impedance spectroscopy flow cytometry for blood and tumor cells." The 11th International Conference on Miniaturized Systems for Chemistry and Life Sciences 2007. MicroTAS 2007, Paris, France, Oct. 7-11, 2007. 11th International Conference on Miniaturized Systems for Chemistry and Life Sciences 2007. MicroTAS 2007, Proceedings, p. 488-490, 2007. , Oct-2007
Patents	US provisional patent. Patent October 2007. Oct-2007 Tai YC, Zheng S, Lin JC-H, Kasdan H. "Fluorescent labeling, sensing and differentiation of leukocytes from undiluted whole blood samples."
Patents	US patent application. Patent October 2007. Oct-2007 Tai YC, Zheng S. "Streamline-based microfluidic device."