



## Cost of Services

For simple applications that are routinely carried out in the unit, such as monitoring of bovine cell types, cell sorting of particular bovine cell populations, cell cycle analysis, etc. few preliminary tests are needed. However, new and more advanced applications will require some initial tests and planning with the operators of the unit, in order to select the machine, the correct reagents, technical help, the cost and the time required.

As a general guide, the cost of services at the Unit is approximately **US\$50 per hour**. For further information on costs, please contact the unit coordinator.

### For further information please contact:

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# Cytometry Unit

## Fluorescence Activated Cell Sorting and Analysis

Cytometry Unit



**ILRI**  
INTERNATIONAL  
LIVESTOCK RESEARCH  
INSTITUTE

**biosciences**  
eastern and central africa



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## *The Cytometry Unit*

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The cytometry unit operates a number of machines designed to provide high quality data on cell populations. The flow cytometers use laser light of a defined frequency to identify fluorescent signals on cells and provide size and fluorescence data of individual cells at a rate of thousands per minute. Researchers use the technique for a range of applications on animal cells, parasites, bacteria and plant cells when they can be brought in suspension. Using different fluorescent labels, the technique can be employed for a range of applications, including determination of intracellular concentrations of chemicals such as  $\text{Ca}^{2+}$ , discrimination of live and dead cells, discovery of pathogen-infected cells, measurement of DNA content, detection of cell multiplication and identification of functional cells.

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## *The Machines*

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The unit has three Becton Dickinson machines: a **FACScan** which is now only used for routine analyses, a **FACScanto II**, which is a recently acquired, high tech instrument that can simultaneously analyse eight different fluorescent wavelengths, and a **FACStar plus**, which is mainly used for cell sorting cells with a particular size and fluorescence pattern, for further use in functional assays (FACS stands for Fluorescence Activated Cell Sorting). In addition, the unit owns a fluorescent microscope which allows scientists to identify and localize biochemical components in tissues and cells. The Zeiss machine provides top quality digital imaging.

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## *Operation*

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The unit is operated by an electronics engineer, who is licensed by Becton Dickinson to carry out maintenance on the machines. This person makes sure that the machines are in working condition and will help scientists with setting up the machine, running samples and carrying out cell sorting. Researchers who frequently use the machine get training to allow them to operate the machines independently. Data are handed over as hard copy print outs, or transferred as soft copy for further analysis Flo-Jo software, which is available on a few computers.

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## *Species-specific reagents*

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A major application at the institute is the identification and monitoring of particular bovine cell types during infections, such as CD4/CD8 ratios. ILRI is unique in the world in that it has developed one of the most extensive collections of monoclonal antibodies specific for bovine leukocyte populations. Many of these are used and distributed by commercial partners. Using indirect staining, these antibodies allow us to identify the different lymphocyte populations from bovine blood and tissues, such as macrophages, neutrophils, B cells, T cells, gamma delta-T cells, CD4 and CD8 subpopulations and many of the different maturation and activation stages of these cell types. For multi-colour analysis, we need directly labeled antibodies and several of these are being prepared. The fluorescent dyes recommended by Becton Dickinson are: fluorescein, rhodamine, al-

