XML based Process Management in Cryo-Biotechnology: The ChameleonLab

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Abstract: In this paper, we introduce a completely new kind of lab management system, ChameleonLab. We present the main ideas, areas of application, of an XML-based implementation of ChameleonLab and a production system in a large-scale cryobank. The main feature of ChameleonLab is its ability to handle new and future proof substrates for cryobanking. These consist of miniaturised multi-wells and unique cryo-tolerant memory-chips. XML-files stored on memory-chips attached to the samples allow complete control of preparation and handling laboratories. Additionally, full documentation relevant to clinical application is stored decentralised in XML-files on the sample-attached memory-chip. We show the unique feature of ChameleonLab is to schedule and control both automated and manual operating steps. This allows scaling from the small lab to high-throughput environments. Device concepts of implementation like generic and human devices are exemplified. We show that management of biotechnological labs is a new and relevant application area for XML.

1 Introduction

The widespread progress in biomedical science requires new techniques and new technologies for coping with new cell uses. Live cells need to be stored for decades for later stem cell therapy, retrospective diagnostics or tissue engineering [KBRAGW04].

Future biomedical science is certain to create many new applications. Diverse medical applications require different cell workflows. A cell workflow consists of both cell treatment steps and cell handling steps. Treatment is defined to be anything that causes an effect on the cell, whereas handling denotes any activity that does not. In the future, biomedical laboratories will need to carry out a large number of different workflows including steps for cell storage.

Already today's biomedical labs need to have a more flexible and dynamic workflow management than conventionally established. Different solutions are explored in [MY04] and [FK04]. The facts mentioned below show the need for further improvement.

Large numbers of cell samples need to be stored now for later use. Estimates range to more than a billion samples in the next few years.

There is currently only one practical long-term storage technique available for living cells: cryopreservation. It is based on freezing cells and storing them at temperatures between –130°C and –196°C. Liquid nitrogen maintains these temperatures. This technique is well established but the old technology is no longer suitable such high sample numbers. Therefore, the Fraunhofer-Institute for Biomedical Engineering (IBMT) is reengineering the technology and improving the cryopreservation techniques and evolving the future standards [ZKIDSF04].

The main differences between the new IBMT technology and the old one are a dramatic reduction of sample size to between 1/50 and 1/2000 of conventional volumes and the physical attachment of a cryo-tolerant memory chip to each sample carrier or stack. This is now possible because of progress in low temperature electronics [ISZ03], [ISZ04] and [ZIS04]. Each sample's data is stored at-sample on-chip to avoid possibly lethal mistakes in data and sample association, especially in situations of sample exchange. The sample always carries its own information. The need for this solution can be seen in detail in [DIHZ04]. Already in the past, there have been efforts for improving structure and management of biological data [RR04] and [YBM04] showing the insufficiency of conventional sample data handling.

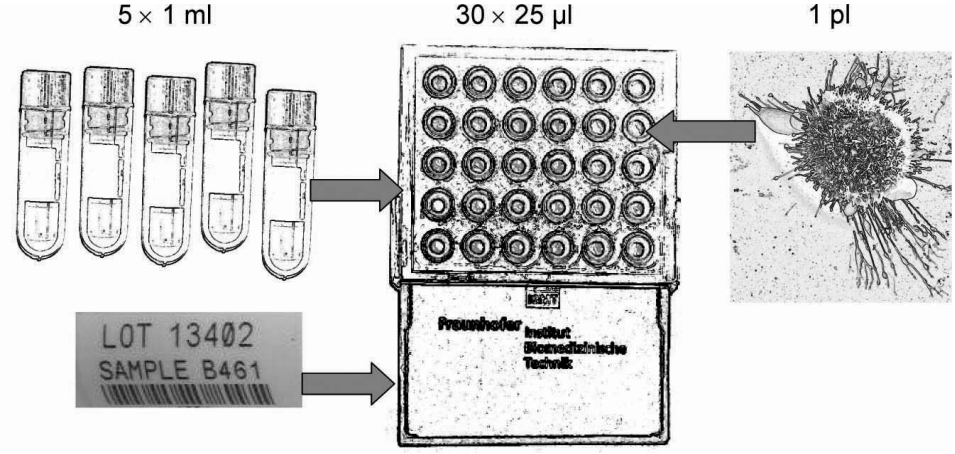

Fig. 1: Comparison of conventional cryotechnology and Fraunhofer IBMT's reengineered cryotechnology. More information can be found in the text.

Whereas a cell‘s volume is 1 pl on average, the smallest conventional cryotubes on the market have a volume of 1 ml. For coping with future cell sample numbers, miniaturization of volume is required and biologically useful. IBMT’s miniaturized substrate contains 30 sample wells of 25 µl volume in a plate of 4x3 cm. Furthermore, in conventional sample storage, sample data are stored in a database to which is referred by a sample identification label, e.g. a barcode. In contrast to that, there is a low temperature tolerant memory chip attached to each single miniaturized substrate or stack with a capacity of up to 1 GB. All sample data are stored at-sample on-chip in an XML file.

The at-sample on-chip data can be backed up in an adaptive sample database for cryobanking [Du03]. Currently, an XML file is used for the on-chip data.

The idea for ChameleonLab has been born from the following facts:

- Lab workflow definitions depend on cell type and on the future purpose of a cell sample. Individual workflow definitions must be allowed for special purposes.

- There will be sample exchange between different labs and cryobanks, e.g. when a cryopreserved cell sample is needed for a therapeutic purpose. This requires exchange of the workflow definitions associated to a sample. Therefore, workflow definitions must be independent of any particular lab. Labs must be able to treat and handle cells purely according to the workflow definitions.

- Mistaking of workflow definitions carries the same risk as errors in sample data.

- Workflow documentation is important. Depending on cell type and biomedical purpose, there are different duties by law for documentation. Cell samples for therapy are defined to be medicines for which e.g. the German medicine act obliges them to be documented in detail for up to 15 years, including each workflow step. Exchange of workflow documentation is important to labs, physicians and science.

2 The ChameleonLab principles

In order to cope with the facts mentioned above, ChameleonLab is based on the following principles.

1) The workflows associated to a sample are defined at-sample.

2) Instead of being a reference to an existing workflow definition, the sample-chip combination is the workflow definition. That means that each single treatment and handling step is defined in detail at-sample on-chip. Thus, labs using ChameleonLab are able to perform workflows unknown before arriving of the sample. The workflow definition is transported in a form suitable for direct apparatus control. Therefore, the sample dictates the lab behaviour spanning the lab type from completely manual to fully automated.

3) Workflow documentation data is stored at-sample.

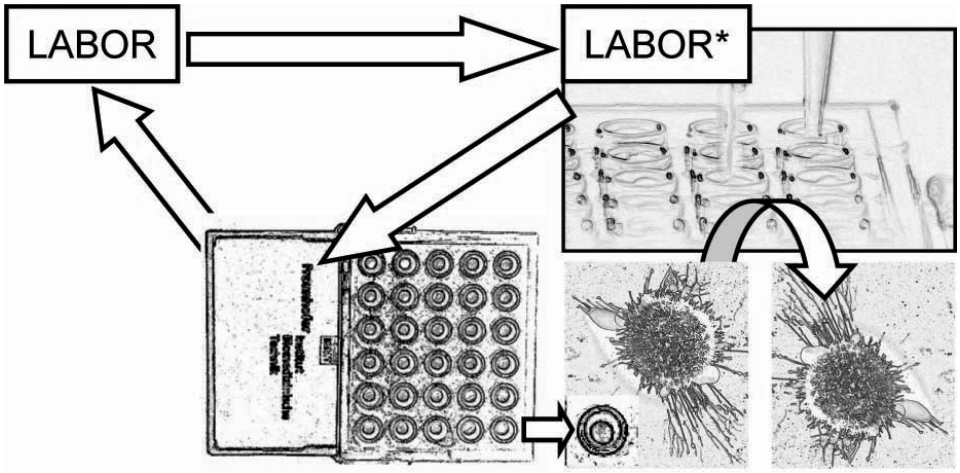

Fig . 2: Schematic view of the ChameleonLab principles showing a sample-chip combination and a cell sample to which the workflow definition is associated.

The at-sample on-chip workflow definition dictates the lab's behaviour. Before execution starts, the lab is configured and initialised accordingly to the workflow definition. Therefore, the lab is transferred from an idle state 'LABOR' to the defined state 'LABOR*'. In this state, the workflow is done. The cell sample changes by workflow. Workflow documentation data is stored at-sample on-chip. After finishing workflow, the lab returns to idle state 'LABOR'.

3 An implementation of ChameleonLab

For a working implementation of the ChameleonLab and a proof-of-concept, we have decided to develop an existing workflow management system to ChameleonLab functionality. Firstly, the at-sample workflow definition and at-sample workflow documentation functions need to be realised. This can be done easily by extending the on-chip XML file by workflow definition tags and workflow documentation tags.

‘Bernstein’ is a well-known workflow management software in the biotechnology market [GB00] controlling fully automated high throughput screening machines for pharmaceutical products. These machines are called HTS modules and consist of different automatic devices like pipette arrays, incubators, screeners, and robotic handling mechanisms for micro-titer plates. In an HTS module, a large number of samples is transported automatically between the different devices carrying out the operational steps defined for a pharmaceutical screening workflow.

The interaction of the different devices within such an HTS module is supervised by Bernstein. Bernstein is a dynamic scheduler suited and adaptable for all screening workflows that can be described as a sequence of operating steps. The operating steps of any screening workflow need to be allocated to the different devices of the HTS module capable of the appropriate step requirements. A Bernstein process is defined to be a subsequence of operating steps which can be fulfilled in direct succession at a particular device. Thus, any screening workflow is a sequence of several Bernstein processes being executed by the appropriately allocated devices.

Each single device is controlled by individual device software. The Bernstein scheduler controls the interaction between the different devices by communicating with each device’s software.

But how can Bernstein be used for development of Chameleon Lab? First of all, any cell workflow is in fact a sequence of operating steps, namely of cell treatment steps and cell handling steps as can be seen by analysing biomedical protocols. Thus far, Bernstein’s functionality matches any biomedical workflow definition well. In an HTS module, Bernstein schedules the operating steps by scheduling those devices to which the operating steps are allocated. This principle implies that all operating steps of any biomedical workflow have to be allocated to appropriate devices in a biomedical lab for being scheduled by Bernstein. Moreover, each lab device needs to be controlled by its own device software for communication with Bernstein. In fact, transferring the function of Bernstein to a biomedical lab regards the biomedical lab as a distributed HTS module. This is the key for our further development.

But in state-of-the-art biomedical labs many of the cell treatment and cell handling steps of a workflow cannot be allocated to devices. This is because there are many simple manual operating steps. Having all of them fulfilled by machines would be a loss of efficiency, require many expensive devices more and would not be cost-effective.

Additionally, there are many small and simple devices that do neither have software nor computer interface and therefore need to be operated manually.

Furthermore, there are also many devices controlled by their own software but having no interface to Bernstein. A standard lab apparatus interface and protocol do not exist so far. Thus, scheduling of manual operating steps must be enabled. This requires the allocation of manual operating steps to an appropriate device.

An appropriate device for manual operating steps is the human device, e.g. a lab employee. To schedule a human device's operating steps we need a communication interface. Therefore, we have developed the so-called 'generic device' software.

Basically, the generic device software has the same functionality as any other device software in an HTS module. There is initialisation by Bernstein and communication with both Bernstein and the device itself. But there is one main difference arising from the abilities of a human device: the 'generic device' software is compatible with all manual operating steps. That means that a single software device can cope with the large number and variety of manual operating steps in a biomedical lab. As with any other device software, the 'generic device' software can control the execution of a sequence of different operating steps. The user interface consists of instructions to the human device in HTML format and of input forms for feedback; this is necessary to get the documentation data for manual operating steps.

Using the human device and the 'generic device' software, any manual operating step can be allocated. Thus, Bernstein can now schedule all operating steps of any cell sample workflow in a biomedical lab that can be regarded as distributed HTS module consisting of different technological devices and the human device.

With this solution, any biomedical lab can be turned into a ChameleonLab simply by using one human device and installing one instance of the 'generic device'software. Also those biomedical labs without any Bernstein compatible technological devices can be turned into ChameleonLabs by having the devices operated manually using the human device and the 'generic device' software.

This solution is fully scalable for any size of lab facility. There can be several human devices in a biomedical lab and several instances of the 'generic device' software, e.g. one in each lab department for achieving higher sample throughput.

We have added XML functionality to both the Bernstein scheduler and the 'generic device' software. Now, Bernstein can read the at-sample on-chip XML file and access the workflow definition for fully automatic initialisation of the ChameleonLab. The 'generic device' software is able to write documentation data into the on-chip XML file while an operating step is being executed. Bernstein, the technological devices and the instances of 'generic device' are equipped with a chip reader for accessing the on-chip XML file.

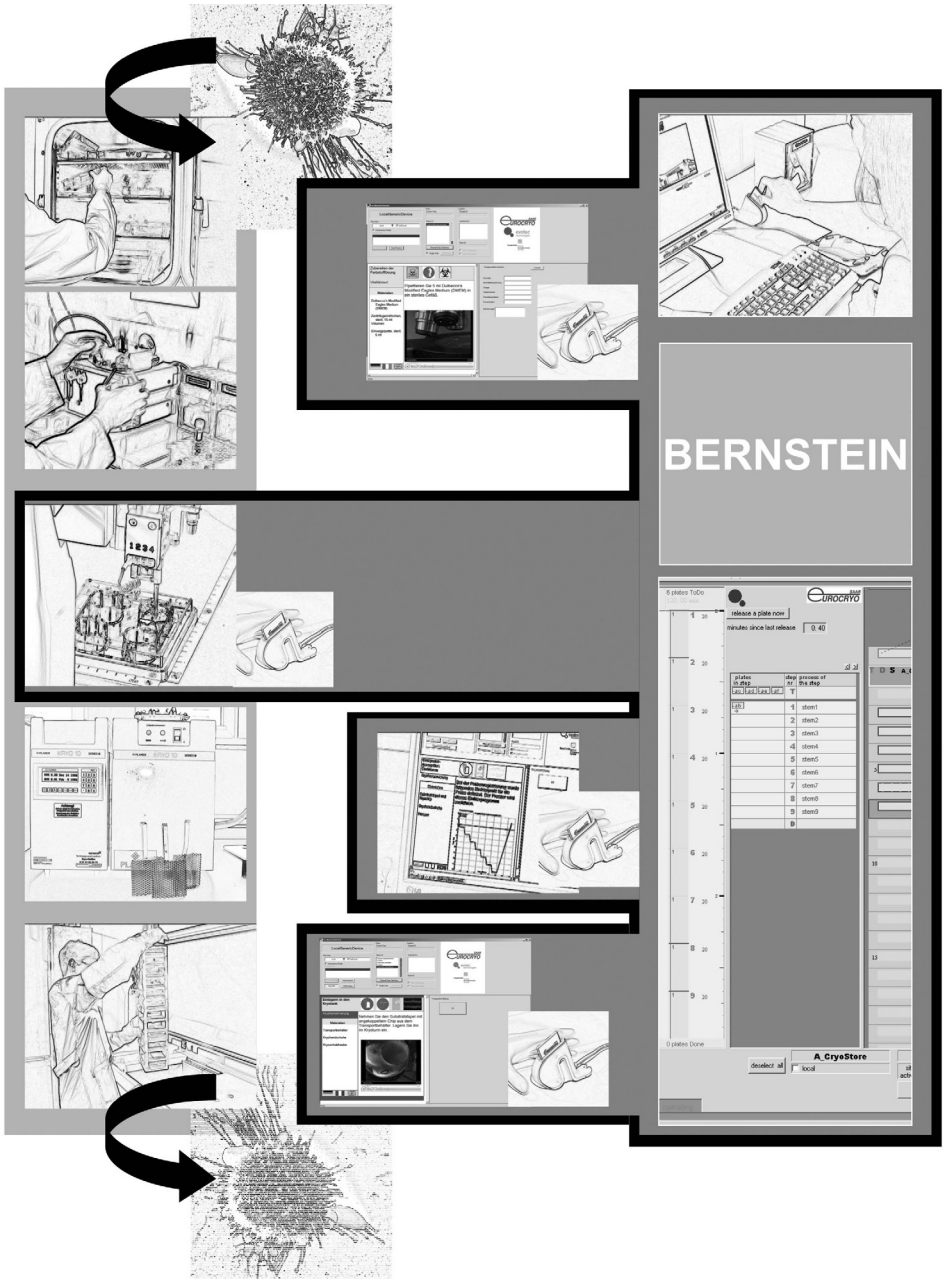

Fig. 3: Visualization of a schematic workflow for L929 fibroblast cells in the ChameleonLab. For further details see text.

Figure 3 shows on the right side the initialisation of ChameleonLab which is done by inserting the at-sample chip into Bernstein's chip reader. The on-chip XML workflow definition is loaded and initialises the lab's devices allocated to the operating steps. During workflow execution, Bernstein controls the interaction of the devices and schedules the processes and operating steps. The chip escorts the sample to each device. Therefore, each device is equipped with a chip reader for supplying the on-chip XML file for writing documentation data into by each device's software. On the left side, some operating steps of the workflow are illustrated from top to bottom. The two uppermost operating steps are fully manual steps which must be fulfilled by the human device. Therefore, both operating steps need to be scheduled by an instance of the 'generic device' software forming the interface between Bernstein and the human device. Taking place in the same department of the lab, the same human device and the same instance of 'generic device' is used for both operating steps. The generic device presents HTML sheets instructing the human device and supplies input forms for documentation data input. The next process is allocated to the nanoplotter, a fully automated device for pipetting small volumes of cryoprotecting solution. This device is controlled directly by Bernstein using a fully compatible device software. The next operating step is a semi-manual sample freezing step. The allocated device is a freezer with non-compatible device software and, therefore, has to be operated manually. Here, the human device is scheduled by the 'generic device' instance of the lab's freezing department. The last operating step is again manual and to be done by the human device as described above. The black border around Bernstein's topography is defined to show the grade of a device's integration. A fully compatible device can communicate directly with Bernstein through a compatible device software and can be scheduled directly. This is indicated by the black border enclosing the device. Human devices or semi-manual devices are not enclosed by the black border because they need scheduling by an instance of the 'generic device' software. Looking at the Bernstein topography marked by the black border, one can see that this topography changes accordingly to the devices allocated to processes. That means that the Bernstein topography adapts to the sample workflow definitions.

4 ChameleonLab as a production system

ChameleonLab is established in the first large-scale research and demonstration cryobank EurocryoSAAR since Autumn 2003 (www.eurocryo.de). EurocryoSAAR currently has 22,000 liters of cooled volume and an infrastructure allowing a maximum of 220,000 liters of nitrogen-cooled storage volume. All cryo preparation labs are controlled and adapted by the stable version of ChameleonLab using XML at-sample on-chip files. In EurocryoSAAR, ChameleonLab is connected and synchronized to a web-based sample database capable of workflow definition and memory chip initialisation.

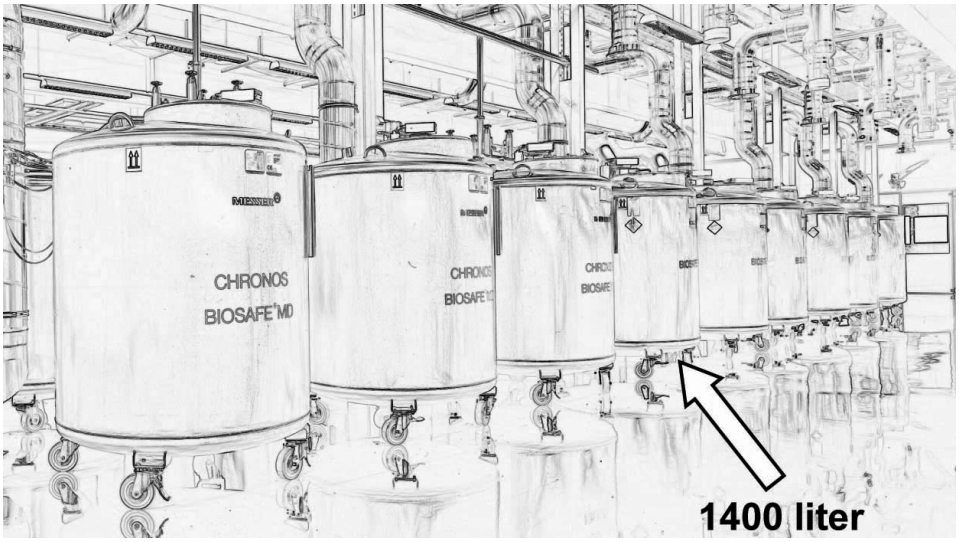

Fig. 4: The EurocryoSAAR large-scale cryobank.

Cell samples are stored long term in the cryotanks using Fraunhofer IBMT's miniaturized substrates with a memory chip attached. Each sample has been prepared in labs equipped with the ChameleonLab implementation. In a tank with capacity of 1400 liters, 15000 microwell plates with attached chips can be stored.

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