Image-based Systems Biology

What You Get is More Than You Can See

"A picture is worth a thousand words" – This well-known quote expresses the common wisdom that a single image can convey a wealth of information. In fact, various areas in the biological sciences experienced a tremendous boost during the last decade owing to the development of ingenious imaging techniques that allow visualizing biological processes with high spatial and temporal resolution.

Today, microscopy experiments are often routinely performed in biological research and provide fascinating insights into the complexity of living systems. In many cases, however, the acquired image data are eventually used for illustrative purposes only, implying that valuable information – e.g. on dynamical, functional and morphological aspects of the biological system under consideration – is disregarded. This is partly due to the fact that manual analyses of image data can be extremely time-consuming and can therefore often not be performed on a statistical level. In principle, this should not be an issue in the computer age that we live in.

"A Computer is Worth a Thousand Hours"

It is obvious that automated processing of images by computers can save many hours of man power. At the same time – and more importantly – analyses by machines are also by far more objective than the view of a researcher being biased by a working hypothesis can ever be. Unfortunately, the development of computer algorithms is a challenging task due to the lack of universal strategies for the appropriate processing of image data. This is mainly attributed to the huge diversity of biological systems reflected by unique image data that typically require situational consideration. Moreover, since image data never show the whole truth of the system's complexity, assumptions about hidden variables may have to be made. Thus, after translating pictures into numbers, the unambiguous interpretation of the readout quantities may still remain a complicated task. Computer simulations of biomathematical models may be an elegant way out and during recent years the biological community is becoming more and more aware of this.

"A Model is Worth a Thousand Hypotheses"

Biomathematical models that are based on image data open up the possibility to systematically study the impact of various model parameters by comparative computer simulations. This enables narrowing down the regime of relevant values for hidden variables and allows formulating hypotheses that may direct further experimental investigations. The art of modeling relies in solving the dilemma of describing complex systems at a level where they become computationally feasible, while at the same time maintaining the model's predictive power to generate new hypotheses. A plethora of different modeling techniques, each with their unique characteristics and defined limitations, exists that have been applied to complex biological systems and have proven successful in formulating new hypotheses.

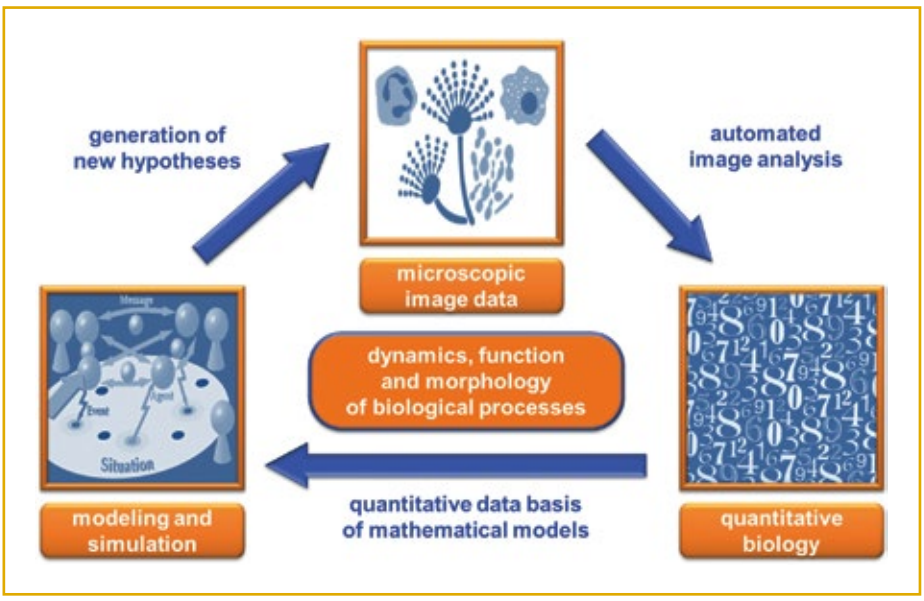

Fig. 1: Iterative cycle between experiment and theory of the Image-based Systems Biology approach. Microscopic image data are translated into numbers of characteristic measures that serve as a data basis for mathematical models. Computer simulations of mathematical models generate new hypotheses that can direct the design of new experiments by narrowing down the regime of relevant parameters.

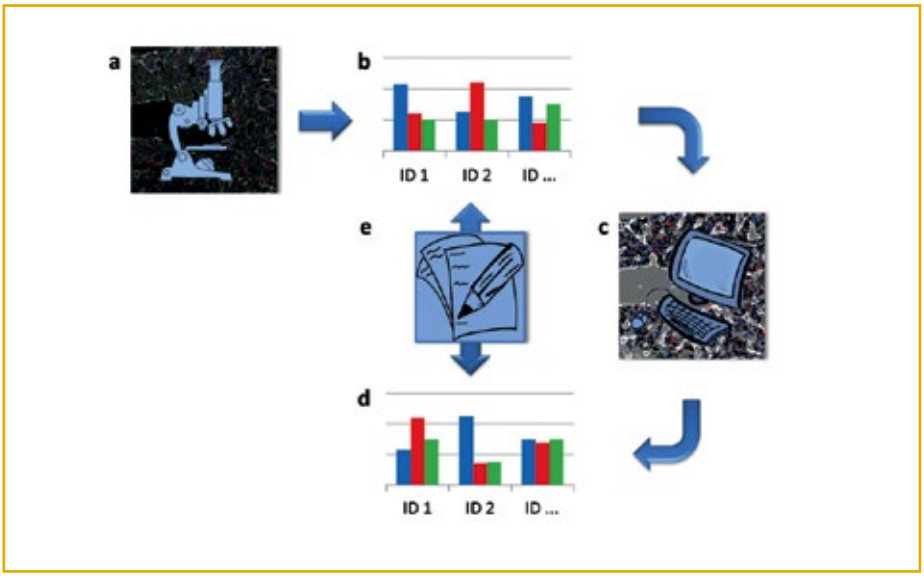

Fig. 2: Workflow of the approach in the bone marrow project. The combination of (a) image acquisition and (b) automated image analysis with (c) mathematical modeling and (d) analysis of the synthetic images enables (e) interpretation of system properties.

The emerging approach of Image-based Systems Biology seeks to take full advantage of the information contained in images and establishes an essential connective link between experimental and theoretical investigations of biological processes at a quantitative level [1]. As indicated in Figure 1, this approach generally combines the three aforementioned elements:

(i) Acquisition and automated analysis of image data for high-content and high-throughput screening,

(ii) Quantitative description of biological processes by appropriate characteristic measures,

(iii) Construction of image-derived spatio-temporal models and predictive computer simulations.

It is obvious that the Image-based Systems Biology approach can actually be applied in virtually all areas of science where image data are generated to visualize the dynamics, function and morphology of system behavior.

To give a typical example, in a recent study we applied an Image-based Systems Biology approach in order to achieve a quantitative interpretation of cell colocalization in respect of plasma cell survival niches [2]. Long-term antibody production is a key property of immunity and involves long-lived plasma cells. They mainly reside in the bone marrow, whose importance as an organ is becoming increasingly evident during recent years. Signals provided by stromal cells and eosinophils in the bone marrow may play an important role for plasma cell maintenance, constituting a survival microenvironment.

To quantify cell colocalization in the bone marrow, we started by generating confocal microscopy images in the bone marrow of mice (fig. 2a). Next, we developed an algorithm for automated image analysis, quantified the spatial distributions of B cells, eosinophils and plasma cells in the image data and computed the contacts that these cells have among each other and with stromal cells in the bone marrow (fig. 2b).

However, it is important to realize that the quantification of spatial correlations from the image data is virtually unusable. Why? – Well, because these numbers alone do not give us a clue how to interpret them. In particular, the distribution of stroma strongly varies from image to image and this makes it impossible to decide whether B cells, eosinophils and plasma cells are distributed in some specific way or whether their spatial positions in the bone marrow are possibly entirely random. This is a quite typical situation and reveals the importance of mathematical modeling and computer simulation as part of this approach.

Consequently, in order to create reference data under defined conditions, we augmented our study by Monte Carlo simulations. Thus, we repeated the actual experiments on the computer, i.e. we performed numerical experiments on synthetic images (fig. 2c). The underlying mathematical model was referred to as random cell position (RCP) model, because cells in the numerical experiments were placed within the image-derived stromal environment in a random fashion. To this end we used the exact experimental distributions of cell numbers and cell sizes per type that were obtained from the preceding analysis for each confocal microscopy image. Next, the spatial distributions of cells and the contacts that cells have among each other were as well computed for the synthetic image data (fig. 2d). By comparing the experimental distributions with the simulated distributions as a reference, it was then possible to interpret the spatial arrangement of cells in the bone marrow (fig. 2e).

Thus, we tested the observed spatial colocalization of cells in the bone marrow against computer simulations based on the hypothesis that cells are uniformly distributed at random positions in the

bone marrow. In this way, we could show that B cells and plasma cells highly colocalize with stromal cells, to an extent larger than in the simulated random situation. While B cells are preferentially in contact with each other, i.e. forming clusters among themselves, plasma cells seem to be solitary or organized in aggregates, i.e. forming loosely defined groups of cells that are not necessarily in direct contact. Our data suggest that the plasma cell bone marrow survival niche facilitates colocalization of plasma cells with stromal cells and eosinophils, respectively, promoting plasma cell longevity.

This example demonstrates that, while automated image analysis alone implies that "what you see is what you get", its combination with mathematical modeling implies "what you get is more than you can see". This is, of course, not only true for the analysis of single microscopy images, but also holds in the context of analyzing time-lapse microscopy videos of system dynamics [3] that often requires derivation of specific measures to characterize and classifiy the system behavior in an automated fashion [4]. Furthermore, the combination of automated image analysis with mathematical modeling naturally establishes a connective link between experimentalists and theoreticians and, by this means, the Image-based Systems Biology approach represents a point of crystallization for interdisciplinary collaborations.

Outlook

In the future, it can be expected that this approach will also be combined with traditional systems biology approaches that are based on so-called "omics"-data, e.g. genomics, transriptomics and proteomics data. While image and video data unravel the dynamics, function and morphology of a biological system, accompanying "omics" data provide information on gene regulatory networks and gene/protein activity. The combination of these approaches may be realized by multi-scale modeling that establishes a connection between the activity patterns of genes/proteins and their actual function in the context of biological processes. Today, we did not yet manage to fully cope with this challenge, but isn't it exciting to have ambitious aims in sight that can only be reached by joining forces in interdisciplinary research? – Well, it surely is.

References

[1] Medyukhina A. *et al.*: Cytometry A, (2015). DOI: 10.1002/cyto.a.22638
[2] Mokhtari Z. *et al.*: Cytometry A, (2015). DOI: 10.1002/cyto.a.22641
[3] Brandes S. *et al.*: Medical Image Analysis 20(1), 34-51 (2015)
[4] Mokhtari Z. *et al.*: PLOS ONE 8(12), e80808 (2013)

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