

2nd European Workshop on Glycolysis

Functional Dynamics Group
Dept. of Chemistry, University of Copenhagen

February 1–2, 2002

Program

Friday, February 1

- 13:00 - 13:15 Preben Graae Sørensen: Introduction
- 13:15 - 14:00 Thomas Mair: Oscillatory glycolysis in an open spatial reactor
- 14:00 - 14:45 Christian Warncke: Dispersion of glycolytic waves
- 14:45 - 15:30 coffee break
- 15:30 - 16:15 Diogo Camacho: Self-induced oscillations
- 16:15 - 17:00 Rolf Wasén: A resilient equation-system from modelling of glycolysis
- 20:30 working dinner

Saturday, February 2

- 9:00 - 10:00 Barbara Bakker: Stability of yeast glycolysis is not controlled by a single oscillophore
- 10:00 - 10:45 coffee break
- 10:45 - 11:30 Francesco d'Ovidio: Precursors of synchronization in glycolytic oscillators
- 11:30 - 12:15 Martin Bertau: *Saccharomyces cerevisiae* as biocatalyst in chemical synthesis and glycolytic NADH oscillations
- 12:15 - 13:30 lunch break
- 13:30 - 14:30 plenary discussion

Abstracts

Barbara M. Bakker: Stability of yeast glycolysis is not controlled by a single oscillophore

Spontaneous biochemical oscillations are often thought to be caused by a so-called oscillophore, a key enzyme that is the primary source of the oscillations. In yeast glycolytic oscillations phosphofructokinase seemed to be such an oscillophore, since removing the positive feedback regulation on this enzyme in a core model of glycolysis, removed the oscillations completely at any combination of kinetic parameter values.

In real glycolysis, however, more than one possible source of oscillations has been identified. In order to quantify to which extent each enzyme contributes to the occurrence of oscillations, we applied the principles of Metabolic Control Analysis. We calculated the control exerted by each enzyme on the real part and the trace of the eigenvalues of stable as well as of instable steady states. We concluded that even in the above mentioned core model, the control on the eigenvalues was distributed among several enzymes. In a detailed and realistic model of yeast glycolysis most, but not all control of the eigenvalues was shared by glucose transport, pyruvate decarboxylase and the utilization of ATP. From these results we conclude that the concept of an oscillophore cannot be applied to complex biochemical networks. Instead, Metabolic Control Analysis provides an unambiguous method to determine the importance of each enzyme for the emergence of oscillations.

Martin Bertau: *Saccharomyces cerevisiae* as biocatalyst in chemical synthesis and glycolytic NADH oscillations

Biotransformations with *Saccharomyces cerevisiae* (baker's yeast) play an important role in modern stereoselective chemical synthesis. With the help of this microorganism reactions are catalysed which classical-chemically can be realised only worse or even not at all.

On the other hand, with living microorganisms the selective conversion of a substrate by a single enzyme to a single product is the ideal case.

Living cells are highly complex systems, and microbial syntheses therefore depend on culture and reaction conditions. These exert a great effect on cell physiology and thus on the outcome of the overall reaction. For these reasons, the elucidation of the processes involved in a whole-cell biotransformation bears a great potential for the development of highly selective biocatalytic

reactions. Also with regard to biotransformations, *S. cerevisiae* is one of the best explored microorganisms.

It is a fact that whole-cell biotransformations may suffer from the opposite-directed stereoselectivities of enzymes which act simultaneously on the same substrate. Latest approaches to inhibit undesired stereoselectivity have been very successful - with the drawback that these methods, although highly effective, are the result of intensive empirical studies.

Further, every biocatalytic method requires very special conditions. The determination of the best process parameters is demanding.

It is therefore necessary to gain a concise understanding of the enzymatic network. For the investigation of stereoselective reductions, time-dependent changes in intracellular NADH concentration are a promising tool for getting deeper insight into how enzymes interact during a biotransformation.

Finally, mathematical methods are expected to provide substantial help in understanding and controlling biotransformations with living cells.

References:

M. Bertau, Biospektrum 2002, **8**, in press.

M. Bertau, Tetrahedron Lett. 2001, **42**, 1267-1268.

M. Bertau, M. Bürli, Chimia 2000, **54**, 503-507.

Diogo Camacho: Self-induced oscillations

The periodic behavior of glycolysis has been observed in various organisms and cell types, namely in yeast. These oscillations occur in yeast when cells are shifted from aerobic to anaerobic metabolism. This is usually achieved by adding D-glucose to starved and aerated yeast cells, followed by cyanide or other mitochondrial respiration inhibitor. It is therefore possible to observe sustained oscillations in the glycolytic NADH concentration.

Are there metabolic products that can induce glycolytic oscillations? If so, what metabolites are those? How do they act, and why? These questions are waiting to be answered in a near future.

Thomas Mair: Oscillatory glycolysis in an open spatial reactor

No abstract available.

Francesco d'Ovidio: Precursors of synchronization in glycolytic oscillators

Macroscopic metabolic oscillations in yeast can be observed experimentally in a CSTR under specific conditions. A question of great importance for modelling yeast metabolism is then how to relate the macroscopic measurements to the individual cells. Using theory for coupled oscillators, we show here that the problem is not trivial, since transitions with very different phenomena at the cellular level can show macroscopically the same features observed in the experiments.

Such different phenomena, however, can be resolved measuring precursors of the oscillations when the system is before the transition, *i.e.*, in the steady state. These precursors appear as deviation from the linear behaviour in the perturbation response and allow to identify the type of the transition without ambiguity and in an experimentally feasible way.

Christian Warncke: Dispersion of glycolytic waves

No abstract available.

Rolf Wasén: A resilient equation-system from modelling of glycolysis

An equation-system from the Michaelis-Menten kinetics for glycolysis following Dan Fraenkel at Harvard Medical School is analyzed. It turns out that it is very difficult or resilient to numeric computation. However using non-linear equation solvers of the kind Groebner basis in computer algebra system the exact parametric solution is found.

This puts lights on the resilience and the explanation for the difficulties is indeed quite remarkable. The reason is the proximity of the datasets to a singularity in the "solution-space". This gives rise to a considerable numerical instability. The remedy seems to be to move further away from equilibrium or simply away from steady state, as Sune Danø has pointed out, in order that the problem should not be that ill-conditioned.

Possibly the Michaelis-Menten kinetics should also be improved or replaced by something better!

A lot of other interesting methodological issues is actualised as well.

Participants

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