as ATP and ADP level of *A. orientalis* with respect to initial glucose concentration to assess the role of the pathway on the production of vancomycin antibiotic whose clinical importance derives from its often unique ability to treat multi-resistant Gram positive infections.

Methods: Enzyme activities and protein levels were determined spectrophotometrically. Nucleotide, glucose and vancomycin analysis were measured with HPLC.

Results: G6PDH, 6PGDH and transaldolase activities of *A. orientalis* increased with increases in glucose concentration from 5 to 15 g/L up to 48th hour as 1.9, 1.7 and 1.8 fold, respectively. 6PGDH activities increased continually with respect to glucose concentration during incubation period although nearly similar values of G6PDH and transaldolase activities were determined before 48th hour of incubation. Intracellular ATP level showed a positive correlation with glucose concentrations while ADP level increased up to 15 g/L. ATP concentration of *A. orientalis* increased rapidly on the 48th hour of incubation as was the case also for G6PDH, 6PGDH and transaldolase activities although the incubation period corresponding to maximum values of ADP shifted to 60th hour. Glycopeptide antibiotic, vancomycin production increased with the increases in glucose concentrations up to 15 g/L by showing coherence the rates of oxidative and nonoxidative parts of pentose phosphate pathway.

Discussion: These results indicate that the both the oxidative and nonoxidative parts of pentose phosphate pathway are anabolic circuit of glucose metabolism and therefore play an important role in precursor supplementation for vancomycin biosynthesis in *A. orientalis*.

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[P-I.187]

Rice oil stimulates high level of L-cysteine and cephalosporin C production in *Acremonium chrysogenum* M35

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Keywords: Cephalosporin C; Rice oil; Cysteine

During the production of cephalosporin C using *Acremonium chrysogenum* on an industrial scale, the fungus was cultivated in a complex medium containing glucose and a various plant oils as the major carbon sources. The addition of low solubility of carbon source such as oil is to avoid carbon catabolic repression. Previously, we found that the addition of 3% rice oil to the medium enhanced cephalosporin C production (4.5 g l^{-1}).

L-Cysteine is a precursor of penicillin and cephalosporin C of β-lactam antibiotics. Cystathionine -γ-lyase, an enzyme that splits cystathionine releasing cysteine, is required for high level of cephalosporin production. Analysis in cystathionine -γ-lyase activity of cultures with different carbon sources showed that the rice oil supplemented culture contained high level of cysteine, whereas rice oil unsupplemented culture contained normal level of cysteine level in *A. chrysogenum* M35. However, cystathionine -γ-lyase activity was decreased in *A. chrysogenum* M35 by rice oil supplemented medium. The results indicated that rice oil was converted into cysteine in *A. chrysogenum* M35 during cephalosporin C production.

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[P-I.188]

Alteration of Mitochondrial NAD Content in Yeast: Physiological Characterization

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Keywords: NAD; mitochondria; yeast; cofactor

In *Saccharomyces cerevisiae* pyridine coenzymes are required in many reactions involving central metabolism and energy generation. Manipulation of their cellular contents as well as of their intracellular compartmentalization is expected to deeply affect overall metabolism, ATP generation and biomass yield. Mitochondria are impermeable to NAD^+ and two proteins, named Ndt1 and Ndt2, are the carriers of NAD^+ from cytoplasm.

In the present paper we investigated the physiological effects of a double *ndt1*Δ *ndt2*Δ deletion and of the overespression of *NDT1* in batch and chemostat cultures. Deletion of both genes resulted in a relevant drop in the NAD^+ mitochondrial level, while the concentration of the coenzyme was sensibly increased in *NDT1* overespressing strain. Consequently, also the NAD^+/NADH mitochondrial ratio was affected in the mutant strains. In batch conditions the double deleted strain showed relevant defects when grown in minimal medium. Moreover, these defects were dependent on catabolite repression of the carbon source used, being light on glucose, and more marked on ethanol. Strikingly, we measured very high respiratory activity for *ndt1*Δ*ndt2*Δ strain, pointing to oxygen consumption rate close to the upper physiological limit for the cell. Chemostat analysis also revealed metabolic alterations for *NDT1* overespressing strain. Despite the high oxygen consumption, both mutants showed low biomass yield values, indicating an impairment in mitochondrial functions. Since glucose consumption rates were higher than the wild type, ethanol was accumulated as a by product, and Crabtree effect was triggered at lower dilution rates. Further evidences confirmed the negative impact of both low and high NAD(H) content onto mitochondrial activity, shedding light on homeostatic mechanisms adopted by the cells to maintain their mitochondrial functionality. This study confirms the importance of coenzyme redistribution on metabolic fluxes, and could help the optimisation of biotechnological processes based on enzymatic reactions involving pyridine cofactors.

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[P-I.189]

The Non Conventional *Z. bailii* Yeast Shows Substantial Differences from *S. cerevisiae* Upon Exposition to Oxidative Stress

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Keywords: yeast; ROS; stress resistence

The industrial exploitation of yeast always requires the attainment of high biomass yield, meaning the instauration of aerobic conditions in the bioreactor. The mitochondrial respiratory chain is the major cellular source of reactive oxygen species (ROS), toxic compounds that can easily damage all cellular components including proteins, lipids and nucleic acids.