

# OptiSet

One plate cultivation set for comparison of EnBase™ cultivation conditions



EnBase™

Note 16 June 2009

## Introduction

The EnBase™ technology based on catalytically controlled glucose delivery results in controlled growth of *E. coli* to high cell densities and enhanced protein production. The 24 deep well plate of the OptiSet enables a direct comparison between the traditional expression method and three different EnBase™ media (mineral salt medium, balanced medium, EnBase™ Flo) and also includes cultivations with an additional boosting step. After only one cultivation optimum conditions for the specific product can be selected from analysis of 3 ml cultures per well.

## Material and Methods

In EnBase™ glucose derives by a biocatalytic reaction from a polymeric substrate, embedded in a gel and slowly diffusing into the medium (columns 1-3) or being directly dissolved in the medium (EnBase™ Flo, columns 4-6). Each of the eight different conditions in the final setup is tested in three parallels in the same plate.

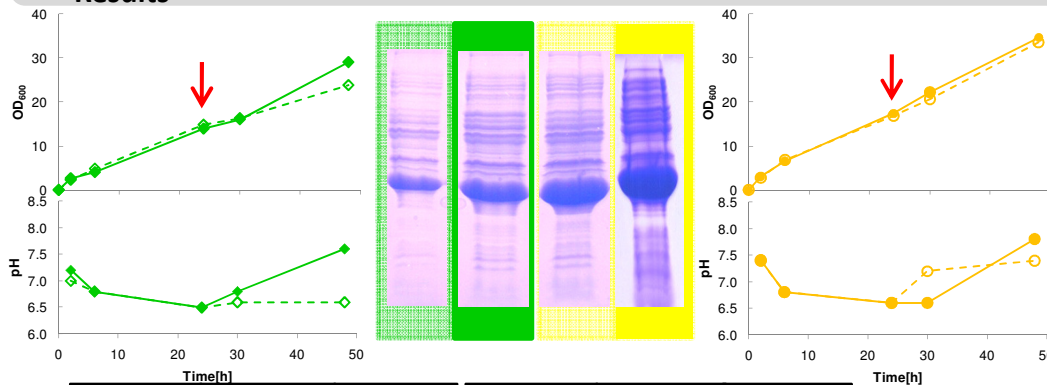
Strain: *E. coli* M2

Induction: 1mM IPTG

Protein: Alcohol dehydrogenase (ADH)

Shaker speed: 200 rpm, Temperature: 30°C

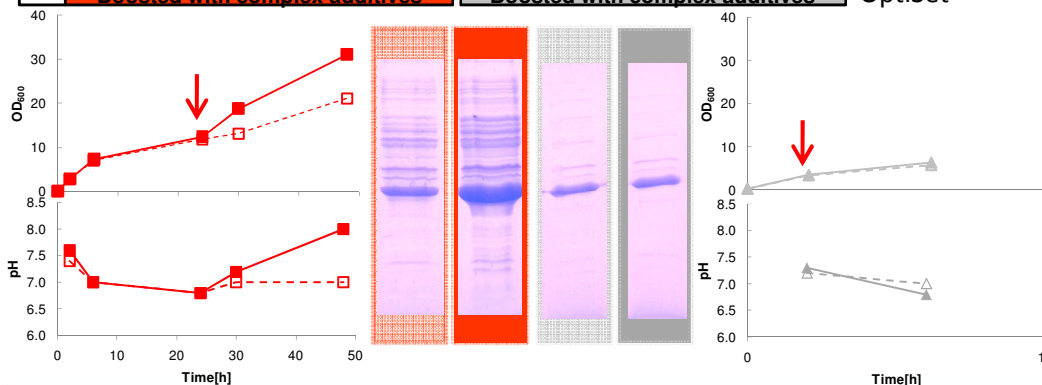
## Results



Recombinant expression of ADH was performed in parallel with four media (labeled with four different colors) without boosting (dashed line, open symbols) and with boosting (solid line, closed symbols). The boosting step at the time of induction is marked by a red arrow. All cultivations performed with EnBase™ media showed at least a final OD of 20; with boosting OD was >30. Analysis of protein samples by SDS PAGE showed highest volumetric protein yield with EnBase Flo.

	1	2	3	4	5	6
A	Mineral Salt Medium			EnBase Flo		
B	Mineral Salt Medium Boosted with complex additives			EnBase Flo Boosted with complex additives		
C	Balanced Medium			User's standard medium		
D	Balanced Medium Boosted with complex additives			User's standard medium Boosted with complex additives		

Plate layout of 24 well OptiSet



## Conclusions

EnBase™ media enable high cell density cultivations. The boosting step increases the pH of the cultivation medium and supports protein production.

