

## Growth Parameter Optimization of Kluyveromyces Marxianus MTCC 4059

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**ABSTRACT**: This study was done to optimize the growth parameters for the yeast *Kluyveromyces marxianus* MTCC 4059 using synthetic MYLP media in shake flask cultures. The conventional methodology was used to investigate the effects of four growth parameters, viz., temperature, pH, incubation time and initial concentration of carbon source on yeast biomass production. It was finally estimated that, for a 10% inoculum size (v/v) of 24h old yeast culture maintained at a constant shaking condition of 150 rpm, a combination of initial lactose concentration 3%, pH 7, temperature 30°C and an incubation time of around 29h yielded optimum biomass (11.25 gm/L) for *K. marxianus* MTCC 4059.

*Keywords*: growth parameter optimization, *Kluyveromyces marxianus*, synthetic media, conventional methodology, yeast biomass production

## 1. INTRODUCTION

A growth medium or culture medium is a liquid or gel designed to support the growth of microorganisms or cells,<sup>[1]</sup> or small plants.<sup>[2]</sup> There are different types of media for growing different types of cells.<sup>[3]</sup>

The strains belonging to the yeast species Kluyveromyces marxianus have been isolated from a great variety of habitats, which results in a high metabolic diversity and a substantial degree of intraspecific polymorphism. As a consequence, several different biotechnological applications have been investigated with this yeast: production of enzymes [ $\beta$ -galactosidase, β-glucosidase, inulinase, polygalacturonases, lipase (He and Tan, 2006; Liu et al. 2006), tannase (Battestin and Macedo, 2007),  $\alpha$ -amylase (Uma Maheswar Rao and Satyanarayana, 2007), β-cyclodextrin glucanotransferase (Ibrahim et al. 2005), dextran dextrinase (Naessens et al. 2004) and chitinase (Nawani and Kapadnis, 2005)<sup>[4]</sup>, among others], of single-cell protein, of aroma compounds, and of ethanol (including high-temperature and simultaneous saccharification-fermentation processes); reduction of lactose content in food products; production of bioingredients from cheese-whey; bioremediation; as an anticholesterolemic agent; and as a host for heterologous protein production. Compared to its congener and model organism, *Kluyveromyces lactis*, the accumulated knowledge on K. marxianus is much smaller and spread over a number of different strains.<sup>[5]</sup>

Despite the importance of these traits, and significant exploitation by the biotechnology sector, fundamental research with *K. marxianus* is just emerging from the shadow of its sister species, *Kluyveromyces lactis*. The

availability of new molecular tools and resources for *K*. *marxianus*, its interesting metabolic and cellular traits, and the potential to become the leading yeast for many biotechnological processes, argue strongly for increased research into this particular species.<sup>[6]</sup>

Increasing industrial demand of this yeast strain requires optimum production methods to ensure the economic viability of the strain at commercial scale. Apart from this, the activity and stability of the products is influenced by the type of strain, cultivation conditions (temperature, pH, aeration, agitation, incubation time) and the growth medium composition (particularly carbon and nitrogen sources) (Schneider et al. 2001; Jurado et al. 2004; Tari et al. 2007). For example, in order to improve the  $\beta$ -galactosidase production, several groups (Fiedurek and Szczodrak, 1994; Bojorge et al. 1999; Furlan et al. 2000 & 2001; A.P. Manera et al. 2008) have made investigations to select microorganisms that have high activity, to evaluate substrates and define optimized fermentation conditions for the chosen microorganism.<sup>[7]</sup>

The "change one factor at a time" method is widely used as a conventional technique for multifactor experimental design. This method involves changing one independent variable while maintaining all others at a fixed level.<sup>[8]</sup>

It is already known that *Kluyveromyces marxianus* has the ability of producing the enzyme  $\beta$ -D-galactosidase which breaks down lactose into glucose and galactose. Because of its gaining popularity as lactose-utilizing yeast, it is the sole purpose of this paper to optimize the growth parameters of *K. marxianus* using lactose as the only carbon source in the synthetic media.

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