The Active Ingredients Used in Galozyme® Probiotic Supplements (Kluyveromyces Marxianus) Vs Competitors Active Ingredients , Saccharomyces (aka brewers yeast)

KLUYVEROMYCES MARXIANUS VERSUS SACCHAROMYCES SP.

For many years the motivations for the use of yeasts in feed reside in their high content in proteins and vitamins (group B). Only recently, the ban of antibiotic growth promoters in feed for production of animal foods in the European Union has increased interest in evaluating the effect of yeast and bacterial products (DFMs – Direct Fed Microbial) on the gastrointestinal ecosystem, rumen microbial populations and overall animal performance.

Feeding yeast (Saccharomyces cerevisiae) has largely increased in Ruminants: lactating dairy cows (for reduction of rumen lactate and prevention of acidosis and improving production efficiency), newborn calves (for reducing enteric disorders), at weaning (for reducing stress and improving intake).

On the other hand some bacterial species have been proposed for monogastric animals, pigs and piglets in particular (ex. Enterococcus faecium is used as a probiotic for piglets and has been shown to modify the porcine intestinal microbiota).

Only one Yeast type, Kluyveromyces marxianus (the strain K.m B0399 in particular), has shown to be equally effective as a health and growth promoter in both monogastric and poligastric animals.

This cumulative effect can in part be explained by unique **immunomodulatory action** of this yeast, unique enzymatic activity **(β-galactosidase and inulinase, but also endopolygalacturonase, carboxypeptidases, and aminopeptidases)** and unique **metabolite complex**. These beneficial metabolites also act as the "active" ingredients themselves and suggest that live K.marxianus cells may not even be required for exerting its effects.

PRINCIPAL DIFFERENTIAL CHARACTERISTICS OF K.MARXIANUS

1. The ability to enzymatically degrade lactose due to the presence of the **enzyme beta-galactosidase** (b-gal) or lactase in the cytosol in its constituents galactose and glucose (Fig.1).

Fig. 1: Beta-galactosidase reaction:

GAL - O - G + H2O = GAL + G

lactose + H2O = galactose + glucose

3. Thanks to its unique enzymatic properties K.marxianus produce valuable bio-ingredients (metabolites),

either as A) fermentation by-products or B) as the result of yeast autolysis:

A1) Polysaccharide Prebiotics (stimulate the growth of endogenous probiotics such as Bifidobacterium sp. and Lactobacillus sp.):

fructooligosaccharides (FOS), oligofructose & inulin), galactooligosaccharides (GOS), transgalacto-oligosaccharides (TOS), mannanoligosaccharides (MOS) and lactulose) which

A2) Oligopeptides with antibacterial function:

"mycocin" - secreted yeast toxin effective against E.coli; the one originating from K.marxianus was shown to have stronger antibacterial effect than S.cerevisiae.

lactoferrin - partially hydrolysed milk/whey protein – (angiotensin-I-converting enzyme (ACE) inhibitory activity); antibacterial effects against E.coli and against Clostridum sp.

A3) Short chain fatty acids (SCFA), in particular lactic and acetic acid - produced by K.marxianus much more efficiently than many of the commercially available products based on Saccharomyces cervisae (Fig.2)

Fig2: Comparative SCFA production between K.marxianus B0399 and Bakery Yeast

SCFA production	Kluyveromyces m. B0399	Saccharomyces cerevisiae
Lactic acid (mg/l)	2152	214
Lactic + Acetic ac. (mg/l)	3483	330

B) Immunomodulators and immunostimulators (activate immune cells such as macrophages, neutrophils, and other immunocompetent cells): yeast cell wall polysaccharides – glucans and mannan

K.marxianus has the capacity of production and secretion of B-glucanase enzyme which cause autolysis of the cell wall and liberation of mannan & B-glucans. The proportion of soluble branched b-1,6–glucan is much higher in Kluyveromyces marxianus than in Saccharomyces cerevisiae;

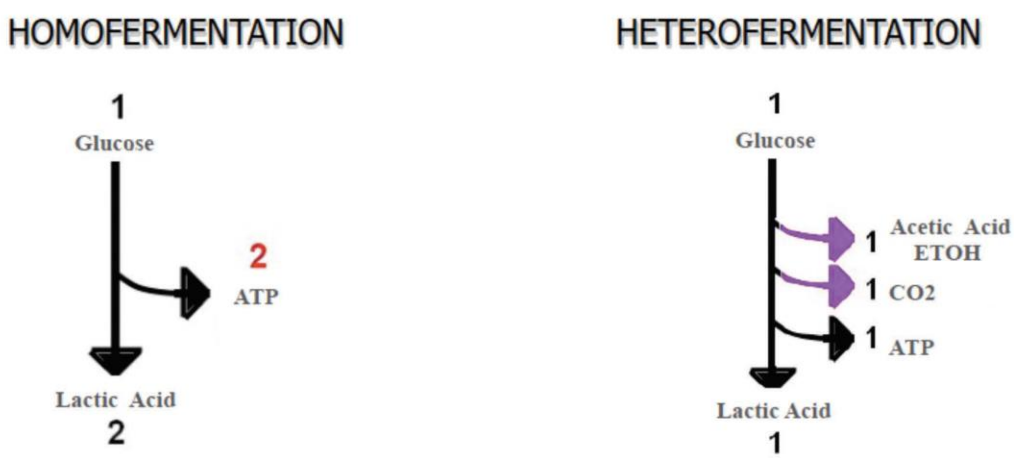
4. Modulation of the host's immune system

Kluyveromyces marxianus was shown to have different **immune-modulatory properties** from Sacharomyces in terms of adaptive immune responses promoting health benefits in conditions characterized by excessive inflammation.

5. K.marxianus increases energy production and energy absorption

It reduces Carbon Footprint as homo-fermenting yeast (particularly in anaerobic conditions): produce 2 ATP for every molecule of glucose which leads to a decrease in CO2 and methane production (Fig3.):

FIG.3: Simplified mechanism of homo- and hetero-fermentation pathway



Since 1994 this yeast has been classified as "Generally Regarded As Safe" (GRAS) by the Food and Drugs Administration (FDA) and since 2005 it has been listed as a microorganism with "Qualified presumption of safety" (QPS) by the European Food Safety Authority (EFSA).

COMPARISON OF THE PROPERTIES AND INDICATIONS of
Kluyveromyces marxianus B0399 and *Sacharomyces sp.*

ROPERTIES AND INDICATIONS	KLUYVEROMYCES B0399	SACCHAROMYCES SP.
Beneficial effects for ANIMAL CATEGORIES:	MONOGASTRIC POLIGASTRIC	POLIGASTRIC
Ability to digest LACTOSE (βgalactosidase activity)	Yes +++	NO
SHFA production (lactic and **A**cetic acid in particular) and positively reduce the intestinal pH	HIGHER	LOWER
Ability to grow on whey and other milk-by products	Yes +++	NO
ANTIBACTERIAL effect	STRONGER (bette**r** mycocin induced antibacterial effect in vitro and in vivo; produce lactoferrin)	WEAKER
Production of valuable METABOLITES: prebiotics, antibacterials, immunomodulators and immunostimulators	HIGHER	WEAKER
Increase the number of good bacteria of th**e** gut natural flora (bifidobacteria)	Demonstrated	NO
CO2 production as a fermentation by product in lower gut	NO CO2 production as a result of homolactic fermentation	2 CO2 molecule from 1 sugar molecule as a result of heterolactic fermentation
ENERGY production	HIGHER (2 ATP molecules from 1 sugar molecule)	LOWER (1 ATP molecule from 1 sugar molecule)
Dosage - Therapeutic minimum and its effect upon gut flora	LOW ~ “natural” 10 million CFU / day	HIGH **–** 10 billion CFU / day drastic impact on the natural microflora