

## STUDY OF SPECIFIC GROWTH RATE AND GENERATION TIME OF TWO *LACTOBACILLUS SALIVARIUS* STRAINS ISOLATED FROM DENTAL ROOT CANAL AND SOME PROBIOTIC STRAINS AT pH 8,0

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### Abstract

In this study we investigated the specific growth rate ( $\mu$ ) and the generation time ( $\Delta t$ ) of two *Lactobacillus salivarius* strains isolated from dental root canal and two probiotic *Lactobacillus* strains by intestinal origin. Differences between values of both parameters were observed, depends to the strains and pH values. All *Lactobacillus* investigated strains presented higher values of specific growth rate ( $\mu$ ) and smaller of generation time ( $\Delta t$ ) at pH 7.0 than pH 8.0.

*Lactobacillus salivarius* strains G1 and G2 isolated from dental root canal had bigger values of specific growth rate ( $\mu$ ) at both pH values than probiotic *Lactobacillus* strains.

Between the specific growth rate ( $\mu$ ) and the generation time ( $\Delta t$ ) there is a close negative correlation. Pearson coefficient was -0,95.

**Keywords:** *Lactobacillus salivarius*, generation time, specific growth rate.

### INTRODUCTION

The lactobacilli are considered the most acidoresistant lactic acid bacteria. They grow best in slightly acidic conditions with an initial pH of 6.5...5.4 and even under 5.0. *Lactobacillus subebicus* grows at pH 2.8 and a similar pH tolerance is also found in *Lactobacillus plantarum*, *Lactobacillus casei* (4) and *Lactobacillus salivarius* strains G1 and G2 isolated from dental root canal (1). In this work we investigated the specific growth rate ( $h^{-1}$ ) and the generation time (h) of these strains with dental origin at pH 8.0.

### MATERIALS AND METHODS

In this study we examined two *Lactobacillus salivarius* strains isolated from dental root canal (G1 and G2) and two probiotic *Lactobacillus* strains by intestinal origin (*Lactobacillus salivarius* probiotic and *Lactobacillus rhamnosus* GG). All strains were grown in MRS medium and were incubated at 37°C for 24h, in 5% CO<sub>2</sub> atmosphere at pH 8.0 and 7.0. The DO<sub>600</sub> values were determined in the moment of inoculation (T0) and than hourly (moment T1 after one hour, T2 after two

hours, T3 after three hours etc). All determinations have been repeated three time (experiment 1,2,3).

The DO<sub>600</sub> values were plotted on logarithmic graphic and the curves growth were obtained. The specific growth rate ( $\mu$ ) and generation time ( $\Delta t$ ) were calculated.

The specific growth rate was calculated using the formula:

$$\mu = \frac{\ln OD_{max} - \ln OD_{min}}{T_{max} - T_{min}}$$

Where:

OD max is the value of DO<sub>600</sub> in moment T<sub>max</sub>;

OD min is value DO<sub>600</sub> in moment T<sub>min</sub>;

The generation time ( $\Delta t$ ) (doubling time) was calculated in this main:

$$\Delta t = \frac{\ln 2}{\mu}$$

## RESULTS AND DISCUSSIONS

The specific growth rate and generation time of investigated strains at pH 8.0 are shown in table 1.

*Lactobacillus salivarius* strains isolated from dental root canal G1 and G2 showed higher average values of specific growth rate ( $0.61 \text{ h}^{-1}$  and  $0.59 \text{ h}^{-1}$ ) compared with *Lactobacillus* probiotic strains ( $0.15 \text{ h}^{-1}$  for *L.salivarius* probiotic and  $0.26 \text{ h}^{-1}$  for *L. rhamnosus* GG) at pH 8.0.

While, the *Lactobacillus* strains by dental origin, had smaller generation time average values, at pH 8.0 (1.15h for G1 and 1.22h for G2) compared with the probiotic strains (4.74h

for *L.salivarius* probiotic and 2.69h for *L. rhamnosus* GG) (table 1).

At pH 7.0, the values of specific growth rate and generation time of the investigated strains are shown in table 2.

Also, at pH 7.0 *Lactobacillus salivarius* strains isolated from dental root canal G1 and G2 showed higher average values of specific growth rate (1.15 and 1.05) compared with *Lactobacillus* probiotic strains (0.85 for *L. salivarius* probiotic and 0.73 for LGG). The *Lactobacillus* strains by dental origin had smaller average values of generation time (0.59h for G1 and 0.66h for G2) compared with the probiotic strains (0.81h for *L.salivarius* probiotic and 1.13h for *L. rhamnosus* GG) (table2).

Table1. The specific growth rate  $\mu$  ( $\text{h}^{-1}$ ) and the generation time  $\Delta t$  (h) at pH 8.0

pH 8,0	Strain							
	<i>L.salivarius</i> probiotic		G1		G2		<i>L. rhamnosus</i> GG	
	$\mu$	$\Delta t$	M	$\Delta t$	M	$\Delta t$	$\mu$	$\Delta t$
Experiment 1	0,16	4,31	0,51	1,35	0,47	1,46	0,21	3,30
Experiment 2	0,1	6,30	0,66	1,04	0,52	1,32	0,31	2,22
Experiment 3	0,19	3,63	0,65	1,06	0,77	0,89	0,27	2,55
Average values	0,15	<b>4,74</b>	0,61	<b>1,15</b>	0,59	<b>1,22</b>	0,26	<b>2,69</b>

Table 2. The specific growth rate  $\mu$  ( $\text{h}^{-1}$ ) and the generation time  $\Delta t$  (h) at pH 7.0

pH 7,0	Strain							
	<i>L.salivarius</i> probiotic		G1		G2		<i>L. rhamnosus</i> GG	
	$\mu$	$\Delta t$	$\mu$	$\Delta t$	M	$\Delta t$	$\mu$	$\Delta t$
Experiment 1	0,77	0,89	1,26	0,54	0,92	0,75	0,69	1,25
Experiment 2	0,81	0,85	1,16	0,59	1,07	0,64	0,88	1,04
Experiment 3	0,96	0,71	1,03	0,66	1,15	0,6	0,62	1,11
Average values	0,85	<b>0,81</b>	1,15	<b>0,59</b>	1,05	<b>0,66</b>	0,73	<b>1,13</b>

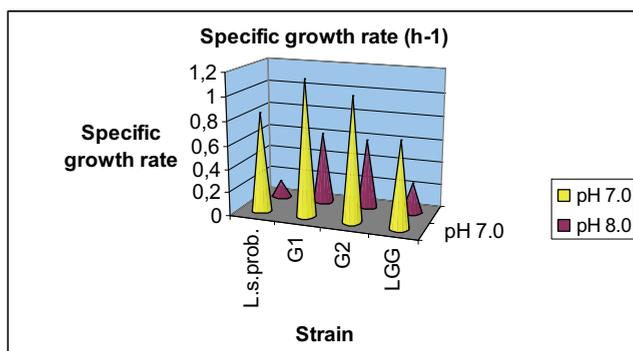


Fig. 1. The specific growth rate  $\mu$  of *Lactobacillus* strains at pH 8.0 and 7.0

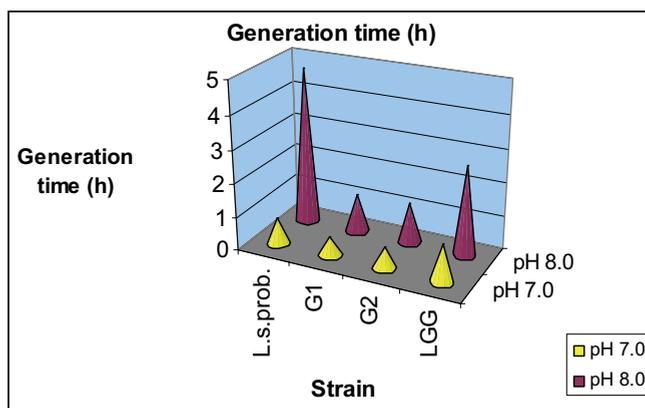


Fig. 2. The generation time  $\Delta t$  (h) of *Lactobacillus* strains at pH 7.0 and 8.0

The values of specific growth rate of *Lactobacillus* strains with dental origin were higher than those of probiotic strains at both pH values. The values of generation time were smaller at *Lactobacillus salivarius* strains isolated from dental root canal than those of probiotic strains at both pH values.

All *Lactobacillus* strains showed smaller values of generation time at pH 7.0 (ranged between 0.59h and 1.13h and the average time was 0,798h) compared with those at pH 8.0 (ranged between 1.15h and 4.74h and the average time was 2.45h).

Table 3

The Pearson coefficient at pH 7.0		
	$\mu(pH=7,0)$	$\Delta t$
$\mu(pH=7,0)$	1	
$\Delta t$	-0,9481	1

At pH 7.0 between specific growth rate and the generation time there is a very strong negative correlation. Pearson Factor  $r=-0.95$ .

The dispersion diagram has a descending tendency. 90% of variation of specific growth rate depends by linear expressed by regression line (decreasing the coefficient of determination is  $R^2= 0,90$ ). Residual variation in specific growth rate is 10%. (Fig. 3).

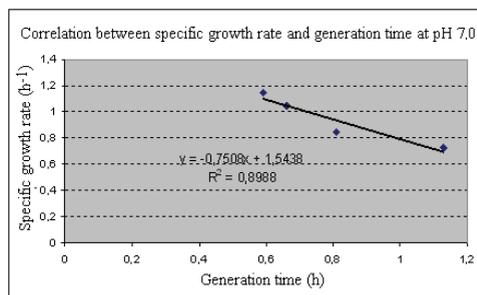


Fig. 3. The dispersion diagram at pH 7.0

Table 4. The Pearson coefficient at pH 8.0

	$M(pH=8,0)$	$\Delta t$
$\mu (pH=8,0)$	1	
$\Delta t$	-0,9473	1

At pH 8.0 between specific growth rate and the generation time there is a very strong negative correlation. Pearson Factor  $r=-0.95$ .

The dispersion diagram has a descending tendency. 90% of variation of specific growth rate depends by linear expressed by regression line (decreasing the coefficient of determination is  $R^2= 0,90$ ). Residual variation in specific growth rate is 10%. (Fig. 4).

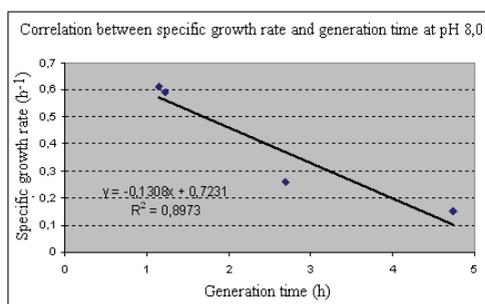


Fig. 4. The dispersion diagram at pH 8.0

Between the specific growth rate and the generation time is a high negative correlation. The correlation Pearson factor  $r= -0,95$ . These data are correlated with those of Nezhad H.M, 2010.

## CONCLUSIONS

After this study we observed that higher values of specific growth rate at 7.0 pH were registered for all *Lactobacillus* investigated strains compared with those at 8.0 pH. The strains with dental origin showed bigger specific growth rate values at both pH values compared with the probiotic *Lactobacillus* strains. The values of generation time for all *Lactobacillus* strains were smaller at pH 7.0 (the average time was 0.798h) than those at pH 8.0 (the average time was 2.45h). A negative strong correlation between specific growth rate and generation time at both pH values was observed. Pearson factor was  $r=-0.95$ .

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