

## An improved method to assess the suitability of lactic acid bacteria as dried starter culture for cereal fermentation

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A majority of traditional cereal based foods consumed in Africa are processed by natural fermentation of maize, sorghum and/or millet and are particularly important as weaning foods for infants and dietary staples for adults (Owusu-Kwarteng et al., 2012). The use of dried lactic acid bacteria (LAB) starter culture to raw materials could be a breakthrough in the processing of fermented cereal based foods, resulting in a high degree of control over the fermentation process and standardization of the end product. LAB are exposed to various environmental stress conditions, such as temperature fluctuations, acid, pH, high osmotic pressure and absence of available nutrients during cereal fermentation as well as their production in the dried state. The ability of LAB to establish them-selves and to dominate the LAB population during cereal fermentation is another important characteristic of a starter culture. The dominance of the starter culture would be exerted by its fast and predominant growth under fermentation conditions and could be characterized by the generation time [ $g$  (h)] (Delignette-Muller, 1998).  $g$  is directly linked to the maximum specific growth rate [ $\mu_m$  ( $h^{-1}$ )], which is the slope of the logarithm of the growth curve in the exponential growth phase. The evaluation of cell viability under different stress conditions and the determination of kinetics growth parameters ( $\mu_m, g$ ) could be a valuable tool to evaluate the potential of lactic acid bacteria to be produce in the dried state.

$\mu_m$  and  $g$  or viability after stress treatments were evaluated for *Lactobacillus plantarum* LP, *Lactobacillus fermentum* LF, *Leuconostoc* spp M1 and *Lactococcus* spp M2, isolated during production of fermented cereal based products from Côte d'Ivoire. Viable count and pH were determinate during cell cultures incubated at 30° C for 22 h.  $\mu_m$  and  $g$  were obtained by directly fitting the logarithmic transformation of plate count versus time:

$$g = \frac{0.693}{\mu_m} \quad (1) \quad \mu_m = \frac{\ln N_t - \ln N_0}{t_1 - t_0} \quad (2) \quad \text{Where } g \text{ (h) is the generation time, } N_0 \text{ the initial population at time } t_0 \text{ (cfu/g DW), } N_t \text{ is the population at time } t \text{ (cfu/g DW), } \mu_m \text{ (h}^{-1}\text{) is the maximal specific growth rate.}$$

Dehydration with glycerol solution of increasing concentration, freezing at -80°C followed by thawing at 50°C and heating in a water bath at 54°C or 75°C were stress treatments applied. The percentage survival of the strains before and after stress treatments was expressed as follows: Survival (%) =  $N_0/N_c \times 100$  where  $N_0$  is the CFU/g DW before stress treatment and  $N_c$  is the CFU/g DW after stress treatment. The water activity ( $a_w$ ) was estimated after each addition of glycerol with Norrish equation:  $a_w = (1-X) e^{-KX^2}$  (3) where  $a_w$  is water activity,  $X$  is the molar fraction glycerol, and  $K = 1.16$  is the correlating constant for glycerol at 25°C,

In general, we observed an increase in cell concentration and a decrease in pH in the culture media until the 16<sup>th</sup> hour. After the 18<sup>th</sup> h of culture, cell concentrations or pH values were, respectively  $\geq 2.71 \times 10^8$  cfu/g DW or between 4.1 and 4.4.  $\mu_m$  or  $g$  ranged between 0.51  $h^{-1}$  and 0.14  $h^{-1}$  or 0.61 h and 1.33 h, respectively. We observed that the strains were sensitive to a change in water activity ( $a_w$  0.32 final), storage temperature (freezing stress) and incubation temperature (heat stress) with survival rates ranging between 12.57% and 20.95 %, 1.20% and 31.03% and 0.04% and 1.71%, respectively. The Strain LF obtained a higher cell concentration (after 18h of culture) or viability (dehydration with glycerol solution and freezing stress) and a lower generation time ( $g$ ) compared to those obtained by the strains, LP, M1 and M2. These results indicate that the kinetic growth parameters ( $\mu_m, g$ ) and viability after stress treatments can be used to predict the potential of Lactic acid bacteria as dried starter culture. Further attempts to select strains as suitable dried starter culture for cereal fermentation must be made

### References

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