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Original Article

Impact of inulin on viability and storage stability of probiotic *Lactobacillus plantarum* TISTR 2075 in fermented rice extract

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ABSTRACT

The influence was determined of various concentrations of inulin as a prebiotic on the growth of probiotic *Lactobacillus plantarum* TISTR 2075 fermented in Plai Ngahm Prachin Buri rice extract. The supplementation of 2% inulin provided the highest viable cell number of 8.90 log colony forming units/mL after fermentation at 37 °C for 24 h. The storage stability of the probiotic strain could be considered in terms of the specific rate of cell death (*k* value). The supplementation of 2% inulin exhibited the lowest *k* value of 2.48×10^{-2}/d (30.16% survival) and 8.03×10^{-2}/d (7.84% survival) after storage at 4 °C for 52 d and 30 °C for 31 d, respectively. The total reducing sugar and free amino nitrogen profiles of all treatments decreased over the storage period.

Introduction

Probiotic foods represent one of the largest sectors in functional food markets (Buruleanu et al., 2013). The growing awareness regarding the improvement of gut health and efficiency of probiotic bacteria are responsible for the development of the global probiotics market which is expected to grow from USD 62.6 billion in 2014 to USD 96 billion in 2020 (Sudip, 2016). The demand for alternatives to dairy products is growing due to problems with lactose intolerance, cholesterol content, allergenic milk proteins and the desire for vegetarian alternatives (Santos et al., 2014). The application of probiotic cultures in non-dairy products represents a great challenge and needs to be researched at the industrial level for commercial productions (Rivera-Espinoza and Gallardo-Navarro, 2010). Beverage supplemented with probiotics could potentially be produced from a multitude of raw materials including fruits, vegetables and cereals, which are considered vehicles for delivering probiotic bacteria to the human gastrointestinal tract (Sharma and Mishra, 2013). In particular, cereals are one of the most suitable substrates for the development of foods containing probiotic products (Lamsal and Faubion, 2009). Cereal grains are rich sources of protein, carbohydrates, vitamins mineral and fiber (Granato et al., 2010). They also consist of non-digestible carbohydrates that have beneficial effects for human health such as stimulating the growth of Lactobacilli and Bifidobacteria present in the colon (Kandylis et al., 2016). Several studies have proved that probiotic microorganisms could grow on cereal-base substrates. For example, Wu et al. (2015) reported that mung bean milk could stimulate the growth of *L. plantarum* B1-6 with the highest viable cell count of 8.88 log colony forming units (CFU)/mL. Furthermore, non-dairy probiotic drinks developed from sprouted wheat, barley, pearl millet and green gram were reported to have viable cell numbers of *L. acidophilus* NCDC14 in the range 10.36–11.51 log CFU/mL and 10.36–11.17 log CFU/mL in samples based on pearl millet and barley, respectively (Mridula and Sharma, 2015). This was in accordance with the results of Salmerón et al. (2014) that *L. acidophilus* NCIMB 8821 and *L. plantarum* NCIMB 8826 cultured in a malt-and-barley-based beverage exhibited good growth of higher than 8 log CFU/mL. Santos et al. (2014) developed a peanut-soymilk beverage as a novel substrate for probiotic *L. acidophilus* (LACA 4) and *Pediococcus acidilactici* (UFLA BFFCX 27.1) providing a cell population above 8 log CFU/mL.

Inulin is a functional food additive due to its prebiotic properties which are beneficial to humans (Mensink et al., 2015). The

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supplement of prebiotic inulin could enhance the viability of probiotics (da Silva Sabo et al., 2015). *L. acidophilus* FTDC 8033 and *L. casei* ATCC 393 grow well in soymilk supplemented with inulin (Yeo and Liong, 2010). Furthermore, da Silva Sabo et al. (2015) also suggested that the presence of 1% inulin in de Man-Rogosa-Sharpe (MRS) medium increased the maximum specific growth rate of *L. plantarum* ST16 Pa. Additionally, the number of probiotic *L. plantarum* 14 was affected by the addition of inulin in soft cheese (Modzelewska-Kapitula et al., 2007).

Therefore, the aim of this study was to determine the applicability of using different concentrations of prebiotic inulin for the growth of probiotic *L. plantarum* TISTR 2075. The stability of the strain in fermented Plai Ngahm Prachin Buri rice extracts supplemented with inulin during storage was evaluated. Additionally, determination of the lactic acid content, free amino nitrogen (FAN) and the total reducing sugar (TRS) of fermented rice extracts was also carried out.

Materials and methods

Microorganism

The probiotic *Lactobacillus plantarum* TISTR 2075 isolated from fermented vegetables was obtained from the Microbiological Resource Center, Thailand Institute of Scientific and Technological Research (TISTR), Thailand. The probiotic strain was preserved in de Man-Rogosa-Sharpe (MRS) broth (Difco; Detroit, MI, USA) with 20% (volume per volume; v/v) glycerol content at −20 °C. The strain was subcultured in MRS broth and was incubated at 37 °C for 24 h under microaerobic-static conditions and then used as inoculum.

Preparation of Plai Ngahm Prachin Buri rice extract fermentation and storage

Fermented Plai Ngahm Prachin Buri rice extract was prepared as described by Wirunpan et al. (2016) with minor modifications. Plai Ngahm Prachin Buri rice was washed, soaked in distilled water (rice:water = 1:10 wt per volume; w/v) for 6 h and comminuted in a blender for 5 min. The resultant slurry was twice filtered through double-layered cheesecloth to yield cereal extracts. The rice extract was dispensed into containers, added with different concentrations (1%, 2% and 3% w/v) of inulin (Orafti®GR, Beneo; Mannheim, Germany) and sterilized by heating at 121 °C for 15 min. Sterilized rice extracts supplemented with various concentrations of inulin were inoculated with an overnight culture of 1% (v/v) *L. plantarum* TISTR 2075. The fermentations were performed under no pH control in Duran screwcapped glass bottles at 37 °C for 24 h (Savedboworn and Wanchaitanawong, 2015). Viable cell counts were determined using the standard plate count method with MRS medium supplemented with 0.5% $CaCO_3$ at 37 °C for 24 h. The pH was measured using a pH meter.

During the storage period, fermented Plai Ngahm Prachin Buri rice extracts containing probiotic *L. plantarum* TISTR 2075 were stored at 4 °C for 52 d and 30 °C for 31 d. The specific rate of cell death per day (k) was calculated as a first-order reaction (Equation (1)):

$$k = \ln(N_0/N)/t \quad (1)$$

where N refers to the bacterial count at a particular storage period (CFU/mL), N_0 represents the bacterial count at the beginning of the storage (CFU/mL) and t is the storage time.

The viable cell counts, pH, lactic acid content, reducing sugar content and free amino nitrogen content of the fermented Plai Ngahm Prachin Buri rice extracts were determined.

Enumeration of viable cell number

Viable cell counts were determined using the standard plate count method on MRS agar plate supplemented with 0.5% (w/v) $CaCO_3$. Briefly, 1 mL of each sample was vortexed aseptically with 9 mL of sterile physiological saline (0.85% NaCl) to make a series of dilutions. The plates were incubated at 37 °C for 24 h. The viable cell counts were expressed as $\log_{10}$ values per milliliter. The specific growth rate (μ) was calculated using Equation (2):

$$\mu = \frac{\ln(x_t) - \ln(x_0)}{t - t_0} \quad (2)$$

where x_0 and x_t are viable cell numbers measured within the exponential phase of growth at times t_0 and t, respectively.

The percentage of cell survival was defined as shown in Equation (3):

$$\text{survival rate}(\%) = \left(\frac{N}{N_0}\right) \times 100 \quad (3)$$

where N represents the number of viable cells count (CFU/mL) at a particular period and N_0 denotes the initial viable cell count (CFU/mL) at the beginning.

Titratable acidity analysis

Titratable acidity expressed as the percentage of lactic acid was determined using a titration method according to Association of Official Analytical Chemists (1990). The supernatant of culture (2 mL) mixed with distilled water (18 mL) was titrated with 0.1 M NaOH. Phenolphthalein (1 mL) was used as an indicator. Each milliliter of 1 N NaOH is equivalent to 90.08 mg of lactic acid.

Reducing sugar analysis

Reducing sugars were determined using the 3, 5-dinitrosalicylic acid colorimetric method (Miller, 1959), with glucose as the standard.

Free amino nitrogen analysis

The concentration of free amino nitrogen (FAN) was estimated using the ninhydrin colorimetric method (Convention, 1987) with glycine solution as the control.

Statistical analysis

All experiments were carried out in duplicate with duplication determination. The data were statistically analyzed for analysis of variance in a completely randomized design. Significant divergences among mean values were established using Duncan's multiple range tests at the 95% confidence interval. All statistical analyses were performed using the SPSS software, version 22 (SPSS Inc.; Armonk, NY, USA).

Results and discussion

Fermentation of Plai Ngahm Prachin Buri rice extract in the presence of different concentrations of inulin

Plai Ngahm Prachin Buri rice extract was used as culture media for the growth of probiotic *L. plantarum* TISTR 2075 in the presence of various concentrations of inulin. Based on the results, *L. plantarum* TISTR 2075 grew well in Plai Ngahm Prachin Buri rice

extract and reached a maximum viable cell number of 8.42 log CFU/mL after 24 h fermentation. A slight improvement in the viable cell numbers of 0.50 log CFU/mL, 0.52 log CFU/mL and 0.43 log CFU/mL was observed with the addition of 1%, 2% and 3% inulin, respectively. As shown in Fig. 1, all treatments generally followed a similar growth pattern. The specific growth rate of the strain was significantly affected by different concentrations of inulin. The results indicated that the extract supplemented with inulin exhibited a higher specific growth rate compared to no inulin addition. This observation was in close agreement with the report of da Silva Sabo et al. (2015) which indicated that the addition of 1% inulin increased the maximum specific growth rate of *L. plantarum* ST16 Pa from 0.37/hr to 0.49/hr. As a result of the partial hydrolysis of the inulin, it was likely that some fructose would be detected in the culture medium. Consequently, the strain could utilize fructose as a carbon source. This was also supported by the observation of Takagi et al. (2014) that *L. plantarum* 22A-3 could be able to utilize fructose; however this strain could not degrade inulin. Nevertheless, Nazzaro et al. (2012) suggested that the presence of inulin did not markedly affect the growth of *L. plantarum* subsp. *plantarum* DSM 20174, because the strain does not have the capability to degrade inulin. The acidification rates influenced the availability of simple sugars such as fructose resulting in the increased level of lactic acid bacteria. At pH 4.0, hydrolysis reaction started to occur at the fructo-oligosaccharide chains and the stability of the chains was lowered by protonic activation of the leaving group resulting in the release of fructose or glucose (Ng et al., 2014). This was supported by Matusek et al. (2009) who noted that the degradation rate of the oligosaccharides would be increased due to the decrease in the pH.

The addition of 2% inulin showed the highest specific growth rate of 0.157/hr. However, the addition of 3% inulin was not significantly different compared with 1% inulin. This may be due to the fact that a decrease in water activity in the system promoted by large amounts of a water-binding substance resulted in an increase in the lag phase and a decrease in the specific growth rate (Shirai et al., 2001; Timbuntam et al., 2006). Moreover, high acid production of the strain at 3% inulin may have adversely affected probiotic growth and viability.

The pH value and titratable acidity content evolved in different profiles depending on the various concentrations of inulin addition. As shown in Fig. 2, the lactic acid content was significantly higher in the presence of 2% and 3% inulin after 24 h. The highest acid production of 0.16% was achieved at 3% inulin addition followed by the

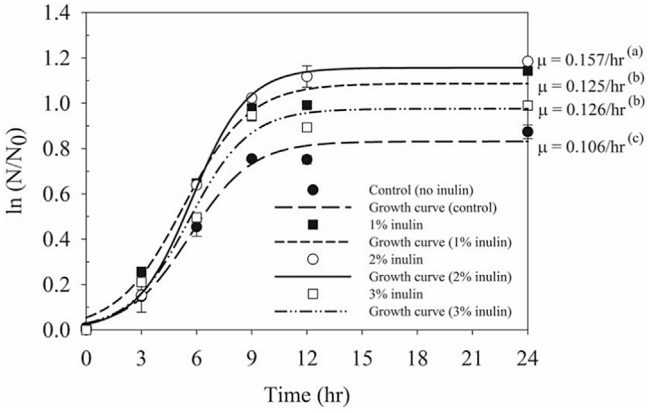

Fig. 1. Cell growth of *L. plantarum* TISTR 2075 in fermented Plai Ngahm Prachin Buri rice extract supplemented with various contents of inulin during fermentation at 37 °C for 24 h (y axis details provided in Equation (1) description; values with different superscripted, lowercase letters (a–c) are significantly different ($p < 0.05$) using Duncan's multiple range test; error bars indicate ± SD).

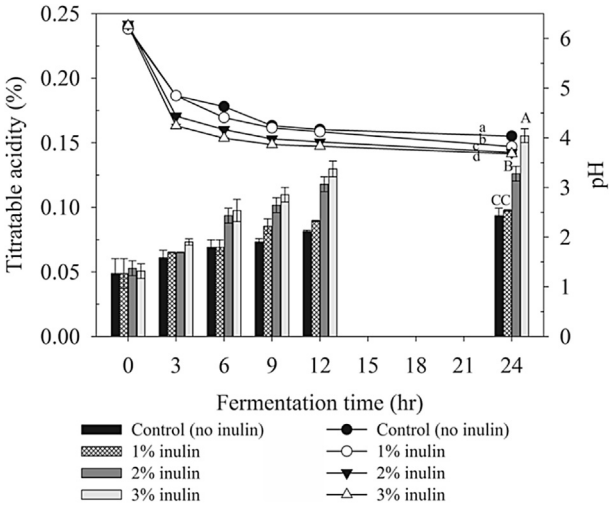

Fig. 2. Changes in pH (line and scatter plot) and titratable acidity (bar chart) of fermented Plai Ngahm Prachin Buri rice extract supplemented with and without inulin at 37 °C for 24 h (values with different capital letters (A–C) of titratable acidity and lowercase letters (a–d) of pH at 24-h fermentation are significantly different ($p < 0.05$) using Duncan's multiple range test; error bars indicate ± SD.).

supplementation of 2% inulin (0.13% lactic acid content). No significant difference in the lactic acid content was detected when no inulin and 1% inulin were added. A significant decrease in the pH value of 3.69–3.83 was achieved in the addition of 1–3% inulin which was lower than that of control (pH = 4.04). This was in agreement with the report of Yeo and Liong (2010) that supplementation of soymilk with inulin significantly increased the production of lactic acid in the soymilk. Additionally, da Silva Sabo et al. (2015) suggested that the addition of 1% inulin in cultures of *L. plantarum* ST16 Pa resulted in a greater amount of lactic acid than with the absence of inulin, probably because fructose monomers released from inulin were assimilated by the Embden-Meyerhof-Parnas pathway, leading to the production of a higher concentration of lactic acid.

The differences in the consumption of reducing sugar and free amino nitrogen (FAN) in fermented Plai Ngahm Prachin Buri rice extract supplemented with various inulin concentrations are shown in Fig. 3. Prior to fermentation, the total reducing sugar in the rice extract without inulin addition was 0.22 g/L. The addition of inulin increased the total reducing sugar profiles in the rice extract. Total reducing sugars of 0.99 g/L, 1.87 g/L and 2.36 g/L were obtained in rice extract supplemented with 1%, 2% and 3% inulin, respectively, perhaps because heating in the preparation process increased the reducing sugar concentration in the samples. Kim et al. (2001) suggested the increase in temperature and the concentration of inulin resulted in higher amounts of reducing sugar. The change in reducing sugar resulted from the hydrolysis of inulin during heating. Additionally, a rice grain contains a variety of carbon sources including glucose, sucrose, maltose and maltotriose (Singh and Juliano, 1977) which might alter the carbohydrate metabolism of the probiotic and subsequently the production of end products. During 24 h, the fermented rice extracts supplemented with various contents of inulin had significantly different amounts in the consumption of reducing sugar. The available sugars were consumed during the exponential phase (at 3–9 h) and this resulted in a significantly different sugar consumption. A decrease in the total reducing sugar concentration could be attributed to the consumption of fermentable sugars as a carbon source during the exponential phase of growth (Rathore et al., 2012). Although fermented rice extract with 3% inulin exhibited

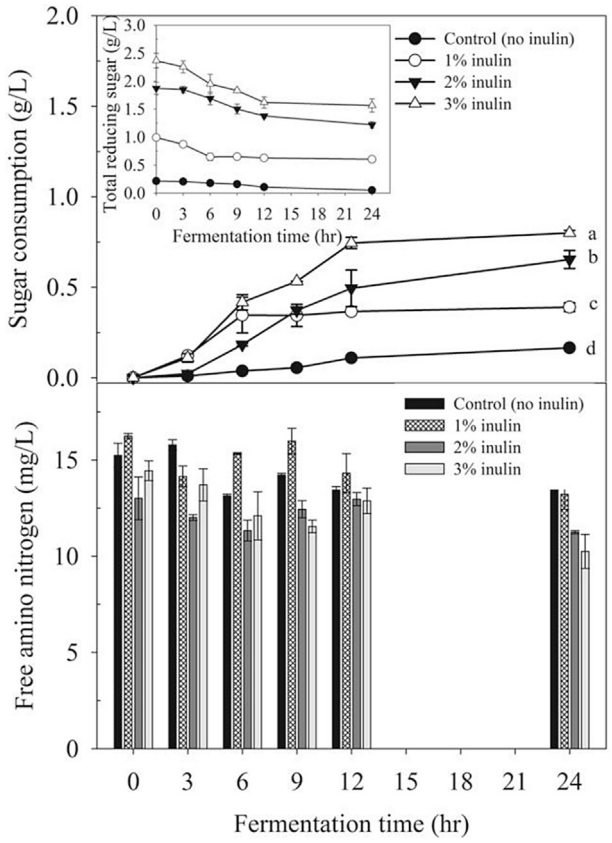

Fig. 3. Variations of sugar consumption and free amino nitrogen (FAN) evolution during the fermentation of Plai Ngahm Prachin Buri rice extract supplemented with various concentrations of inulin at 37 °C for 24 h (curves with different lowercase letters (a–d) are significantly different ($p < 0.05$) using Duncan's multiple range test; error bars indicate ± SD.).

the highest sugar consumption during 3–9 h fermentation, the specific growth rate at this concentration was significantly lower than for 2% inulin probably because the probiotic cells not only consumed fermentable sugars as a carbon source for growth but also produced organic acids. The production of organic acids via the metabolic pathways progressively decreased the pH of the medium from 6.27 to 3.86 until 9 h of fermentation, in the fermented rice extract supplemented with 3% inulin. In a low pH environment, cells may utilize metabolizable sugar to provide adenosine triphosphate (ATP) to F_0F_1-ATPase via glycolysis, enabling proton exclusion and thereby maintaining their viability (Hutkins and Nannen, 1993).

Lactic acid bacteria are generally fastidious organisms requiring complex nutrients for cell growth, such as amino acids. The nitrogen uptake in the fermentation was monitored by measuring free amino nitrogen. The initial FAN in all treatments was 13.01–16.24 mg/L. The reduction of FAN in fermented rice extract was found to decrease until the end of 24 h fermentation. This result was in accord with the report of Salmerón et al. (2014) that the FAN of fermented *L. plantarum* NCIMB 8826 in oat and barley extract decreased through 24 h. Furthermore, the reduction of free amino nitrogen during 24 h was also observed in fermented *L. plantarum* TISTR 2075 in white kidney bean, red kidney bean and black glutinous rice extracts (Savedboworn et al., 2014). In the current study, fermented rice extract supplemented with 3% inulin exhibited the highest reduction in FAN of 4.19 mg/L followed by 1% inulin (3.03 mg/L reduction), 2% inulin (1.76 mg/L reduction) and the control without inulin addition (1.40 mg/L reduction). The results revealed that the consumption of FAN was not related to the specific growth rate. This result was consistent with the observation by Champagne et al. (2009) that the growth of lactic cultures in soy beverages was independent of the free amino compound concentration. It was possible that the amino acid composition and not its concentration may have been responsible for growth stimulation of the strain.

Storage stability of L. plantarum TISTR 2075 in fermented Plai Ngahm Prachin Buri rice extract supplemented with different concentrations of inulin

The shelf-life of probiotic *L. plantarum* TISTR 2075 fermented in Plai Ngahm Prachin Buri rice extract supplemented with different concentrations of inulin was determined during storage at 4 °C and 30 °C. After storage at 4 °C, the number of probiotic *L. plantarum* TISTR 2075 was still high (more than 7.7 log CFU/mL) then it slightly declined over time until the end of storage at 52 d. It might be possible that the probiotic could effectively use nutrients such as fermentable sugars and amino acids in the medium to sustain its viability. The survival rate of the strain remained in the range 10.64–30.16% (0.52–0.97 log reduction) which was well above the recommended therapeutic minimum of 6 log CFU/mL at the time of consumption (Food and Agriculture Organization of the United Nations and World Health Organization, 2001). There was a significant difference in the survival rate among all treatments during storage at 4 °C. The addition of 2% inulin resulted in the highest survival rate of 30.16% followed by 3% inulin (28.73% survival rate; 0.54 log reduction) and 1% inulin (12.15% survival rate; 0.92 log reduction), respectively, whereas, a survival rate of 10.64% with 0.97 log reduction was observed in fermented Plai Ngahm Prachin Buri rice extract without inulin addition. Fig. 4 suggests that the storage temperature was a crucial parameter affecting the survival of the probiotic strain. A high storage temperature led to a great decrease in the viable cell number of microorganisms. During storage at 30 °C for 31 d, a viability loss of 1.11 log CFU/mL, 1.41 log CFU/mL and 1.48 log CFU/mL was observed in fermented Plai

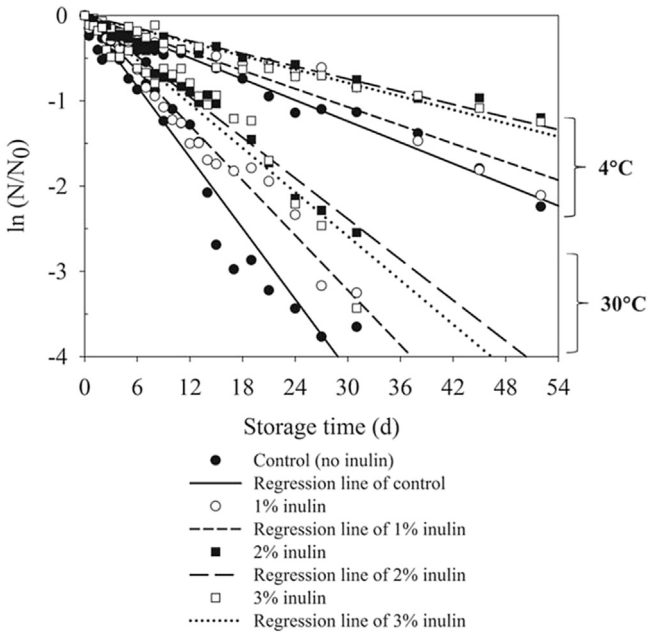

Fig. 4. Storage stability of *L. plantarum* TISTR 2075 expressed as natural logarithmic values of relative survival fraction ln (N/N_0) against storage time at 4 °C for 52 d and at 30 °C for 31 d (y axis details provided in Equation (1) description).

Ngahm Prachin Buri rice extract supplemented with 2%, 1% and 3% inulin, respectively, while the extract without inulin addition exhibited the highest loss of viable cell number of 1.59 log CFU/mL. These results were consistent with the report of Valero-Cases and Frutos (2015) that extrusion microencapsulated *L. plantarum* CECT 220 (ATCC 8014) in the presence of 2% inulin exhibited the highest survival value of 6.66 log CFU/g in refrigerated storage at 4 °C for 30 d.

The influence of various concentrations of inulin on the survival of probiotic *L. plantarum* TISTR 2075 in fermented Plai Ngahm Prachin Buri rice extract could be considered in terms of a specific rate of cell death (*k* value). As shown in Table 1, inulin efficiently improved the storage stability of the strain with lower *k* values compared with the control at both storage temperatures. The addition of 2% inulin was found to be relatively effective with the lowest *k* value of 2.48×10^{-2}/d and 8.03×10^{-2}/d after storage at 4 °C and 30 °C, respectively.

Some sugar consumption and free amino nitrogen were still present until the end of storage. The total reducing sugar profiles in all treatments exposed similar trends. Small amounts of the available sugars were consumed during storage at 4 °C and 30 °C. As shown in Fig. 5, the sugar consumption profiles significantly increased throughout storage period. The sugar consumption in fermented rice extracts kept at refrigerated temperature was lower than that at a non-refrigerated temperature. The sugar consumption of 0.32–0.84 g/L in the presence of inulin was significantly higher than in the absence of inulin (0.07 g/L) during subsequent storage at 30 °C. The same tendency was also observed at a storage temperature of 4 °C with sugar consumption of 0.23–0.72 g/L in inulin supplemented with fermented rice extract probably because the low culture pH resulted in increased proton levels and consequently inhibited the uptake of carbohydrate (Lv et al., 2016).

The FAN consumption of 31.92% was obtained in fermented rice extract without inulin addition after storage at 4 °C for 52 d (Fig. 6). Furthermore, the addition of 1–3% inulin produced a 27.78–32.13% FAN reduction. The same tendency was also achieved at the non-refrigerated temperature of 30 °C for 31 d. A FAN reduction of 24.52–29.26% was observed in fermented rice extract with and without inulin addition. According to Rozada-Sánchez et al. (2008), the concentration of FAN decreased with the exponential phase of growth of *Bifidobacterium infantis* but increased from then on. They attributed this to the lyses of cells which lose their viability and release amino acids to the media.

As illustrated in Fig. 7, fermented *L. plantarum* TISTR 2075 in fermented rice extract supplemented with inulin had a similar acidification performance mainly associated with the release of organic acid. The amount of titratable acidity in fermented rice extract supplemented with inulin ranged from 0.26% to 0.30% at the end of storage at 4 °C for 52 d and pH values of 3.40–3.52 were achieved, whereas titratable acidity in rice extract without inulin addition (0.1%) tended to slightly increase with a final pH value of 3.88. This could indicate that this strain is capable of producing acid even at a refrigerated temperature. At a non-refrigerated temperature of 30 °C, the evolution in titratable acidity ranged from 0.62% to 0.71% with final pH values of 3.0–3.2 being obtained from fermented rice extract supplemented with inulin. However, the pH is one of the most important factors affecting the survival of probiotics. The accumulation of acids during storage may be responsible for the decline in viable cell number (Rozada-Sánchez et al., 2008). The production of lactic acid and organic acids inhibits microbial growth in their undissociated form, dissociated form or indirectly by releasing protons (H^+) in the medium (Rathore et al., 2012). Additionally, Pereira et al. (2011) suggested that the pH could reach 4.0 without having a detrimental effect on the viability of the probiotic bacteria during shelf life.

In summary, 2% inulin as a prebiotic could be considered as the optimum concentration for supporting the growth of probiotic *L. plantarum* TISTR 2075 fermented in Plai Ngahm Prachin Buri rice extract, giving a viable cell number above 8.94 log CFU/mL during fermentation at 37 °C for 24 h. Furthermore, the storage stability of the probiotic strain in fermented Plai Ngahm Prachin Buri rice

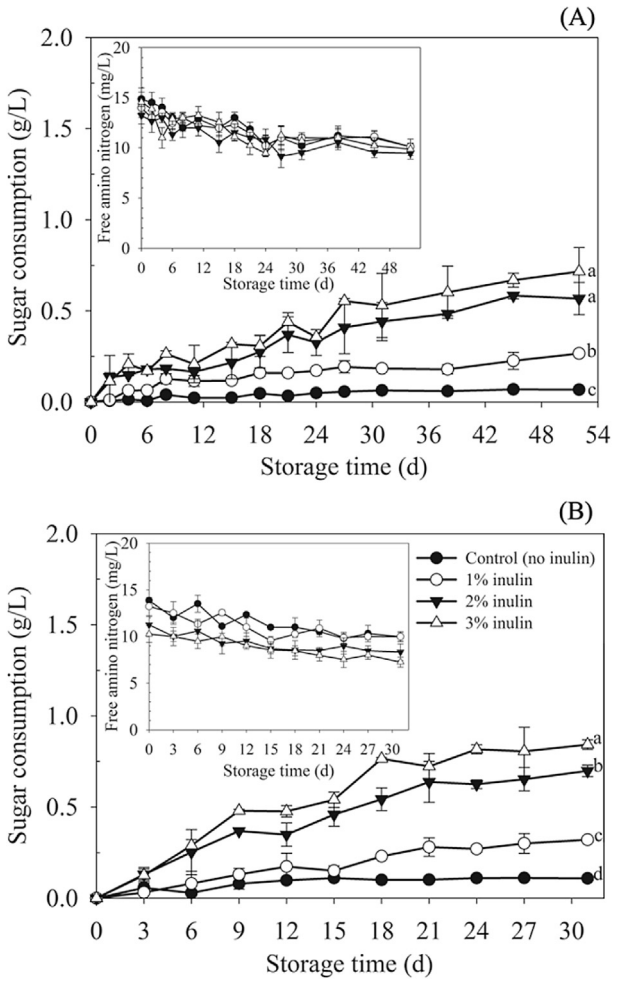

Fig. 5. Sugar consumption and free amino nitrogen concentration of fermented Plai Ngahm Prachin Buri rice extract supplemented with various concentration of inulin during storage at: (A) 4 °C for 52 d; (B) 30 °C for 31 d (values with different lowercase letters of sugar consumption at each storage temperature are significant different ($p < 0.05$) using Duncan's multiple range test; error bars indicate ± SD.).

Table 1
Specific rate of daily cell death (*k*) of *L. plantarum* TISTR 2075 in fermented Plai Ngahm Prachin Buri rice extract supplemented with different concentrations of inulin during storage at 4 °C for 52 d and 30 °C for 31 d.

Inulin content (%)	Specific rate of cell death of *L. plantarum* TISTR 2075 kept at 4 °C		Specific rate of cell death of *L. plantarum* TISTR 2075 kept at 30 °C	
	k_4 (per day)[a]	R^2[b]	k_{30} (per day)[a]	R^2[b]
Control (No inulin addition)	4.13×10^{-2}	0.942	1.40×10^{-1}	0.945
1% Inulin	3.58×10^{-2}	0.923	1.09×10^{-1}	0.978
2% Inulin	2.48×10^{-2}	0.900	8.03×10^{-2}	0.980
3% Inulin	2.63×10^{-2}	0.940	8.71×10^{-2}	0.907

[a] Slopes of the regression lines, as shown in Fig. 4, were taken as the inactivation rates.
[b] R^2 coefficient of determination.

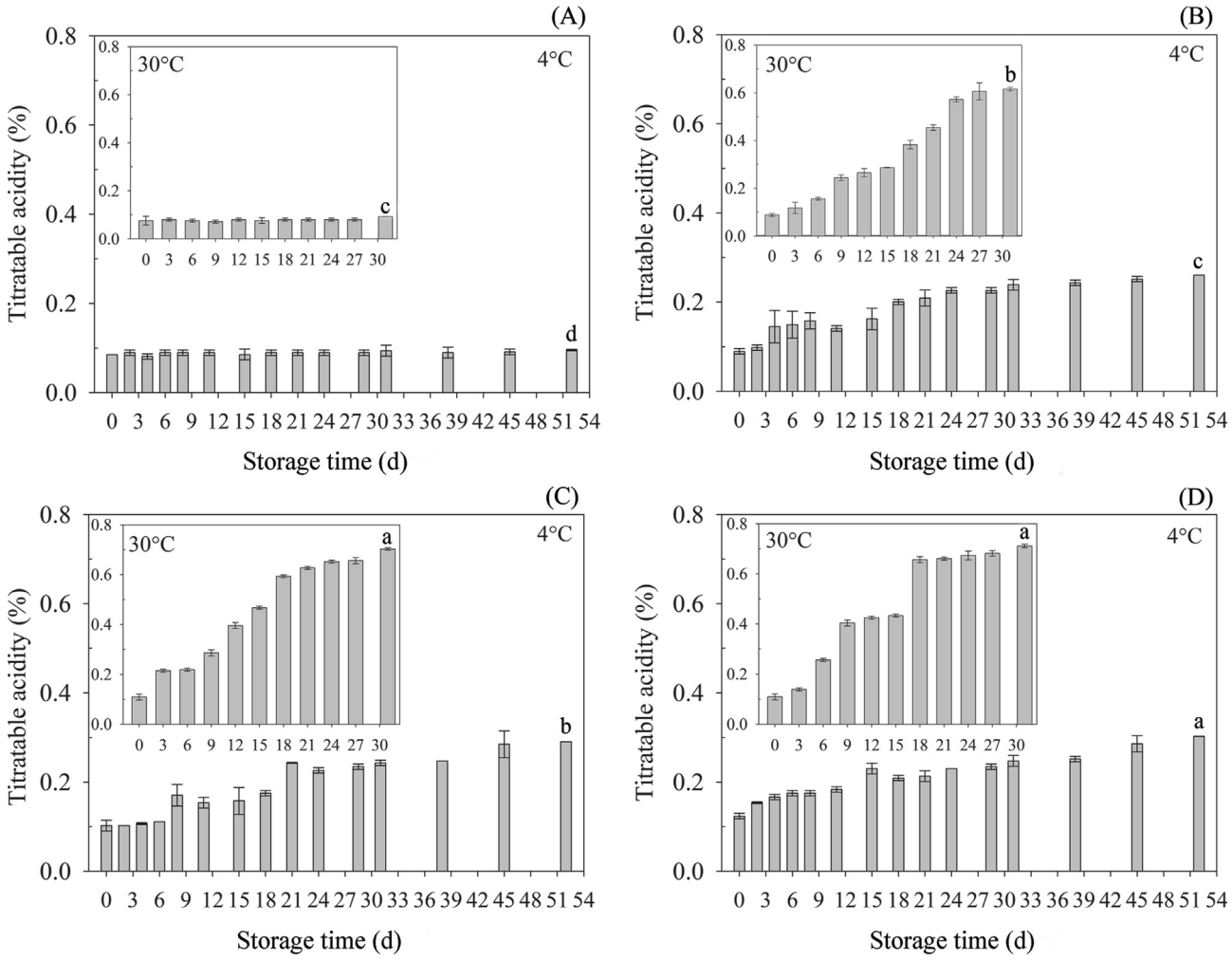

Fig. 6. Changes in titratable acidity of fermented Plai Ngahm Prachin Buri rice extract during storage at 4 °C for 52 d and at 30 °C for 31 d (plot insert) supplemented with: (A) no inulin; (B) 1% inulin; (C) 2% inulin; (D) 3% inulin (values with different lowercase letters (a–d) for each storage temperature are significant different ($p < 0.05$) using Duncan's multiple range test; error bars indicate ± SD.).

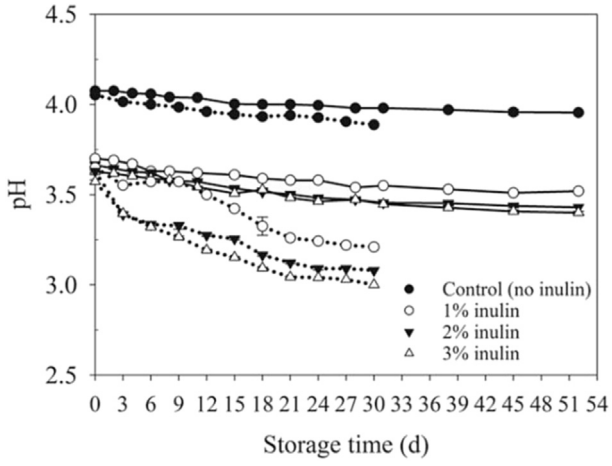

Fig. 7. Changes in pH and lactic acid content of fermented Plai Ngahm Prachin Buri rice extract supplemented with various concentrations of inulin during storage at 4 °C for 52 d (solid lines) and 30 °C for 31 d (dotted lines).

extract supplemented with 2% inulin had the lowest specific rate of cell death of 2.48 × 10^{-2}/d and 8.03 × 10^{-2}/d after storage at 4 °C for 52 d and 30 °C for 31 d, respectively. It could be expected that the use of inulin as a prebiotic might relatively increase the survival of the assayed probiotic *Lactobacillus* strain. These findings may help to expand the application of prebiotic inulin in the design of new functional food ingredients.

Conflict of interest

No conflicts of interest influenced this research.

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