



Usage of Inulin, Lemon, Carrot and Pea Fibers as Prebiotic for *Bifidobacterium Animalis Subsp. Lactis* (BB-12)



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Abstract

Prebiotics, contain oligosaccharides, lignin, resistant starch, tannins and associated plant substances, stimulate growth of probiotic organisms. A lot of dietary fibers (DF) are used as prebiotics in foods. In this study, it was investigated if inulin, lemon, carrot and pea fibers have prebiotic effect or not on *Bifidobacterium animalis subsp. lactis* (*Bifidobacterium* BB-12). For this purpose skimmed milk was inoculated with *Bifidobacterium* BB-12 fortified with different DF, such as inulin, lemon, carrot and pea fibers. According to the results inulin and pea fiber stimulated *Bifidobacterium* BB-12, whereas lemon and carrot fibers had no prebiotic effect on *Bifidobacterium* BB-12. The highest prebiotic effect of inulin and pea fiber were found at a level of 2%.

Keywords: *Bifidobacterium* BB-12; Inulin; Lemon; Carrot; Pea fiber

Abbreviations: DF: Dietary Fiber; BB-12: *Bifidobacterium*; MRS: Man Ragosa Sharpe; LSD: Least Significant Difference

Introduction

Prebiotics are defined as non digestible food ingredients that beneficially affect the host by selectively stimulating the growth and/or activity of one or more desired bacterial species in the colon, thus improving host health. Many oligosaccharides, are added to food as prebiotic, mainly to allow the preferential growth of probiotic organisms and called as prebiotics [1]. Dietary fiber (DF), which includes oligosaccharides, lignin, resistant starch, tannins and associated plant substances, is a remnant of the edible part of plant; it is analogous carbohydrates that are resistant to digestion and absorption in the human small intestine and undergo complete or partial fermentation in the human large intestine. It improves human health by the prevention, reduction and treatment of some diseases, such as diverticular and coronary heart diseases [2,3]. In this study it was investigated whether or not inulin, lemon, carrot and pea fibers have prebiotic effect on *Bifidobacterium animalis subsp. lactis* (*Bifidobacterium* BB-12).

Materials and Methods

Bifidobacterium BB-12 (Chr. Hansen, Turkey), inulin (Sigma, İstanbul), lemon, carrot and pea fibers (Arosel Food, İstanbul, Turkey) and skim milk powder (Pınar Dairy, İzmir, Turkey) were used as materials. Man Ragosa Sharpe (MRS) agar, nalidixic acid, neomycine sulphate, lithium chloride and paramomycine sulphate

obtained from Sigma chemicals (İstanbul, Turkey). All other reagents used were of analytical grade. 13 different reconstituted skim milk, with 12% dry matter, were prepared. For propagation of *Bifidobacterium* BB-12, each sterile reconstituted skim milk was supplemented with 0.50%, 1% and 2% inulin, lemon, carrot and pea fiber. *Bifidobacterium* BB-12 was grown until pH reach to 4.7 at 37°C, using a 0.05% (w/v) inoculum. The milks were kept cooled at 4°C until for enumeration. Bacterial counts were determined after 8h fermentation. Fermented milks were decimally diluted in 100mL sterile peptone water (0.1%) and 1mL aliquot dilutions were poured onto plates of the various selective and differential agars in triplicate. *Bifidobacterium* BB-12 were incubated anaerobically by using MRS-NNLP agar [4]. The plates were incubated at 37°C for 72h. Anaerobic conditions were created using Anaerocult A sochets (Merck). The results were expressed as colony-forming units per gram (cfu g⁻¹) of sample. Statistical analyses were carried out using SPSS Version 5.0 (SPSS Inc. Chicago, IL, USA). Statistically different groups were determined by the LSD (Least Significant Difference) test [5].

Results and Discussion

Table 1 show the variations in *Bifidobacterium* BB-12 counts in reconstituted skim milk with and without DF. *Bifidobacterium*

BB-12 counts were found to be in the range of 7.35-7.96 log cfu g⁻¹, respectively. As seen, inulin and pea fiber had prebiotic effect for *Bifidobacterium* BB-12, whereas lemon fiber and carrot fiber did not have (p<0.05). Even if a slight increasing was found in the *Bifidobacterium* BB-12 counts in the skimmed milk fortified with

lemon and carrot fibers, they were not significant (p>0.05). Pea fiber led to slight increasing in the number of *Bifidobacterium* BB-12, while inulin caused the noticeable increasing. Guler-Akın, Göncü, Akın & Akın, & Güler-Akın [1,6,7] also reported that probiotics had the ability to ferment DFs.

Table 1: Viable counts of *Bifidobacterium* BB-12 in reconstituted skim milk with or without dietary fiber addition (log cfu g⁻¹).

Milk with Fiber* (%)	Inulin	Lemon Fiber	Carrot Fiber	Pea Fiber
0	7.43±0.12 ^{a2}	7.43±0.12 ^{a1}	7.43±0.12 ^{a1}	7.43±0.12 ^{a2}
0.5	7.63±0.10 ^{a2}	7.44±0.09 ^{a1}	7.56±0.11 ^{a1}	7.58±0.08 ^{a2}
1	7.95±0.06 ^{a1}	7.52±0.22 ^{b1}	7.52±0.08 ^{b1}	7.85±0.09 ^{a1}
2	8.06±0.15 ^{a1}	7.45±0.18 ^{b1}	7.48±0.20 ^{b1}	7.97±0.12 ^{a1}

*Different letters in the same line indicate significant differences among the samples depending on DF type and different numbers in the same column indicate significant differences among the samples depending on DF level (p<0.05).

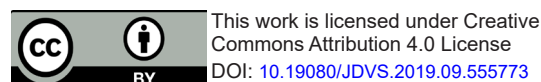
Fiber level affected the number of *Bifidobacterium* BB-12 statistically (p<0.05). The highest prebiotic effect of inulin and pea fiber were found at a level of 2%. The higher inulin and pea fiber added, the more *Bifidobacterium* BB-12 obtained. The numbers of *Bifidobacterium* BB-12 decreased as lemon and carrot fiber increased (p<0.05) due to the possible decreasing of water activity of milk. As known DF is highly hygroscopic and had a high water binding capacity [8].

Conclusion

Results showed that lemon fiber and carrot fiber didn't affect of *Bifidobacterium* BB-12 counts and they had no prebiotic effect for *Bifidobacterium* BB-12. In contrast, inulin and pea fiber stimulated *Bifidobacterium* BB-12 and they could be used as potential prebiotic for *Bifidobacterium* BB-12.

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