

# *Lactobacillus casei* strain Shirota (Yakult)

Research Update for Healthcare Professionals

Issue 7, December  
2016



# Research Update

## 1. Habitual intake of fermented milk products containing *Lactobacillus casei* strain Shirota and a reduced risk of hypertension in older people. Aoyagi Y et al; Benef Microbes. 2016 Dec 1:1-8.

**Aim:** This study investigated relationships between the frequent intake of fermented milk products containing *Lactobacillus casei* strain Shirota (LcS) and the onset of hypertension (resting systemic pressure  $\geq 140$  mmHg [systolic]/ $\geq 90$  mmHg [diastolic]).

**Methods:** A 5-year period of study was conducted in 352 Japanese who were in the age group of 65 to 93 years, (125 men and 227 women). The subjects were divided into two groups (n=254 and n=98) on the basis of their intake of fermented milk products (less than 3 times a week or more than 3 times a week).

**Results:** The incidence of hypertension over the 5-year interval was significantly lower in those who took fermented milk products more than 3 times rather than less than 3 times/week (6.1 vs 14.2%,  $P=0.037$ ). A multivariate-adjusted proportional hazards model predicted that blood pressures were significantly more likely to remain normal over 5 years in subjects who took more than 3 times fermented milk products rather than less than 3 times/ week (relative risk 0.398 [95% confidence interval 0.167-0.948],  $P=0.037$ ).

**Conclusions:** These results suggest that after adjustment for potential confounders, the risk of developing hypertension is substantially lower in elderly people who take fermented milk products containing LcS at least 3 times a week.

## 2. Immune response of healthy adults to the ingested probiotic *Lactobacillus casei* Shirota.

Harbige LS et al; Scand J Immunol. 2016 Oct 8. doi: 10.1111/sji.12495.

**Aim:** To determine the immune response of healthy adults to the ingested Probiotic.

**Methods:** Daily ingestion of a probiotic drink containing *Lactobacillus casei* Shirota (LcS;  $1.3 \times 10^{10}$  live cells) by healthy adults for (i) 4-weeks LcS, (ii) 6-weeks discontinuation of LcS, and (iii) final 4-weeks of LcS, was investigated.

**Results:** There was a significant increase in expression of the T-cell activation marker  $CD3^+ CD69^+$  in ex vivo unstimulated blood cells at weeks 10 and 14 and a significant increase in the NK cell marker  $CD3^+ CD16/56^+$  in ex vivo unstimulated blood cells at weeks 4, 10 and 14. Expression of the NK cell activation marker  $CD16/56^+ CD69^+$  in

ex vivo unstimulated blood cells was 62% higher at week 10 and 74% higher at week 14. Intracellular staining of IL-4 in ex vivo unstimulated and PMA/ionomycin-stimulated CD3<sup>+</sup> β7<sup>+</sup> integrin blood cells was significantly lower at week 10 and 14. Intracellular staining of IL-12 in ex vivo unstimulated and LPS-stimulated CD14<sup>+</sup> blood cells was significantly lower at weeks 4, 10 and 14. Intracellular staining of TNF-α in LPS-stimulated CD14<sup>+</sup> blood cells was significantly lower at weeks 4, 10 and 14. Mucosal salivary IFN-γ, IgA1 and IgA2 concentrations were significantly higher at week 14 but LcS did not affect systemic circulating influenza A-specific IgA or IgG and tetanus specific IgG antibody levels.

**Conclusions:** In addition to the decrease in CD3<sup>+</sup> β7<sup>+</sup> integrin cell IL-4 and a CD14<sup>+</sup> cell anti-inflammatory cytokine profile, at week 14 increased expression of activation markers on circulating T cells and NK cells and higher mucosal salivary IgA1 and IgA2 concentration indicated a secondary boosting effect of LcS.

### **3. The effect of probiotic treatment on elderly patients with distal radius fracture: a prospective double-blind, placebo-controlled randomised clinical trial.**

Lei M et al; Benef Microbes. 2016 Sep 16:1-8.

**Aim:** To assess the effect of probiotic treatment on functional recovery in elderly patients with a distal radius fracture.

**Methods:** A total of 417 elderly patients with an acute distal radius fracture were enrolled in this double-blind placebo-controlled clinical trial. They were randomised to receive skimmed milk containing either a commercial probiotic (*Lactobacillus casei* strain Shirota) or placebo daily for a period of 6 months after the fracture. Treatment outcomes were the DASH (disabilities of the arm, shoulder and hand) score, pain, complex regional pain syndrome (CRPS) score, active range of motion and grip strength, all of which were measured on a monthly basis.

**Results:** Throughout the duration of the study, DASH score, pain, CRPS score, wrist flexion and grip strength of patients receiving probiotics exhibited a significantly faster pace of improvement than those on placebo, with treatment outcomes of patients receiving *Lactobacillus casei* Shirota at month 4 at comparable levels with those of patients receiving placebo at month 6.

**Conclusions:** Probiotic treatment has been shown to improve bone formation, increase bone mass density and prevent bone loss. In elderly patients with a fracture of the distal radius, administration of the probiotic could greatly accelerate the healing process.

#### 4. Evaluation of acrylamide-removing properties of two *Lactobacillus* strains under simulated gastrointestinal conditions using a dynamic system.

Rivas-Jimenez L et al; Microbiol Res. 2016 Sep;190:19-26

**Aim:** The aim of this study was to evaluate the capability of *Lactobacillus reuteri* NRRL 14171 and *Lactobacillus casei* Shirota to remove dietary acrylamide (AA) under simulated gastrointestinal conditions using a dynamic system.

**Method:** The effects of different AA levels or bacteria concentration on toxin removal by *Lactobacillus* strains were assessed. Thereafter, AA-removing capability of bacteria strains under either fasting or postprandial simulated gastrointestinal conditions was evaluated. Commercial potato chips were analyzed for their AA content, and then used as a food model. Average AA content (34,162 $\mu$ g/kg) in potato chips exceeded by ca. 34-fold the indicative values recommended by the EU.

**Results:** Toxin removal ability was dependent on AA content and bacterial cell concentration. A reduction in bacterial viability was observed in the food model and at the end of both digestive processes evaluated. However, bacteria survived in enough concentrations to remove part of the toxin (32-73%). Both bacterial strains were able to remove AA under different simulated gastrointestinal conditions, *L. casei* Shirota being the more effective (ca. 70% removal).

**Conclusions:** These findings confirmed the risk of potato chips as dietary AA exposure for consumers, and that strains of the genus *Lactobacillus* could be employed to reduce the bioavailability of dietary AA.

#### 5. Intra-gastric injection of *Lactobacillus casei* strain Shirota suppressed spleen sympathetic activation by central corticotrophin-releasing factor or peripheral 2-deoxy-d-glucose in anesthetized rats.

Tanida M et al; Neurosci Lett. 2016 Apr 21;619:114-20.

**Aim:** To examine the effects of intra-gastric administration of *Lactobacillus casei* strain Shirota (LcS) on sympathetic activation induced by an intra cerebro ventricular (ICV) injection of corticotrophin-releasing factor (CRF) and an intravenous (IV) injection of 2-deoxy-d-glucose (2DG) or interleukin (IL)-1 $\beta$  in urethane-anesthetized rats.

**Method:** Male Wistar rats were used (weighing 245–290 g) in the present experiment. Animals were housed in a temperature-controlled room with a 12 h light-dark cycle (07:00—19:00 h). Food and water were freely available. Animals were adapted to the experimental environment for at least 1 week prior to the experiment. The autonomic nervous data during each 5 min period after injection of all agents were analyzed by

digital signal processing and appropriate statistical analyses. All data were expressed as mean  $\pm$  SEM.  $P < 0.05$  was considered as statistically significant.

**Results:** Intragastric (IG) administration of probiotic strain *Lactobacillus casei* Shirota (LcS) decreases the sympathetic nerve outflow of anesthetized rats in a tissue-specific manner. In the present study, The IG administration of LcS differently affected the stimulatory responses of sympathetic nerve outflow to CRF. LcS suppressed the increase in splenic sympathetic nerve activity (Spleen-SNA), induced by central CRF, in a dose-dependent manner; however, it did not alter adrenal sympathetic nervous activity (ASNA). In contrast, LcS did not affect spleen-SNA and ASNA following an IV injection of IL-1 $\beta$ . On the other hand, IG administration of LcS suppressed the activation of ASNA following an IV injection of 2DG.

**Conclusions:** These findings suggest that the suppression of central CRF-induced sympathetic activation by LcS is tissue-specific. Moreover, it can suppress the 2DG-induced sympathetic activation. Furthermore, it was also found that stomach-specific vagotomy attenuates the suppressive effect of LcS on CRF-mediated spleen-SNA activation. Thus, the present study suggests that LcS administered to the stomach may act on the afferent vagal nerve and send afferent signals to the brain to regulate efferent SNA induced by sympathetic stimulators.