## The efficacy of a potential new probiotic, *Propionibacterium jensenii* 702, to correct vitamin B<sub>12</sub> levels in an *in vivo* deficient animal model

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For a bacterium to be considered probiotic, it must exert a beneficial effect when consumed by the desired host. *In vitro* methods examining specific traits are useful for screening new strains, but do not necessarily reflect the efficacy of the strain when exposed to a complex ecological system such as the gastrointestinal tract of humans and animals.

*Propionibacterium jensenii* 702 is a novel potential probiotic bacterium. Unlike most probiotics that tend to a member of the genera *Lactobacilli* and *Bifidobacteria*, *P. jensenii* 702 is a member of the dairy propionibacteria group, that to date have not been exploited in the probiotic market. Although it is recognised that propionibacterium do possess a number of traits that make them worthy candidates for probiotics, dairy propionibacteria were chosen for this study due to their ability to produce uniquely high concentrations of vitamin  $B_{12}$  (1). Vitamin  $B_{12}$  deficiency is reported to be one of the risk factors for heart disease, multiple sclerosis, stroke, breast cancer, Alzheimer's disease, some psychiatric syndromes and accelerated aging (2,3).

Twenty-one weanling male Wistar rats were divided into three groups and fed for a period of three months on commercially prepared and pelleted Vitamin  $B_{12}$  Deficient Diet Modified (ICN). The groups, each consisting of seven rats, were all provided with fresh excess sterile drinking water daily which, from week two, was supplemented with either 1µg/mL of cyanocobalamin in 0.85%NaCl (Vitamin  $B_{12}$  group), 0.85% NaCl (Deficiency group), or ~ 10<sup>10</sup> cfu/mL of *P. jensenii* 702 in 0.85% NaCl (Bacteria group). Blood and faces were collected prior to water supplementation and at month one, two and three. Tissue samples were collected at the completion of the study. The samples were analysed for a number of probiotic and safety criteria including serum vitamin  $B_{12}$  and homocysteine.

Group	Month 0 B <sub>12</sub> pmol/L	Homocysteine µmol/L	Month 2 B <sub>12</sub> pmol/L	Homocysteine µmol/L	Month 3 B <sub>12</sub> pmol/L	Homocysteine µmol/L
Vitamin B <sub>12</sub>	$464 \pm 50^{a}$	$18.14 \pm 2.44^{a}$	$*1096 \pm 338^{d}$	$16.77 \pm 2.44^{d}$	*949 ± 347 <sup>g</sup>	$16.39 \pm 1.89^{f}$
Deficiency	$444 \pm 28^{a}$	$15.39 \pm 1.45^{a}$	*135 ± 47 <sup>e</sup>	$*23.05 \pm 4.92^{\text{e}}$	$*105 \pm 34^{h}$	$*26.41 \pm 6.82^{\text{g}}$
Bacteria	$387 \pm 62^{a}$	$13.34 \pm 1.93^{a}$	$241 \pm 94^{f}$	$*18.90 \pm 2.07^{de}$	$*216 \pm 67^{i}$	$16.70 \pm 2.40^{\rm f}$

Values are means  $\pm$  SD (n = 7). Values within a given column with the same superscripts are not significantly different (P > 0.05, Student t-test, two tailed). \* indicates significant difference (P < 0.05) to Month 0 value for equivalent measure (Student t-test, two tailed).

After two months of feeding with *P. jensenii* 702, the serum vitamin  $B_{12}$  of the Bacteria group was comparable to those at day zero (P > 0.05). After three months, the homocysteine levels of the Bacteria group were at the same level as that of day zero and that of the Vitamin  $B_{12}$  group. This demonstrates that *P. jensenii* 702 could be used to provide an *in vivo* vitamin  $B_{12}$  source that could supplement daily food intake. This could have significant impact for those on low vitamin  $B_{12}$  diets who are not willing to take supplements or fortified foods. Other probiotic features of this bacterium (data not shown), such as affects on circulating lipid levels, may also make it an attractive alternative to the genera currently available in probiotic foods.

## References

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