

Apples increased the bifidobacteria population in human *in vitro* colonic gut model – preliminary results

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Apples are among the most frequently consumed fruits and a rich source of polyphenols and soluble fibre, pectin. A major proportion of apple polyphenols escape absorption in the stomach and small intestine, and together with apple non-digestible polysaccharides reach the colon and may serve as substrates for bacterial fermentation⁽¹⁾. Animal studies suggest a synergistic interaction between apple polyphenols and pectin which could increase their biological activity and beneficial effects compared to the individual compounds⁽²⁾. However the effects of whole apples on the human gut microbiota are less extensively studied.

Four commercial apple varieties – Gold Rush, Renetta, Golden Delicious and Pink Lady – were digested⁽³⁾ and fermented *in vitro* using a batch culture model (pH 5.5–6.0, 37 °C), inoculated with human faeces from 3 healthy donors. A 5 ml sample from each vessel was removed immediately for analysis (T0), similarly samples were taken at 5 (T5), 10 (T10), and 24 (T24) hours. Inulin and cellulose were used as positive and negative control respectively. *Bifidobacterium spp* and total bacteria enumeration was performed by real time polymerase chain reaction (RT-PCR)^(4,5).

Table 1. Bacterial populations (log₁₀ cells/ml batch culture fluid) determined by RT-PCR in anaerobic, stirred, pH and temperature controlled faecal batch cultures (n = 3 healthy adults). (*) Mean value was significantly different from that at 0 h: P < 0.05. (#) Mean value was significantly different from Blank at 24 h: P < 0.05. (§) Mean value was significantly different from inulin at 24 h: P < 0.05. (°) Mean value was significantly different from cellulose at 24 h: P < 0.05

	Bifidobacteria													
	Blank		Inulin		Cellulose		Gold rush		Renetta		Golden delicious		Pink lady	
Treatment	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
T0	7.33	0.3	7.43	0.2	7.36	0.2	7.51	0.1	7.39	0.2	7.55	0.2	7.39	0.3
T5	7.56	0.3	7.80	0.4	7.66	0.4	8.05*	0.4	8.29	0.8	8.52*	0.1	8.23	0.2
T10	7.71	0.5	7.74	0.6	7.77	0.6	8.37*	0.2	8.38*	0.2	8.65*	0.2	8.34*	0.1
T24	7.42	0.1	7.84	0.5	7.56	0.1	8.49*#°	0.1	8.62*#§°	0.1	8.51*#°	0.3	8.47*#°	0.1

Bifidobacteria population increased significantly at 10 h and 24 h after a treatment with Gold Rush (P = 0.007 and P = 0.013, respectively), Renetta (P = 0.008 and P = 0.013, respectively) and Pink Lady (P = 0.015 and P = 0.038, respectively) (Table 1); in the case of Golden Delicious, there was an increase after 10 h (P = 0.001), while a trend was observed after 24 h (P = 0.063) (Table 1). There were no significant changes in total bacteria population for any of the fermentations (data not shown). Our preliminary results have shown that apples may beneficially modulate the human gut microbiota composition by increasing the levels of bifidobacteria *in vitro*. Renetta was associated with the highest bifidogenic effect. Ongoing work includes fuller characterisation of the gut microbiota and the end products of their metabolism of apples, short chain fatty acids and polyphenolic catabolites. A randomized controlled human intervention study is required to determine the effects of the Renetta variety on gut microbiota *in vivo* and potential benefits to other health outcomes.

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