Effects of the gut microbiota on colonic secretomotor function

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Introduction: The microbiota is known to influence intestinal myoelectric activity, and germ-free rodents have proven to be a useful tool in determining the divergent role of specific microbial species on intestinal function (Husebye et al., 2001). However, little is known about the influence of the microbiota on mucosal and submucosal sectromotor activity. Given that the microbiota is disturbed in functional gastrointestinal (GI) disorders, such as irritable bowel syndrome (Salonen et al., 2010), examining such interactions may help in understanding how the microbiota contribute to the pathophysiology of functional GI disorders.

Aims: The aim of this study was to investigate the influence of the microbiota on colonic secretomotor activity and to determine whether its absence alters the functional response to selected bacterial species, *Bifidobacterium sp* and *Lactobacillus sp in vitro.*

Methods: Following seromuscular stripping, segments of descending colon from female germ-free (GF) or conventional (CON) Swiss Webster mice (25-35g) were mounted in Ussing chambers (exposed tissue area 0.12cm^2). The segments were bathed with Krebs buffer solution and bubbled with carbogen gas (95% O_2, 5% CO_2).Tissues were voltage clamped at 0mV and changes in short-circuit current (I_{sc}) were continuously recorded. Transepithelial resistance (TER) was calculated using Ohm's Law. Veratridine (VER; 30µM) and capsaicin (CAPS 3µM) were used as pan- and sensory-neural stimulants respectively. Bethanechol (BCh;100µM) and forskolin (FSK;10µM) were used to activate calcium- and cAMP-stimulated changes in I_{sc}. All additions were to the basolateral reservoir, with the exception of bacterial suspensions (1 x 10^9 cfu/ml; bacteria and supernatants) which were added to the luminal reservoir. Data sets were tested for normality using Kolmogorov-Smirnov Test. Following this, a one-way anova followed by post-hoc analysis using Bonferroni's multiple comparison test or a t-test was carried out as appropriate. Data sets are presented as mean ± SEM and a $P<0.05$ was considered statistically significant.

Results: Basal I_{sc} and TER (I_{sc}, GF, 39.1 ± 6.0 µA.cm^{-2}, N=5 vs. CON, 40.2 ± 4.7 µA.cm^{-2}, N=7; TER, data not shown) were comparable between GF and CON tissues, as were neurally-evoked responses (VER, GF, 98.5 ± 14.1 µA.cm^{-2}, N=8 vs. CON, 100.3 ± 12.4 µA.cm^{-2} , N= 7; CAPS, GF, 9.9 ± 2.4 µA.cm^{-2} , N= 8 vs. CON, 9.7 ± 3.8 µA.cm^{-2} , N= 5). However, cAMP-induced changes in I_{sc} were significantly increased in GF compared to CON colon (131.6 ± 16.9 µA.cm^{-2}, N=6 vs. 78.9 ± 13.6 µA.cm^{-2}, N=8, $P<0.05$). No differences in BCh-induced responses were observed between GF and CON tissues. Acute exposure to either *Lacobacillus sp* (GF, 22.6 ± 4.7 µA.cm^{-2}, N=7 vs. CONV, 17.3 ± 3.5 µA.cm^{-2}, N=7) or *Bifidobacterium sp* (GF, 12.1 ± 1.7 µA.cm^{-2}, N=8 vs. CONV, 8.1 ± 3.7 µA.cm^{-2}, N=7) resulted in a comparable I_{sc} response in both groups.

Conclusion: These data suggest that the lack of microbiota does not alter neurally-induced changes in I_{sc}, but significantly enhances sensitivity to cAMP-mediated changes in I_{sc}. However, whether this occurs at a neural or epithelial level remains unknown. Furthermore, the absence of a host flora does not appear to influence the I_{sc} response to two bacterial species following acute exposure.

Husebye et al., Am. J. Physiol. Gastrointest. Liver Physiol 280: G368-G380, 2001.
Salonen et al., Microbiology 156: 3205-3215, 2010.

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