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Butyrate responses in the ascending colon are neuronally mediated, involve PYY but not GLP-1

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Introduction: Butyrate is a short-chain fatty acid produced by fermentation, with mouse colonic luminal concentrations of ~10-20 mM. Free fatty acid receptors (FFA)2 and 3 are located on enteroendocrine L-cells (containing PYY and GLP-1), with FFA2 (Gq-coupled) additionally present on enteric leukocytes and FFA3 (Gi-coupled) on neurons (1). Whilst acting on both receptors, butyrate shows FFA3 preference (2). This study aimed to determine the mechanisms underpinning the functional butyrate response in the ascending colon (AC).

Method: AC mucosa, with intact submucosal innervation, was dissected from male and female wild-type (WT) or PYY-/- (KO) mice (>10 weeks, C57Bl6/129Sv). Mucosae were voltage-clamped and short-circuit current (Isc) measured prior to basolateral tetrodotoxin (TTX; 100 nM) or vehicle pre-treatment, as described previously (3). Following vasoactive intestinal polypeptide (10 nM) stimulation, 5 mM butyrate (the maximal concentration established between 1-20 mM) was added apically or basolaterally. Endogenous GLP-1 mediation of butyrate responses was determined using the GLP-1 antagonist, exendin 9-39 (1 µM), with subsequent GLP-1 agonist, exendin 4 (100 nM) added as a control. Mean+/-1sem were obtained from 5-6 different mice and statistical significance determined using unpaired Student's t-test or one-way ANOVA.

Results: Biphasic 5 mM butyrate responses were observed in WT AC (primary increase in Isc, +2.4±1.6 µA.cm^{-2}, followed by a secondary decrease of -9.5±2.5 µA.cm^{-2},n=5), which were significantly larger than 1 mM (+1.2±0.8 µA.cm^{-2} followed by -1.4±0.4 µA.cm^{-2},n=5, P<0.05) and no different to 10 or 20 mM. There were no significant differences between control apical and basolateral butyrate responses. The decreases in Isc showed TTX-sensitivity: inhibited to 24.8±15.0% of apical responses, $P<0.05$ and 6.5±5.2% of basolateral responses, $P<0.001$. In KO, the residual reductions in Isc seen in WT were abolished, whilst the increases (+12.7±3.2 µM.cm^{-2},n=5) showed partial TTX-sensitivity (+5.6±0.7 µA.cm^{-2},n=5). Exendin 9-39 did not inhibit butyrate-induced increases in Isc, despite inhibiting subsequent exendin 4 responses.

Conclusion: Butyrate inhibitory responses in the AC are neuronally-mediated, possibly via FFA3 inhibition of secretomotor neurones (1), with PYY playing a role presumably via FFA2-mediated signalling (4). However, the Isc increase is not GLP-1-mediated, leaving this phase of the butyrate response unresolved.

References:

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