

PAS/Alcian blue analysis

The entire colon segment of the gastrointestinal tract was dissected and fixed overnight at room temperature in 10% formalin. For each mouse, six 2.5 mm segments were collected every 10 mm along the length of the colon and randomly embedded in the same paraffin block. For each paraffin block, 5  $\mu$ m sections were cut at three different depths (levels), at an average of 400  $\mu$ m apart. At each level, one section was collected and stained with the Richard-Allen Scientific Periodic Acid-Schiff kit, which stains acidic and neutral mucins (*Muc*) for goblet cell visualization and quantification (31). All tissue sections were scanned using a Revolve microscope (ECHO Laboratories, San Diego, CA).