

Title	The impact of complexation or complex coacervation of lactoferrin and osteopontin on simulated infant gastrointestinal digestion, intestinal inflammation, and in vivo bone development
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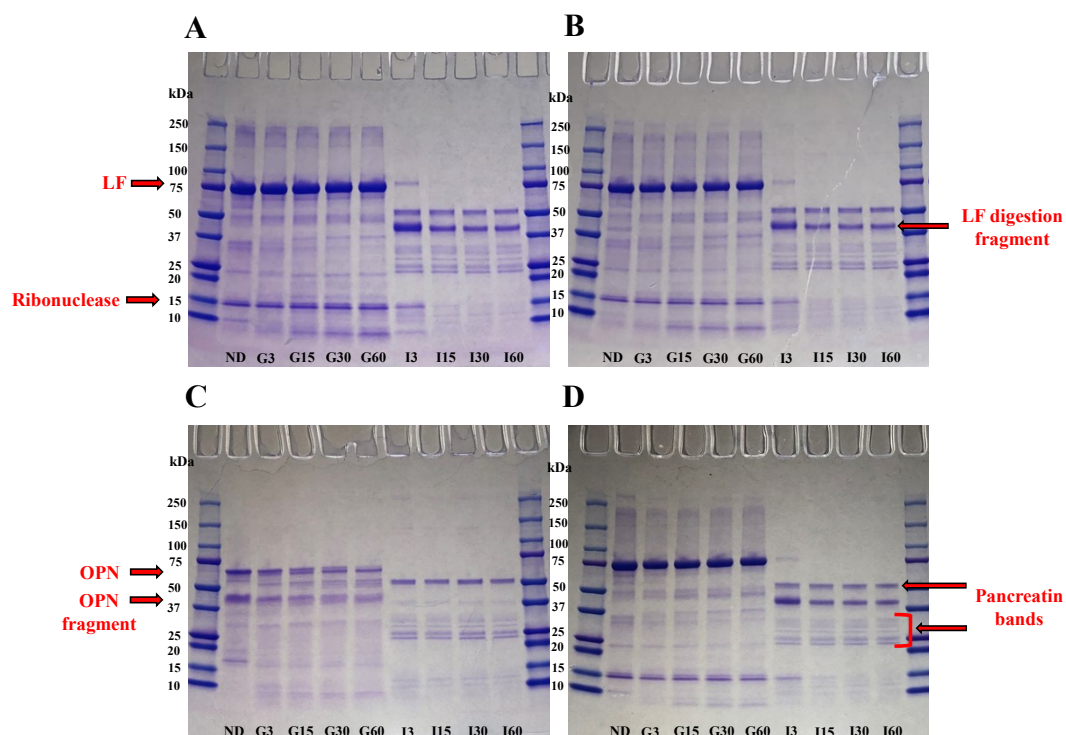


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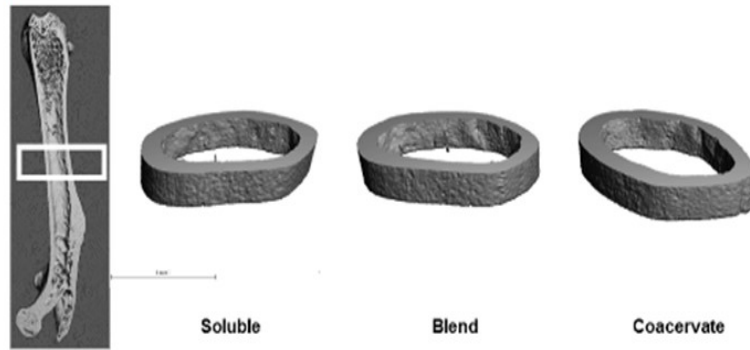
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Supplementary information

**The impact of complexation or complex coacervation of lactoferrin and osteopontin on simulated infant gastrointestinal digestion, intestinal inflammation, and in vivo bone development**



**Supplementary Figure 1.** Proteolysis of uncomplexed and complexed lactoferrin (LF) and osteopontin (OPN) during simulated infant gastrointestinal digestion at pH 5.3 (gastric) and pH 6.6 (intestinal). Panels A-D show the apparent molecular weight profile of gastric and intestinal digestates of (A) LF, (B) LF-OPN soluble complexes (SC), (C) OPN, and (D) LF-OPN complex coacervates (CC), as analysed by reducing SDS-PAGE. Loading quantity was 6  $\mu$ g protein/lane. Each lane contains a digestate collected at a specific timepoint (in minutes) during either the gastric (G) or intestinal (I) phases of simulated digestion, which is annotated at the bottom of each lane. ND is the non-digested sample.



**Supplementary Figure 2.** The impact of lactoferrin-osteopontin soluble complexes (SC), lactoferrin-osteopontin complex coacervates (CC), and a mixed blend of uncomplexed lactoferrin and osteopontin (LF & OPN mix) on cortical bone structure. Presented is a micro-CT image of bone cortical structures following supplementations. Data are shown as mean values  $\pm$  standard error of triplicate measurements. ‘\*’ represents statistically significant differences ( $P < 0.05$ ) between treatments, other p-values compare CC to SC or to LF & OPN mix, as indicated on the figure.