

Bone loss prevention in ovariectomized rats using stable amorphous calcium carbonate*

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ABSTRACT

In assessing the relationship between calcium supplementation and maintaining bone mass or reducing the risk of fracture, the effectiveness of calcium supplementation has never been decisive. Freshwater crayfish rely on amorphous calcium carbonate (ACC), an instable polymorph of calcium carbonate, as the main mineral in the exoskeleton and in the temporary storage organ, the gastrolith. Inspired by the crayfish model, we have previously shown an increase in calcium bioavailability in rats administered with synthetic stable ACC vs. crystalline calcium carbonate (CCC). The current study compared the effects of amorphous calcium derived from either gastrolith or synthetic ACC with those of crystalline calcium, found in commercial CCC or calcium-citrate supplements, in a bone loss prevention model. Rats were subjected to either sham or ovariectomy (OVX) operation (n~20/ group) followed by administration of food pellets supplemented with 0.5% calcium from either source over 12 weeks. Micro-computed tomography (μCT) and histomorphometric analyses revealed bone loss prevention by both gastrolith and ACC treatments, manifested by an increase in morphometric bone parameters, compared to both

CCC- and calcium citrate-treated groups. Both gastrolith and ACC treatments resulted in bone formation in the tibia cancellous bone, indicated by dynamic histomorphometry parameters, compared to either the CCC or calcium citrate treatments. Levels of urine deoxypyridinoline (DPD), suggested an anti-resorptive effect of ACC, which was also the only treatment that led to a significant increase in vertebral mechanical strength, as supported by μCT analysis of topology and orientation parameters of the vertebral trabeculae. To our knowledge, such levels of bone loss prevention by calcium supplements have never been reported. These findings thus suggest the potential of both natural (crayfish gastrolith) and, to a greater extent, synthetic ACC sources for the prevention of metabolic bone disorders and possibly of osteoporotic processes.

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Micro-CT image following 90 days treatment post ovariectomy

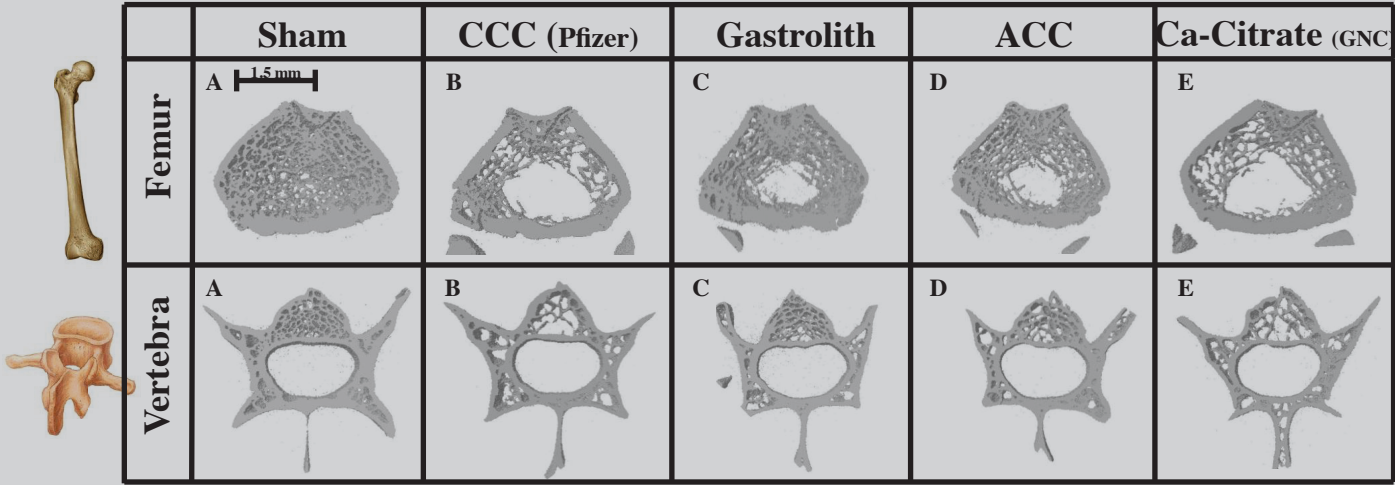


Figure 1. Three-dimensional reconstruction of representative distal femoral and 4th lumbar vertebral cross sections. The trabecular bone region from the (A) sham-operated group treated with crystalline CaCO₃(sham). OVX-treated groups treated with: (B) crystalline CaCO₃(control), (C) gastrolith powder, (D) amorphous calcium carbonate (ACC) or (E) calcium citrate (Ca-citrate). Scale bar represents 1.5 mm. For the illustration, the femoral volume of interest (VOI) was set to 1 mm below the growth plate (most proximal boundary with the metaphysis) and extended proximally for 0.4 mm. The vertebral VOI was set to a 0.4 mm thick cross-section starting 0.14 mm below the cranial growth plate.

ACC Maintains Bone Mechanical Strength

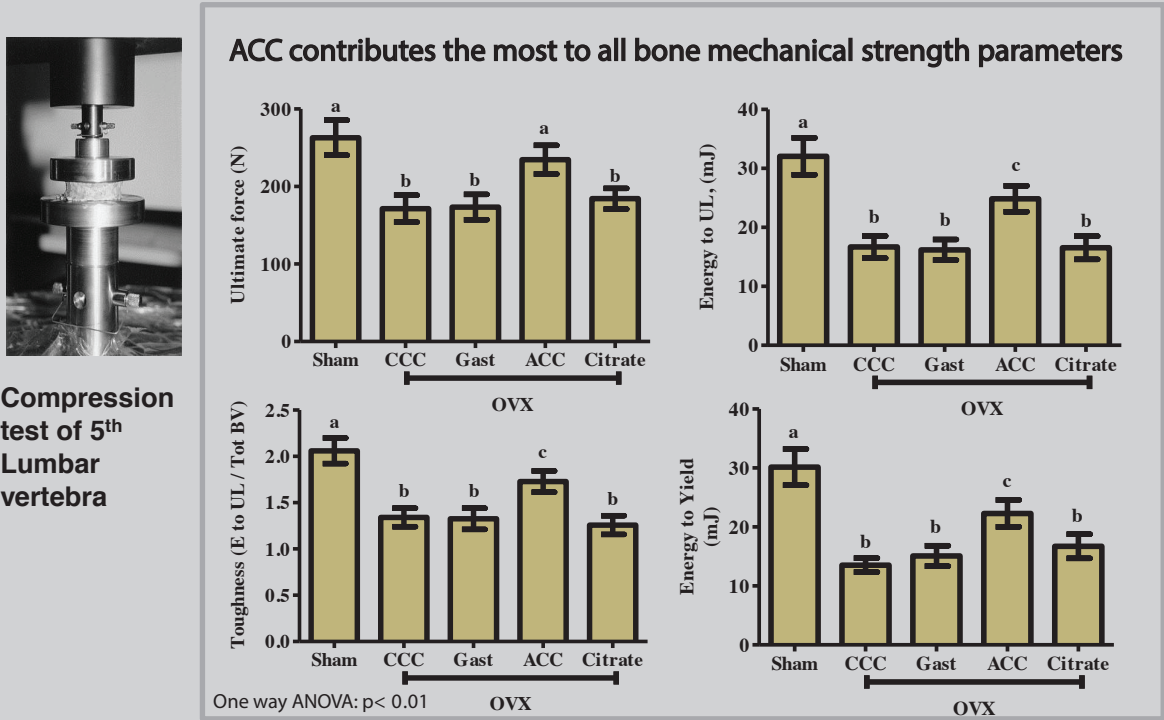


Figure 2. Trabecular bone mineral density (Tb.BMD) and bone volume from total bone tissue (BV/TV) assessed by μCT. (A) Tb. BMD in Distal femur. (B) Tb.BMD in 4th lumbar vertebra. (C) BV/TV in Distal femur. (D) BV/TV 4th lumbar vertebra. Sham-operated group treated with crystalline CaCO₃(Sham). OVX-treated groups treated with: crystalline CaCO₃(Control), gastrolith powder (Gast), amorphous calcium carbonate (ACC) or calcium citrate (Citrate).