Uptake and transport of iron across Caco-2 cell monolayers over time. Intact cells monolayers were established by seeding Caco-2 cells at a density of 30,000 cells/cm² in Transwell® permeable support plates (Costar®, Corning Inc., New York, USA) and culturing in normal medium for 21 days, refreshing the medium every 2 to 3 days. On day 21, (A) total iron uptake (calculated as the sum intracellular iron and iron transported across the cells to the basolateral medium) into the cells from apical medium and (B) iron transport across the cell layer to the basolateral medium were determined at different time point as described in material and methods. Error bars indicate mean ± standard deviation (derived from measurements obtained from 6 separate wells set up in parallel for each time point).