

Co-culture of Caco2 and HT-29 cells as an innovative method to mimic in vitro the morphology and permeability properties of human intestinal epithelium

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For investigating the complexity of the human intestinal epithelium, a valid experimental approach is represented by co-culture. In the present study an intestinal co-culture Caco2/HT-29 (70/30) was set up starting from the parental populations of differentiated cells as previously described [1, 2]. Co-culture was harvested at 0 (T0), 6 (T6), and 14 (T14) days of post confluence after plating. Transmission electron microscopy was carried out to monitor the morphological features of cell differentiation. Alkaline Phosphatase (ALP), Aminopeptidase N (APN) and Dipeptidyl Peptidase IV (DPP IV) activity were assayed as known markers of intestinal cell differentiation. The measure of TEER and the apparent permeability of Lucifer Yellow allows to monitor the integrity of the tight junctions and the permeability of the cell layer formed. At T0 a classical monolayer is present, with a mixed population of immature absorptive elements and secretive cells. At T6 and T14, cells are progressively organized in a multilayer with a parallel growth of microvilli. At T6, co-culture demonstrates good properties of permeability and barrier components, such as mucus, representing an appropriate model for absorption study. At T14, the brush border is even more developed respect to T6 and, together with the increase of the specific activity of ALP, APN, and DPP IV, indicate co-culture as a good model for digestion study. The advantage of this co-culture described is the use of the whole cell population without particular inducers of subclones and growth support. In conclusion, the morphological and biochemical features of co-cultured parental cells change with time, strongly supporting i) an active interaction between the two parental cell lines and ii) the versatility of this model, with more than one prevalent cell type depending on the post confluent stage.

References

- [1] Ferraretto et al. (2007) New methodological approach to induce a differentiation phenotype in Caco-2 cells prior to post confluence stage. *Anticancer. Res.* 27, 3919 – 3925.
- [2] Gravaghi et al. (2007) Casein phosphopeptide promotion of calcium uptake in HT-29 cells - relationship between biological activity and supramolecular structure. *FEBS J.* 247, 4999 – 5011. [10.1111/j.1742-4658.2007.06015.x](https://doi.org/10.1111/j.1742-4658.2007.06015.x)

Keywords

Differentiation; transepithelial electrical resistance; Transmission Electron Microscopy.