

Colorectal cancer: HT-29 Xenograft Mouse Model

Introduction

Colorectal (colon, rectum and anus) cancer is the second leading cause of cancer-related deaths and the third most common cancer worldwide, accounting for approximately 10% of all cancer cases and is the second leading cause of cancer-related deaths worldwide, according to WHO¹. The statistics from 2020 indicate that over 1.9 million new cases of colorectal cancer were reported, with more than 930,000 deaths worldwide. According to global cancer statistics in 2022, there is an approximately 10-fold variation in colon cancer incidence rates by world region in men and women, respectively, with the highest rates in Europe, Australia/New Zealand, and Northern America². The projections for colorectal cancer by 2040 are concerning with an expected increase to 3.2 million new cases and 1.6 million deaths per year¹.

Several lifestyle factors significantly contribute to an increased risk of colon cancer. Strong evidence links alcohol consumption, smoking, diet, and body fatness to a higher likelihood of developing the disease³. Treatments for colon cancer are based on the type and progression of the cancer and the person's medical history. Early detection of colorectal cancer can lead to better treatments and outcomes. Treatments include surgery, radiotherapy, chemotherapy, targeted therapy and immunotherapy.

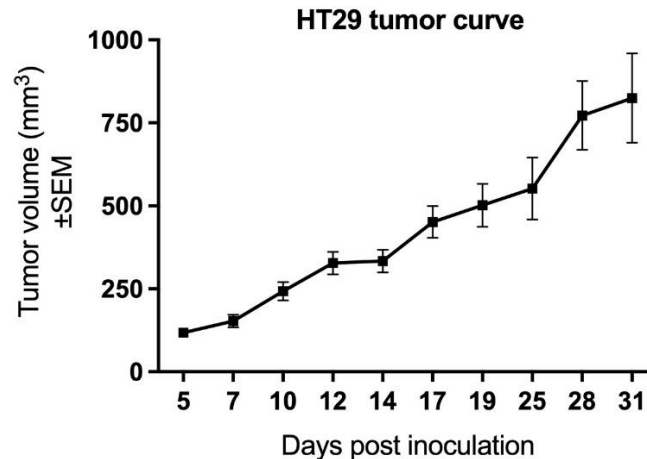
Short description of the cell line

HT-29 cells are a widely used human colorectal adenocarcinoma cell line that exhibit enterocyte-like differentiation under certain conditions⁴. This cell line has an epithelial like morphology and is a suitable transfection host. The ability to form tight monolayers with an apical brush border makes these cells particularly valuable for applications in cancer and toxicology research for studying intestinal permeability, drug absorption and gastrointestinal diseases.

HT-29 cell line was isolated from a primary tumor from a 44-year-old, white, female patient with colorectal adenocarcinoma grade II. It forms a well-differentiated adenocarcinoma consistent with colony primary, grade I.

Growth of Tumor

In BIOEMTECH's laboratories, a mouse model utilizing HT-29 cell line through heterotopic transplantation has been developed. Female immunodeficient nude (CD1-Foxn1nu) mice of 16 weeks old were used for the transplantation. Specifically, 5×10^6 cells/mouse in 100ul PBS solution were inoculated via subcutaneous injection at the lower right flank of the mice. The size of the tumor was evaluated with an electronic caliper. The curve of the tumor's volume is represented in the figure below.



Available Literature Data

Various models have been developed for the identification and pre-clinical validation of novel therapeutic targets in colorectal cancer. Cell line xenograft models have been extensively used for rapid, reductionist, target validation studies, including validation of candidate cancer genes. These studies have shown that the colorectal carcinoma cell lines HCT-116 or HT-29 provide the highest success rate⁵.

Orthotopic xenografts of colorectal cell lines have been reported for improving tumor-stromal cell crosstalk and the reproducibility of liver metastases. Genetically modified mouse models of colorectal cancer are useful for studying species-specific tumor-stroma-immune interactions. They provide a realistic and immune-competent setting that can be used to explore tumor progression, metastasis, and immunotherapy. Moreover, patient-derived xenografts, retain the genetic mutations, molecular profiles, and histological features of the original patient tumor⁶. However, due to the altered tumor microenvironment and lack of an immune system, they are less suitable for studying tumor microenvironment interactions and immune responses.

Regarding the xenograft mouse models, a plethora of them have been developed using HT-29 cells⁷. For example, Tachibana et al. developed a xenograft mouse model of a submillimeter subcutaneous tumor using SCID mice (severe combined immunodeficiency). They transplanted subcutaneously a single spheroid formed by HT-29 cells expressing red fluorescent protein⁸. The spheroid was created in radiation-crosslinked gelatin hydrogel microwells.

Moreover, there is considerable progress on the development of radioligands targeting markers of inflammation which is highly connected with colorectal cancer. Among the identified markers of colorectal inflammation, cyclooxygenase-2 (COX-2) expression is often linked to lesions with a poor prognosis⁹. Various radioligands targeting COX-2 have been developed for SPECT or PET applications including *in vivo* imaging of HT-29 xenografted mice.

The field of colorectal cancer models is evolving, and new models are under development including advanced *in vitro* methods combined with *in silico* studies for more accurate models for drug discovery and screening¹⁰.

References

1. <https://www.who.int/news-room/fact-sheets/detail/colorectal-cancer>.
2. Bray, F. et al. *CA Cancer J Clin* **74**, 229–263 (2024).
3. Chu, A. H. Y. et al. *Am J Clin Nutr* (2025) doi:10.1016/j.ajcnut.2025.01.014.
4. Cohen, E., Ophir, I. & Shaul, Y. Ben. *J Cell Sci* **112**, 2657–2666 (1999).
5. McIntyre, R. E., Buczacki, S. J. A., Arends, M. J. & Adams, D. J. *BioEssays* **37**, 909–920 (2015).
6. Bürtin, F., Mullins, C. S. & Linnebacher, M. *World Journal of Gastroenterology* vol. 26 1394–1426 Preprint at <https://doi.org/10.3748/WJG.V26.I13.1394> (2020).
7. Leigh, A. B. et al. *Integr Cancer Ther* **16**, 339–350 (2017).
8. Tachibana, T. et al. *Applied Sciences (Switzerland)* **11**, (2021).
9. Dagallier, C. et al. *Front Med (Lausanne)* **8**, (2021).
10. Golovko, D., Kedrin, D., Yilmaz, Ö. H. & Roper, J. *Expert Opinion on Drug Discovery* vol. 10 1217–1229 Preprint at <https://doi.org/10.1517/17460441.2015.1079618> (2015).

Please note that all animal care and use was conducted at an authorized user establishment in compliance to European legislation on the protection of animals used for scientific purposes (Directive 2010/63/EU).

Author:

Dr. Maria Kitsara

Science and Innovation Manager



Date: 9/4/2025