

## References

American Cancer Society. (2018). What Is Colorectal Cancer? Retrieved from <https://www.cancer.org/cancer/colon-rectal-cancer/about/what-is-colorectal-cancer.html>

Arnold, M., Sierra, M. S., Laversanne, M., Soerjomataram, I., Jemal, A., & Bray, F. (2016). Global patterns and trends in colorectal cancer incidence and mortality. *Gut*, *66*(4), 683-691. doi:10.1136/gutjnl-2015-310912

ASCO. (2017, December 22). Hereditary Mixed Polyposis Syndrome. Retrieved from <https://www.cancer.net/cancer-types/hereditary-mixed-polyposis-syndrome>

Beck D. E. (2015). The importance of colorectal cancer screening. *The Ochsner journal*, *15*(1), 11-2.

Berridge MV, Herst PM, and Tan AS. Tetrazolium dyes as tools in cell biology: new insights into their cellular reduction. *Biotechnology Annual Review*, *11*: 127-152 (2005)

Bienz, M., & Clevers, H. (2000). Linking Colorectal Cancer to Wnt Signaling. *Cell*, *103*(2), 311-320. doi:10.1016/s0092-8674(00)00122-7

Brazil, D. P., Church, R. H., Surae, S., Godson, C., & Martin, F. (2015). BMP signalling: Agony and antagonism in the family. *Trends in Cell Biology*, *25*(5), 249-264. doi:10.1016/j.tcb.2014.12.004

Cao, X. (2005). Mapping of hereditary mixed polyposis syndrome (HMPS) to chromosome 10q23 by genomewide high-density single nucleotide polymorphism (SNP) scan and identification of BMPR1A loss of function. *Journal of Medical Genetics*, *43*(3). doi:10.1136/jmg.2005.034827

Campos, F. G. (2014). Surgical treatment of familial adenomatous polyposis: Dilemmas and current recommendations. *World Journal of Gastroenterology*,20(44), 16620. doi:10.3748/wjg.v20.i44.16620

*Cell culture manual: 2006-2007*. (2006). St Louis, MO: Sigma-Aldrich.

Church, R. H., Krishnakumar, A., Urbanek, A., Geschwinder, S., Meneely, J., Bianchi, A., ... Brazil, D. P. (2015). Gremlin1 preferentially binds to Bone Morphogenetic Protein-2 (BMP-2) and BMP-4 over BMP-7. *Biochemical Journal*, 466(1), 55-68. <https://doi.org/10.1042/BJ20140771>

Davis H, Irshad S, Bansal M, Rafferty H, Boitsova T, Bardella C, Jaeger E, Lewis A, Freeman-Mills L, Giner FC, Rodenas-Cuadrado P, Mallappa S, Clark S, Thomas H, Jeffery R, Poulson R, Rodriguez-Justo M, Novelli M, Chetty R, Silver A, Sansom OJ, Greten FR, Wang LM, East JE, Tomlinson I, Leedham SJ *Nat Med*. 2015 Jan; 21(1):62-70.

Dukes, C. E. (1932). The classification of cancer of the rectum. *The Journal of Pathology and Bacteriology*,35(3), 323-332. doi:10.1002/path.1700350303

Faussadier, X. (2017, November 28). HeLa cells: Origin of this important cell line in life science research. Retrieved from <https://www.tebu-bio.com/blog/2017/11/28/hela-cells-the-first-cell-line/>

Fitzmaurice, C., Akinyemiju, T. F., Lami, F. H., Alam, T., Alizadeh-Navaei, R., Allen, C., . . . Naghavi, M. (2018). Global, Regional, and National Cancer Incidence, Mortality, Years of Life Lost, Years Lived With Disability, and Disability-Adjusted Life-Years for 29 Cancer Groups, 1990 to 2016. *JAMA Oncology*,4(11), 1553. doi:10.1001/jamaoncol.2018.2706

Grela, E., Ząbek, A., & Grabowiecka, A. (2015). Interferences in the Optimization of the MTT Assay for Viability Estimation of *Proteus mirabilis*. *Avicenna journal of medical biotechnology*, 7(4), 159–167.

Haggar, F. A., & Boushey, R. P. (2009). Colorectal cancer epidemiology: incidence, mortality, survival, and risk factors. *Clinics in colon and rectal surgery*, 22(4), 191-7.

Harris R. Global epidemiology of cancer. Burlington, MA: Jones Bartlett; 2016.

He, Y., Zhu, Q., Chen, M., Huang, Q., Wang, W., Li, Q., ... Di, W. (). The changing 50% inhibitory concentration (IC50) of cisplatin: a pilot study on the artifacts of the MTT assay and the precise measurement of density-dependent chemoresistance in ovarian cancer. *Oncotarget*, 7(43), 70803–70821. doi:10.18632/oncotarget.12223

H. Q. Yang, Y. H. Wang, J. X. Chen, et al., “Efficacy of Proliferation of HeLa Cells under Three Different Low-Intensity Red Lasers Irradiation,” *International Journal of Photoenergy*, vol. 2012, Article ID 290796, 5 pages, 2012. <https://doi.org/10.1155/2012/290796>.

Jaeger E, Leedham S, Lewis A, et al. Hereditary mixed polyposis syndrome is caused by a 40-kb upstream duplication that leads to increased and ectopic expression of the BMP antagonist GREM1. *Nat Genet* 2012; 44: 699-703

Karagiannis, G. S., Musrap, N., Saraon, P., Treacy, A., Schaeffer, D. F., Kirsch, R., . . . Diamandis, E. P. (2015). Bone morphogenetic protein antagonist gremlin-1 regulates colon cancer progression. *Biological Chemistry*, 396(2). doi:10.1515/hsz-2014-0221

Kim J. H. (2015). Chemotherapy for colorectal cancer in the elderly. *World journal of gastroenterology*, 21(17), 5158-66.

Kim, M., Yoon, S., Lee, S., Ha, S. A., Kim, H. K., Kim, J. W., & Chung, J. (2012). Gremlin-1 Induces BMP-Independent Tumor Cell Proliferation, Migration, and Invasion. *PLoS ONE*, 7(4). doi:10.1371/journal.pone.0035100

Kim, T. K., & Eberwine, J. H. (2010). Mammalian cell transfection: the present and the future. *Analytical and bioanalytical chemistry*, 397(8), 3173–3178. doi:10.1007/s00216-010-3821-6

Kišonaitė, M., Wang, X., & Hyvönen, M. (2016). Structure of Gremlin-1 and analysis of its interaction with BMP-2. *The Biochemical journal*, 473(11), 1593–1604. doi:10.1042/BCJ20160254

Klabunde, C. N., Cronin, K. A., Breen, N., Waldron, W. R., Ambs, A. H., & Nadel, M. R. (2011). Trends in Colorectal Cancer Test Use among Vulnerable Populations in the United States. *Cancer Epidemiology Biomarkers & Prevention*, 20(8), 1611-1621. doi:10.1158/1055-9965.epi-11-0220

Kopelson, G. (1983). Adjuvant postoperative radiation therapy for colorectal carcinoma above the peritoneal reflection I. Sigmoid colon. *Cancer*, 51(9), 1593-1598. doi:10.1002/1097-0142(19830501)51:93.0.co;2-8

Koroglu, N., Mathyk, B. A., Tola, E. N., Cetin, B. A., Yuksel, I. T., Dag, I., & Yildirim, G. Y. (2019). Gremlin-1 and gremlin-2 levels in polycystic ovary syndrome and their clinical correlations. *Gynecological Endocrinology*, 35(7), 604-607. doi:10.1080/09513590.2019.1566452

Kosinski, C., Li, V. S., Chan, A. S., Zhang, J., Ho, C., Tsui, W. Y., . . . Chen, X. (2007). Gene expression patterns of human colon tops and basal crypts and BMP antagonists as intestinal stem cell niche factors. *Proceedings of the National Academy of Sciences*, 104(39), 15418-15423. doi:10.1073/pnas.0707210104

Li, J., Yi, C. H., Hu, Y. T., Li, J. S., Yuan, Y., Zhang, S. Z., ... Ding, K. F. (2016). TNM Staging of Colorectal Cancer Should be Reconsidered According to Weighting of the T Stage: Verification Based on a 25-Year Follow-Up. *Medicine*, 95(6), e2711. doi:10.1097/MD.0000000000002711

Lieberman, S., Walsh, T., Schechter, M., Adar, T., Goldin, E., Beerli, R., . . . Goldberg, Y. (2017). Features of Patients With Hereditary Mixed Polyposis Syndrome Caused by Duplication of GREM1 and Implications for Screening and Surveillance. *Gastroenterology*,*152*(8). doi:10.1053/j.gastro.2017.02.014

Lockhart-Mummery, J. P. (1926). Two hundred cases of cancer of the rectum treated by perineal excision. *British Journal of Surgery*,*14*(53), 110-124. doi:10.1002/bjs.1800145312

Macrae, F. A. (2018, November 26). Colorectal cancer: Epidemiology, risk factors, and protective factors. Retrieved December 18, 2018, from <https://www.uptodate.com/contents/colorectal-cancer-epidemiology-risk-factors-and-protective-factors#H3>

Mahmood, T., & Yang, P. C. (2012). Western blot: technique, theory, and trouble shooting. *North American journal of medical sciences*, *4*(9), 429–434. doi:10.4103/1947-2714.100998

Maxton, D. (2018, May 09). Familial polyposis coli (familial adenomatous polyposis or FAP). Retrieved from <https://www.netdoctor.co.uk/conditions/digestive-health/a4923/familial-polyposis-coli-familial-adenomatous-polyposis-or-fap/>

Meerlo, J. V., Kaspers, G. J., & Cloos, J. (2011). Cell Sensitivity Assays: The MTT Assay. *Methods in Molecular Biology Cancer Cell Culture*,*237*-245. doi:10.1007/978-1-61779-080-5\_20

Mittal, S., Mifflin, R., & Powell, D. W. (2009). Cancer Stem Cells: The Other Face of Janus. *The American Journal of the Medical Sciences*,*338*(2), 107-112. doi:10.1097/maj.0b013e3181ad5865

Namkoong, H., Shin, S. M., Kim, H. K., Ha, S. A., Cho, G. W., Hur, S. Y., ... Kim, J. W. (2006). The bone morphogenetic protein antagonist gremlin 1 is overexpressed in human cancers and interacts with YWHAH protein. *BMC cancer*, *6*, 74. doi:10.1186/1471-2407-6-74

Pelli A, Väyrynen JP, Klintrup K, et al. Gremlin1 expression associates with serrated pathway and favourable prognosis in colorectal cancer. Page 17 of 30 <http://mc.manuscriptcentral.com/jpath> The Journal of Pathology 123456789 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47 48 49 50 51 52 53 54 55 56 57 58 59 60 For Peer Review Page | 18 Histopathology 2016; 69: 831-838

Recillas-Targa, F. (2006). Multiple Strategies for Gene Transfer, Expression, Knockdown, and Chromatin Influence in Mammalian Cell Lines and Transgenic Animals. *Molecular Biotechnology*, 34(3), 337-354. doi:10.1385/mb:34:3:337

Retting, K. N., Song, B., Yoon, B. S., & Lyons, K. M. (2009). BMP canonical Smad signaling through Smad1 and Smad5 is required for endochondral bone formation. *Development (Cambridge, England)*, 136(7), 1093–1104. doi:10.1242/dev.029926

Sato, M., Kawana, K., Fujimoto, A., Yoshida, M., Nakamura, H., Nishida, H., . . . Fujii, T. (2015). Clinical significance of Gremlin 1 in cervical cancer and its effects on cancer stem cell maintenance. *Oncology Reports*, 35(1), 391-397. doi:10.3892/or.2015.4367

Sigma Aldrich. (2016). Cell Proliferation Kit I (MTT) 11465007001. Retrieved from <https://www.sigmaaldrich.com/catalog/product/roche/11465007001?lang=en&ion=GB>

Sneddon JB, Zhen HH, Montgomery K, van de Rijn M, Tward AD, West R, Gladstone H, Chang HY, Morganroth GS, Oro AE, Brown PO. Proc Natl Acad Sci U S A. 2006 Oct 3; 103(40):14842-7.

Tasca, A., Stemig, M., Broege, A., Huang, B., Davydova, J., Zwijsen, A., ... Mansky, K. C. (2015). Smad1/5 and Smad4 expression are important for osteoclast differentiation. *Journal of cellular biochemistry*, 116(7), 1350–1360. doi:10.1002/jcb.25092

Topol LZ, Modi WS, Koochekpour S, Blair DG (2000) DRM/GREMLIN (CKTSF1B1) maps to human chromosome 15 and is highly expressed in adult and fetal brain. *Cytogenet Cell Genet* 89: 79–84.

Wee, P., & Wang, Z. (2017). Epidermal Growth Factor Receptor Cell Proliferation Signaling Pathways. *Cancers*, 9(5), 52. doi:10.3390/cancers9050052

Western Blot Protocol: Immunoblotting Protocol. (n.d.). Retrieved from <https://www.sigmaaldrich.com/technical-documents/protocols/biology/western-blotting.html>

Wordinger, R. J., Zode, G. and Clark, A. F. (2008) Focus on molecules: gremlin. *Experimental eye research*. 87, 78-79

Wu J. S. (2007). Rectal cancer staging. *Clinics in colon and rectal surgery*, 20(3), 148–157. doi:10.1055/s-2007-984859