**Bone Marrow Derived Mesenchymal Stem Cells Versus Astaxanthin in Treatment of Toxic Effect of 5- Fluorouracil on Gastric mucosa of Male Albino Rats**

Amal M. ElSafy Elshazly1, Neama Mahmoud Taha 2, Naglaa

A.S. Sarg1, Asmaa Y.A. Hussein 3 ,Yasmeen Mohammed Ismail El Sayed 4, Haidy M.Fakher3,and Ali Mohamed Ali 1.

Department of Anatomy and Embryology Faculty of Medicine, Benha University 1.

Physiology Department, Umm Al-Qura University, KSA 2.

Department of Forensic Medicine and Clinical Toxicology Faculty of Medicine, Benha University 3.

Department of pharmacology Faculty of Medicine, Benha University4.

Corresponding author: Amal Elshazly,

**Mobile**:(+20)1222924128,**Emai**[**l:**amal.elshazly79@yahoo.com](mailto:amal.elshazly79@yahoo.com)

https://orcid.org/0000-00031051-7241

**Abstract:** 5-fluorouracil (5-FU) is an effective chemotherapeutic medication. It is commonly implicated in treatment of various malignancies; this research aimed to investigate potential therapeutic effect of the BMMSCs and Astaxanthin on 5-FU toxic effect on albino rat's stomach. 40 rats were divided into 4 groups: Group I (control group): received no treatment Group II (5-fluorouracil group): for 5 consecutive days rats were intravenously injected by 50 mg/kg 5-FU. Group III (5- Fluorouracil group and Astaxanthin): got 5-FU as in group II, followed by astaxanthin 50mg/kg for 10 consecutive days. Group IV (BMMSCs): received 5-FU as in group II then intravenously injected by 106 of BMMSCs once in rat tail. All rats were sacrificed on the 35th day from the beginning of the experiment, to be examined microscopically, stomach sections were stained by H&E to evaluate histopathological changes, stained also with PAS to evaluate mucosal glycoprotein production, transmission electron microscopic was used to evaluate ultra-structural changes and Immunohistochemistry using Ki67 was used to evaluate level of cell proliferation. examination of stomach Sections of group II showed separation in the gastric glands, congested blood capillary, vacuolations of parietal cells, while (Gr III) and (Gr IV) showed marked stomach histological structure improvement and down regulated expression of Ki67, compared to the (GrII). electron microscope supported these results. Both BMMSCs and Astaxanthin have good impact in ameliorating the toxic effect of 5-FU on albino rats stomach, with no significant difference between both techniques.

**Key words:** Astaxanthin,5FU,stomach,MSCs.

### Introduction:

Chemotherapy includes the use of cytotoxic agents to eradicate neoplastic cancer cells in different organs (Ferlay et al., 2015). 5-fluorouracil (5-FU) is an effective antimetabolite chemotherapeutic medication. It is commonly implicated in various malignancies treatment as colorectal, liver, head, neck skin, and breast cancers (Bachmeier et al., 2019). Regarding the National Comprehensive Cancer Network and

the European Society for Medical Oncology guidelines, 5-FU is a crucial medicine for both adjuvant therapy and metastatic colorectal cancer (Benson et al., 2017). 5FU is activated by thymidine phosphrylase enzyme into fluoro - deoxyuridylate which is capable of suppressing thymidylate synthetase, resulting in direct inhibition of DNA synthesis. (Kataoka et al., 2010)

There is a consistent and long history of 5-FU in mucosal injury induction. It causes lesions in the form of inflammation and ulceration at any region of the gastrointestinal system, causing a variety of symptoms that significantly influence the tolerance of chemotherapy and quality of life is the definition of mucositis (Gawish et al., 2013). Astaxanthin is a carotenoid pigment with a powerful antioxidative and anti- inflammatory effects. It is the most potent and safest antioxidant known to exist in nature. Numerous types of algae, plants, and types of seafood, such as shrimp and salmon fish, are typical natural sources of Astaxanthin. It is derived from Haematococcus pluvialis microalgae (Mosaad et al., 2016).

Astaxanthin, has been found to be highly effective in mopping up free radicals as it possesses anti-oxidative, anti-inflammatory, anti-apoptotic, and other beneficial pharmacological properties. Many chemical reactions produce free radicals which are injurious to body cells, as they are the causes of many diseases, disabilities, and death. Antioxidants suppress and mop up these circulating free radicals (Kim et al., 2018). Stem cell therapy refers to any technique that uses or stimulates stem cells. Typically, it serves to repair or replace damaged cells or tissues (Rashed et al., 2016). Bone marrow-mesenchymal stem cells (BMMSCs) were reported by multiple studies to had a valuable role in the gut damage healing and contribute to the gastrointestinal tissue's development (Liu et al., 2015).

Askarov et al., (2008) found that multipotent mesenchymal stromal cells derived from autologous bone marrow may promote gastric mucosa microcirculation, stimulate angiogenesis, and enhance gastric ulcer healing in rats

This study aimed to assess 5-FU effect on gastric mucosa and compare between the beneficial regenerative effects of BMMSCs and Astaxanthin on 5-FU- induced gastric toxic lesions.

### References:

**Aldo, C., Ítalo, M., Marília, L., et al. (2018):** Effects of simvastatin on 5- fluorouracil-induced gastrointestinal mucositis in rats. Rev Col Bras Cir 45(5):1968- 1980.

**Alexandra, K., Tanja, A., Svenja, W., et al. (2017):** Labeling Mesenchymal Stromal Cells with PKH26 or Vybrant Dil Significantly Diminishes their Migration, but does not affect their Viability, Attachment, Proliferation and Differentiation Capacities. Journal of Tissue Science & Engineering, 8:2

**Askarov, M., Vostrikova, O., Vorobjova, N., et al. (2008):** Effects of autologous bone marrow cells on apoptosis and regeneration of non-healing autoimmune gastric ulcers. Bull. Exp. Biol. Med., 146: 647–651.

**Bachmeier, E., López, M., Linares, J. et al. (2019):** 5-Fluorouracil and Cyclophosphamide modify functional activity in submandibular gland of rats. J. Oral Res, 8: 363–369.

**Bancroft, J., Suvarna, K., Layton, C., (2018):** Bancroft, sTheory and Practice of Histological Techniques, eighth ed.

**Benson, A., Venook, A., Cederquist L., et al (2017):** Colon cancer, version 1.2017, NCCN clinical practice guidelines in oncology. J Natl Compr Canc Netw. 15:370– 398.

**Bruno, S., Grange, F., Collino, M., et al. (2012):** Microvesicles derived from mesenchymal stem cells enhance survival in a lethal model of acute kidney injury, PLoS One 7 (3).

**Camera, E., Matrofrancesco, A., Fabbri, F., et al. (2008)**: Astaxanthin, canthaxanthin and beta-carotene differently affect UVA-induced oxidative damage and expression of oxidative stress-responsive enzymes, Exp. Dermatol., in press. **Chang, Q., Yan, L., Wang, C., et al. (2012)**: In vivo transplantation of bone marrow mesenchymal stem cells accelerates repair of injured gastric mucosa in rats. Chin. Med. J. 125, 1169–1174 [PMID: 22613549]

**Chi-Chang, H., Yi-Ming, C., Dean-Chuan, W., et al. (2014):** Cytoprotective effect of American ginseng in a rat ethanol gastric ulcer model. Molecules, 19, 316–326.

**El-Bermawy, M., (2015):** Light and scanning electron microscopic study of 5- fluorouracil-induced mucosal injury in the gastric fundus and the possible protective

role of omeprazole in adult male albino rat. The Egyptian Journal of Histology., 38 (3

): 415–426.

**Ferlay, J., Soerjomataram, I., Dikshit, R., et al (2015):** Cancer Incidence and Mortality Worldwide: Sources, methods and major patterns in GLOBOCAN 2012. Int. J. Cancer 2015, 136, E359–E386.

**Gawish S., Nosseir D., Omar N., et al. (2013):** Histological and Ultra Structural Study of 5-fluorouracil-induced Small Intestinal Mucosal Damage inRats. Asian Journal of Cell Biology 8: 1-21.

**Gorkem A., Huseyin E., Levent T., et. al. (2018):** The protective effect of astaxanthin against cisplatin-induced nephrotoxicity in rats. Biomedicine & Pharmacotherapy 100 575-582

**Guangxin, J., Gongcai, Q., Dequan, H., et al. (2013):** Allogeneic bone marrow- derived mesenchymal stem cells attenuate hepatic ischemia-reperfusion injury by suppressing oxidative stress and inhibiting apoptosis in rats. Int. J. Mol. Med. 31 (6): 1395–1401, 1340.

**Hix, L., Lockwood, J., Bertram, (2004)**: Upregulation of connexin 43 protein expression and increased gap junctional communication by water soluble disodium disuccinate astaxanthin derivatives, Cancer Lett., 211 (1): 25–37.

**Ikeda, S., Tsuji, A., Satoh, M., et al. (2008):** Protective effects of astaxanthin on 6- hydroxydopamine-induced apoptosis in human neuroblastoma SH-SY5Y cells, J. Neurochem. 107 (6): 1730–1740.

**Iwamoto, T., Hosoda, K., Hirano, R., et al. (2000):** Inhibition of low-density lipoprotein oxidation by astaxanthin. J. Atheroscler. Thromb., 7: 216–222.

**Jacobo, T., Tihomir, G., Mariano, G., et al. (2013):** Regenerative medicine and cell therapy. Stem Cell Biol. Regener. Med. 24, 5–277.

**Kataoka Y., Iimori M., Niimi S., et al. (2010):** Cytotoxicity of trifluridine correlates

with the thymidine kinase 1 expression level. Sci Rep., 9:79642019.

**Khan, S., Malinski, T., Mason, R., et al. (2010):** Novel astaxanthin prodrug (CDX-

085) attenuates thrombosis in a mouse model. Thromb. Res. 126: 299–305.

**Kim S., Christopher L., John D., et al (2019):** Theory and practice of histological techniques. 8th ed. China, Churchill Livingstone, Elsevier P. 126 .

**Kim, S., Kim, H. (2018):** Inhibitory effect of astaxanthin on oxidative stress-induced mitochondrial dysfunction-A mini-riview. Nutrients 10: 1137.

**Kuznietsova N. and Luzhenetska V. (2014):** the state of rat gastric mucosa under the influence of cytostatics: dihydropropyrrol derivative 5-FU and their compination. Studia Biologica •Том 8/No1 • С. 85–92

**Liu, L., Chiu, P., Lam, P., et al. (2015):** Effect of local injection of mesenchymal stem cells on healing of sutured gastric perforation in an experimental model. Br. J. Surg., 102 (2):158–68.

**Manabe, E., Handa, O., Naito Y., et al. (2008)**: Astaxanthin protects mesangial cells from hyperglycemia-induced oxidative signaling, J. Cell Biochem. 103 (6) : 1925– 1937.

**Manar, A., and Dalia, A., (2020) :**Light and Electron Microscopic Studies on the Possible Protective Effect of Ginger on the Gastric Fundic Mucosa of Adult Male Albino Rats Treated with 5- Fluorouracil E GYPTIAN JOURNAL OF HISTOLOGY Article 3, [Volume 43, Issue 1](https://ejh.journals.ekb.eg/issue_14348_14349_.html)Page 36-50

**Mosaad, Y., Abd El Khalik G., Hussein, M., et al. (2016);** a Astaxanthin promising protector against gentamicin-induced nephrotoxicity in rats, Curr. Pharm. Biotechnol. 17 (13): 1189–1197.

**Nakao, R., Nelson, O., Park, J., et al. (2010):** Effect of astaxanthin supplementation on inflammation and cardiac function in BALB/c mice. Anticancer Res., 30: 2721– 2725.

**Ohgami, K., Shiratori, K., Kotake, S., et al. (2003)**: Effects of astaxanthin on lipopolysaccharide-induced inflammation in vitro and in vivo. Investig. Ophthalmol. Vis. Sci., 44, 2694–2701.

**Rashed, L., Gharib, M., Hussein, E., et al. (2016)**: Combined effect of bone marrow derived mesenchymal stem cells and nitric oxide inducer on injured gastric mucosa in a rat model. Tissue and Cell, 48(6): 644-652.

**Santocono M., Zurria, M., Berrettini, M., et al. (2006):** Influence of astaxanthin, zeaxanthin and lutein on DNA damage and repair in UVA-irradiated cells, J Photochem. Photobiol. B 85 (3): 205–215.

**Takkem, A., Barakat, C., Zakaraia, S., et al. (2018):** Ki-67 prognostic value in different histological grades of oral epithelial dysplasia and oral squamous cell carcinoma. Asian Pacific Journal of Cancer Prevention, 19(11): 3279– 3286.

**Yujiro, H., Shingo, T., Masahiko, T., et al. (2008):** Sunao Kawano Topical transplantation of mesenchymal stem cells accelerates gastric ulcer healing in rats. Am. J. Physiol. Gastrointest. Liver Physiol., 294, 778–786.

**Zhang, C., Helmsing, S., Zagrebelsky, M., et al. (2012):** Suppression of p75 neurotrophin receptor surface expression with intrabodies influences Bcl-xL mRNA expression and neurite outgrowth in PC12Cells. PLoS One 7:30684.

# لباقم ماظعلا

**يبرعلا صخلملا**

**عاخن نم هقتشملا ةطيسولا ةيعذجلا ايلاخلا**

**-5 راقع نع**

**مجانلا ماسلا**

**ريثأتلا**

**جلاع يف**

**نيثنازاتسأ**

**ناذرجلا**

**روكذ**

**ةدعمل**

**هيطاخملا هيشغلاا ءاضيبلا**

**يلع ليسارويورولف**

## عيشي

ثيح

## يئايميكلا

جلاعلا

يف لاعف

ءاود

ليسارويورولف-5

راقع ربتعي

:ةصلاخلا

يف قيقحتلا

ىلإ

## ثحبلا

اذه

## فدهي

.ةفلتخملا

ةثيبخلا

مارولأا

جلاع

يف همادختسا

راقعو ماظعلا عاخن نم هقتشملا هطيسولا هيعذجلا ايلاخلل ـل لمتحملا يجلاعلا ريثأتلا

ةدعم

ىلع

## ليساريورولف

5لا راقع هببسي يذلا

ماسلا

ريثأتلا

ىلع

## نيثنزاتسلاا

ةعومجملا

:تاعومجم

هعبرا

ىلإ

## نيعبرلاا نازرجلا

ميسقت مت

. ءاضيبلا

ناذرجلا

-5 ةعومجم(

ةيناثلا

ةعومجملا

جلاع

يأ ىقلتت

لا :)ةطباضلا

ةعومجملا(

ىلولأا

/ مجم 50

رادقمب

ديرولا

قيرط

نع اهنقح مت

ةيلاتتم

مايأ 5

ةدمل

:)ليسارويورولف

:)نيثنازاتسأو ليسارويورولف-5 ةعومجم( ةثلاثلا ةعومجملا .ليساروي ورولف-5 مجك

/ مجم 50 نيثنازاتسأ اهيلي ، ةيناثلا ةعومجملا يف امك ليساري ورولف-5 ىلع تلصح

:هطيسولا هيعذجلا ايلاخلا هعومجم :ةعبارلا ةعومجملا .ةيلاتتم مايأ هرشع ةدمل مجك

ديرولا

قيرط

نع اهنقح

مت مث

ةيناثلا

ةعومجملا

يف امك

ليساري ورولف-5

تقلت

ةيحضتلا مت

.نارئفلا

ليذ يف

ةدحاو

ةرم

## هطيسولا

هيعذجلا ايلاخلا

نم 106

## ةطساوب

، ايً رهجم اهصحف متيل ، ةبرجتلا ةيادب نم نيثلاثلاو سماخلا مويلا يف نارئفلا عيمجب

جاتنإ

مييقتلو

نيسويلااو نيلسكوتاميهلا

هغبص

ةطساوب

ةدعملا

ماسقأ

هغابص

متو

## ينورتكللإا

رهجملا

، PAS

لا هغبصـب

اهتغابص مت

يطاخملا

يركسلا

نيتوربلا

مت Ki67

مادختساب

ةيعانملا

ءايميكلاو

ةقئافلا

ةيلكيهلا

تارييغتلا

مييقتل

لاسرلإل

ةعومجملا

نم ةدعملا

ماسقأ

صحف

## رهظأ

.ايلاخلا

رثاكت

ىوتسم

مييقتل

اهمادختسا

ايلاخلا

تاغجفتو

، ةيومدلا

تاريعشلا

ناقتحاو

، ةيوعملا

ددغلا

يف ًلااصفنا

ةيناثلا

بيكرتلا

يف اظً

وحلم

انً سحت

هعبارلاو هثلاثلا هعومجملا ت

رهظأ

امنيب

، ةيرادجلا

معد و هيناثلا هعومجملاب ةنراقم ،

Ki67ريبعت يف امً

ظنم اضً

افخناو ةدعملل يجيسنلا

.جئاتنلا هذه ينورتكللإا رهجملا

امهل

نيثنازاتسلاا

راقع و

هطيسولا هيعذجلا ايلاخلا

نم لك

نأ جئاتنلا تتبثا دقو

يف ليساريورولف-5

نع مجانلا

ةدعملا

باهتللا

ماسلا

ريثأتلا

فيفخت

يف ديج

ريثأت

.نيتقيرطلا اتلك نيب ريبك قرف دوجو مدع عم ، ءاضيبلا ناذرجلا