

# MUCOSITIS PATHOLOGY IN CHEMOTHERAPY INDUCED DIGESTIVE TOXICITY

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**ABSTRACT.** Cytostatic chemotherapy can produce toxicity in the digestive tract at standard doses, in regimens given for various malignancies. Mucositis or gastrointestinal mucosal chemotherapy toxicity decreases the oncology patient's quality of life and often leads to chemotherapy dose-reduction or even its discontinuation. This paper has two objectives: to emphasize a synthesis of data from the literature on the manifestation and pathogenesis of mucositis and, secondly, to present a study on the clinical mucositis on a group of patients receiving chemotherapy. The results showed that 61% of applications of chemotherapy are accompanied by digestive toxicity and each patient has at least one episode of nausea during cytostatic therapy. Understanding the mechanisms of mucositis allow the clinician to have a complex approach to the oncological patient receiving chemotherapy and find adjuvant substances that prevent or reduce this type of toxicity.

**KEYWORDS:** mucositis, chemotherapy, gastrointestinal tract, intestinal toxicity, histopathology

## INTRODUCTION

Normal digestive function is a balance between oral ingestion, secretions of the digestive tract, reabsorption and metabolism. Chemotherapy generally refers to antineoplastic therapy using cytotoxic drugs in order to eliminate the neoplastic cells formed in an individual organism. The cancers are varied in terms of pathogenesis, malignancy grade, so the chemotherapeutic regimens are multiple and are administered according to international standards and protocols. Besides chemotherapy, antineoplastic treatment can associate radiation therapy, immunological therapy, hormonal therapy and other novel therapies introduced in medical practice in recent decades. The routes for cytostatics administration are varied, single or multiple in the same patient: oral, intravenous (perfusion), intraarterial, intraperitoneal, intrathecal, intrapleural, topically local, intramuscular, subcutaneous, intravesical, intralesional (within the tumor). Chemotherapy is indicated in the treatment of malignancies as induction therapy, consolidation / intensification therapy, adjuvant chemotherapy, neoadjuvant chemotherapy, maintenance therapy, salvage therapy, palliative therapy (Wintrobe, 2014).

This paper proposes two research objectives: first, a synthesis of data from the literature on the manifestation and pathogenesis of mucositis and, secondly, to present a study on the clinical mucositis in a group of patients receiving chemotherapy.

## MATERIAL AND METHODS

I. Collecting of data on mucositis from the literature was carried out over a 6 month period between January and June 2016; there were analyzed a total of 150 articles found in electronic archives by accessing Anelis-Plus.

II. A total of 182 applications of chemotherapy were included in the study, from a total of 30 patients followed in the Hematology Clinic, Emergency Clinical County Hospital of Arad. Data were taken from the patients's records, a retrospective and prospective study, between January-June 2016.

The patients included in the study had the following characteristics: age over 18 years, diagnosis of non-Hodgkin's lymphoma or Hodgkin's disease, chemotherapy type CHOP (cyclophosphamide, epirubicin, vincristine, prednisone), R-CHOP (rituximab-

CHOP) or ABVD (epirubicin, bleomycin, vinblastine, dacarbazine).

Patient's data included description of early symptoms or signs of digestive toxicity, namely: nausea, vomiting, diarrhea.

## RESULTS AND DISCUSSION

The symptoms of nausea were observed in 112 (61%) of chemotherapy applications from 30 patients (all patients), grade 2-3 (according to CTCAE, see below). The frequency of nausea was higher in the first cycle of therapy; ABVD therapy induced nausea in every application. Each patient had at least one episode of nausea during any chemotherapy. Regarding chemotherapy induced diarrhea, it was recorded in 10% of patients, grade 1-2.

Chemotherapy are drugs having a direct cytotoxic activity, so any normal tissue from the body is potentially vulnerable and can be affected, earlier or later. The toxic effects of cytostatics are classified into groups depending on their severity in Common Terminology Criteria for Adverse Events (CTCAE), a classification made and updated by the National Institute of Health and the National Cancer Institute of USA (CTCAE, 2009). Thus, toxicity is grouped as mild (grade I), moderate (grade 2), severe (Grade 3) and life-threatening (grade 4), based on specific parameters, depending on the organ involved. Toxicity type depends on the chemotherapy protocol used and the patient's individual susceptibility (Nie et al., 2013; Huri et al., 2010).

Chemotherapy can cause structural or functional changes in the digestive tract (Keefe et al., 2000; Gibson et al., 2002; Cunningham et al., 1985), due to the lack of high selectivity on tumor tissue. The antineoplastic drug can't discriminate between cancer cells and cells of rapidly dividing normal tissue, such as digestive epithelial tissue (Boussios et al., 2012). The modality by which the chemotherapy drugs act is through cellular apoptosis.

The volume of distribution of lipo-soluble drugs may increase in a patient having an increased total body fat mass, and other changes, such as decreasing the amount of total body water and low concentrations of plasma protein or hemoglobin can reduce the volume of distribution for hydrophilic drugs and therefore the plasma drug levels can increase (Beijnen et al., 2004).

Mucositis, starting from oral cavity and pharynx to the colon is a complication due to the toxic and non-selective effect of chemotherapy, on the epithelial cells (Keefe, 2004; Duncan et al., 2003). It is believed that mucositis is a unitary pathology in the entire alimentary tract, so it is also called alimentary mucositis.

Common risks for alimentary mucositis fall into two categories: therapy-related and patient-related. Variables associated with therapy include: the drug itself (anthracyclines are frequently involved in the occurrence

of oral mucositis) and the chemotherapeutic regimen, concomitant use of other therapies, type of radiation therapy associated. Chemotherapy drugs don't have an identical risk for mucositis, there is even some dispute about the toxicity of some chemotherapy drugs. Patient-related risks include: age, body weight, body mass, kidney and liver function, local oral factors (the hygiene and integrity of mouth mucosa) and pharmacogenetics (Gibson et al., 2008). It has been found that elevated serum levels of tumor necrosis factor alpha (TNF) and interleukins 1 and 6 correlate to the degree of non-hematologic toxicity after chemotherapy. Similarly, the mucous concentration of interleukin 1 and TNF gene expression are associated with the development of mucositis in the animal model (Sonis et al., 2004). Gastrointestinal toxicities and infections by leukopenia, were linked by Kishi to certain polymorphisms of genes involved in tumor development (Kishi et al., 2007). Physiological changes that may increase the toxicity of chemotherapy are low reserves of stem cells, decreased ability to repair cellular injuries, progressive loss of body protein, body fat accumulation. Some research tried to identify predictive biomarkers for chemotherapy induced gastrointestinal toxicity, but without consistent results (Rosmarin et al., 2014; Boige et al., 2010; McLeod et al., 2010; Simon, 2013). A study published in 2007 in the *Blood* journal showed the link between genetic polymorphisms and susceptibility to gastrointestinal toxicity after chemotherapy. The authors described genetic polymorphisms for vitamin D receptor, cytochrome P4503A5 or folate transporter, which may increase the risk of gastrointestinal toxicity during chemotherapy in leukemia patients (Kishi et al., 2007).

Early digestive toxicity is clinically manifested by nausea and vomiting, which can have an early onset 24 hours after cytostatics administration, later at 24 interval or even anticipatory, before chemotherapy. The pathophysiologic mechanism is initiated by chemotherapy through serotonin release from the enterochromaffin cells of the digestive tract. Through specific receptors, serotonin further stimulates the nervous pathways up to the brain emesis center. The type of chemotherapy, the dose and the sequence of administration have an impact on the severity and risk of emesis. The emetogenic potential of cytostatics is quantified into a scale of intensity (Freter et al., 2008). Loss of appetite, changes in smell may occur early after administration of cytostatics, single or multi-agent chemotherapy. All symptoms occur as a result of injuries caused by cytostatic agents at different levels (Duncan and Grant, 2003).

Chemotherapy drugs studied as having a potential gastrointestinal toxicity are taxanes, platinum salts, anthracyclines, fluoropyrimidines, alkylating agents, topoisomerase inhibitors. Taxanes are associated with colitis, especially ischemic ones, having severe abdominal pain, diarrhea and neutropenia. Of platinum salts, cisplatin causes acute and delayed severe emesis,

diarrhea, increased serum transaminases. Carboplatin can also cause vomiting, diarrhea, constipation, abdominal pain. Oxaliplatin produces nausea, vomiting, abdominal pain, stomatitis, anorexia, xerostomia, rectal hemorrhage, proctitis, rectal tenesmus, pancreatitis, hepatic sinusoids dilatation, colitis. Regarding the mitotic inhibitors, the most common digestive toxicity consists of nausea and vomiting in 20-85% of patients (Boussios et al., 2012). Stomatitis occurs in up to 80% of patients and is dose-related (Wintrobe, 2014). Ulceration of the esophagus or the colon, diarrhea, anorexia can also occur. Antimetabolites produce severe digestive toxic events such as stomatitis, esofago-pharyngitis, diarrhea, anorexia, nausea, vomiting, mucositis, especially oral mucositis, abdominal pain. Capecitabine may cause liver failure. 6-mercaptopurine can cause intestinal ulcers, diarrhea, pancreatitis. Cytarabine produces oral and anal mucositis, with ulceration and inflammation, intestinal necrosis, hepatic dysfunction. Gemcitabine is associated with elevated transaminases. Methotrexate frequently causes mouth ulcerations and diarrhea, vomiting, with subsequent dehydration. Other effects are gingivitis, stomatitis, pharyngitis, digestive hemorrhage, gastrointestinal ulcers, less frequent are the colitis or toxic megacolon. Alkylating agents cause nausea and vomiting, anorexia, diarrhea, mouth ulcers, rarely hepatitis or veno-occlusive disease. Cyclophosphamide may be associated with hemorrhagic colitis. Nitrozureas produce nausea and vomiting, liver dysfunction. Dacarbazine produce early emesis, anorexia, procarbazine can cause granulomatous hepatitis. The anthracyclins produce nausea, vomiting, anorexia, diarrhea, esophageal and colon ulcerations (Hoffman, 2013).

Other clinical manifestations of chemotherapy digestive toxicity can be: abdominal distension, early satiety, dyspepsia, dysphagia, cheilitis, gastric hemorrhage, gastric necrosis, gastritis, gastroesophageal reflux, gastrointestinal pain, gastroparesis, enterocolitis, flatulence, ileus, malabsorption, pancreatic necrosis, colic bleeding (colon), rectal necrosis, rectal pain, rectal ulceration, typhlitis, liver failure, hepatic necrosis, hepatic hemorrhage and portal hypertension. Diarrhea induced by cytostatics administration, particularly irinotecan, fluorouracil, capecitabine or oxaliplatin, as single agents or combination regimens, is a common complication in cancer patients, with an incidence between 40-80% (Wintrobe, 2014). Pathophysiology of diarrhea after chemotherapy is complex, it can have many underlying mechanisms: secretory, osmotic, malabsorption, exudative and by dysmotility (Engelking et al., 1998). There can also be other causes: inflammatory causes, infectious causes or steatorrhea. Diarrhea is due to alterations in the absorption function of intestinal cells, changes in goblet cells, changes in the composition and distribution of mucin, interaction of bacteria with these cells (Stringer et al., 2009) as well as by toxic metabolites produced by the degradation of

cytostatics (Ikuno and et al, 1995; Takasuna et al, 2006). Because of the alteration of normal absorption function, patients with mucositis often have weight loss and malnutrition. Keefe et al have shown that therapy with irinotecan in rats induces diarrhea as early as 2 hours post-chemotherapy in 23% of treated rats, the highest prevalence was reached in 44% of the animals at 24 hours (Al-Dasooqi et al., 2010). The mechanism that causes postchemotherapy constipation is not completely defined, it is considered to be frequently due to secondary adjuvant antiemetic or opioid medication (Boussios et al., 2012). Abdominal pain is caused by extensive damage in the abdomen as a whole.

One of the most common questions in clinical practice is: how can we know which patient will develop mucositis? The question is very pertinent because mucositis is one of the reasons most frequently invoked in postponing the chemotherapy, in chemotherapy dose reduction or even discontinuation of therapy in oncologic patient. All these carry a risk in reducing the percentage of disease remission, or the patient survival scores.

Mucositis research methods are complex, referring not only to structural changes, but also to genetic changes, by searching for certain suppressed or activated genes in the patient receiving chemotherapy.

Mucositis pathogenesis is complex. If 20 years ago it was believed that the mechanism of mucositis was a singular one, through direct toxicity on the target cell, modern research revealed complex mechanisms for alimentary tract injuries - a multifactorial process and a cascade of events in multiple tissue structures (Al-Dasooqi et al., 2011). The pathogenesis of the alimentary tract mucositis is considered to be the same throughout the gastrointestinal tract (Stringer et al., 2009), although each segment of digestive tract has its own anatomical, morphological and functional particularities.

Sonis et al have described five stages for mucositis mechanism: initiation, regulation with mediators generation, signaling and amplification, ulceration and inflammation, and finally healing (Sonis et al., 2004). The first phase in the onset of mucositis occurs rapidly after administration of cytostatic and is activated by the simultaneous effects of deoxyribonucleic acid (DNA) and non DNA damage within the epithelial and submucosal cells and also by generation of reactive oxygen species (ROS). Overexpression and message generation phase is characterized by breaking the DNA strands and the activation, through ROS, of multiple transcription factors, including p53 and NF- $\kappa$ B (nuclear factor kappa-light-chain-enhancer of activated B cells). They in turn regulate a great number of genes involved in mucous toxicity. Changes in non-DNA injury occur in parallel, including production of ceramide and matrix metalloproteinases. All these lead to apoptosis and tissue destruction. The third phase, of signaling and amplification follows shortly thereafter. Following activation of transcription factors, the proinflammatory

cytokines accumulate and target submucosal tissues; these injuries also act as a positive feedback signal to amplify this reaction. Furthermore, certain cytokines can also activate initial transcription factors and increase overexpression of metalloproteinases and ceramides. Ulceration phase is considered to be the most clinically significant in mucositis, and is characterized by a loss of mucosal integrity with superficial bacterial colonization of the new lesions. This results in the activation of inflammatory mononuclear cells that release additional proinflammatory cytokines and in pro-apoptotic genes overexpression, leading to tissue damage. Increased activity of the transcription factor and the high levels of cytokines and other mediators lead to local responses, including angiogenesis. Finally, healing starts once the healthy epithelium migrates from the lesion edges, according to signals coming from the submucosa (Rubenstein et al., 2004). Generally, mucositis heals after the completion of antineoplastic therapy (Gibson, Bowen, Keefe, 2008).

Research on apoptosis became a priority in the assessment of mucositis pathogenic pathways. Thus, by using apoptosis markers by staining with specific antibodies in immunohistochemistry, studies have shown that apoptosis is increased in the basal layer in the first 3 days post chemotherapy, begins to decline in the next 6 days but the normal level is not reached until 11 days after chemotherapy. Apoptosis precedes the intestinal crypt hypoplasia in cancer patients receiving chemotherapy, these initial changes consist of decreased morphometric size of the crypt and decreased rate of proliferation, without mucosal destruction (Keefe, Brealey et al., 2000).

An interesting aspect was observed in patients previously treated with chemotherapy, namely, the level of apoptosis in these patients was lower on days 1 and 2 post-chemotherapy compared to biopsies that were taken prechemotherapy (which were taken on day 21 of the previous therapy cycle) (Gibson et al., 2006).

The main mediator in apoptosis is a proteolytic system involving a family of cysteinyl proteases called caspases. The initiation of apoptotic cascade is also made by chemotherapeutic agents, and leads to caspase-dependent cleavage of a set of regulatory proteins, to cellular DNA damage and complete disintegration of the cell. Changes in apoptosis were observed both in patients who had symptoms of mucositis, and also in those who were asymptomatic (Gibson et al., 2006).

The role of metalloproteinases (MMP) was also studied as possible key mediators in chemotherapy induced mucositis (increase in 2 and 3 metalloproteinases has been associated with inflammatory infiltrate and severe tissue damage) (Al-Dasooqi, Gibson et al., 2010). Sonis's study on metalloproteinases involvement revealed that they can have a role in recruiting inflammatory cells and immune mediators. They also can initiate the mucosa damage by disrupting interactions between cells and between cells and the extracellular

matrix. The highest histological mucosa injury level within the jejunum and colon was associated with elevated levels of MMP-2, MMP-3, MMP-9 and MMP-12 (Al-Azri et al., 2012). These are produced by the leukocytes infiltrating the tissue to facilitate their transendothelial migration to mucosa (Al-Dasooqi, Gibson et al., 2010).

Oral mucositis is a common complication of chemotherapy (Wang et al., 2015), which may occur 5-14 days after chemotherapy and can last for 7-14 days (Di Palma et al., 2009), while the intestinal mucositis has a peak at 3-7 days after chemotherapy. Most commonly it manifests as stomatitis, with erosive and ulcerative oral lesions, but it can also manifest as altered taste (Kalaskar et al., 2014) or xerostomia. The symptoms most frequently described by the patients are oral pain, oral mucosal enantema, difficulty in opening the mouth, in eating, in talking, cheilitis. Assessing the lesion degree can be done according to two scales, the most used being the Oral Toxicity Scale WHO (World Health Organisation) (Di Palma et al., 2009). Through its manifestations and the infectious complications it can induce, oral mucositis has a significant impact on cancer patient quality of life (Bressana et al., 2016). Mucositis also alters the nutritional status of the patient receiving chemotherapy (Jin Won Kim et al., 2012).

Within the oral cavity, being the mucositis site most easily observed and investigated, there are numerous studies that described changes at the macroscopic level to the intimate, ultrastructural level, by electron microscopy. Optical microscopy revealed an aspect of mild to moderate chronic inflammation. Ultrastructural changes include increased number of intercellular fibers within the basal cellular layer, cytoplasmic vacuolisation, loss of membrane contact with neighboring cells, multinucleated cells in the suprabasal layer, loss of cell cytoplasm. These changes persist in pre-chemotherapy biopsies taken from patients who had previously received cytostatic therapy. Increasing in the intercellular filaments as a result of cytotoxic injury can be attributed to a cellular attempt to "protect" themselves against injury by increasing the structural support (Gibson et al., 2006). The ultrastructural changes were seen in early stages of apoptosis, including nuclear separation and pyknotic nuclei on days 1 and 2 post-chemotherapy (Gibson et al., 2006).

The changes of the esophagus include abnormalities of dividing and proliferating cells, leading to epithelial thinning and ulceration. Chemotherapy also affects the rate of connective tissue cell proliferation within the lamina propria, leading to increased vascular permeability and inflammatory infiltrate (Sonis et al., 2004).

Concerning research on mucositis in the stomach, fewer data exist compared with those related to oral cavity or small intestine. This segment is also involved in cytostatics digestive toxicity, the first

evidence being the nausea and vomiting occurring early after administration of certain cytostatics (Triantafyllou et al., 2008).

Histopathological changes associated with alimentary tract mucositis are well described. Research studies on small intestine chemotherapy toxicity were conducted mainly on the jejunum. Mucosal barrier is affected by epithelial cell injury, with flattening of the villi and exposure of mucosa to luminal contents. Mucositis changes have been observed in the jejunum and colon to 6 hours post irinotecan chemotherapy. Other studies have shown that the flattening of villi occurred in the jejunum at 48 hours after chemotherapy, at 72 hours it has been described the complete ablation of crypts in the colon, and after 144 hours the restitution takes place in both segments of the digestive tract (Al-Dasooqi, Gibson et al., 2010). In clinical trials, Keefe et al have conducted research on patients receiving chemotherapy with sequential duodenal pre and post therapy biopsies (Keefe et al., 2000). Anticancer drugs act at different levels of intestinal crypt (Ijiri and Potten 1983, Potten and Ijiri, 1987). The injury outcome consists of intestinal crypt hypoplasia followed by crypt rebound hyperplasia (after the 5<sup>th</sup> day), then by restoration of normal epithelium (Keefe et al, 2000; Gibson et al, 2003; Keefe et al, 2004). Duncan and Grant have also noticed that cytostatic chemotherapy produces direct toxic effects on proliferating epithelial cells from crypts, resulting in destruction of the mucosal barrier (Duncan and Grant, 2003).

In the small intestine, the stem cells are most likely found in positions 3-5, in the colon they are in positions 1-2. Potten et al. proved that at small intestine level, apoptosis tends to be highest in positions 3-5, thus destroying stem cells. Apoptosis in the colon also acts at the level 3-5, thus sparing the stem cells (Keefe, Brealey et al., 2000). After chemotherapy, epithelial stem cells within the small intestine are altered and therefore no longer divide or no longer differentiate toward specific cell lines (Inomata et al, 2002; Gibson et al, 2003). So the cell regeneration becomes abnormal, vilous mucosa is not renewed, which quickly leads to loss of normal structure and functionality (Duncan and Grant, 2003).

The postchemotherapy structural changes also include the presence of an intense inflammatory infiltrate within the mucosa, seen at 48-72 hours after chemotherapy (Al-Dasooqi, Gibson et al., 2010). Another type of tissue injury in mucositis is the damage of submucosal vessels (Al-Dasooqi, Bowen et al., 2011).

Rubenstein et al. observed that the first histological effect at small intestine level is the increase in apoptosis, by 7 times in the intestinal crypts during the first day after chemotherapy (Rubenstein et al., 2004). The immediate consequences were reduction in villi area, the crypts length and the mitotic index, with the highest intensity at 3 days after chemotherapy. Studies by Keefe have shown that in the small intestine there are two peaks of apoptosis, one early after chemotherapy corresponding

to the top cellular injury and the second after healing has started, in order to correct cell proliferation so that the tissue returns to equilibrium (Gibson, Cummings et al., 2006).

Research on rats showed that Bak and Bax (pro-apoptotic markers belonging to Bcl-2 family) are increased in the small intestine after chemotherapy (Bowen et al, 2005). Research studies have shown that the ratio between pro-apoptotic and anti-apoptotic genes of bcl-2 family change along the gastrointestinal tract. This ratio is higher in the small intestine than the colon, which may help to explain the differences in the occurrence of mucositis in the two segments (Sonis, Elting et al., 2004). Studies on laboratory animals treated with doxorubicin showed the significant induction of proapoptotic Bad protein expression, the decreasing of antiapoptotic factor Bcl-2 and of the mitochondrial membrane potential, the increase of cytochrome c and the increase in caspase 9 and 3 cleavage (Das et al., 2012).

Gobson reported that small intestine goblet cell doesn't seem affected by chemotherapy (Gibson et al, 2003).

Studies in rats treated with methotrexate revealed an increased production of ROSs and of myeloperoxidase activity in rats small intestine (Maeda et al., 2010). This study demonstrated an increase in paracellular permeability, process started with the augmented production of ROSs. This cytostatic drug can alter intestinal barrier function due to inhibition of cell proliferation, after the invasion of xenobiotics and endotoxins. Consequently, phagocytes such as macrophages and neutrophils infiltrate the tissue and produce ROSs. Given the context of mucositis mechanisms, the synthesis of ROSs by chemotherapy is the first step that leads to intestinal inflammation. It has been suggested that ROSs can activate the nuclear factor NF- $\kappa$ B, which is a transcription factor that stimulates the inflammatory cytokines and adhesion molecules. ROSs in low concentrations can modulate cell function and act as transmission signals for apoptosis, cell proliferation, cell adhesion, and chemotaxis by regulating the nuclear transcription factors. Doxorubicin can lower the activity of antioxidant enzymes (Das et al., 2012).

A feature of the development of mucositis as a result of primary tissue injury is the initiation of an acute inflammatory response. Chemotherapy induced mucositis is associated with overexpression of stress genes and activation of signaling mitogen activated protein kinase (MAPK), activation of NF- $\kappa$ B (Al-Dasooqi, Gibson et al., 2010).

Studies on laboratory animals post administration of methotrexate demonstrated the opening of tight junctions by ROSs. The barrier function of tight junctions is maintained during transepithelial neutrophil migration, but migration of an increased population of neutrophils can damage the integrity of the epithelial cells. Paracellular epithelial permeability is altered by

changes in occludin and ZO-1, which compose the tight junctions (Maeda et al., 2010).

It has been shown that systemic administration of chemotherapy also produces changes in the colon. Crypt cells undergo intense apoptosis, causing crypt hypoplasia (Keefe et al., 2004). Lamina propria becomes infiltrated with mononuclear cells and an increased number of eosinophils. Glandular and surface epithelium loses its normal columnar morphology and becomes cuboid (Verburg et al, 2000). Comparing to the changes that occur in the small intestine, the colon goblet cells undergo important changes, with increased mucus production. As more severe architectural changes occur, the number of goblet cells decreases within the areas of crypt severe hypoplasia (Al-Dasooqi, Gibson et al., 2010), but the amount of mucus secreted is higher (Gibson et al, 2003).

Extracellular matrix components play a vital role in maintaining the integrity of the mucosal barrier by regulating cell apoptosis, proliferation and differentiation of upper epithelial cells. Special staining can show an important increase in collagen and other components of the basal membrane around the crypts after 24 hours post chemotherapy. After irinotecan administration, a significant alteration occurs in the adhesion protein expression both in the jejunum and colon, with direct effect on the mucous layer integrity loss. It has been found that the cytostatics can halve the jejunum and colon cell proliferation after 48 hours and 24 hours post therapy, respectively. Significant impairment of jejunum and colon epithelial cell kinetics after administration of irinotecan was correlated with altered expression of components of the extracellular matrix, which can lead to loss of integrity of the mucosal barrier in mucositis (Al-Dasooqi, Bowen, Gibson et al., 2011). After chemotherapy injury, collagen increase plays a role in cell migration along the crypt in the small intestine and colon. Each extracellular matrix protein has a distinct function in cell kinetics and therefore their spatial organization is essential to maintaining the integrity of the mucosal layer. This spatial expression is regulated by metalloproteinases, in order to ensure an effective turnover of gastrointestinal tissue (Al-Dasooqi, Bowen et al., 2011).

Studies in rats treated with methotrexate have revealed severe mucositis, with deep vilous atrophy, epithelial damage, increasing levels of mucosal myeloperoxidase (41.5 times) and decrease of citrullinemia (6.6 times) compared to the control group. Permeability of the intestine was also modified (Russo et al., 2013). Functional changes such as permeability for sugars persists after symptoms and morphological changes resolution (Rubenstein et al., 2004; Keefe et al., 1997). Changing of intestinal permeability allows the mucositis be diagnosed by the sucrose test (Tooley et al., 2010).

Mucosal barrier damage and ulcerations in mucositis lead to systemic infections in over 70% of

patients receiving myeloablative or myelosuppressive therapy. Changes in the composition of microflora lead to alterations in intestinal absorption and other intestinal functions. Stringer et al. published data on alteration of the microflora of the stomach, small intestine and faeces after treatment with 5-fluorouracil in mice (Stringer et al., 2009). This study also revealed intestinal bacteria translocation towards mesenteric lymph nodes, increasing the risk of secondary infections (Stringer et al., 2009). Changes in intestinal microflora after chemotherapy can lead to failure of its functions, namely the mucosa protection, metabolism of internal biological products, nutrient processing, regulation of intestinal angiogenesis, immune functions. Susceptibility to mucosal injury in mucositis is based on certain specific tissue components, including local microbial environment (Stringer et al., 2009). Severe intestinal mucositis with citrullinemia <10 micromol / L defines the period of risk for bacteremia better than neutropenia in patients after chemotherapy (Tooley et al., 2009; Herbers et al., 2014).

Mechanisms underlying mucositis are complex and follow or parallel each other and culminate in mucosal injury.

## CONCLUSIONS

Mucositis is a common complication of chemotherapy, which dramatically reduces the quality of life of cancer patients. Each segment of the digestive tract can be affected, and mucositis mechanisms are similar, despite the morphological or structural differences between different segments of the alimentary tract. Mucositis pathogenesis is complex, involving events taking place in various tissues and time sequences. Apoptosis, inflammation and production of reactive oxygen species, changes in extracellular matrix components have multiple local consequences: impaired cell proliferation, cell metabolic dysfunction, cell death, local inflammatory infiltration. These processes generally end with injury healing, yet studies show that patient receiving chemotherapy, though not all patients have mucositis symptoms, may present structural changes even 21 days after chemotherapy. Therefore, the need to develop therapeutic strategies to prevent or improve mucositis is mandatory for medical practice.

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