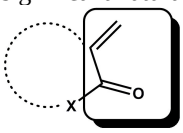
Cancer is the leading cause of death in developed countries. The search for novel drug candidates is a priority goal for cancer therapy. Plants have always been an excellent source of biologically active agents used in traditional medicine and continue to be viewed as major sources for the development of new anticancer drugs. On the other hand, natural products isolated from plants are often used as valuable leads for the synthesis of analogs with improved pharmacological properties. A vast number of biologically significant natural products is characterized by an exo-methylene moiety conjugated with a carbonyl group (Fig. 1).



X = O, NR

Fig. 1. Structure of exo-methylene moiety conjugated with a carbonyl group.

Such products are abundant in plants of the Asteraceae (Compositae) family and have a broad spectrum of biological activities, ranging from antiinflammatory, allergenic, phytotoxic, antibacterial and antifungal to cytotoxic and/or anticancer. The most common class of compounds with such moiety are α-methylene-γ-lactones (1), but α-methylene-δ-lactones (2) and α-methylene-γ- and δ-lactams (3, 4 respectively) are also found in the nature (Fig. 2).

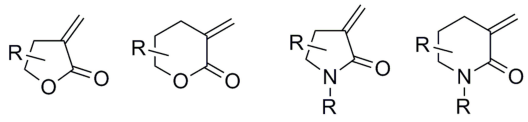


FIg. 2. General structures of α-methylene-γ- and -δ-lactones and lactams.

Compounds containing an exocyclic methylene moiety conjugated with a carbonyl group which can react via the Michael-type reaction with mercaptyl groups in cysteine residues of enzymes, other functional proteins, and free intracellular glutathione, leading to the formation of covalent adducts. Such alkylation of cellular thiols may interfere with key biological processes, leading to the controlled cell death, apoptosis.

The best known and the most studied representative of the compounds with exo-methylene motif conjugated to the carbonyl group is parthenolide, isolated from feverfew (Tanacetum parthenium). High cytotoxicity of this compound was proven in various cancer cell lines. Water soluble derivative of parthenolide, dimetyloaminopartenolid (DMAPT) has already entered clinical trials in the United Kingdom for treatment of leukemia. At the molecular level, the cytotoxic action parthenolide involves different signaling pathways. However, the main mechanism of its action is associated with the inhibition of the transcription factor nuclear factor NF-κB activity. Constitutive activation of NF-κB or its induction in a response on chemotherapeutics is the major reason of the drug resistance development in many types of cancer. A new promising paradigm for anticancer therapy is combination chemotherapy, involving at least two drugs that work by different mechanisms, thereby decreasing the likelihood that resistant cancer cells will develop. Due to pleiotropic action, parthenolide recently often draws attention to its potential new use in combination therapy as a factor sensitizing cancer cells to chemo- and radiotherapy.

In the Department of Biomolecular Chemistry for some time now we have been investigating anticancer activity and structure-activity relationships of heterocycles containing exo-methylene group conjugated with a carbonyl, synthesized at the Institute of Organic Chemistry, Technical University of Lodz.

Screening experiments (MTT assay) performed by our team on the breast cancer cell lines (MCF-7, MDA-MB-231), and leukemia cell lines (HL-60, NALM-6), enabled us to select compounds with the highest cytotoxicity. Further, more detailed experiments showed that these compounds induce apoptosis, inhibit migration of cancer cells and decrease levels of the markers responsible for cancer metastasis.

The aim of this study is to investigate the anticancer activity of two series of new synthetic α-methylene-δ-lactones, synthesized at the Institute of Organic Chemistry, Technical University of Lodz. First, in order to select compounds with the best therapeutic index, the screening experiments will be performed on two cancer lines (MCF-7 and HL-60), as well as on two non-cancerous cell lines (HUVEC and MCF-10Afor comparison. Then, the anticancer activity of selected compounds will be evaluated. The specific aims of this part will include: evaluation of the effects of these compounds on cell proliferation and cell cycle regulation, induction of apoptosis, oxidative stress and DNA damage, changes in mitochondrial membrane potential and GSH/GSSG ratio., and tumor suppressor p53 and transcription nuclear factor NF-κB activity.The last step of this project will be will be evaluation of possible synergistic activity of α-methylene-δ-lactones, used in combination with well-known anticancer drugs, that act through different mechanisms. Combined experiments with use of α-methylene-δ-lactones and 5-fluorouracil (5-FU), taxol (Tx) and oxaliplatin (Ox) or X-irradiation will be performed. We want to check if α-methylene-δ-lactones can sensitize cancer cells on chemotherapeutics.

This is a basic science study with possibly high significance of outcomes. The obtained results will increase our understanding of the molecular mechanisms by which compounds with α-methylene-δ-lactone motif can exert their anticancer activity. The project might result in finding an analog with a remarkable pharmacological profile and help direct future structure-activity relationship studies. Thus, it will have an impact on academic research and development. In further perspective the project might result in finding an analog with potential anticancer activity, that can be used alone or in combination therapy to potentiate effect of well-known anti-cancer drugs and may draw attention of pharmaceutical industry. Thus the project may also have measurable practical aspect.