Publications


Role of cytokines and Jak3/Stat3 signaling in the 1,2-dimethylhydrazine dihydrochloride-induced rat model of colon carcinogenesis: early target in the anticancer strategy.
Saini MK, Vaish V, Sanyal SN.

Source
Department of Biophysics, Panjab University, Chandigarh, India.

Abstract
The molecular mechanisms by which colon cancer cells regulate the expression of various proinflammatory and anti-inflammatory cytokines and transcription factors resulting in tumor progression have not been well clarified. The present study thus explores the effect of cancer cell-derived cytokines and transcription factors on the chemoprevention of a rat model of early colon carcinogenesis. Elevated expression of proinflammatory cytokines [interleukin-1β (IL-1β), IL-2, interferon γ, and tumor necrosis factor-α] and the transcription factors [Janus kinase 3 (Jak3) and signal transducer and activator of transcription 3 (Stat3)] was found in the 1,2-dimethylhydrazine dihydrochloride (DMH) group; however, this elevated expression was reversed by the individual and combination treatment with piroxicam, a traditional nonsteroidal anti-inflammatory drug [inhibiting both cyclooxygenase-1 (COX-1) and COX-2] and c-phycocyanin, a cyanobacterium-derived biliprotein from Spirulina platensis (selective COX-2 inhibitor). In the DMH group, low expression of IL-4, an anti-inflammatory cytokine, was further observed with respect to the other groups. Expression of inducible nitric oxide synthase and nitric oxide/citrulline levels was also analyzed and was found to be elevated with DMH treatment. Increased apoptotic index and stimulated levels of Bcl-2-associated death promoter (Bad), a proapoptotic protein, were observed in piroxicam-treated and c-phycocyanin-treated rats. In-silico molecular docking of piroxicam as a ligand with several regulatory proteins was performed, indicating that, except inducible nitric oxide synthase, it effectively binds with COX-1, COX-2, Jak3, and Stat3. Piroxicam and c-phycocyanin perhaps showed chemopreventive properties by inhibiting proinflammatory cytokines and Jak3/Stat3 signaling while promoting apoptosis. In addition, a combination regimen was found to be more beneficial than monotherapy.
Piroxicam and C-Phycocyanin Mediated Apoptosis in 1,2-Dimethylhydrazine Dihydrochloride Induced Colon Carcinogenesis: Exploring the Mitochondrial Pathway

Manpreet Kaur Saini and Sankar Nath Sanyal
Department of Biophysics, Panjab University, Chandigarh, India

Kim Vaiphei
Department of Histopathology, Post Graduate Institute of Medical Education and Research, Chandigarh, India

INTRODUCTION

Apoptosis is a synchronized procedure of cell death that is regulated by caspases and proapoptotic proteins. During apoptosis, translocation of cytochrome c, an electron carrier, from mitochondria into the cytosol is regulated by Bcl-2 family members. Cytochrome c in association with an apoptotic protein activating factor (ApaF), a proapoptotic protein essential for cell differentiation and procaspase-9 form the apoptosome complex, which consecutively activates effector caspase-3, and coordinate the implementation of apoptosis. In the current study, an attempt has been made to gain insight into piroxicam, a traditional non-steroidal antiinflammatory drug and c-phycoeyanin, a biliprotein from Spirulina platensis (cyanobacterium) mediated apoptosis in DMBH-induced colon cancer. Male Sprague-Dawley rats were segregated into 5 groups: control, DMH, DMH + piroxicam, DMH + c- phycoeyanin, and DMH + piroxicam + c-phycoeyanin. Results illustrated that piroxicam and c-phycoeyanin treatments stimulate cytochrome c release by downregulating the Bcl-2 (an antiapoptotic protein) expression significantly, while promoting the level of Bax (a proapoptotic protein), thereby activating caspases (caspases-9 and -3) and Apaf-1. The outcomes of the present study clearly signify that piroxicam and c-phycoeyanin mediate mitochondrial-dependent apoptosis in DMH-induced colon cancer. Moreover, apoptosis induction was more apparent in the combination regimen of piroxicam and c-phycoeyanin than the individual drugs alone.

Department of Biophysics. Panjab University. Chandigarh 160 014. India E-mail: sanyalpr@rediffmail.com

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Address correspondence to Sankar Nath Sanyal, Department of Biophysics, Panjab University, Chandigarh 160 014, India. E-mail: sanyalpr@rediffmail.com

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thus forms an inescapable end-effect in cancer cell killing in their apoptic interventions (2). Inflammation is the primary process in carcinogenesis, and among all the cell mediators in the development of cancer cells, prostaglandin E2 stands out as the most important proinflammatory molecule (3). Recent evidence has described that cyclooxygenase (COX) or prostaglandin H2 (PGH2) synthase is the key enzyme in the biosynthesis of the prostaglandins mediating inflammation and other important physiological processes (4). It is also stated that COX-2 (one of the inducible forms of COX) may modulate the cell proliferation and apoptosis mainly in solid tumors, including colorectal cancer. Further, a large body of evidence suggests that non-steroidal antiinflammatory drugs (NSAID) proved to be a very important chemical group in prevention of colon and other cancers and in NSAID-induced changes the induction of apoptosis is considered the main action mechanism of the antineoplastic effect, involving multiple signal transducing molecules (5,6). All the related events, cytochrome c release one of the important respiratory-chain proteins from mitochondria to the cytosol is a significant step and a hallmark of the cells undergoing apoptosis (7). Moreover, cytochrome c release can be regulated by pro- and antiprotective members of Bcl-2 family and in the execution of apoptosis, the activation of cysteine proteases (caspases) plays a pivotal role. In cytoplast, cytochrome c is known to become associated with caspase-9 and Apaf-1 (apoptotic protease activating factor 1) to form the apoptosome complex which in turn activates the effector caspase, caspase-3/-7.

Evidence has also illustrated that both proliferation and apoptosis are critical determinants of tumor growth (8). Thus, aberrant proliferation and impairment of apoptosis, with its defining morphologic and biochemical characteristics including caspase activation and nuclear fragmentation, are both critical to oncogenesis and pathogenesis of cancer (9). Consequently, in the current study, we investigated the role of piroxicam, a traditional NSAID, which is a dual inhibitor of COX-1 and COX-2
Chemoprevention of DMH-induced rat colon carcinoma initiation by combination administration of piroxicam and C-phycocyanin

Manpreet Kaui Saini · Kim Vaiphei · Sankar Nath Sanyal

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Abstract Cancer research illustrated that combinatorial studies can provide significant improvement in safety and effectiveness over the monotherapy regimens. A combination of two drugs may restrain precancerous colon polyps, opening a new possible opportunity for chemoprevention of colon cancer. In this context, chemopreventive efficacy of a combination regimen of C-phycocyanin, a biliprotein present in Spirulina platensis, a cyanobacterium, which is a selective cyclooxygenase-2 (COX-2) inhibitor and piroxicam, a traditional non-steroidal anti-inflammatory drug was considered in 1,2 dimethylhydrazine (DMH)-induced colon carcinogenesis in rats. Western blotting, immunohistochemistry, DNA fragmentation, fluorescent staining, PGE2 enzyme immunoassay, and carrageenan-induced paw edema test were performed along with morphological and histological analysis. DMH treatment showed a rich presence of preneoplastic lesions such as multiple plaque lesions, aberrant crypt foci, and well-characterized dysplasia. These features were reduced with piroxicam and C-phycocyanin administration. The number of apoptotic cells was featured prominently in all the groups compared with DMH. DMH treatment revealed intact high molecular weight genomic DNA with no signs of laddering/DNA fragmentation while it was noticeable significantly in control and DMH + piroxicam + C-phycocyanin. DMH group showed highest COX-2 expression and PGE2 level in comparison with other groups.

Keywords Apoptosis · COX-2 · C-phycocyanin · Piroxicam · PGE2

Introduction Colorectal cancer is a major cause of cancer death worldwide and one of the main features of colon cancer is the overexpression of the inducible enzyme cyclooxygenase-2 (COX-2) that catalyzes the first two steps in the biosynthesis of prostaglandins from arachidonic acid [1]. Evidence illustrated that COX has two isoforms, i.e., COX-1 and COX-2, where COX-1 is constitutively found in most tissue while COX-2 is an inducible early response gene product. Both these isoforms can synthesize prostaglandins but prostaglandins metabolized by COX-1 are mainly responsible for tissue homeostasis and protection of gastrointestinal lining, whereas those produced by COX-2 are associated with pain and inflammation [2]. Earlier studies from the present laboratory had clearly shown the antineoplastic effects of specific COX-2 inhibitors in the experimental colon cancer [3, 4]. Non-steroidal anti-inflammatory drugs (NSAIDs) are a class of compounds that block the prostaglandin generation particularly prostaglandin E2 (PGE2), the most abundant COX-2 metabolite in colorectal tumor tissue [5, 6]. In this context piroxicam, a traditional NSAID (NSAID) which is the non-selective
PTEN regulates apoptotic cell death through PI3-K/Akt/GSK3β signaling pathway in DMH induced early colon carcinogenesis in rat

Manpreet Kaur Saini, Sankar Nath Sanyal *
Department of Science, Health and Environment, Chandigarh, India

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ABSTRACT

PTEN, the most well-established target of NSAlDs, which catalyze the synthesis of prostaglandins from arachidonic acid (Tegeder et al., 2001; Williams et al., 1997). The enzyme COX exists in two isoforms, COX-1 which is constitutien to most of the tissues, COX-2 is known to be over expressed in many cancer types including colon (Tegeder et al., 2001). Moreover, natural chemopreventive agents such as c-phycocyanin and piroxicam, a traditional non steroidal anti-inflammatory drug and c-phycocyanin, a biliprotein from Spindino platensis (cyanobacterium) as the chemopreventive agents. Western blotting and immunofluorescence results indicated that the expression of PI3 K and Akt was promoted in the DMH group while least apoptotic cell death in this group as analysed by Hoechst 33342 and PI3-K and Akt expressions as compared to other groups. Piroxicam and c-phycocyanin treatment resulted significant apoptotic cell death while showing low PI3-K and Akt expressions. Microtubules and microfilaments, microtubules and microfilaments, 4G-8 labeling of calcein, and fluorescence intensity measurement of ROS level, were also analyzed showing the raised 3G, while reduced ROS levels in DMH group, however, piroxicam and c-phycocyanin treatment resulted in falling of Akt although both stimulated the ROS production as analysed by flow cytometry. The present study thus elucidated that piroxicam, a traditional NSAlD, and c-phycocyanin, a newly discovered COX 2 selective inhibitor, constitute irreplaceable chemopreventive targets in modulating apoptosis in the DMH induced early rat colon carcinogenesis via regulating PI3-K/Akt/GSK3β/PTEN signaling pathways further, a combination of these drugs provides a better therapeutic option than the monotherapy regimen.

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