ANTICANCER RESEARCH *40*: 4843-4856 (2020)
doi:10.21873/anticanres.14487

Review

Inhibitors of Fatty Acid Synthesis and Oxidation as Potential Anticancer Agents in Colorectal Cancer Treatment

PAULINA MOZOLEWSKA[1], KATARZYNA DUZOWSKA[1], ALICJA PAKIET[2], ADRIANA MIKA[1,2] and TOMASZ ŚLEDZIŃSKI[1]

[1]*Department of Pharmaceutical Biochemistry, Faculty of Pharmacy, Medical University of Gdansk, Gdansk, Poland;*
[2]*Department of Environmental Analytics, Faculty of Chemistry, University of Gdansk, Gdansk, Poland*

Abstract. *Aberrant fatty acid (FA) metabolism has long been recognized in colorectal cancer (CRC) cells. Since de novo lipogenesis is required for CRC tumour growth and survival, the inhibition of FA metabolism is a promising potential therapeutic target. Inhibition of the opposite process, β-oxidation of FAs, has also showed promising results in many CRC models. For patients with CRC, both FA synthesis and β-oxidation inhibitors are promising potential therapeutic options as monotherapies or as combination therapies with other anticancer agents. In this review, we discuss recent reports concerning inhibitors of FA synthesis and β-oxidation in various CRC models. The exact mechanisms of action of the selected compounds described in this review remain unknown and require precise evaluation before the development of new successful therapies for CRC is possible.*

Colorectal cancer (CRC) is among the most common types of cancer, accounting for approximately 10% of cancer incidence and mortality in both sexes (1, 2). Early detection owing to advanced screening methods (3, 4) has led to a reduction in death rates over the past several decades (5). However, some modifiable risk factors, such as poor diet and inactive lifestyle, are projected to contribute to increased incidence rates globally (5), especially affecting people in developing nations and underprivileged social classes (6, 7). Advances in understanding the pathophysiological alterations underlying this disease have led to a wide array of treatment options being available for the management of CRC. In addition to endoscopic and surgical excision, radiotherapy, and chemotherapy, drugs are used for the treatment of CRC and include biologicals and immunotherapeutics (7).

Recently, for the development of successful therapeutic strategies in the treatment of cancer, lipid metabolism has been considered an avenue worthy of pursuit (8, 9). Metabolic reprogramming of cancer cells facilitates enhanced proliferation and, in later stages cell dissemination, processes in which alterations of lipid metabolism play pivotal roles. Lipids serve as structural components of membranes, fuel sources for rapidly growing and dividing cells, and secondary messengers (9, 10). In particular, researchers focus on fatty acid (FA) metabolism because FAs are the main building blocks for other, more complex classes of lipids, such as phospholipids, sphingolipids, glycosphingolipids or triglycerides, and therefore contribute to the regulation of many different biochemical processes.

Aberrant FA metabolism has long been recognized in CRC cells (11), thus providing potential targets for therapeutics. A considerable body of work shows that *de novo* lipogenesis is required for CRC tumour growth and survival (12-15). FA synthase (FASN) provides palmitate for cancer cells for intensive membrane formation, enhancing cell resistance to oxidative damage and to chemotherapeutics due to increased lipid saturation (16), provides fuel for FA oxidation, and promotes the metastatic capacity of CRC (12, 17, 18). The palmitate from FASN-driven lipogenesis can also be modified *via* elongases (ELOVLs) or desaturated by stearoyl-CoA desaturase (SCD1), and these modifications may be targets of new therapy design (9). Indeed, CRC exhibits both the enhanced elongation of FAs (19, 20) and up-regulation of SCD1 activity (21, 22). Upon activation by an enzyme from the long-chain acyl-CoA synthetase family (ACSL), endogenously synthesized or exogenous FAs can be incorporated into triglycerides and cholesterol esters, which

Correspondence to: Tomasz Sledzinski, Department of Pharmaceutical Biochemistry, Faculty of Pharmacy, Medical University of Gdansk, Gdansk, Poland. E-mail: tomasz.sledzinski@gumed.edu.pl

Key Words: Colorectal cancer, fatty acid synthesis, fatty acid oxidation, inhibitors, review.

can be stored in lipid droplets as energy depots that are available to generate ATP *via* β-oxidation of FA (9). In CRC cells, a decrease in triglycerides and an up-regulation of enzymes has been observed in association with β-oxidation (23), conferring unique tumour advantages, such as evasion of anoikis, which is imperative for cancer metastasis (24). Although some changes in the lipid profiles of CRC samples are contradictory (11) and not straightforward because they are affected by many whole-body relationships between different tissues and by diet (8, 25), the contribution of altered lipid metabolism to the development and progression of CRC warrants the effort to pursue inhibitors of FA metabolism as potential drugs. In this review, we cover inhibitors associated with FA synthesis and oxidation. The potential targets for the inhibitors described below are presented in Figure 1.

FA Synthesis

FA synthesis is a multistep process leading to the formation of palmitic acid. The simplest way to reduce FA levels in cancer cells is to block FA synthesis. By inhibiting enzymes and reducing FA availability from the diet, cancer cell proliferation can be limited. Inhibitors of *de novo* FA synthesis may slow cancer cell proliferation, since they rely mostly on the *de novo* synthesis of FA, and they have minimal effect on healthy cells, which access FA mostly from dietary sources (26).

ATP-citrate lyase (ACLY) inhibitors. The first synthesis step links the tricarboxylic cycle (TCA) to lipogenesis by transforming citrate originating in the mitochondria into oxaloacetate and acetyl-CoA through the cytosolic enzyme ATP-citrate lyase (ACLY). Inhibition of ACLY was shown to reduce the ability of cells to metabolize the glucose needed to generate lipids (27). The up-regulation of ACLY, the first-step enzyme of *de novo* lipogenesis, has been found in several types of cancer, including CRC (28). GSK165 is a novel small-molecule compound that inhibits ACLY activity in a concentration-dependent manner. It had an antiproliferative effect on HT29 CRC cells. Furthermore, GSK165 sensitized these cells to the antineoplastic drug SN38 (28).

Acetyl-CoA carboxylase (ACC) inhibitors. The second step of FA synthesis, the carboxylation of acetyl-CoA to generate malonyl-CoA, is catalysed by ACC. ACC inhibitors have recently gained increased interest as potential anticancer drugs. The effect of 5-(tetradecyloxy)-2-furancarboxylic acid (TOFA), an allosteric inhibitor of ACC, has been examined in CRC cells. The results showed that TOFA is cytotoxic to HCT-8 and HCT-15 CRC cells and induced apoptosis in a dose-dependent manner. Supplementing cells with palmitic acid prevented the cell death induced by TOFA (29). These results suggest that TOFA should be further analysed in the context of CRC; however, it is noteworthy that TOFA did not affect cancer cell survival in some cancer types, such as breast and ovarian cancer (30, 31). *Citrus limon*-derived nanovesicles are naturally formed vesicles that exhibit ACC inhibitory properties. They inhibited SW480 cell proliferation without affecting normal cells; however, more studies are needed to verify whether *C. limon* nanovesicles can serve as supplementary compounds in CRC treatment (32, 33). To the best of our knowledge, other ACC inhibitors, such as PF-05175157, soraphen A and ND-646, were not tested in any CRC model. Altogether, novel, more-sensitive ACCA inhibitors for use as antitumour agents still need to be developed.

FASN inhibitors. FASN converts one acetyl-CoA and seven malonyl-CoA molecules into palmitic acid through a series of reactions. FASN is the key enzyme in *de novo* lipogenesis, which is up-regulated in CRC cells compared to normal colonic mucosal cells (34). It has been shown that a higher expression of FASN is correlated with a worse CRC prognosis; however, some reports show that this correlation is more complex and that the influence of FASN expression on patient outcome depends on body mass index (nonobese patients with CRC and tumoural FASN overexpression have better survival than obese patients) (35, 36). FASN is a 270-kDa cytosolic enzyme composed of seven functional domains: three *N*-terminal domains (ketoacyl synthase, malonyl/acetyltransferase, and dehydratase), and four C-terminal domains (enoyl reductase, ketoacyl reductase, acyl carrier protein, and thioesterase (37). FASN seems to be a promising therapeutic because of its tissue distribution. FASN inhibitors can target only colonic cancer cells, and the normal tissue thus remains unaffected. Moreover, FASN is elevated in aberrant crypt foci; thus, it may be important in the very early stage of colorectal tumorigenesis and a worthy target for possible chemoprevention (38). Several pharmacological FASN inhibitors appear promising for clinical use in the future. Cerulenin [(2S,3R)-2,3-epoxy-4-oxo-7,10-dodecadienamide] was the first FASN inhibitor discovered. It is an antibiotic isolated from *Cephalosporium caerulens* that has strong noncompetitive affinity for the active site of the ketoacyl synthase domain (39). Cerulenin suppresses the proliferation of many CRC cell lines *in vitro* by inducing apoptosis (15, 40-42). Furthermore, cerulenin reduced the number and size of liver metastases based on Colon 26 and CMT 93 murine CRC cell lines (41). Chang *et al*. showed that the inhibition of FASN by cerulenin in HT-29 and LoVo cells suppressed their malignant phenotype by attenuating energy metabolism and down-regulating the mammalian target of rapamycin (mTOR) signalling pathway (15). Another study revealed the potentiated effect of a

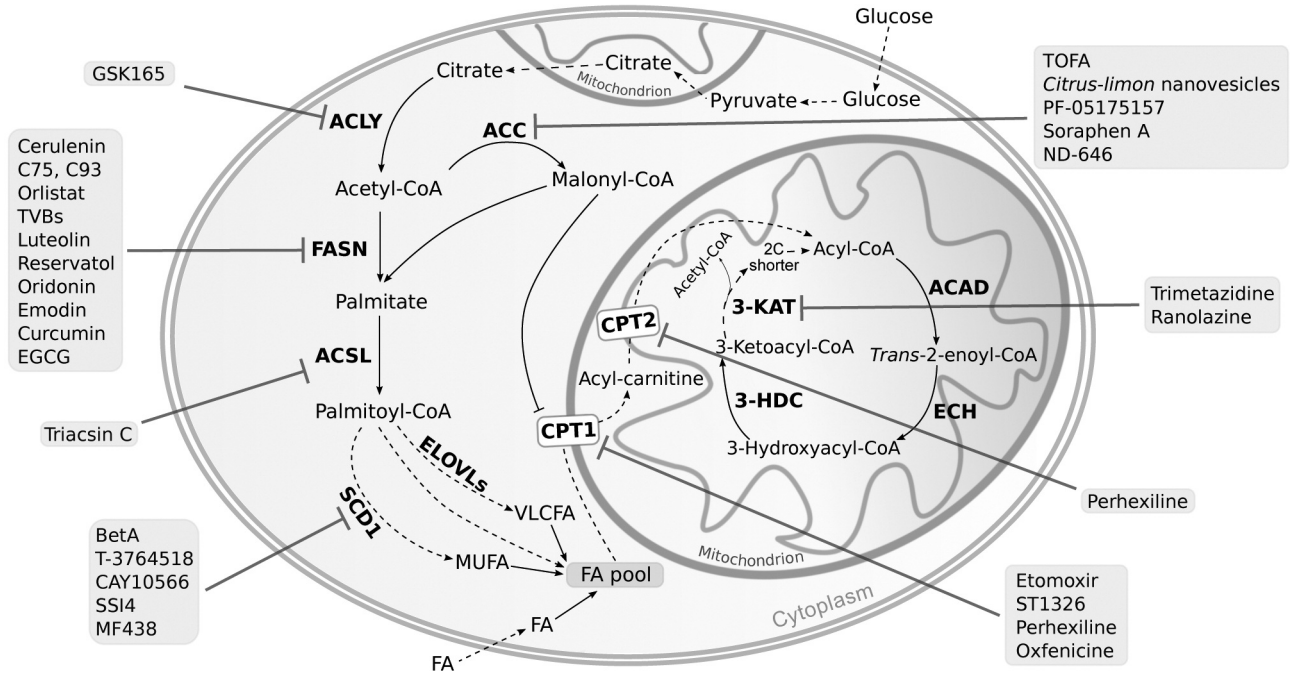

Figure 1. *Potential targets for colorectal cancer treatment by inhibitors of fatty acid (FA) synthesis or FA oxidation. The enzymes of FA synthesis and β-oxidation are shown in bold. The inhibitors of these enzymes are shown with the straight lines outside the cell scheme. 3-HDC: 3-Hydroxyacyl-CoA dehydrogenase; 3-KAT: 3-ketoacyl-coenzyme A thiolase; ACAD: acyl-CoA dehydrogenase; ACC: acetyl-CoA carboxylase; ACLY: ATP-citrate lyase; ACSL: long-chain acyl-CoA synthetase; CPT: carnitine palmitoyl transferase; ECH: enoyl-CoA hydratase; EGCG: epigallocatechin gallate; ELOVL: long-chain fatty acid elongase; FASN: fatty acid synthase; MUFA: monounsaturated fatty acid; SCD1: stearoyl-CoA desaturase; VLCFA: very-long chain fatty acid.*

combination treatment consisting of cerulenin and oxaliplatin, which led to the reduction of oxaliplatin dosage and promoted longer-lasting chemotherapy in mice (42). Tetrahydro-4-methylene-2R-octyl-5-oxo-3S-furancarboxylic acid (C75) is a competitive irreversible FASN inhibitor with a structure similar to that of cerulenin. It has been proposed as an antitumour and anti-obesity agent. C75 inactivates the enoyl reductase, thioesterase and ketoacyl synthase activities of FASN (43). C75 and cerulenin share several mechanisms of action and have similar side-effects. Both of these compounds are critical for the rapid and profound inhibition of DNA replication and S-phase cell-cycle progression. Study showed these effects to be detected 90 min after drug exposure of RKO colon carcinoma cells, thus they preceded the apoptotic changes observed 6 h after drug exposure (44). C75 and cerulenin both directly or indirectly activate carnitine palmitoyltransferase-1 (CPT1), which is the rate-limiting enzyme for β-oxidation (45, 46). CPT1 activation causes anorexia and weight loss in mice, which is one of the main obstacles in the clinical use of these compounds (47, 48). To address issues of weight loss, another FASN inhibitor, C93, has been designed to avoid activating CPT1. Cancer cell proliferation was significantly inhibited in mice bearing xenografts of the Colo680N oesophageal squamous cell carcinoma cell line when were treated with C93, without anorexia or weight loss (49). Orlistat is an anti-obesity drug with good patient tolerance (50). It inhibits FASN and exhibits antiproliferative effects on CRC cancer lines *in vitro* (Caco-2 and SW480 cells) (51) and in HT-29/*tk-luc* human colorectal carcinoma-bearing animals *in vivo* (52). Orlistat induces cell-cycle arrest at the G_1 phase in a dose- and time-dependent manner and triggers apoptosis *via* the activation of caspase-3 (52). Taken together, these results allowed us to formulate the hypothesis that the oral formulation of orlistat may be beneficial in treating tumours such as colon cancer; however, our recent study showed that exogenous palmitate can reverse the antiproliferative effects of orlistat in HT-29 CRC cells (53). Novel potent FASN inhibitors (TVBs) were developed by 3-V Biosciences, and they demonstrate antitumour activity *in vitro* and *in vivo* and are being evaluated in phase I/II clinical trials. TVB-3166 inhibits the ketoacyl reductase enzymatic domain of FASN. TVB-3166 is a reversible, potent, and selective FASN inhibitor that interferes with cell proliferation, and viability by inducing apoptosis. TVB-3166 disrupted lipid raft architecture and inhibited signal transduction in the protein kinase B (AKT)–mTOR and

β-catenin pathways in COLO-205 and HT-29 cells (54). Another study evaluated the effect of TVB-3664 on tumour growth in nine CRC patient-derived xenografts (55). More than 90% of the tumours in that study were positive for FASN expression; however, treatment with TVB-3664 led to a significant decrease in tumour volume in only one-third of the cases. The authors suggested that the presence of FASN expression does not predict the response to FASN inhibitors. Moreover, it has been proposed that the antitumour activity of TVB-3664 is associated with reduced activation of the AKT and extracellular signal-regulated protein kinases 1 and 2 (ERK1/2) oncogenic pathways and significant alteration of the lipid composition of tumours. The authors also noted that *PI3K* mutational status and activation of AKT downstream signalling may affect the response of CRC cells to FASN inhibition and suggested that the combination of FASN inhibitors with inhibitors of the AKT or mitogen-activated protein kinase (MAPK) pathway may be a potential therapeutic strategy for CRC (55). Currently, TVB-3664 and TVB-3166 are undergoing preclinical evaluation for use in patients with CRC (54, 55), and TVB-2640 is in phase I clinical trials for the treatment of resectable colon cancer (56). Luteolin (3',4',5,7-tetrahydroxyflavone) is a potent FASN inhibitor that has been shown to reduce DNA synthesis and inhibit HT-29 cell proliferation by inducing G_1 and G_2/M cell-cycle arrest and apoptosis in a concentration-dependent manner (57). These findings were confirmed by the increased percentage of G2/M-phase SW480 colon cancer cells when treated with luteolin (58). Moreover, luteolin displayed a dose-dependent apoptotic effect on Caco-2 cells with negligible or no effect on normal cells. It has been suggested that the mechanisms accounting for this are AKT phosphorylation and sphingosine kinase 2 (SPHK2) inhibition. As a consequence, the endoplasmic reticulum–Golgi trafficking of ceramide is impaired, and finally, the sphingolipid rheostat is unbalanced. An observation of these reports inspired the suggestion that supplementing the diet with luteolin may be a potential clinical strategy for treating CRC (59). This hypothesis is supported by the observation that supplementation with luteolin in addition to aspirin showed a better effect than aspirin alone in rats with dimethylhydrazine-induced carcinogenesis (60). Resveratrol (3,5,4'-trihydroxystilbene) is a potent FASN inhibitor that binds reversibly to the ketoacyl reductase domain (61). A number of studies have reported that resveratrol suppresses CRC cell proliferation and elevates the apoptosis rate. CaCo-2 cells treated with resveratrol exhibit 70% inhibition of proliferation with no signs of cytotoxicity or apoptosis (62). The data suggest that resveratrol suppresses cell proliferation by inhibiting the insulin-like growth factor type 1 receptor and its downstream signalling pathways and enhances apoptosis *via* the activation of the p53 pathway in HT-29 and SW480 cells (63). Resveratrol has been shown to have ameliorating effects on CRC in different mouse and rat models (64, 65). Resveratrol has been shown to reduce tumorigenesis associated with colitis in C57BL/6 mice (64). Moreover, prolonged daily administration of resveratrol significantly reduced the number of aberrant crypt foci and crypt multiplicity in the colorectal mucosa (65). Resveratrol has moved into clinical trials; however, whether resveratrol is beneficial in CRC treatment is still debated. Because of its poor bioavailability, high doses have been used in clinical trials, with some side-effects being thus observed, and its use has not yet been translated into clinical practice (66, 67). Some hypotheses have suggested that resveratrol derivatives may be better cancer chemopreventive candidates than resveratrol itself. For example, the resveratrol analogue (E)-*N*-(2-(4-methoxystyryl) phenyl) furan-2-carboxamide was shown to induce G_2/M cell-cycle arrest through the activation of p53-p21 in HCT116 cells (68). Nevertheless, resveratrol has a wide range of potential targets for chemoprevention. The detailed regulatory mechanisms for resveratrol-induced inhibition of CRC cellular proliferation were presented in a recent review by Honari *et al*. (69). Oridonin is a natural compound isolated from *Rabdosia rubescens* that effectively inhibits *FASN* and sterol regulatory element-binding protein 1 mRNA and protein expression in CRC contexts. It induces apoptosis and reduces cellular palmitic acid and stearic acid levels (70). Furthermore, oridonin inhibited the proliferation of LoVo cells partially by disrupting the transforming growth factor-β1–SMAD protein–plasminogen activator inhibitor-1 signalling pathway (71). Despite promising preclinical results, its poor solubility and bioavailability have slowed further research. HAO472 is an oridonin derivative that may be more applicable. It inhibits the proliferation and activation of T-cells and has been advanced into phase I clinical trials in China for the treatment of acute myelogenous leukaemia (72). Emodin (1,3,8-trihydroxy-6-methylanthraquinone) was shown to down-regulate the expression of FASN, inhibit intracellular FASN activity and FA biosynthesis, and induce the apoptosis of HCT116 cells in a dose- and time-dependent manner (73). Moreover, the combined treatment of emodin and cerulenin resulted in an additive effect (74). Curcumin inhibits FASN *in vitro* (75). Curcumin inhibited the proliferation of HT-29 and HCT-116 human colon cancer-derived cell lines. At least partially, this effect was due to peroxisome proliferator-activated receptors activation and the inhibition of epidermal growth factor receptor activation (76, 77). A clinical study on patients with adenocarcinoma of the colon or rectum showed a beneficial role of curcumin supplementation; however, no definite conclusions have been drawn to date (78). A plant polyphenol, epigallocatechin gallate (EGCG), present in green tea, has been tested on CRC cell lines with positive results. *In vivo* and *in vitro* studies showed that EGCG treatment induced apoptosis, affected the cell cycle

and reduced the volume of tumours (79). EGCG has also been shown to induce apoptosis and suppress liver metastasis of CRC in SCID mice (80). Moreover, EGCG induced sensitization to 5-fluorouracil by targeting cancer stem cells in CRC and may serve as a supportive treatment for patients with CRC (81). A human study showed that 1 year of EGCG supplementation by increasing green tea consumption was safe and significantly prevented metachronous colorectal adenomas in Japanese patients (82). Promising preclinical results have led to clinical trials of EGCG supplementation as a potential therapeutic option for patients with CRC. The results support the hypothesis that EGCG may display some chemopreventive effects for patients with CRC; however, more studies with a larger group of patients and longer study periods are needed to confirm this effect (83-85).

ACSL inhibitors. To be further elongated or desaturated, a FA must be transformed into an acyl-CoA molecule by ACS, which are classified according to the length of the acyl chain of their substrates: - short-chain, medium-chain and long-chain ACSs. In this review, we focus on ACSL isoforms, as they are up-regulated in CRC (86). In mammals, five ACSL isoforms (ACSL1-6) have been identified. Four main ACSL isoforms are up-regulated in CRC: ACSL1, ACSL4, ACSL5, and ACSL6 (87-89). High levels of ACSL1 or ACSL4 expression in patient tumour samples were found to correlate with worse prognoses for patients with CRC (90). In CRC cells, ACSL1 and ACSL4 overexpression causes a shift of energy metabolism towards glucose utilization (90). Despite promising results obtained from analysing ACSL expression in CRC, there is only one chemical ACSL inhibitor, triacsin C, which inhibits ACSL1, -3, and -4 (91). Unfortunately, at high concentrations, triacsin C is toxic to cells, and achieving effective pharmacokinetic effects and cell penetration is challenging (92).

Inhibitors of SCD1. The desaturation of a FA is catalysed by SCD1, which introduces a double bond at the n-9 position. SCD1 is highly expressed in CRC tissues and has a negative correlation with patient prognosis; however, the SCD1 expression level was found to be different in five different human CRC cell lines: SW620, HCT116, Caco-2, SW116, and HT29 cells. Moreover, SCD1 enhanced the migration and invasion ability of CRC cells (93). Betulinic acid (BetA) is a pentacyclic triterpenoid extracted from birch trees, with a wide range of biological properties, including SCD inhibition. BetA inhibited the proliferation of HCT116, SW480, and DLD-1 cells *in vitro* in a time- and dose-dependent manner. Moreover, BetA induced cell apoptosis through the mitochondrial-mediated pathway and inhibited the metastasis of cancer cells. In vivo, BetA suppressed the growth of tumours in an HCT-116 xenograft tumour mouse model (22). In another study, BetA induced rapid CSC cell death *via* the elimination of cell clonogenic capacity (94). Other SCD inhibitors have some interesting properties; however, only a few reports are available. For example, T-3764518 is a novel and orally available small-molecule SCD inhibitor that successfully inhibited HCT-116 cell proliferation (95), and CAY10566, a chemical SCD1 inhibitor, significantly reduced cell viability and spheroid formation in a WiDr 3D culture system (96). Other SCD1 inhibitors, such as SSI-4 and MF-438, have not been tested in CRC models.

ELOVL inhibitors. The elongation of palmitate and other long-chain FAs requires a unique set of enzymes. Elongation takes place in the endoplasmic reticulum and is catalysed by ELOVLs. ELOVL isoforms can be divided into two groups: ELOVLs 1, 3, 6 and 7, which take part in the elongation of saturated and monounsaturated FAs; and ELOVLs 2, 4, and 5, which catalyse the elongation of polyunsaturated FAs (97). ELOVLs have not been thoroughly studied in the context of CRC; however, it is known that CRC specimens show enhanced FA elongation. The results from a study by our team showed a dramatic increase in *ELOVL1* and *ELOVL6* mRNA levels in CRC tissue, suggesting that these enzymes may play critical roles in the development and metabolism of CRC cells (19). Moreover, higher ELOVL6 activity was observed in patients with adenomas compared to those with no colonic polyps, and this characteristic was associated with an increased risk of colonic adenoma (98). To the best of our knowledge, no studies have investigated the consequences of ELOVL knockdown in CRC cell lines. Attempts have been made to discover ELOVL6 inhibitors; however, to date, no candidates have been thoroughly studied, and none have been tested on CRC cells (99, 100).

Since FA synthesis is important for cancer cells to survive, enzymes taking part in this process may be potential therapeutic targets for clinical application. The inhibition of many enzymes mentioned in this review reduces the proliferation and diminishes the metastatic capabilities of cancer cells in multiple CRC models. The role of the overexpression of these enzymes in CRC cells and the mechanisms by which exposure to different inhibitors induces cell death are still not fully understood due to significant variability in responses to these inhibitors in CRC. A better understanding of the mechanism(s) of by which various compounds inhibit different enzymes will advance the development of new potential therapeutic approaches to target CRC.

β-Oxidation of FAs

FAs are catabolized during β-oxidation (also known as FA oxidation) in the mitochondrial matrix by the sequential removal of two-carbon units (acetyl-CoA) from the carboxyl end of the FA chain in repetitive cycles. Before actual oxidation, FA must be activated by converting the FA into a

fatty acyl-CoA ester in an irreversible reaction catalysed by ACSs on the outer mitochondrial membrane. After activation, the FAs are transported from the cytosol to the mitochondrial matrix. The mitochondrial inner membrane is impermeable to long-chain FAs. To cross the membrane, long-chain FAs must be conjugated with carnitine by carnitine palmitoyltransferase 1 (CPT1), which is present on the outer surface of the inner mitochondrial membrane. The conjugate is then transported across the membrane by translocase, and carnitine acyltransferase II (CPT2), located on the inner mitochondrial membrane on the matrix side, removes the carnitine (101). This is the rate-limiting step of FA oxidation. β-Oxidation involves repeated cycles of four reactions: Dehydrogenation (by acyl-CoA dehydrogenase), hydration (*via* enoyl-CoA hydratase), a second dehydrogenation (by 3-hydroxyacyl-CoA dehydrogenase), and thiolysis [by 3-ketoacyl-coenzyme A thiolase (*3-KAT*)]. This process is accompanied by the reduction of NAD to NADH and FAD to $FADH_2$, which then transfer electrons to the respiratory chain, where they enable the production of cellular energy in the form of ATP. Moreover, the final product of β-oxidation – acetyl CoA – is also oxidized in mitochondria, leading to further production of ATP (102). FA oxidation plays a crucial role in cancer cells and is necessary for cell proliferation, survival, stemness, drug resistance and metastasis (103, 104). Rapidly growing cancer cells have high energy demands and require FAs to produce cell membranes. In CRC, FA oxidation may be critical to cancer development. Therefore, it seems that the inhibition of this process in CRC cells may be a possible treatment option for the inhibition of cancer growth.

CPT1 inhibitors. CPT1 is the key rate-limiting enzyme of FA oxidation. Its inhibition prevents the formation of acylcarnitines and makes the transport of FA chains from the cytosol into the mitochondria impossible. Currently, three different isoforms of CPT1 have been identified, which exhibit tissue-specific distribution: CPT1A in the liver, CPT1B in the muscle, and CPT1C expressed exclusively in the brain (105). Inhibitors are specific for the different CTP1 isoforms. Etomoxir {ethyl 2-[6-(4-chlorophenoxy)hexyl]oxirane-2-carboxylate} is an irreversible pharmacological inhibitor of CPT1 that is used in clinical studies for the treatment of heart failure (106). A study on MCA-38 tumour-bearing mice (colon adenocarcinoma) treated with etomoxir indicated that this drug caused a significant delay in tumour growth with minimal necrosis and a higher number of cells undergoing apoptosis. However, an *in vitro* study on MCA-38 cells treated with etomoxir did not show a delay in cell proliferation or a change in the number of cancer stem cells (107). Moreover, Hossain and co-workers noted that etomoxir blocked the immune inhibitory pathways and immunosuppressive function of myeloid-derived suppressor cells in MCA-38 tumour-bearing mice. These cells promote tumour progression through multiple mechanisms in CRC (107). Furthermore, etomoxir significantly inhibited FA uptake and ATP production and reduced oxygen consumption and extracellular acidification rates (107). In the HCT116 colon carcinoma cell line, etomoxir also reduced the cellular ATP level (108). Inhibition of FA oxidation by etomoxir enhanced the antitumour effect of low-dose chemotherapy, especially cisplatin, on HCT116 colon carcinoma cells (108). Another study showed that combining etomoxir with radiation improved its therapeutic efficacy in H460 human lung epithelial carcinoma cells and LNCaP prostate carcinoma cells. The authors hypothesized that administering etomoxir immediately after cell irradiation may be a promising alternative for solid tumour treatment, including for CRC (109). In light of the work by Wang *et al*. (24), the inhibition of CTP1A may be a potential strategy for preventing CRC metastasis. CPT1A was one of the top up-regulated genes in detached CRC cells, compared to its expression in attached CRC cells. The inhibition of *CPT1A* by etomoxir inhibited CRC cell proliferation and promoted anoikis in detached CRC cells (24).

It is noteworthy that some data suggest that etomoxir has off-target effects at high concentrations. For example, this CPT1 inhibitor may also inhibit complex I of the electron transport chain and reduce cell proliferation independently of FA oxidation (110). Referenced data suggest that etomoxir may be a promising treatment for many cancer types, including CRC as a monotherapy or in combination with other anticancer agents, such as cisplatin or radiation; however, cancer cachexia and possible off-target effects must be taken into account during further studies. Because of the possible toxicity induced by etomoxir, a more selective CPT1 inhibitor ST1326 (teglicar) was developed. This aminocarnitine derivative is a liver-specific reversible inhibitor and probably does not share a toxicity profile similar to that of etomoxir (111). Its efficiency in CRC has yet to be investigated. Perhexiline 2-(2,2-dicyclohexylethyl)piperidine is another partial inhibitor of the carnitine shuttle of long-chain FAs into the mitochondria. Perhexiline is a potential dual CPT1/CPT2 inhibitor. In vitro studies demonstrated that the cardiac isoform of CPT1 is more sensitive than the hepatic isoform to inhibition by perhexiline (112). Perhexiline has been approved for the treatment of heart disease in some countries; however, it may induce toxicity, especially in long-term therapy, and recently, its use has thus declined (113, 114). The results of studies by Wang and co-workers from 2020 indicate that using perhexiline to block FA catabolism sensitizes CRC cells to the antitumour effect of oxaliplatin, as demonstrated by the enhanced apoptosis of treated HCT116 and DLD-1 CRC cells (115). Similar to other CPT inhibitors, such as etomoxir, perhexiline-induced inhibition of FA oxidation and intensified ROS accumulation enables classic chemotherapeutic drugs to kill more CRC cells (115). Oxfenicine (*S*-2-(4-hydroxyphenyl)glycine) is an effective

inhibitor of carnitine-palmitoyl transferase 1B (CPT1B) in the heart but not in the liver. It is still in a preclinical stage, and no data concerning its effect on CRC are available (116).

3-KAT inhibitors. 3-KAT is the final enzyme activated in the FA β-oxidation process, and it releases acetyl-CoA. Trimetazidine and ranolazine are 3-KAT inhibitors that directly inhibit FA oxidation. Trimetazidine is used as an anti-anginal therapy in Europe and Asia. Ranolazine suppresses FA oxidation in rat cardiac and skeletal muscle cells and serves as a therapeutic agent in heart disease in Europe and the United States (113). Ranolazine is a clinically used inhibitor of voltage-gated Na+ channels, and has also been shown to inhibit breast cancer metastasis *in vivo* (117, 118). Sodium channel protein type 5 subunit alpha is a voltage-gated Na+ channel isoform that is expressed in colon cancer. It is a key regulator of a gene transcriptional network that controls the invasion of CRC, although the mechanisms are still not fully clear (119). The most recent study placed some inhibitors of FA oxidation mentioned in this review in a completely different light. Ma and co-workers pointed out that ranolazine and trimetazidine did not show any inhibitory effects on FA oxidation in established cell lines, primary cells, or mice; therefore, they formulated a hypothesis that these two molecules may not be adequate FAO inhibitors of β-oxidation at the cellular level. Similarly, they did not detect anti-FAO activity of perhexiline, an outcome that the authors explained may be related to its poor specificity as a dual CPT1/CPT2 inhibitor. Only etomoxir and oxfenicine at higher concentrations were able to significantly inhibit FA oxidation in MCF-7 and T47D cells (120).

α-Methylacyl CoA racemase (AMACR) inhibitors. The enzyme AMACR, also known as P504S, is involved in converting an R-configurated branched FA to an *S*-configuration before β-oxidation (121). AMACR protein overexpression was found in a number of types of cancer, including CRC (122-124). Several inhibitors of AMACR have been indicated, for instance ebselen, the seleno-organic compound ebselen oxide, pyrazoloquinolines, pyrazolopyrimidines, 2-trifluoromethyl tetradecanoyl CoA and both the R and S isomers of ibuprofenoyl CoA (125-127); however, due to their modest affinity, there is still a need to discover more potent inhibitors in order to evaluate the possibility of targeting AMACR for inhibition in CRC treatment.

Non-steroidal anti-inflammatory drugs (NSAIDs) and FA oxidation. Inflammation increases the risk of CRC, thus, it is widely assumed that NSAIDs may reduce this risk (128). Substantial available evidence showed that NSAIDs reduced the proliferation of CRC cell lines in several experimental models. NSAIDs (particularly aspirin) have been reported to reduce the risk of CRC by approximately 20% in the general population or even by 50% in those with a first-degree relative with a history of colon cancer (129-131). Although NSAIDs are among the drugs most used worldwide, their mechanism of action is still not fully understood. It is known that these molecules inhibit the cyclo-oxygenases COX1 and COX2, which are involved in the synthesis of eicosanoids, such as prostaglandins, from arachidonic acid. Prostaglandins are engaged in numerous biological responses and play roles in maintaining both normal and cancer cell viability and proliferation (132). Although CRC is characterized by overexpression of COX2 but not of COX1, studies concerning the inhibitory effects of NSAIDs on CRC cells suggest that some inhibit CRC cell proliferation, cause cell-cycle arrest and induce apoptosis independently of the COX1 or COX2 pathway (131, 133). These findings led to the assumption that other inhibitory pathways must be critical for NSAID effects (134-136). Some NSAIDs may inhibit long-chain FA oxidation in mouse liver mitochondria *in vivo* and *in vitro* (137-139). The first documented evidence of this by NSAIDs in a CRC cell line was published by Yang et aI. (136). LIM 1215, LIM 1899 and HT-29 CRC cell lines were treated with aspirin, ibuprofen, indomethacin, meclofenamate, sulindac sulfide, and sulindac sulfone. These tested NSAIDs inhibited long-chain FA oxidation and thus contributed to the inhibition of cell proliferation (136). The detailed mechanism of β-oxidation inhibition by different NSAIDs remains unclear and needs to be determined. Although daily aspirin consumption may have beneficial effects on CRC risk and taking particular NSAIDs seems to be a promising therapeutic option for CRC, complications and side-effects should be taken into consideration. The inhibition of β-oxidation leads to the accumulation of long-chain FAs, some of which may induce toxicity. In further studies, a more complex analysis of the long-term intake of NSAIDs on diverse colon cancer types should be performed, taking into account other factors such as body mass index, comorbidities, and stage of the disease (140).

Other FA oxidation inhibitors. Dipropylacetic acid (VPA) is a branched-chain FA that directly or indirectly affects FA oxidation. There are many hypotheses concerning the mechanism of action of VPA in the case of inhibition of FA oxidation, such as the generation of reactive metabolites that irreversibly inactivate oxidative enzymes, but the pleiotropic effects of the drug on mitochondrial metabolism are the most likely contributors to its effects on CRC (141, 142). VPA is also considered a first-generation histone deacetylase inhibitor (143). VPA administered with fluoropyrimidine-based chemoradiotherapy had antiproliferative effects on CRC cell lines expressing P53 (wild-type or mutant) (144). Thia FAs are FAs with a sulfur atom inserted into the FA backbone. Saturated 3-thia tetradecylthioacetic acid (TTA) and 4-thia tetradecylthiopropionic acid (TTP) induce nearly opposite effects on FA metabolism in the liver. TTA increases the catabolism of FA, and TTP is a powerful inhibitor of FA

oxidation (145). Thia FA with the sulfur atom in an even carbon position, such as TTP, blocked FA oxidation in rat liver and heart, probably by inducing the accumulation of alkylthioacryloyl-CoA in mitochondria (146, 147). Interestingly, it has TTA was observed to inhibit SW620 CRC cell proliferation, probably by inducing endoplasmic reticulum stress and activating the unfolded protein response pathway independently of FA oxidation (148).

FA oxidation inhibitors are promising therapeutic options for some types of cancer, including CRC, when used in monotherapy or in combination with other anticancer agents. Inhibition of FA oxidation may also be an attractive strategy for overcoming drug resistance or metastasis of tumours. Their potential for affecting these outcomes is extremely important because more than 20% of patients with CRC exhibit distant metastases at initial diagnosis (149). The exact mechanisms of action of the selected inhibitors described in this review remain unclear, and their effects on CRC growth are also unknown. Therefore, there is an urgent need to appropriately evaluate their β-oxidation-inhibitory and antiproliferative functions at the cellular level in CRC models. Understanding the specific mechanisms of action of these inhibitors is key for the development of new successful therapies of CRC.

Conclusion

The balance between *de novo* FA synthesis and β-oxidation is crucial for cell homeostasis, and dysregulation of these opposing processes can lead to serious consequences. In the case of cancer cells, the chemical inhibition of the synthesis or β-oxidation of FAs often leads to slower proliferation and an increased rate of apoptosis of cancer cells without affecting normal cells. These outcomes suggest that targeting enzymes engaged in FA synthesis and oxidation is a promising therapeutic approach for CRC; however, notably, more research is needed to confirm their beneficial effects in human patients with CRC.

Conflicts of Interest

The Authors declare that there are no conflicts of interest regarding the publication of this article.

Authors' Contributions

P.M., A.M. and TS conceived and designed the review; P.M., K.D. and A.P. studied the literature and wrote the article; T.S. and A.M. verified the article. All Authors agreed on the final version of the review.

Acknowledgements

This research was supported by the National Science Centre of Poland (grant no. 2016/22/E/NZ4/00665) and Medical University of Gdansk (grants no. ST-40).

References

1 Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA and Jemal A: Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin 68: 394-424, 2018. PMID: 30207593. DOI: 10.3322/caac.21492

2 Siegel RL, Miller KD and Jemal A: Cancer statistics, 2020. CA Cancer J Clin *70*: 7-30, 2020. PMID: 31912902. DOI: 10.3322/caac.21590

3 Hadjipetrou A, Anyfantakis D, Galanakis CG, Kastanakis M and Kastanakis S: Colorectal cancer, screening and primary care: A mini literature review. World J Gastroenterol *23*: 6049-6058, 2017. PMID: 28970720. DOI: 10.3748/wjg.v23.i33.6049

4 Schreuders EH, Ruco A, Rabeneck L, Schoen RE, Sung JJY, Young GP and Kuipers EJ: Colorectal cancer screening: A global overview of existing programmes. Gut *64*: 1637-1649, 2015. PMID: 26041752. DOI: 10.1136/gutjnl-2014-309086

5 Rawla P, Sunkara T and Barsouk A: Epidemiology of colorectal cancer: incidence, mortality, survival, and risk factors. Gastroenterol Rev *14*: 89-103, 2019. PMID: 31616522. DOI: 10.5114/pg.2018.81072

6 Siegel RL, Miller KD, Goding Sauer A, Fedewa SA, Butterly LF, Anderson JC, Cercek A, Smith RA and Jemal A: Colorectal cancer statistics, 2020. CA Cancer J Clin *70*: 145-164, 2020. PMID: 32133645. DOI: 10.3322/caac.21601

7 Dekker E, Tanis PJ, Vleugels JLA, Kasi PM and Wallace MB: Colorectal cancer. Lancet *394*: 1467-1480, 2019. PMID: 31631858. DOI: 10.1016/S0140-6736(19)32319-0

8 Koundouros N and Poulogiannis G: Reprogramming of fatty acid metabolism in cancer. Br J Cancer *122*: 4-22, 2020. PMID: 31819192. DOI: 10.1038/s41416-019-0650-z

9 Chen M and Huang J: The expanded role of fatty acid metabolism in cancer: new aspects and targets. Precis Clin Med *2*: 183-191, 2019. PMID: 31598388. DOI: 10.1093/pcmedi/pbz017

10 DeBerardinis RJ and Chandel NS: Fundamentals of cancer metabolism. Sci Adv *2*: e1600200, 2016. PMID: 27386546. DOI: 10.1126/sciadv.1600200

11 Pakiet A, Kobiela J, Stepnowski P, Sledzinski T and Mika A: Changes in lipids composition and metabolism in colorectal cancer: A review. Lipids Health Dis *18*: 29, 2019. PMID: 30684960. DOI: 10.1186/s12944-019-0977-8

12 Zaytseva YY, Rychahou PG, Gulhati P, Elliott VA, Mustain WC, O'Connor K, Morris AJ, Sunkara M, Weiss HL, Lee EY and Evers BM: Inhibition of fatty acid synthase attenuates CD44-associated signaling and reduces metastasis in colorectal cancer. Cancer Res *72*: 1504-1517, 2012. PMID: 22266115. DOI: 10.1158/0008-5472.CAN-11-4057

13 Li N, Bu X, Tian X, Wu P, Yang L and Huang P: Fatty acid synthase regulates proliferation and migration of colorectal cancer cells *via* HER2-PI3K/AKT signaling pathway. Nutr Cancer *64*: 864-870, 2012. PMID: 22860766. DOI: 10.1080/01635581.2012.701704

14 Zhan Y, Ginanni N, Tota MR, Wu M, Bays NW, Richon VM, Kohl NE, Bachman ES, Strack PR and Krauss S: Human cancer biology control of cell growth and survival by enzymes of the fatty acid synthesis pathway in HCT-116 colon cancer cells. Clin Cancer Res *14*: 5735-5743, 2008. PMID: 18794082. DOI: 10.1158/1078-0432.CCR-07-5074

15 Chang L, Wu P, Senthilkumar R, Tian X, Liu H, Shen X, Tao Z and Huang P: Loss of fatty acid synthase suppresses the malignant phenotype of colorectal cancer cells by down-regulating energy metabolism and mTOR signaling pathway. J Cancer Res Clin Oncol *142*: 59-72, 2016. PMID: 26109148. DOI: 10.1007/s00432-015-2000-8

16 Rysman E, Brusselmans K, Scheys K, Timmermans L, Derua R, Munck S, Van Veldhoven PP, Waltregny D, Daniëls VW, Machiels J, Vanderhoydonc F, Smans K, Waelkens E, Verhoeven G and Swinnen J V: *De novo* lipogenesis protects cancer cells from free radicals and chemotherapeutics by promoting membrane lipid saturation. Cancer Res *70*: 8117-8126, 2010. PMID: 20876798. DOI: 10.1158/0008-5472.CAN-09-3871

17 Jafari N, Drury J, Morris AJ, Onono FO, Stevens PD, Gao T, Liu J, Wang C, Lee EY, Weiss HL, Evers BM and Zaytseva YY: *De novo* fatty acid synthesis-driven sphingolipid metabolism promotes metastatic potential of colorectal cancer. Mol Cancer Res *17*: 140-152, 2019. PMID: 30154249. DOI: 10.1158/1541-7786.MCR-18-0199

18 Wang H, Xi Q and Wu G: Fatty acid synthase regulates invasion and metastasis of colorectal cancer *via* Wnt signaling pathway. Cancer Med *5*: 1599-606, 2016. PMID: 27139420. DOI: 10.1002/cam4.711

19 Mika A, Kobiela J, Czumaj A, Chmielewski M, Stepnowski P and Sledzinski T: Hyper-elongation in colorectal cancer tissue – cerotic acid is a potential novel serum metabolic marker of colorectal malignancies. Cell Physiol Biochem *41*: 722-730, 2017. PMID: 28214830. DOI: 10.1159/000458431

20 Kondo Y, Nishiumi S, Shinohara M, Hatano N, Ikeda A, Yoshie T, Kobayashi T, Shiomi Y, Irino Y, Takenawa T, Azuma T and Yoshida M: Serum fatty acid profiling of colorectal cancer by gas chromatography/mass spectrometry. Biomark Med *5*: 451-460, 2011. PMID: 21861667. DOI: 10.2217/bmm.11.41

21 Chen L, Ren J, Yang L, Li Y, Fu J, Li Y, Tian Y, Qiu F, Liu Z and Qiu Y: Stearoyl-CoA desaturase-1 mediated cell apoptosis in colorectal cancer by promoting ceramide synthesis. Sci Rep *6*: 19665, 2016. PMID: 26813308. DOI: 10.1038/srep19665

22 Ran H, Zhu Y, Deng R, Zhang Q, Liu X, Feng M, Zhong J, Lin S, Tong X and Su Q: Stearoyl-CoA desaturase-1 promotes colorectal cancer metastasis in response to glucose by suppressing PTEN. J Exp Clin Cancer Res *37*: 54, 2018. PMID: 29530061. DOI: 10.1186/s13046-018-0711-9

23 Mika A, Pakiet A, Czumaj A, Kaczynski Z and Liakh I: Decreased triacylglycerol content and elevated contents of cell membrane lipids in colorectal cancer tissue: a lipidomic study. J Clin Med *9*: 1-11, 2020. PMID: 32290558. DOI: 10.3390/jcm9041095

24 Wang YN, Zeng ZL, Lu J, Wang Y, Liu ZX, He MM, Zhao Q, Wang ZX, Li T, Lu YX, Wu QN, Yu K, Wang F, Pu HY, Li B, Jia WH, Shi M, Xie D, Kang TB, Huang P, Ju HQ and Xu RH: CPT1A-mediated fatty acid oxidation promotes colorectal cancer cell metastasis by inhibiting anoikis. Oncogene *37*: 6025-6040, 2018. PMID: 29995871. DOI: 10.1038/s41388-018-0384-z

25 Wen Y-A, Xing X, Harris JW, Zaytseva YY, Mitov MI, Napier DL, Weiss HL, Mark Evers B and Gao T: Adipocytes activate mitochondrial fatty acid oxidation and autophagy to promote tumor growth in colon cancer. Cell Death Dis *8*: e2593, 2017. PMID: 28151470. DOI: 10.1038/cddis.2017.21

26 Swinnen J V, Brusselmans K and Verhoeven G: Increased lipogenesis in cancer cells: New players, novel targets. Curr Opin Clin Nutr Metab Care *9*: 358-365, 2006. PMID: 16778563. DOI: 10.1097/01.mco.0000232894.28674.30

27 Khwairakpam A, Shyamananda M, Sailo B, Rathnakaram S, Padmavathi G, Kotoky J and Kunnumakkara A: ATP Citrate lyase (ACLY): A promising target for cancer prevention and treatment. Curr Drug Targets *16*: 156-163, 2015. PMID: 25537655. DOI: 10.2174/1389450115666141224125117

28 Zhou Y, Bollu LR, Tozzi F, Ye X, Bhattacharya R, Gao G, Dupre E, Xia L, Lu J, Fan F, Bellister S, Ellis LM and Weihua Z: ATP citrate lyase mediates resistance of colorectal cancer cells to SN38. Mol Cancer Ther *12*: 2782-2791, 2013. PMID: 24132143. DOI: 10.1158/1535-7163.MCT-13-0098

29 Wang C, Xu C, Sun M, Luo D, Liao D fang and Cao D: Acetyl-CoA carboxylase-α inhibitor TOFA induces human cancer cell apoptosis. Biochem Biophys Res Commun *385*: 302-306, 2009. PMID: 19450551. DOI: 10.1016/j.bbrc.2009.05.045

30 Zhou W, Wan FH, Landree LE, Thupari JN, Pinn ML, Bililign T, Eun KK, Vadlamudi A, Medghalchi SM, El Meskini R, Ronnett GV, Townsend CA and Kuhajda FP: Fatty acid synthase inhibition activates AMP-activated protein kinase in SKOV3 human ovarian cancer cells. Cancer Res *67*: 2964-2971, 2007. PMID: 17409402. DOI: 10.1158/0008-5472.CAN-06-3439

31 Pizer ES, Thupari J, Han WF, Pinn ML, Chrest FJ, Frehywot GL, Townsend CA and Kuhajda FP: Malonyl-coenzyme-A is a potential mediator of cytotoxicity induced by fatty-acid synthase inhibition in human breast cancer cells and xenografts. Cancer Res *60*: 213-218, 2000. PMID: 10667561.

32 Raimondo S, Naselli F, Fontana S, Monteleone F, Lo Dico A, Saieva L, Zito G, Flugy A, Manno M, Di Bella MA, De Leo G and Alessandro R: *Citrus limon*-derived nanovesicles inhibit cancer cell proliferation and suppress CML xenograft growth by inducing TRAIL-mediated cell death. Oncotarget *6*: 19514-19527, 2015. PMID: 26098775. DOI: 10.18632/oncotarget.4004

33 Raimondo S, Saieva L, Cristaldi M, Monteleone F, Fontana S and Alessandro R: Label-free quantitative proteomic profiling of colon cancer cells identifies acetyl-CoA carboxylase alpha as antitumor target of *Citrus limon*-derived nanovesicles. J Proteomics *173*: 1-11, 2018. PMID: 29197582. DOI: 10.1016/j.jprot.2017.11.017

34 Rashid A, Pizer ES, Moga M, Milgraum LZ, Zahurak M, Pasternack GR, Kuhajda FP and Hamilton SR: Elevated expression of fatty acid synthase and fatty acid synthetic activity in colorectal neoplasia. Am J Pathol *150*: 201-208, 1997. PMID: 9006336.

35 Ogino S, Nosho K, Meyerhardt JA, Kirkner GJ, Chan AT, Kawasaki T, Giovannucci EL, Loda M and Fuchs CS: Cohort study of fatty acid synthase expression and patient survival in colon cancer. J Clin Oncol *26*: 5713-5720, 2008. PMID: 18955444. DOI: 10.1200/JCO.2008.18.2675

36 Notarnicola M, Tutino V, Calvani M, Lorusso D, Guerra V and Caruso MG: Serum levels of fatty acid synthase in colorectal cancer patients are associated with tumor stage. J Gastrointest Cancer *43*: 508-511, 2012. PMID: 21727995. DOI: 10.1007/s12029-011-9300-2

37 Smith S, Witkowski A and Joshi AK: Structural and functional organization of the animal fatty acid synthase. Prog Lipid Res *42*: 289-317, 2003. PMID: 12689621. DOI: 10.1016/S0163-7827(02)00067-X

38 Kearney KE, Pretlow TG and Pretlow TP: Increased expression of fatty acid synthase in human aberrant crypt foci: Possible target for colorectal cancer prevention. Int J Cancer *125*: 249-252, 2009. PMID: 19358283. DOI: 10.1002/ijc.24356

39 Ōmura S: Cerulenin. Methods Enzymol *72*: 520-532, 1981. PMID: 7031426. DOI: 10.1016/S0076-6879(81)72041-X

40 Huang PL, Zhu SN, Lu SL, Dai ZS and Jin YL: Inhibitor of fatty acid synthase induced apoptosis in human colonic cancer cells. World J Gastroenterol *6*: 295-297, 2000. PMID: 11819582. DOI: 10.3748/wjg.v6.i2.295

41 Murata S, Yanagisawa K, Fukunaga K, Oda T, Kobayashi A, Sasaki R and Ohkohchi N: Fatty acid synthase inhibitor cerulenin suppresses liver metastasis of colon cancer in mice. Cancer Sci *101*: 1861-1865, 2010. PMID: 20491775. DOI: 10.1111/j.1349-7006.2010.01596.x

42 Shiragami R, Murata S, Kosugi C, Tezuka T, Yamazaki M, Hirano A, Yoshimura Y, Suzuki M, Shuto K and Koda K: Enhanced antitumor activity of cerulenin combined with oxaliplatin in human colon cancer cells. Int J Oncol *43*: 431-438, 2013. PMID: 23754252. DOI: 10.3892/ijo.2013.1978

43 Rendina AR and Cheng D: Characterization of the inactivation of rat fatty acid synthase by C75: Inhibition of partial reactions and protection by substrates. Biochem J *388*: 895-903, 2005. PMID: 15715522. DOI: 10.1042/BJ20041963

44 Pizer ES, Chrest FJ, DiGiuseppe JA and Han WF: Pharmacological inhibitors of mammalian fatty acid synthase suppress DNA replication and induce apoptosis in tumor cell lines. Cancer Res *58*: 4611-4615,1998. PMID: 9788612.

45 Aja S, Landree LE, Kleman AM, Medghalchi SM, Vadlamudi A, McFadden JM, Aplasca A, Hyun J, Plummer E, Daniels K, Kemm M, Townsend CA, Thupari JN, Kuhajda FP, Moran TH and Ronnett GV: Pharmacological stimulation of brain carnitine palmitoyl-transferase-1 decreases food intake and body weight. Am J Physiol Regul Integr Comp Physiol *294(2):* R352-361, 2008. PMID: 18056987. DOI: 10.1152/ajpregu.00862.2006

46 Tu Y, Thupari JN, Kim EK, Pinn ML, Moran TH, Ronnett GV and Kuhajda FP: C75 alters central and peripheral gene expression to reduce food intake and increase energy expenditure. Endocrinology *146*: 486-493, 2005. PMID: 15498887. DOI: 10.1210/en.2004-0976

47 Loftus TM, Jaworsky DE, Frehywot CL, Townsend CA, Ronnett GV, Lane MD and Kuhajda FP: Reduced food intake and body weight in mice treated with fatty acid synthase inhibitors. Science *288*: 2379-2381, 2000. PMID: 10875926. DOI: 10.1126/science.288.5475.2379

48 Kumar MV, Shimokawa T, Nagy TR and Lane MD: Differential effects of a centrally acting fatty acid synthase inhibitor in lean and obese mice. Proc Natl Acad Sci USA *99*: 1921-1925, 2002. PMID: 11854492. DOI: 10.1073/pnas.042683699

49 Orita H, Coulter J, Tully E, Abe M, Montgomery E, Alvarez H, Sato K, Hino O, Kajiyama Y, Tsurumaru M and Gabrielson E: High levels of fatty acid synthase expression in esophageal cancers represent a potential target for therapy. Cancer Biol Ther *10*: 549-554, 2010. PMID: 20657182. DOI: 10.4161/cbt.10.6.12727

50 Drent ML and van der Veen EA: First clinical studies with orlistat: A short review. Obes Res *3*: 623S-625S, 1995. PMID: 8697067. DOI: 10.1002/j.1550-8528.1995.tb00236.x

51 Kridel SJ, Axelrod F, Rozenkrantz N and Smith JW: Orlistat is a novel inhibitor of fatty acid synthase with antitumor activity. Cancer Res *64*: 2070-2075, 2004. PMID: 15026345. DOI: 10.1158/0008-5472.CAN-03-3645

52 Chuang HY, Chang YF and Hwang JJ: Antitumor effect of orlistat, a fatty acid synthase inhibitor, is *via* activation of caspase-3 on human colorectal carcinoma-bearing animal. Biomed Pharmacother *65*: 286-292, 2011. PMID: 21723078. DOI: 10.1016/j.biopha.2011.02.016

53 Czumaj A, Zabielska J, Pakiet A, Mika A, Rostkowska O, Makarewicz W, Kobiela J, Sledzinski T and Stelmanska E*: In vivo* effectiveness of orlistat in the suppression of human colorectal cancer cell proliferation. Anticancer Res *39*: 3815-3822, 2019. PMID: 31262909. DOI: 10.21873/anticanres.13531

54 Ventura R, Mordec K, Waszczuk J, Wang Z, Lai J, Fridlib M, Buckley D, Kemble G and Heuer TS: Inhibition of *de novo* palmitate synthesis by fatty acid synthase induces apoptosis in tumor cells by remodeling cell membranes, inhibiting signaling pathways, and reprogramming gene expression. EBioMedicine *2*: 808-824, 2015. PMID: 26425687. DOI: 10.1016/j.ebiom.2015.06.020

55 Zaytseva YY, Rychahou PG, Le AT, Scott TL, Flight RM, Kim JT, Harris J, Liu J, Wang C, Morris AJ, Sivakumaran TA, Fan T, Moseley H, Gao T, Lee EY, Weiss HL, Heuer TS, Kemble G and Evers M: Preclinical evaluation of novel fatty acid synthase inhibitors in primary colorectal cancer cells and a patient-derived xenograft model of colorectal cancer. Oncotarget *9*: 24787-24800, 2018. PMID: 29872506. DOI: 10.18632/oncotarget.25361

56 FASN inhibitor TVB-2640 in treating patients with colon or other cancers that can be removed by surgery. Available from: https://www.cancer.gov/about-cancer/treatment/clinical-trials/search/v?id=NCI-2016-01710&r=1 [Last accessed June 8, 2020]

57 Lim DY, Jeong Y, Tyner AL and Park JHY: Induction of cell cycle arrest and apoptosis in HT-29 human colon cancer cells by the dietary compound luteolin. Am J Physiol Liver Physiol *292*: G66-G75, 2007. PMID: 16901994. DOI: 10.1152/ajpgi.00248.2006

58 Wang W, VanAlstyne PC, Irons KA, Chen S, Stewart JW and Birt DF: Individual and interactive effects of apigenin analogs on G_2/M cell-cycle arrest in human colon carcinoma cell lines. Nutr Cancer *48*: 106-114, 2004. PMID: 15203384. DOI: 10.1207/s15327914nc4801_14

59 Abdel Hadi L, Di Vito C, Marfia G, Ferraretto A, Tringali C, Viani P and Riboni L: Sphingosine kinase 2 and ceramide transport as key targets of the natural flavonoid luteolin to induce apoptosis in colon cancer cells. PLoS One *10*: e0143384, 2015. PMID: 26580959. DOI: 10.1371/journal.pone.0143384

60 Osman NHA, Said UZ, El-Waseef AM and Ahmed ESA: Luteolin supplementation adjacent to aspirin treatment reduced dimethylhydrazine-induced experimental colon carcinogenesis in rats. Tumor Biol *36*: 1179-1190, 2015. PMID: 25342594. DOI: 10.1007/s13277-014-2678-2

61 Liang Y, Tian W and Ma X: Inhibitory effects of grape skin extract and resveratrol on fatty acid synthase. BMC Complement Altern Med *13*, 2013. PMID: 24341420. DOI: 10.1186/1472-6882-13-361

62 Schneider Y, Vincent F, Duranton B, Badolo L, Gossé F, Bergmann C, Seiler N and Raul F: Anti-proliferative effect of resveratrol, a natural component of grapes and wine, on human colonic cancer cells. Cancer Lett *158*: 85-91, 2000. PMID: 10940513. DOI: 10.1016/S0304-3835(00)00511-5

63 Vanamala J, Reddivari L, Radhakrishnan S and Tarver C: Resveratrol suppresses IGF-1 induced human colon cancer cell proliferation and elevates apoptosis *via* suppression of IGF-1R/Wnt and activation of p53 signaling pathways. BMC Cancer *10*: 238, 2010. PMID: 20504360. DOI: 10.1186/1471-2407-10-238

64 Cui X, Jin Y, Hofseth AB, Pena E, Habiger J, Chumanevich A, Poudyal D, Nagarkatti M, Nagarkatti PS, Singh UP and Hofseth LJ: Resveratrol suppresses colitis and colon cancer associated with colitis. Cancer Prev Res *3*: 549-559, 2010. PMID: 20332304. DOI: 10.1158/1940-6207.CAPR-09-0117

65 Tessitore L, Davit A and Sarotto I: Resveratrol depresses the growth of colorectal aberrant crypt foci by affecting BAX and p21 CIP expression. Carcinogenesis *21*: 1619-1622, 2000. PMID: 10910967. DOI: 10.1093/carcin/21.5.619

66 Patel KR, Scott E, Brown VA, Gescher AJ, Steward WP and Brown K: Clinical trials of resveratrol. Ann N Y Acad Sci *1215*: 161-169, 2011. PMID: 21261655. DOI: 10.1111/j.1749-6632.2010.05853.x

67 Chow HHS, Garland LL, Hsu CH, Vining DR, Chew WM, Miller JA, Perloff M, Crowell JA and Alberts DS: Resveratrol modulates drug- and carcinogen-metabolizing enzymes in a healthy volunteer study. Cancer Prev Res *3*: 1168-1175, 2010. PMID: 20716633. DOI: 10.1158/1940-6207.CAPR-09-0155

68 Cheah FK, Leong KH, Thomas NF, Chin HK, Ariffin A and Awang K: Resveratrol analogue (*E*)-*N*-(2-(4-methoxystyryl) phenyl) furan-2-carboxamide induces G_2/M cell cycle arrest through the activation of p53-p21 CIP1/WAF1 in human colorectal HCT116 cells. Apoptosis *23*: 329-342, 2018. PMID: 29754265. DOI: 10.1007/s10495-018-1457-8

69 Honari M, Shafabakhsh R, Reiter RJ, Mirzaei H and Asemi Z: Resveratrol is a promising agent for colorectal cancer prevention and treatment: Focus on molecular mechanisms. Cancer Cell Int *19*: 180, 2019. PMID: 31341423. DOI: 10.1186/s12935-019-0906-y

70 Kwan HY, Yang Z, Fong WF, Hu YM, Yu ZL and Hsiao WLW: The anticancer effect of oridonin is mediated by fatty acid synthase suppression in human colorectal cancer cells. J Gastroenterol *48*: 182-192, 2013. PMID: 22722903. DOI: 10.1007/s00535-012-0612-1

71 Bu H-Q, Shen F and Cui J: The inhibitory effect of oridonin on colon cancer was mediated by deactivation of TGF-β1/SMADs-PAI-1 signaling pathway *in vitro* and *vivo*. Onco Targets Ther *12*: 7467-7476, 2019. PMID: 31686852. DOI: 10.2147/OTT.S220401

72 Ding Y, Ding C, Ye N, Liu Z, Wold EA, Chen H, Wild C, Shen Q and Zhou J: Discovery and development of natural product oridonin-inspired anticancer agents. Eur J Med Chem *122*: 102-117, 2016. PMID: 27344488. DOI: 10.1016/j.ejmech.2016.06.015

73 Xie MJ, Ma YH, Miao L, Wang Y, Wang HZ, Xing YY, Xi T and Lu YY: Emodin-provoked oxidative stress induces apoptosis in human colon cancer HCT116 cells through a p53-mitochondrial apoptotic pathway. Asian Pacific J Cancer Prev *15*: 5201-5205, 2014. PMID: 25040975. DOI: 10.7314/APJCP.2014.15.13.5201

74 Lee KH, Lee MS, Cha EY, Sul JY, Lee JS, Kim JS, Park JB and Kim JY: Inhibitory effect of emodin on fatty acid synthase, colon cancer proliferation and apoptosis. Mol Med Rep *15*: 2163-2173, 2017. PMID: 28260110. DOI: 10.3892/mmr.2017.6254

75 Zhao J, Sun XB, Ye F and Tian WX: Suppression of fatty acid synthase, differentiation and lipid accumulation in adipocytes by curcumin. Mol Cell Biochem *351*: 19-28, 2011. PMID: 21221723. DOI: 10.1007/s11010-010-0707-z

76 Reddy S, Rishi AK, Xu H, Levi E, Sarkar FH and Majumdar APN: Mechanisms of curcumin- and EGF-receptor related protein (ERRP)-dependent growth inhibition of colon cancer cells. Nutr Cancer *55*: 185-194, 2006. PMID: 17044774. DOI: 10.1207/s15327914nc5502_10

77 Chen A and Xu J: Activation of PPARγ by curcumin inhibits Moser cell growth and mediates suppression of gene expression of cyclin D1 and EGFR. Am J Physiol Liver Physiol *288*: G447-G456, 2005. PMID: 15486348. DOI: 10.1152/ajpgi.00209.2004

78 Sharma RA, Euden SA, Platton SL, Cooke DN, Shafayat A, Hewitt HR, Marczylo TH, Morgan B, Hemingway D, Plummer SM, Pirmohamed M, Gescher AJ and Steward WP: Phase I clinical trial of oral curcumin: Biomarkers of systemic activity and compliance. Clin Cancer Res *10*: 6847-6854, 2004. PMID: 15501961. DOI: 10.1158/1078-0432.CCR-04-0744

79 Jin H, Gong W, Zhang C and Wang S: Epigallocatechin gallate inhibits the proliferation of colorectal cancer cells by regulating Notch signaling. Onco Targets Ther *6*: 145-153, 2013. PMID: 23525843. DOI: 10.2147/OTT.S40914

80 Maruyama T, Murata S, Nakayama K, Sano N, Ogawa K, Nowatari T, Tamura T, Nozaki R, Fukunaga K and Ohkohchi N: Epigallocatechin-3-gallate suppresses liver metastasis of human colorectal cancer. Oncol Rep *31*: 625-633, 2014. PMID: 24337301. DOI: 10.3892/or.2013.2925

81 Toden S, Tran HM, Tovar-Camargo OA, Okugawa Y and Goel A: Epigallocatechin-3-gallate targets cancer stem-like cells and enhances 5-fluorouracil chemosensitivity in colorectal cancer. Oncotarget *7*: 16158-16171, 2016. PMID: 26930714. DOI: 10.18632/oncotarget.7567

82 Shimizu M, Fukutomi Y, Ninomiya M, Nagura K, Kato T, Araki H, Suganuma M, Fujiki H and Moriwaki H: Green tea extracts for the prevention of metachronous colorectal adenomas: A pilot study. Cancer Epidemiol Biomarkers Prev *17*: 3020-3025, 2008. PMID: 18990744. DOI: 10.1158/1055-9965.EPI-08-0528

83 Shin CM, Lee DH, Seo AY, Lee HJ, Kim SB, Son WC, Kim YK, Lee SJ, Park SH, Kim N, Park YS and Yoon H: Green tea extracts for the prevention of metachronous colorectal polyps among patients who underwent endoscopic removal of colorectal adenomas: A randomized clinical trial. Clin Nutr *37*: 452-458, 2018. PMID: 28209333. DOI: 10.1016/j.clnu.2017.01.014

84 Stingl JC, Ettrich T, Muche R, Wiedom M, Brockmöller J, Seeringer A and Seufferlein T: Protocol for minimizing the risk of metachronous adenomas of the colorectum with green tea extract (MIRACLE): A randomised controlled trial of green tea extract versus placebo for nutriprevention of metachronous colon adenomas in the elderly population. BMC Cancer *11*: 360, 2011. PMID: 21851602. DOI: 10.1186/1471-2407-11-360

85 Chemopreventive Effects of Epigallocatechin Gallate (EGCG) in Colorectal Cancer (CRC) Patients. Available at: https://clinicaltrials.gov/ct2/show/NCT02891538 [Last accessed June 8, 2020]

86 Mashek DG, Li LO and Coleman RA: Long-chain acyl-CoA synthetases and fatty acid channeling. Future Lipidol *2*: 465-476, 2007. PMID: 20354580. DOI: 10.2217/17460875.2.4.465

87 Chen W-C, Wang C-Y, Hung Y-H, Weng T-Y, Yen M-C and Lai M-D: Systematic analysis of gene expression alterations and clinical outcomes for long-chain acyl-coenzyme a synthetase family in cancer. PLoS One *11*: e0155660, 2016. PMID: 27171439. DOI: 10.1371/journal.pone.0155660

88 Gassler N, Herr I, Schneider A, Penzel R, Langbein L, Schirmacher P and Kopitz J: Impaired expression of acyl-CoA synthetase 5 in sporadic colorectal adenocarcinomas. J Pathol *207*: 295-300, 2005. PMID: 16110457. DOI: 10.1002/path.1831

89 Cao Y, Dave KB, Doan TP and Prescott SM: Fatty acid CoA ligase 4 is up-regulated in colon adenocarcinoma, Cancer Res *61*: 8429-8434, 2001. PMID: 11731423.

90 Sánchez-Martínez R, Cruz-Gil S, García-Álvarez MS, Reglero G and De Molina AR: Complementary ACSL isoforms contribute to a non-Warburg advantageous energetic status characterizing invasive colon cancer cells. Sci Rep *7*: 1-15, 2017. PMID: 28894242. DOI: 10.1038/s41598-017-11612-3

91 Shimabukuro M, Zhou Y-T, Levi M and Unger RH: Fatty acid-induced cell apoptosis: A link between obesity and diabetes. Proc Natl Acad Sci USA *95*: 2498-2502, 1998. PMID: 9482914. DOI: 10.1073/pnas.95.5.2498

92 Rossi Sebastiano M and Konstantinidou G: Targeting long chain acyl-CoA synthetases for cancer therapy. Int J Mol Sci *20*: 3624, 2019. PMID: 31344914. DOI: 10.3390/ijms20153624

93 Ran H, Zhu Y, Deng R, Zhang Q, Liu X, Feng M, Zhong J, Lin S, Tong X and Su Q: Stearoyl-CoA desaturase-1 promotes colorectal cancer metastasis in response to glucose by suppressing PTEN. J Exp Clin Cancer Res *37*: 54, 2018. PMID: 29530061. DOI: 10.1186/s13046-018-0711-9

94 Potze L, di Franco S, H. Kessler J, Stassi G and Paul Medema J: Betulinic acid kills colon cancer stem cells. Curr Stem Cell Res Ther *11*: 427-433, 2016. PMID: 26647913. DOI: 10.2174/1574888x11666151203223512

95 Ono A, Sano O, Kazetani K, Muraki T, Imamura K, Sumi H, Matsui J and Iwata H: Feedback activation of AMPK-mediated autophagy acceleration is a key resistance mechanism against SCD1 inhibitor-induced cell growth inhibition. PLoS One *12*: e0181243, 2017. PMID: 28704514. DOI: 10.1371/journal.pone.0181243

96 Qin X-Y and Kojima S: Inhibition of stearoyl-CoA desaturase-1 activity suppressed SREBP signaling in colon cancer cells and their spheroid growth. Gastrointest Disord *1*: 191-200, 2019. DOI: 10.3390/gidisord1010014

97 Jump DB: Mammalian fatty acid elongases. Methods Mol Biol *579*: 375-389, 2009. PMID: 19763486. DOI: 10.1007/978-1-60761-322-0_19

98 Pickens CA, Lane-Elliot A, Comstock SS and Fenton JI: Altered saturated and monounsaturated plasma phospholipid fatty acid profiles in adult males with colon adenomas. Cancer Epidemiol Biomarkers Prev *25*: 498-506, 2016. PMID: 26721667. DOI: 10.1158/1055-9965.EPI-15-0696

99 Shimamura K, Nagumo A, Miyamoto Y, Kitazawa H, Kanesaka M, Yoshimoto R, Aragane K, Morita N, Ohe T, Takahashi T, Nagase T, Sato N and Tokita S: Discovery and characterization of a novel potent, selective and orally active inhibitor for mammalian ELOVL6. Eur J Pharmacol *630*: 34-41, 2010. PMID: 20045404. DOI: 10.1016/j.ejphar.2009.12.033

100 Zheng J, Wu Z, Dai M, Xu Z, Li X, Zhu S, Lin C, Hu P, Zhang L, Huang H, Zhao S, Zhang K and Sun P: Quantitative structure–activity relationship studies on a novel indolediones as long chain fatty acid elongase 6 (ELOVL6) inhibitors. Lett Drug Des Discov *8*: 422-429, 2011. DOI: 10.2174/157018011795514168

101 Kerner J and Hoppel C: Fatty acid import into mitochondria. Biochim Biophys Acta Mol Cell Biol Lipids *1486*: 1-17, 2000. PMID: 10856709. DOI: 10.1016/s1388-1981(00)00044-5

102 Carracedo A, Cantley LC and Pandolfi PP: Cancer metabolism: Fatty acid oxidation in the limelight. Nat Rev Cancer *13*: 227-232, 2013. PMID: 23446547. DOI: 10.1038/nrc3483

103 Xiong J: Fatty acid oxidation in cell fate determination. Trends Biochem Sci *43*: 854-857, 2018. PMID: 2973539. DOI: 10.1016/j.tibs.2018.04.006

104 Ma Y, Temkin SM, Hawkridge AM, Guo C, Wang W, Wang X-Y and Fang X: Fatty acid oxidation: An emerging facet of metabolic transformation in cancer. Cancer Lett *435*: 92-100, 2018. PMID: 30102953. DOI: 10.1016/j.canlet.2018.08.006

105 Schreurs M, Kuipers F and van der Leij FR: Regulatory enzymes of mitochondrial β-oxidation as targets for treatment of the metabolic syndrome. Obes Rev *11*: 380-388, 2010. PMID: 19694967. DOI: 10.1111/j.1467-789X.2009.00642.x

106 Holubarsch CJF, Rohrbach M, Karrasch M, Boehm E, Polonski L, Ponikowski P and Rhein S: A double-blind randomized multicentre clinical trial to evaluate the efficacy and safety of two doses of etomoxir in comparison with placebo in patients with moderate congestive heart failure: the ERGO (etomoxir for the recovery of glucose oxidation) study. Clin Sci *113*: 205-212, 2007. PMID: 17319797. DOI: 10.1042/CS20060307

107 Hossain F, Al-Khami AA, Wyczechowska D, Hernandez C, Zheng L, Reiss K, Del Valle L, Trillo-Tinoco J, Maj T, Zou W, Rodriguez PC and Ochoa AC: Inhibition of fatty acid oxidation modulates immunosuppressive functions of myeloid-derived suppressor cells and enhances cancer therapies. Cancer Immunol Res *3*: 1236-1247, 2015. PMID: 26025381. DOI: 10.1158/2326-6066.CIR-15-0036

108 Hernlund E, Ihrlund LS, Khan O, Ates YO, Linder S, Panaretakis T and Shoshan MC: Potentiation of chemotherapeutic drugs by energy metabolism inhibitors 2-deoxyglucose and etomoxir. Int J Cancer *123*: 476-483, 2008. PMID: 18452174. DOI: 10.1002/ijc.23525

109 Dheeraj A, Agarwal C, Schlaepfer IR, Raben D, Singh R, Agarwal R and Deep G: A novel approach to target hypoxic cancer cells *via* combining beta-oxidation inhibitor etomoxir with radiation. Hypoxia *6*: 23-33, 2018. PMID: 30175155. DOI: 10.2147/HP.S163115

110 Yao CH, Liu GY, Wang R, Moon SH, Gross RW and Patti GJ: Identifying off-target effects of etomoxir reveals that carnitine palmitoyltransferase I is essential for cancer cell proliferation independent of β-oxidation. PLoS Biol *16*, 2018. PMID: 29596410. DOI: 10.1371/journal.pbio.2003782

111 Conti R, Mannucci E, Pessotto P, Tassoni E, Carminati P, Giannessi F and Arduini A: Selective reversible inhibition of liver carnitine palmitoyl-transferase 1 by teglicar reduces gluconeogenesis and improves glucose homeostasis. Diabetes *60*: 644-651, 2011. PMID: 21270274. DOI: 10.2337/db10-0346

112 Kennedy JA, Unger SA and Horowitz JD: Inhibition of carnitine palmitoyltransferase-1 in rat heart and liver by perhexiline and amiodarone. Biochem Pharmacol *52*: 273-280, 1996. PMID: 8694852. DOI: 10.1016/0006-2952(96)00204-3

113 Lopaschuk GD, Ussher JR, Folmes CDL, Jaswal JS and Stanley WC: Myocardial fatty acid metabolism in health and disease. Physiol Rev *90*: 207-258, 2010. PMID: 20086077. DOI: 10.1152/physrev.00015.2009

114 Ashrafian H, Horowitz JD and Frenneaux MP: Perhexiline. Cardiovasc Drug Rev *25*: 76-97, 2007. PMID: 17445089. DOI: 10.1111/j.1527-3466.2007.00006.x

115 Wang Y, Lu JH, Wang F, Wang YN, He MM, Wu QN, Lu YX, Yu HE, Chen ZH, Zhao Q, Liu J, Chen YX, Wang DS, Sheng H, Liu ZX, Zeng ZL, Xu RH and Ju HQ: Inhibition of fatty acid catabolism augments the efficacy of oxaliplatin-based chemotherapy in gastrointestinal cancers. Cancer Lett *473*: 74-89, 2020. PMID: 31904482. DOI: 10.1016/j.canlet.2019.12.036

116 Stephens TW, Higgins AJ, Cook GA and Harris RA: Two mechanisms produce tissue-specific inhibition of fatty acid oxidation by oxfenicine. Biochem J *227*: 651-660, 1985. PMID: 4004784. DOI: 10.1042/bj2270651

117 Bugan I, Kucuk S, Karagoz Z, Fraser SP, Kaya H, Dodson A, Foster CS, Altun S and Djamgoz MBA: Anti-metastatic effect of ranolazine in an *in vivo* rat model of prostate cancer, and expression of voltage-gated sodium channel protein in human prostate. Prostate Cancer Prostatic Dis *22*: 569-579, 2019. PMID: 30894674. DOI: 10.1038/s41391-019-0128-3

118 Driffort V, Gillet L, Bon E, Marionneau-Lambot S, Oullier T, Joulin V, Collin C, Pagès J-C, Jourdan M-L, Chevalier S, Bougnoux P, Le Guennec J-Y, Besson P and Roger S: Ranolazine inhibits NaV1.5-mediated breast cancer cell invasiveness and lung colonization. Mol Cancer *13*: 264, 2014. PMID: 25496128. DOI: 10.1186/1476-4598-13-264

119 House CD, Wang BD, Ceniccola K, Williams R, Simaan M, Olender J, Patel V, Baptista-Hon DT, Annunziata CM, Gutkind JS, Hales TG and Lee NH: Voltage-gated Na+ channel activity increases colon cancer transcriptional activity and invasion *via* persistent MAPK signaling. Sci Rep *5*, 2015. PMID: 26096612. DOI: 10.1038/srep11541

120 Ma Y, Wang W, Devarakonda T, Zhou H, Wang XY, Salloum FN, Spiegel S and Fang X: Functional analysis of molecular and pharmacological modulators of mitochondrial fatty acid oxidation. Sci Rep *10*: 1-13, 2020. PMID: 31996743. DOI: 10.1038/s41598-020-58334-7

121 Flanagan VP, Ferretti A, Schwartz DP and Ruth JM: Characterization of two steroidal ketones and two isoprenoid alcohols in dairy products. J Lipid Res *16*: 97-101, 1975. PMID: 1168685.

122 Jiang Z, Fanger GR, Banner BF, Woda BA, Algate P, Dresser K, Xu J, Reed SG, Rock KL and Chu PG: A dietary enzyme: α-Methylacyl-CoA racemase/P504S is overexpressed in colon carcinoma. Cancer Detect Prev *27*: 422-426, 2003. PMID: 14642549. DOI: 10.1016/j.cdp.2003.07.003

123 Zhou M, Chinnaiyan AM, Kleer CG, Lucas PC and Rubin MA: Alpha-methylacyl-CoA racemase: A novel tumor marker overexpressed in several human cancers and their precursor lesions. Am J Surg Pathol *26*: 926-931, 2002. PMID: 12131161. DOI: 10.1097/00000478-200207000-00012

124 Jun L, Zha S, Gage WR, Sauvageot J, Saria EA, Putzi MJ, Ewing CM, Faith DA, Nelson WG, De Marzo AM, Isaacs WB, Trent JM, Isaacs WB and Marzo AM De: α-Methylacyl-CoA racemase, a new molecular marker for prostate cancer. Cancer Res *61*: 8617-23, 2001. PMID: 11751373.

125 Petrova YD, Wadda K, Nathubhai A, Yevglevskis M, Mitchell PJ, James TD, Threadgill MD, Woodman TJ and Lloyd MD: Identification of novel small-molecule inhibitors of α-methylacyl-CoA racemase (AMACR; P504S) and structure–activity relationships. Bioorg Chem *92*: 103264, 2019. PMID: 31536955. DOI: 10.1016/j.bioorg.2019.103264

126 Wilson BAP, Wang H, Nacev BA, Mease RC, Liu JO, Pomper MG and Isaacs WB: High-throughput screen identifies novel inhibitors of cancer biomarker α-methylacyl coenzyme A racemase (AMACR/P504S). Mol Cancer Ther *10*: 825-838, 2011. PMID: 21441411. DOI: 10.1158/1535-7163.MCT-10-0902

127 Carnell AJ, Hale I, Denis S, Wanders RJA, Isaacs WB, Wilson BA and Ferdinandusse S: Design, synthesis, and *in vitro* testing of α-methylacyl-CoA racemase inhibitors. J Med Chem *50*: 2700-2707, 2007. PMID: 17477519. DOI: 10.1021/jm0702377

128 Janakiram NB and Rao CV: The role of inflammation in colon cancer. Adv Exp Med Biol *816*: 25-52, 2014. PMID: 24818718. DOI: 10.1007/978-3-0348-0837-8_2

129 Ruder EH, Laiyemo AO, Graubard BI, Hollenbeck AR, Schatzkin A and Cross AJ: Non-steroidal anti-inflammatory drugs and colorectal cancer risk in a large, prospective cohort. Am J Gastroenterol *106*: 1340-1350, 2011. PMID: 21407185. DOI: 10.1038/ajg.2011.38

130 Kuo CN, Pan JJ, Huang YW, Tsai HJ and Chang WC: Association between nonsteroidal anti-inflammatory drugs and colorectal cancer: A population-based case-control study. Cancer Epidemiol Biomarkers Prev *27*: 737-745, 2018. PMID: 29695380. DOI: 10.1158/1055-9965.EPI-17-0876

131 Hixson LJ, Alberts DS, Krutzsch M, Einsphar J, Brendel K, Gross PH, Shipp Paranka N, Baier M, Emerson S, Pamukcu R and Burt RW: Antiproliferative effect of nonsteroidal antiinflammatory drugs against human colon cancer cells. Cancer Epidemiol Biomarkers Prev *3*: 433-438, 1994. PMID: 7920212.

132 Eberhart CE and Dubois RN: Eicosanoids and the gastrointestinal tract. Gastroenterology *109*: 285-301, 1995. PMID: 7797026. DOI: 10.1016/0016-5085(95)90296-1

133 Eberhart CE, Coffey RJ, Radhika A, Giardiello FM, Ferrenbach S and Dubois RN: Up-regulation of cyclooxygenase 2 gene expression in human colorectal adenomas and adenocarcinomas. Gastroenterology *107*: 1183-1188, 1994. PMID: 7926468. DOI: 10.1016/0016-5085(94)90246-1

134 Hanif R, Pittas A, Feng Y, Koutsos MI, Qiao L, Staiano-Coico L, Shiff SI and Rigas B: Effects of nonsteroidal anti-inflammatory drugs on proliferation and on induction of apoptosis in colon cancer cells by a prostaglandin-independent pathway. Biochem Pharmacol *52*: 237-245, 1996. PMID: 8694848. DOI: 10.1016/0006-2952(96)00181-5

135 Murphy VJ, Yang Z, Rorison KA and Baldwin GS: Cyclo-oxygenase-2-selective antagonists do not inhibit growth of colorectal carcinoma cell lines. Cancer Lett *122*: 25-30, 1998. PMID: 9464487. DOI: 10.1016/S0304-3835(97)00361-3

136 Yang Z, Hollande F and Baldwin GS: Blockade of long chain fatty acid oxidation by non-steroidal anti-inflammatory drugs may contribute to inhibition of proliferation of human colorectal carcinoma cell lines. Cancer Lett *124*: 187-191, 1998. PMID: 9500209. DOI: 10.1016/S0304-3835(97)00476-X

137 Geneve J, Hayat-Bonan B, Labbe G, Degott C, Letteron P, Freneaux E, Dinh TL, Larrey D and Pessayre D: Inhibition of mitochondrial beta-oxidation of fatty acids by pirprofen. Role in microvesicular steatosis due to this nonsteroidal anti-inflammatory drug. J Pharmacol Exp Ther *242*: 1133-1137, 1987. PMID: 3116197.

138 Freneaux E, Fromenty B, Berson A, Labbe G, Degott C, Letteron P, Larrey D and Pessayre D: Stereoselective and nonstereoselective effects of ibuprofen enantiomers on mitochondrial beta-oxidation of fatty acids. J Pharmacol Exp Ther *255*: 529-535, 1990. PMID: 2123005.

139 Zhao B, Geisslinger G, Hall I, Day RO and Williams KM: The effect of the enantiomers of ibuprofen and flurbiprofen on the β-oxidation of palmitate in the rat. Chirality *4*: 137-141, 1992. PMID: 1586584. DOI: 10.1002/chir.530040302

140 Hamoya T, Fujii G, Miyamoto S, Takahashi M, Totsuka Y, Wakabayashi K, Toshima J and Mutoh M: Effects of NSAIDs on the risk factors of colorectal cancer: a mini review. Genes Environ *38*: 6, 2016. PMID: 27350826. DOI: 10.1186/s41021-016-0033-0

141 Silva MF, Ruiter JP, IJlst L, Jakobs C, Duran M, de Almeida IT and Wanders RJ: Differential effect of valproate and its Delta2- and Delta4-unsaturated metabolites, on the beta-oxidation rate of long-chain and medium-chain fatty acids. Chem Biol Interact *137*: 203-12, 2001. PMID: 11566289. DOI: 10.1016/s0009-2797(01)00234-4

142 Silva MFB, Aires CCP, Luis PBM, Ruiter JPN, IJlst L, Duran M, Wanders RJA and Tavares de Almeida I: Valproic acid metabolism and its effects on mitochondrial fatty acid oxidation: A review. J Inherit Metab Dis *31*: 205-216, 2008. PMID: 18392741. DOI: 10.1007/s10545-008-0841-x

143 Kuendgen A and Gattermann N: Valproic acid for the treatment of myeloid malignancies. Cancer *110*: 943-954, 2007. PMID: 17647267. DOI: 10.1002/cncr.22891

144 Terranova-Barberio M, Pecori B, Roca MS, Imbimbo S, Bruzzese F, Leone A, Muto P, Delrio P, Avallone A, Budillon A and Di Gennaro E: Synergistic antitumor interaction between valproic acid, capecitabine and radiotherapy in colorectal cancer: critical role of p53. J Exp Clin Cancer Res *36*: 177, 2017. PMID: 29212503. DOI: 10.1186/s13046-017-0647-5

145 Asiedu DK, Al-Shurbaji A, Rustan AC, Björkhem I, Berglund L and Berge RK: Hepatic fatty acid metabolism as a determinant of plasma and liver triacylglycerol levels. Eur J Biochem *227*: 715-722, 1995. PMID: 7867630. DOI: 10.1111/j.1432-1033.1995.0715p.x

146 Dyroy E, Wergedahl H, Skorve J, Gudbrandsen OA, Songstad J and Berge RK: Thia fatty acids with the sulfur atom in even or odd positions have opposite effects on fatty acid catabolism. Lipids *41*: 169-177, 2006. PMID: 17707983. DOI: 10.1007/s11745-006-5085-7

147 Skrede S, Sørensen HN, Larsen LN, Steineger HH, Høvik K, Spydevold ØS, Horn R and Bremer J: Thia fatty acids, metabolism and metabolic effects. Biochim Biophys Acta Lipids Lipid Metab *1344*: 115-131, 1997. PMID: 9030189. DOI: 10.1016/s0005-2760(96)00138-5

148 Lundemo AG, Pettersen CH, Berge K, Berge RK and Schønberg SA: Tetradecylthioacetic acid inhibits proliferation of human SW620 colon cancer cells – gene expression profiling implies endoplasmic reticulum stress. Lipids Health Dis *10*: 190, 2011. PMID: 22027281. DOI: 10.1186/1476-511X-10-190

149 Howlader N, Noone AM, Krapcho M, Miller D, Brest A, Yu M, Ruhl J, Tatalovich Z, Mariotto A, Lewis DR, Chen HS, Feuer EJ and Cronin KA (eds): SEER Cancer Statistics Review, 1975-2017, National Cancer Institute. Bethesda, MD. Available at: https://seer.cancer.gov/csr/1975_2017/, based on November 2019 SEER data submission, posted to the SEER web site, April 2020.

Received June 17, 2020
Revised July 10, 2020
Accepted July 13, 2020