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REVIEW ARTICLE

Dihydropyrimidine dehydrogenase in the metabolism of the anticancer drugs

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Abstract
Cancer caused by fundamental defects in cell cycle regulation leads to uncontrolled growth of cells. In spite of the treatment with chemotherapeutic agents of varying nature, multiple resistance mechanisms are identified in cancer cells. Similarly, numerous variations, which decrease the metabolism of chemotherapeutics agents and thereby increasing the toxicity of anticancer drugs have been identified. 5-Fluorouracil (5-FU) is an anticancer drug widely used to treat many cancers in the human body. Its broad targeting range is based upon its capacity to act as a uracil analogue, thereby disrupting RNA and DNA synthesis. Dihydropyrimidine dehydrogenase (DPD) is an enzyme majorly involved in the metabolism of pyrimidines in the human body and has the same metabolising effect on 5-FU, a pyrimidine analogue. Multiple mutations in the DPD gene have been linked to 5-FU toxicity and inadequate dosages. DPD inhibitors have also been used to inhibit excessive degradation of 5-FU for meeting appropriate dosage requirements. This article focusses on the role of dihydropyrimidine dehydrogenase in the metabolism of the anticancer drug 5-FU and other associated drugs.

Keywords Cancer · Anticancer drugs · Dihydropyrimidine dehydrogenase (DPD) · 5-Fluorouracil (5-FU) · Drug resistance · Drug metabolism

Introduction

Cancer is the abnormal transformation and proliferation of cells triggered by underlying genetic anomalies. Oncogenic cells grow indefinitely and invade other tissues and organs leading to cancer. Multiple mechanisms of oncogenic activation exist and multiple subtypes of cancers exist [1]. Anticancer drugs are a class of drugs that show effect in combating malignant cancers by either killing or inhibiting the growth of such cells. Administration of these anticancer drugs is done as single drug therapy or as a multidrug therapy/combination therapy [1, 2]. 5-Fluorouracil is the most commonly used anticancer drug for solid cancers. Functioning as a pyrimidine analogue, 5-Fluorouracil acts as an antimetabolite and disrupts RNA and DNA synthesis, and thereby combats cancerous cells. It is activated inside the cells by multiple enzymes and degraded by Dihydropyrimidine dehydrogenase (DPD) through the pyrimidine degradation pathway. This catabolic activity displayed by the enzyme on 5-FU plays a crucial role in determining its toxicity and efficiency towards 5-FU-based cancer therapies.

5-Fluorouracil

Cancer cells divide rapidly by utilising cellular metabolites. Antimetabolites target this characteristic and act by competing with normal metabolites for the same targets and displacing them competitively [3]. 5-Fluorouracil (5-FU), one such antimetabolite being used in cancer treatment since 1957, is a heterocyclic aromatic pyrimidine analogue with a Fluorine atom at the C-5 position of Uracil [4]. Also known by its trade name Efudex, or Carac, 5-FU is a key anticancer drug used for broad-spectrum antitumor activity and is commonly used in the chemotherapeutic treatments as a sole remedy for solid tumours such as breast, colorectal, lungs, and head and neck cancers [5]. It interferes with DNA synthesis by acting as Uracil analogue and inhibits the essential biosynthesis process such as DNA and RNA

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synthesis by incorporating it into them using the same facilitated transport mechanisms as Uracil and gets converted into several active metabolites intracellularly [3, 6]. These active metabolites disrupt not only RNA synthesis, but also inhibit the action of thymidylate synthase. The half-life of 5-FU is around 4–5 min after an intravenous bolus infusion [7]. The appropriate dosage varies between cases and depending upon the regimen followed and patient's clinical status. It also lowers the infection-fighting blood cells and aids in blood clotting. Hence, administration of any live vaccines while using 5-FU increases the chances of getting infected. Some of the antimetabolites and other anticancer drugs and their mechanism of actions are listed in the Table 1.

Metabolism of 5-fluorouracil

5-Fluorouracil is metabolised by dihydropyrimidine dehydrogenase, which catabolises the drug 5-FU into dihydrofluorouracil (DHFU). DPD catabolizes more than 80% of the 5-FU in the liver mononuclear cells [8]. The activity of DPD is widely distributed in a variety of organs, such as the small intestinal mucosa, and hence, is termed as rate-limiting enzyme for 5-FU. The remaining 5-FU gets converted into fluorouridine monophosphate (FUMP) directly by orotate phosphoribosyltransferase (ORPT) with phosphoribosylpyrophosphate (PRPP) as a cofactor, or indirectly by fluorouridine (FUR) under the action of uridine phosphorylase (UP) and uridine kinase (UK). Furthermore, phosphorylation of FUMP to FUDP occurs, which subsequently phosphorylated into an active metabolite (FUTP), or to FdUDP by ribonucleotide reductase (RR). Phosphorylation or dephosphorylation of FdUDP leads to the formation of the active metabolites FdUTP and FdUMP, respectively. Another pathway, which involves the catalytic conversion of 5-FU to FUDR by thymidine phosphorylase (TP), further phosphorylates it by thymidine kinase (TK) to form FdUMP. The converted FdUMP causes gastrointestinal and myelotoxicity, respectively. FdUMP inhibits thymidylate synthase, leading to activation of DNA damage response pathways [9]. A meta-analysis of six randomised clinical trials performed on patients assigned to bolus 5-FU infusion also showed more haematological toxicity as compared to individuals with continuous intravenous infusion [3]. Moreover, tegafur and capecitabine are the antineoplastic drugs that are metabolised to 5-FU and are given orally for metastatic colorectal cancer [2].

RNA misincorporation

Disruption of normal RNA function occurs due to the misincorporation of the metabolite FUTP. In human colon cancer, a significant correlation was found between lost colonogenic potential and the misincorporation of 5-FU in RNA [10, 11]. This even leads to the toxicity to RNA at multiple levels and inhibition of the maturation of pre-rRNA to mature rRNA [12]. It is also found disrupting the post-transcriptional modification of tRNAs and the activation of the snRNA/protein complexes, which stops the splicing of the pre-mRNA. Even at the low concentration of 5-FU, polyadenylation of mRNA is inhibited [13, 14]. The in vitro studies indicate that by misincorporation of 5-FU, the RNA processing is disrupted, thereby affecting the cellular metabolism and viability.

Thymidylate Synthases are involved in the conversion of deoxythymidine monophosphate (dTMP) from deoxyuridine monophosphate (dUMP) by acting as a catalyst using 5, 10 methylene-tetrahydrofolate (5, 10-CH_2-THF) as the methyl donor. This reaction provides the sole de novo source of thymidylate for DNA replication and repair mechanism. TS functions as the site for both CH2THF and nucleotide binding. FdUMP, the metabolite of 5-FU binds to the nucleotide binding site of the TS, thereby forms a stable ternary complex with the enzyme, and inhibits dTMP synthesis. Depletion of deoxythymidine triphosphate occurs due to

Table 1 Tabular representation of the various types of anticancer drugs, their mechanism of action along with the examples

Type of anticancer drug	Mechanism of action	Examples
Antimetabolites	Disrupting the essential biosynthesis pathways by incorporating the structural analogues	5-Fluorouracil and mercaptopurine
DNA-alkylating agents	Alkylating both the major as well as minor groves of the DNA	Dacarbazine, procarbazine, and temozolomide
DNA cross-linking agents	Inhibits tumour growth by creating a cross-link between the inter- or intra-strands of the DNA	Platinum complexes (cisplatin, carboplatin, oxaliplatin) and nitrogenous mustards (cyclophosphamide, ifosfamide)
DNA-intercalating agents	By directly binding to the bases of the DNA	Anthracyclines (doxorubicin, epirubicin, mitoxantrone, and actinomycin-D)
Topoisomerase inhibitors	Inhibit topoisomerase activity	As irinotecan and etoposide compounds
Antitubulin	Interferes with the dynamics of the microtubules thereby blocking cell division followed by the cell death	Taxanes and vinca alkaloids

subsequent depletion of dTMP, and this leads to the deviation in the levels of other deoxynucleotides via the various mechanisms [15, 16]. This imbalance disrupts the DNA synthesis and repair, and hence, results in lethal DNA damage. Also, the accumulation of dUMP due the TS inhibition leads to increasing the level of deoxyuridine triphosphate (dUTP), which causes the misincorporation of both dUTP and FdUTP into the DNA, and hence, leading to DNA strand breaks and cell death [17, 18].

5-Fluorouracil catabolism in the body

The breakdown of 5-fluorouracil requires DPD enzyme, abundantly present in liver and intestinal mucosa. DPD catabolises 5-FU to 5, 6-dihydro-5-fluorouracil (FUH_2), also known as Dihydrofluorouracil, which then gets converted into α-fluoro-*β*-ureido propionic acid and α-fluoro-*β*-alanine (FBAL), also known as fluoro-ureidopropionate, and fluoro-alanine, respectively as shown in Fig. 1. Upon an investigation of the kinetics of 5-FU and its metabolites in cancer patients by radiolabelled 5-FU, it was identified that more than 60% of the administered drug was excreted out as FBAL in urine within 24 h of administration [7]. Moreover, several patients with the deficiency of DPD have shown symptoms of being at high risk for severe toxicity including diarrhoea and neurotoxicity [3].

Modifications of 5-FU

The overall response rate for this drug as a single agent is quite low. Important modulation strategies, which have, hence, been developed to increase its anticancer activity, also help in overcoming its current resistance. Hence, the strategies, such as decreasing 5-FU degradation, increasing 5-FU activation, increasing the TS inhibition by FdUMP, and using multidrug therapies, make it the ideal drug for the treatment. For optimal binding of FdUMP to Thymidylate synthase (TS), it is necessary to have a high level of intercellular reduced folate CH2THF. In both in vitro [19, 20] as well as in vivo [21] conditions, Leucovorin (LV) has expanded the intercellular concentration of CH2THF in many cell lines identified to have 5-FU toxicity. LV is anabolised to CH2THF after entering the cell through the reduced folate carrier, and is further polyglutamated by folylpolyglutamate synthetase [22, 23]. This enhances the ternary complex stabilised with TS and FdUMP. It helps in enhancing the Thymidylate synthase inhibition by increasing the 5, 10-methylene tetrahydrofolate pool. Seemingly, the degradation of the drug by the DPD enzyme is major in tumours as compared to the anabolic or activating pathways. Therefore, by inhibiting the activity of the DPD enzyme, more drugs can be made to enter the anabolic pathway which may show a satisfactory result. Therefore, the inhibition of DPD activity increases the antitumor potential of the drug which is very vital for the patients whose tumours are resistant due to an increase in the intratumoral DPD activity. Several DPD inhibitors have been introduced clinically.

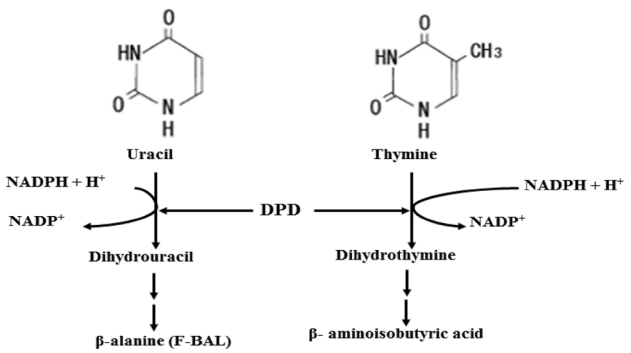

Fig. 1 Degradation pathways of the pyrimidine bases uracil and thymine catalysed by DPD

Inhibitors of DPD

The availability of 5-FU is reduced as it gets degraded into DHFU by DPD. To inhibit this degradation by DPD, a combination of uracil with the 5-FU prodrug ftorafur, in a combination of 4:1 is used, i.e., UFT (Uracil/Ftorafur) [24]. The Eniluracil and 5-chlorodihyropyrimidine, which can be used in inhibiting the degradation of 5-FU by DPD, improve the tumour response rate from 13 to 94% in a rat model [25]. Capecitabine, an oral fluoropyrimidine, which remains unchanged even after absorption through the GI walls, is available to 5′-deoxy-5-fluorouridine (5′DFUR) in the liver by the action of carboxylesterase and cytidine deaminase. 5-DFUR is then converted into 5-FU majorly by thymidine phosphorylase or by uridine phosphorylase as shown in Fig. 2 [26, 27]. Capecitabine has a much higher response rate to colorectal cancer than 5-FU/LV, i.e., 24.8% vs 15.5%, and hence, is preferred more [28].

Methotrexate (MTX) is another drug identified to be converting dihydrofolate (DHF) into tetrahydrofolate (THF) and acting as an antifolate inhibitor of DHFR by inhibiting both purine and thymidine biosynthesis by synergising with 5-FU when administered before 5-FU [29]. The combination of both 5-FU with MTX is more effective in the treatment of colorectal cancer in comparison to bolus single-agent 5-FU [30].

Interferons have shown a negative regulatory effect on the growth of both the normal and malignant cells in the in vitro as well as the in vivo environment, and have found to produce much higher cytotoxicity in various cell lines.

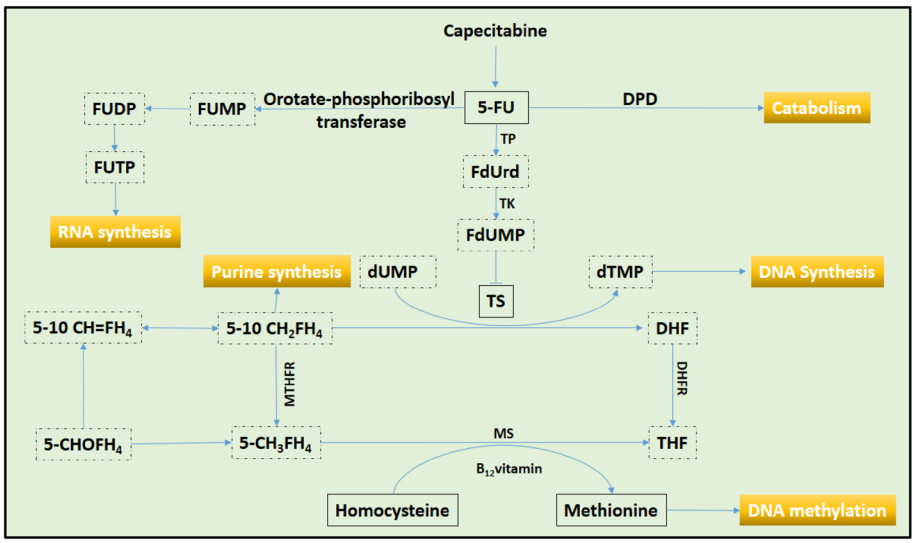

Fig. 2 Metabolic pathways of capecitabine and 5-fluorouracil. 5-Fluorodeoxyuridine (FdUrd), 5–10 methylene-tetrahydrofolate (5–10 CH2FH4), 5-methyltetrahydrofolate (5-CH3FH4), methionine synthase (MS), dihydrofolate reductase (DHFR), 5-formyltetrahydrofolate (folinic acid) (5-CHOFH4), and 5–10 methenyltetrahydrofolate (5–10 CH = FH4)

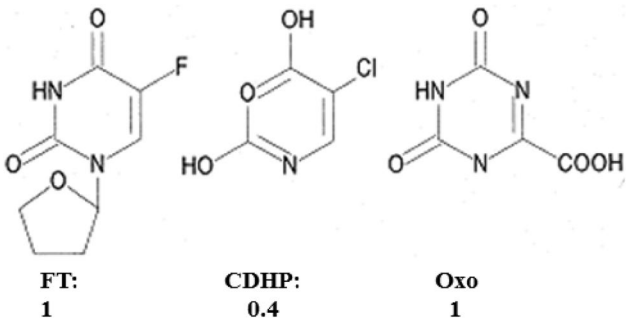

Fig. 3 Composition of S-1 drug

Studies reported that the response rate of the combination of both 5-FU with IFN-α is 42–54% in Phase II clinical trials 57–58; moreover, IFN-α and 5-FU/LV can be given as adjuvant therapy to the patients who have been suffering from the colon cancer [31].

S-1 is a combination of the three drugs (Fig. 3) consisting of the prodrug Tegafur, along with 5-chloro-2, 4-dihydroxypyridine (CDHP)—a DPD inhibitor, and potassium oxonate, in a molar ratio of 1:0.4:1 [32, 33]. S-1 helps in continuous release of 5-FU, with more potent DPD modulation in this combination. Potassium oxonate decreases the potential for typical 5-FU gastrointestinal toxicity, particularly diarrhoea, which is seen in most fluoropyrimidine drugs [34]. CDHP is known to competitively inhibit DPD about 200 times more effectively than uracil as identified in the in vitro studies. The chemical structures of few DPD inhibitors are shown in the Fig. 4. Oxonate (oxo) is an inhibitor of Orotate phosphoribosyltransferase (OPRT) and is distributed majorly in the GI mucosa. After oral administration, oxo selectively improves the gastrointestinal toxicity of the drug by decreasing the production of FdUMP in the GI mucosa. Hence, TS-1 acts as the most promising anticancer drug for advanced or metastatic gastric carcinoma [35].

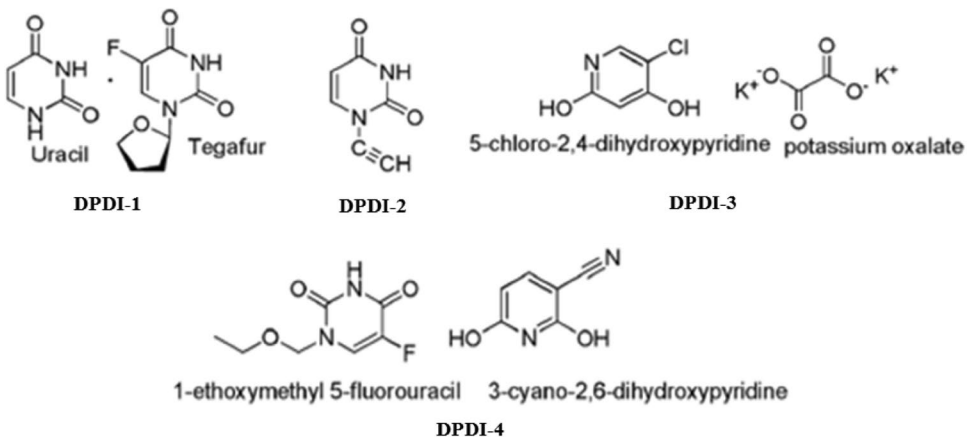

Fig. 4 Chemical structure of DPD inhibitors

Side effects of 5-FU

5-FU has a narrow therapeutic index; 10–26% of patients treated with fluoropyrimidine-based drugs develop an early-onset severe life-threatening fluoropyrimidine toxicity [36, 37]. Some of the side effects caused by 5-fluorouracil, when administered intravenously, are inflammation of the mouth, loss of appetite, and low blood cell count. In severe cases, it can also lead to hair loss and inflammation of the skin (see Fig. 5). Irritation at the site of the application is common when administered as a cream [38, 39].

Dihydropyrimidine dehydrogenase

DPD is an enzyme encoded by the gene DPYD [40]. This enzyme functions by metabolising the two endogenous nitrogen-containing pyrimidines: Thymine and Uracil, and hence, is also known as dihydrothymine dehydrogenase or uracil reductase [41]. In addition to metabolising naturally occurring pyrimidines, it is also known for metabolising the anticancer drug 5-fluorouracil [39]. More than 80% of the metabolism of the administered anticancer drug 5-FU is performed by DPD. The DPD is a rate-controlling enzyme of endogenous pyrimidine and fluoropyrimidine catabolism. Its activity shows a wide range of individual variation [42–44] which is known to have resulted in a broad range of enzymatic deficiencies from partial loss of enzymatic activity, which is seen in 3–5% of the population, to complete loss of the same, seen in 0.2% of the population, which can consequently lead to severe polyvisceral 5-FU-induced toxicity [45, 46].

Mechanism of action of DPD on 5-FU

The administered 5-FU get catabolised into an inactive pharmacological molecule, 5-fluoro-5, 6-dihydrouracil (5-FUH_2), which further gets converted into fluoro-beta-ureidopropionate (FUPA) and subsequently to fluoro-beta-alanine (FBAL) by dihydropyrimidinase (DPYS) and beta-ureidopropionase (UPB1) enzymes, respectively. This anabolic conversion of the 5-FU into a metabolically active nucleotide form is an essential step in the action of 5-FU. However, DPD reduces the availability of 5-FU. Hence, an increased expression of DPD in tumours usually results in the resistance against nitrogen heterocycle containing anticancer agents such as 5-FU [47]. The absence of these enzymes, or any single nucleotide mutations in the gene may show an absolute deficiency in its enzymatic level. DPD polymorphisms, hence, shall result in deficient phenotypes depending upon the mutation and linking them to its polymorphic activity with a total frequency of 3–5%. Insufficient production of this enzyme further leads to the over-accumulation followed by the toxicity of the drug (5-FU). An extensive variety of clinical manifestation and abnormal metabolism of 5-FU is the primary defect found in most of the population.

Function and characterisation of dihydropyrimidine dehydrogenase

Human DPYD gene encodes sequence for an enzyme dihydropyrimidine dehydrogenase, the first rate-limiting enzyme in a three-step metabolic pathway for the degradation of pyrimidine bases [48]. For the synthesis of β-alanine in mammals, this pathway is the major route [49]. Deficiency of the DPD enzyme in the pediatric patients leads to an association with congenital thymine-uraciluria, which is a complex hereditary disease, that shows multiple symptoms such

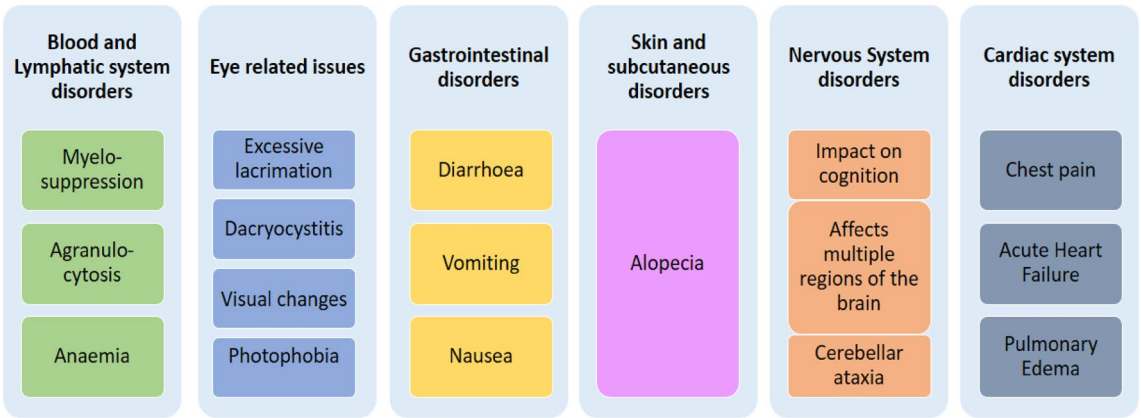

Fig. 5 Various side effects of 5-FU

as epilepsy, slow development, and microcephaly [50, 51]. DPYD gene has been mapped on a 1p22 chromosome and has been found to contain 4400 nucleotides with 23 coding exons which span 950 Kbs. The activity of this enzyme is controlled both at transcription as well as translation level by the transcription factor SP1, SP3, and by microRNA-27a (miR-27a) and microRNA-27b, respectively [52]. Naguib and his colleagues, who studied on DPD activity in humans, and in tumour xenografts, concluded that the activity of these enzymes was high in human liver; however, it is highly variable in tumour patients. Moreover, DPD is known to show a circadian pattern both in human as well as in animals [39, 53]. PMNC-DPD activity is also recognised to display circadian variations and to be involved in differences in blood 5-FU concentrations through an intravenous infusion [54]. A few studies have revealed that DPD shows a variable expression during tumour which justifies the variance in pharmacological responses of tumour towards 5-FU. Those patients having increased expression of DPD have shown resistance to the 5-FU drug even when the level of thymidylate synthase expression was too low [55]. Several studies have proved that there is a positive correlation among the levels of tumoral DPD and resistance against the anticancer agents, while alcohol and smoking have found to decrease 5-FU potential [56].

Polymorphisms of DPYD

Recent studies have indicated that DPD deficiency leads to the genetic aberration in the DPYD comprising of exon skipping and deletion, and missense mutations give the DPD a deficient phenotype [35]. 5-FU-treated patients have been found to be having SNP-associated DPD variants which come under grade 3 and grade 4 toxicities. More than 50 polymorphisms in DPYD have been recognised. Studies have identified multiple novel variations of DPD enzyme in Korean population influencing the effects of 5-FU. Among the novel genetic variants − 832 G > A, − 474 C > T, − 131 C > A, and − 106 G > A are the major SNP mutations. Additionally, it has been found that DPD enzyme activity in the Korean population is found to be slightly higher than those of the Japanese and white population [57]. Similar studies have identified a common relevant polymorphism in the DPYD gene which are DPYD*5A (1627A > G, I543 V) and DPYD*9A along with DPYD*2A (IVS14p 1G > A) in the Chinese population. All of these mutants reduce the enzymatic activity of the DPYD enzyme followed by the increased toxicity of 5-FU [58]. Moreover, among Dutch patients, it has been reported that sequence analysis of the coding exon of DPD has three novel mutations and four rare variants including three missense mutations (c 851G > T, c 1280 T > C, c 2843 T > C), and one intrinsic mutation c2766 + 87 G > A. There are three main DPYD genetic variations, which have been consistently associated with the toxicity of the 5-FU: 2*A rs3918290 G > A, which lead to the skipping of entire exon 14; *13rs5588602 T > G which changes an Ile56Ser amino acid to a flavin-binding domain of the enzyme; and the rs67376798 A > T which causes a change in a Asp949 Val amino acid to an iron–sulphur motif [59, 60]. DPYD IVS14 + 1G > A mutation causes a 165-bp deletion in the mRNA, and thus is formed the DPYD*2A allele followed by the newly truncated protein product, which is formed because this mutation has deficient catalytic activity. Several analyses related to the DPD polymorphism have showed that IVS14 + 1G > A polymorphism is the most frequent among the cancer patients which leads to severe 5-FU toxicity especially in the Caucasian people [61, 62]; DPYD*13 (rs55886062, 1679T > G), DPYD*9A (rs1801265, 85T > C), DPYD*2A (rs3918290, IVS14 + 1G > A), and 2846A > T (rs67376798, D949 V). DPYD 85T > C polymorphism causes the conversion of Cys29Arg with the formation of the DPYD*9A allele (Fig. 6). Patients with DPYD*2A homozygous mutation have shown a complete deficiency of the activity of this enzyme, while in heterozygous case, there are a 50% absences of the enzymatic activity which leads to the life-threatening condition due to 5-FU accumulation in the body [40], whereas DPYD 85T > C polymorphism was 29.4% as a heterozygote mutation and 7.1% as a homozygous mutation [63, 64].

Recent research done on population analysis of DPD catalytic activity has indicated that at least 3–4% of the healthy population could be partially deficient to the DPD enzyme activity. In the absence of the drug treatment, these individual do not show any symptoms, but they could develop a high risk of toxicity if exposed to 5-FU during chemotherapy [65]. Research has shown that 3% of the Caucasian population carries DPYD*2A polymorphism in the exon region, while the Japanese population did not have this variation. However, the mutation in allele DPYD*5 (rs1801159) in 13 exon region was reported to be in 14% of Caucasian, and 15% and 12% in Egyptian and Tunisians, respectively. Among them all, the Japanese population has found to be having the highest frequency of DPYD*5 (rs1801159), i.e., 35% followed by Taiwanese, 21% and African-American, 22.7% [66]. Substitution at 1679T > G in the DPYD gene results in the DPYD*13 allele with the change I560S. Similarly, DPYD 2846A > T mutation causes D949 V change. Both of these polymorphisms have been reported to be associated with low enzyme activity. However, these have been observed to be rare [67, 68].

Clinical manifestation due to the deficiencies

DPD deficiency is a disorder inherited in an autosomal recessive manner. It shows a broad range of phenotypic variability, which ranges from no symptoms to intellectual

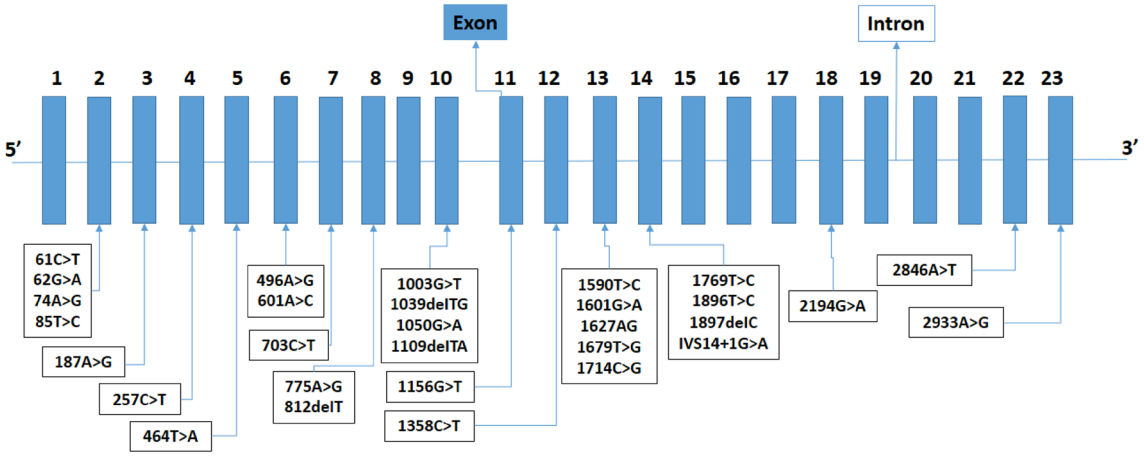

Fig. 6 Schematic representation of the DPD gene structure and polymorphism

disabilities, motor retardation, and convulsions. Additionally, homozygous and heterozygous individuals carrying mutations in this gene can develop severe 5-FU toxicity. The lack of genotype–phenotype correlations and a possibility of other factors playing yet undetected roles in the manifestation of the neurological abnormalities shall make the management and education of asymptomatic DPD-deficient individuals more challenging [69, 70]. Deficiencies in the DPD enzyme may lead to the toxicities of grade 3 or be even more significant due to the 5-FU toxicity. Since the symptom for the toxicity has appeared after the administration of the drug, intensive care and medical interventions are required [71]. Some clinical manifestations that have occurred due to the toxicity of 5-FU are fever, mucositis, stomatitis, nausea, vomiting and diarrhoea. In severe cases, it may lead to neurological abnormalities such as cerebellar ataxia and changes in cognitive functions. Several cases have been recorded in which patients have gone to coma, leukopenia, neutropenia, and possibly thrombocytopenia and anaemia [5]. IVS14 + 1G > A, the DPYD* 2A allele is considered as the most frequent SNP. A G–A mutation in the GT splice donor site during DPD pre-mRNA splicing leads to exon 14 being skipped which is immediately upstream of the mutated splice donor site [72, 73]. Another mutation, 2846A > T, causing D949V change, is located on exon 22 and is known to interfere directly with cofactor binding or electron transport [70, 74]. 1156 G > T, causing E386T change is located on exon 11, resulting in a premature termination codon leading to the formation of a truncated protein, a non-functional enzyme [75]. 2657G > A change, also known as DPYD* 9B allele, is known to cause R886H change leading to a missense mutation on exon 21. This reduces DPD function to 25% of the normal [76]. A 1590T > C change on the DPYD gene promoter, located in the IFNc-binding site, potentially results in lowered DPYD expression [72].

Managing the deficiencies and toxicity

The dosage of 5-FU can be reduced or increased in the following sessions according to the enzymatic status of the DPD observed during the first treatment, and depending on the tolerance level of the patient [77]. If the patient is suspected with the toxicity of 5-FU occurred due to the DPD deficiency, then administration of the drug should be stopped immediately followed by the utilisation of haemodialysis and haemoperfusion to rapidly remove any remaining traces of the drug from the body [71, 78]. Additionally, tumours have also been noted to express variable levels of DPD activity. This may explain the various tumour responses observed in 5-FU. Tumours with relatively low levels of DPD indicate a sensitivity to 5-FU, while tumours with relatively high levels of DPD indicate resistance towards 5-FU. Both the conditions should be considered while devising proper dosage administration. Other alternatives are unblocking the thymidylate synthesis, with the help of the administration of thymidine or uridine, which competitively binds to the drug target. Additionally, to boost the WBC count, colony-stimulating growth factor can be administered in the patients with toxicity. Mainly, an aggressive, holistic, and supportive care is the only way to treat it [71].

Conclusions

Cancer, a disease caused by abnormal proliferation of cells is combated by anticancer drugs, a class of drugs that shows effect in combating these cells by either killing or inhibiting their growth. 5-Fluorouracil, a chemotherapeutic antimetabolite, is the most commonly used anticancer drug for many types of cancers. This Uracil analogue interferes with the essential biosynthetic processes such as DNA and RNA

synthesis and leads to their inhibition. The enzyme DPD catabolises more than 80% of the drug in the liver mononuclear cells into F-ß-alanine. DPD deficiencies are caused by the mutations in the DPYD gene, such as exon skipping and deletions, and missense mutations give a DPD-deficient phenotype. The major DPYD genetic variations that have been consistently associated with 5-FU toxicity are the DYPD 2*A allele (IVS14 + 1G > A mutation in intron 14 coupled with exon 14 deletion) consisting of a G > A variation (rs3918290), the DPYD*13 allele in exon 13 consisting of 167T > G variation (rs5588602), and in exon 22 consisting of 2846A > T variation (rs67376798). Protein products formed due to these mutations have extremely low catalytic activity. At least 3–4% of the healthy population is partially deficient to the DPD enzyme activity which stays undetected in the absence of the 5-FU administration. However, they stay at high risk of developing toxicity towards 5-FU during chemotherapy. Such deficiencies may lead to grade 3 or even higher level toxicity often leading to the death of the patient in homozygous mutants. Multiple studies have identified that high intratumoral activity of DPD markedly decreases the cytotoxic effect of 5-FU. Major modulators to decrease its catabolic activity have also been implemented. Such drugs are used in combination with 5-FU to act as a substrate for DPD, instead of targeting 5-FU.

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Compliance with ethical standards

Conflict of interest Authors declare no conflicting interests.

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