MSAC Application 1760

DPYD genotyping to predict fluoropyrimidine-induced toxicity

PICO Confirmation

Summary of PICO/PPICO criteria to define question(s) to be addressed in an Assessment Report to the Medical Services Advisory Committee (MSAC)

Table 1 PICO for *DPYD* genotyping to predict fluoropyrimidine-induced toxicity in patients with cancer: PICO Set 1

Component	Description
Population	Patients who are about to commence systemic fluoropyrimidine (FP)-based chemotherapy treatment for solid tumour cancer including, but not limited to, colorectal, upper gastrointestinal, head and neck, breast, and pancreatic cancers. Patients who are about to commence systemic FPs as radio-sensitising agents for radiotherapy.
Prior test	No prior test(s).
Intervention	*DPYD* genotyping before the commencement of systemic FP-based chemotherapy (as treatment for solid tumour or as a radio-sensitiser) (to identify patients at risk of severe FP-related toxicity).
Comparator	No pre-FP-based treatment *DPYD* genotyping.
Reference standard	Clinical utility standard: Ability to predict FP-related toxicity.
Outcomes	Safety outcomes: • Adverse events (AEs) related to *DPYD* genotyping. • AEs (or avoided AEs) from any change in patient management (e.g., treatment modifications, monitoring). Test performance: • Prognostic accuracy: Sensitivity, specificity, positive predictive value, and negative predictive value of *DPYD* genotyping to predict toxicity outcomes (e.g., AEs). • Any differences in prognostic accuracy by patient characteristics (e.g., age, sex, ancestry) and cancer characteristics (e.g., type, stage). Change in management: • Change in patient management (e.g., modification of chemotherapy regimen, monitoring). • Any differences in patient management by patient characteristics (e.g., age, sex, ethnicity) and cancer characteristics (e.g., type, stage). Clinical effectiveness outcomes: • Direct: Change in patient-relevant health outcomes (e.g., AEs, the effectiveness of cancer treatment, mortality, morbidity, quality of life) comparing patients who received pre-treatment *DPYD* genotyping versus those who did not receive pre-treatment *DPYD* genotyping. • Indirect: Change in patient-relevant health outcomes (e.g., AEs, the effectiveness of cancer treatment, mortality, morbidity, quality of life) in patients who experienced severe FP-related toxicity and received

Component	Description
	treatment modifications versus patients who received the standard dose treatment. • Any harm from *DPYD* genotyping (e.g., false negatives, test turn-around time (TAT) resulting in potential delay in commencing treatment). • Any differential clinical effectiveness outcomes by patient characteristics (e.g., age, sex, ancestry), and cancer characteristics (e.g., type, stage). Cost-effectiveness outcomes: • Cost per patient with a *DPYD* variant identified. • Cost per patient avoiding severe (≥Grade 3) FP-related toxicity. • Cost per quality-adjusted life year (QALY) gained. • Any differential results by patient characteristics (e.g., age, sex, ancestry), and cancer characteristics (e.g., location, stage). Health system resources: • Cost of *DPYD* genotyping. • Change in the costs associated with the investigation, monitoring, and management of FP-related toxicity (e.g., drugs, hospitalisation) and other AEs, if applicable. • Change in the cost of treatment because of a change in clinical management (e.g., chemotherapy dose modification, alternative non-FP-based treatment). • Total Australian Government healthcare costs.
Assessment question	What is the comparative safety, effectiveness, and cost-effectiveness of pre-treatment *DPYD* genotyping versus no pre-treatment *DPYD* genotyping in patients with solid tumour cancer who are about to commence FP-based chemotherapy?

AE = Adverse events; DPD = Dihydropyrimidine dehydrogenase; FP = Fluoropyrimidine; QALY = Quality-adjusted life year.
MBS = Medicare Benefits Schedule; PASC = PICO Advisory Sub-Committee.

Purpose of application

An application requesting Medicare Benefits Schedule (MBS) listing of *DPYD* genotyping to predict fluoropyrimidine (FP)-induced toxicity in patients with solid tumours who are about to commence a treatment protocol that includes oral or intravenous FP was received from the Royal College of Pathologists of Australasia (RCPA) by the Department of Health and Aged Care.

Clinical claim:

The use of pre-treatment *DPYD* genotyping in patients with solid tumours who are about to commence FP-based chemotherapy as treatment or as a radio-sensitiser results in superior health outcomes compared to no pre-treatment *DPYD* genotyping (p15 of the [application](#)).

PICO criteria

Population

The proposed target population for this application was all patients[1] with solid tumours who are about to commence a treatment protocol that includes oral or intravenous FP. Solid tumours include but are not limited to colorectal, upper gastrointestinal, head and neck, breast, and pancreatic cancers. Population also included all patients requiring FPs as radio-sensitising agents for radiotherapy.

Fluoropyrimidines and dihydropyrimidine dehydrogenase (DPD) activity

FPs are chemotherapy agents used to treat solid tumours. Commonly used FPs are:
- fluorouracil, or 5-fluorouracil (5-FU), administered intravenously, and
- capecitabine, a precursor of 5-FU, administered orally.

Historically, 5-FU was first approved by the United States Food and Drug Administration (US FDA) in 1962 for the treatment of adenocarcinomatous cancers of the colon, rectum, breast, stomach, and pancreas (Longley et al. 2003). Systemic 5-FU is now used to treat solid tumours of the colon, gastrointestinal tract, and carcinomas of the bladder, breast, head, and neck (Jurczyk et al. 2021). The applicant advised (during the PICO development process) that 5-FU may sometimes be used as a radiation sensitiser, with carboplatin, before initiating radiation therapy, especially in gastrointestinal cancers (pre-PASC teleconference, 12 October 2023)(Desilets et al. 2022). Studies have shown superior survival outcomes in head and neck cancer patients treated with concurrent chemoradiotherapy compared with those on radiotherapy alone (Calais et al. 1999; Denis et al. 2003). There was a discussion at the pre-PASC Teleconference that patients requiring radiosensitisation with 5-FU may constitute a second patient population for this testing, and further consultation would be informative regarding the prevalence of the use of 5-FU as a radiation sensitiser, and the radiotherapy modifications required after *DPYD* genotyping.

PASC noted that patients exposed to an FP agent in the context of radiation sensitisation would also potentially experience severe toxicity if they were carriers of the DPYD genetic variants described in this application, and considered that carriers of DPYD variants should be offered the same intervention as other recipients of FP-based therapy. PASC also noted the applicant expert advice that only a small proportion of all patients commencing FP-based therapy would do so for radio-sensitisation. PASC, therefore, advised that the population for pre-treatment DPYD genotyping should also include patients receiving FPs as radio-sensitising agents prior to radiotherapy, and this small subpopulation did not require a separate PICO set.

Lower-dose topical FP is also a treatment for skin cancers. However, *DPYD* genotyping is not proposed for patients about to receive topical FP treatments, only systemic FP (i.e. intravenous or orally administered). Therefore, where this PICO refers to 5-FP-based chemotherapy, this is systemic chemotherapy only.

---

[1] The application defined the population as "all patients" with the characteristics defined in this section but did not restrict the patient group to adults or children. However, the title of the application document suggested that the application aimed to propose *DPYD* genotyping only for adults (application document title "Adults undergoing fluoropyrimidine-based treatment for cancer").

Capecitabine was first approved by the US FDA in 2001 as first-line treatment of metastatic colorectal cancer (Oncology 2001). Capecitabine is now used to treat colorectal, pancreatic, gastric and breast cancer (US FDA 2022). FPs can be used alone or in combination with other treatments, including radiotherapy.

The chemical breakdown of the two commonly used FPs— 5-FU and capecitabine — involves the same metabolic pathway, except that capecitabine first undergoes a three-step enzymatic process to convert into 5-FU in the liver. The breakdown of 5-FU into inactive metabolites requires the dihydropyrimidine dehydrogenase (DPD) enzyme, responsible for the catabolism of 80% of 5-FU in the liver (Longley et al. 2003; Gentile et al. 2016). The remaining 5-FU is either converted to toxic metabolites (about 5%) or excreted in urine (10-20%). If DPD enzyme activity is lower than normal, less 5-FU is converted to the inactive metabolite, and more of the active metabolite accumulates, increasing the risk of FP-related toxicity. The therapeutic effect of 5-FU is mediated by a tiny fraction (1-3%) of the administered dose that is anabolised (smaller fractions converted to complex molecules) into cytotoxic metabolites. These metabolites incorporate into both nuclear and cytoplasmic RNA and DNA. This step exerts a chemotherapeutic effect on tumour cells and normal tissues via the inhibition of RNA processing and function, and DNA synthesis and repair (Ontario Health 2021; White et al. 2021; Dean and Kane 2021 (Update)). The catabolism process evidently plays a significant role in determining the clearance of 5-FU from the system. Therefore, a reduction in the DPD enzyme activity increases drug exposure and 5-FU toxicity (Mattison et al. 2002; Longley et al. 2003; Amstutz et al. 2011).

DPD deficiency and *DPYD* gene variants

The conversion of 5-FU into inactive metabolites by the DPD enzyme, encoded by the dihydropyrimidine dehydrogenase (*DPYD*) gene on chromosome 1, is the rate-limiting step in fluorouracil metabolism. Some *DPYD* gene variants may lead to reduced or completely absent levels of DPD enzymatic activity, known as DPD deficiency (Alsanosi et al. 2014; White et al. 2021). In the case of DPD deficiency, more 5-FU is available for conversion to cytotoxic metabolites, resulting in severe toxicity. Modifications to chemotherapy treatment (e.g., FP dose reduction, switch to alternative treatments) may reduce the risk of severe FP-related toxicity in patients with *DPYD* variant(s) identified through pre-treatment *DPYD* genotyping.

Four well-characterised and prevalent *DPYD* variants are recognised to demonstrate decreased DPD enzymatic function (Gentile et al. 2016; Henricks et al. 2018; White et al. 2021):

No function variants:
- c.1905+1G>A (also known as rs3918290, also known as *DPYD**2A, DPYD:IVS14 1 1G>A), and
- c.1679T>G (also known as rs55886062, *DPYD**13, p.I560S).

Decreased function variants:
- c.2846A>T (also known as rs67376798, p.D949V), and
- c.1129–5923C>G (also known as rs75017182).

Note the variant c.1236G>A was previously thought to be in complete linkage disequilibrium with the c.1129–5923C>G variant and so was considered to be a suitable proxy (Dean and Kane 2021 (Update)). However, a newly published paper raised in the applicant's post-PASC comment reported c.1236G>A is not in complete linkage disequilibrium (Turner et al, 2024) so is not a suitable proxy for the c.1129-5923C>G

variant causally linked to decreased DPD function. The CPIC also updated its guidelines[2] in January 2024 to reflect this new evidence.

DPD deficiency is inherited in an autosomal recessive manner, such that both copies of the *DPYD* gene in each cell contain variants that predispose patients to FP-related toxicity. DPD deficiency has highly variable disease penetrance, meaning that a proportion of people with a *DPYD* variant will exhibit toxicity while some will not (Amstutz et al. 2018). People are categorised as normal, intermediate, or poor *DPYD* metabolisers depending on the combination of the alleles present:

- *DPYD* normal metabolisers carry two 'normal' function alleles resulting in fully functional, normal DPD enzyme activity.
- *DPYD* intermediate metabolisers have reduced DPD enzyme activity as they carry either:
  - one normal function allele plus one no function allele (heterozygous with decreased function variant), or
  - two decreased function alleles (homozygous with decreased function alleles).
- *DPYD* poor metabolisers carry either:
  - two no function alleles (homozygous with no function alleles), or
  - one no function allele plus one decreased function allele (compound heterozygous with no function and decreased function alleles) (Amstutz et al. 2018).

Some patients may carry multiple *DPYD* variants simultaneously. Homozygous patients carrying identical alleles can suffer from reduced or absent DPD enzyme activity. Compound heterozygous patients carrying two or more variants on the same or different alleles can also have variable DPD enzyme activity - more severe when multiple *DPYD* variants are present on different alleles (Knikman et al. 2021).

According to the Clinical Pharmacogenetics Implementation Consortium (CPIC), an international consortium that produces peer-reviewed clinical management guidelines for clinical implementation of pharmacogenetic tests, DPD enzyme activity is reduced by 50% in patients with a decreased function variant (Amstutz et al. 2018).

DPD enzyme activity can be predicted using the *DPYD* gene activity score, which uses the normal/intermediate/poor metaboliser classification as above to calculate a score ranging from 0 (no DPD enzyme activity) to 2 (normal DPD enzyme activity) (Amstutz et al. 2018):

- Normal metabolisers have a *DPYD* gene activity score of 2.
- Intermediate metabolisers have a *DPYD* gene activity score of 1 or 1.5.
- Poor metabolisers have a *DPYD* gene activity score of 0 or 0.5.

The prevalence of poor or intermediate metabolisers varies with the ancestral makeup of the population and occurs in approximately 3-5% of individuals of European descent (Innocenti et al. 2020; Ragia et al. 2023). No other phenotype is associated with *DPYD* intermediate metaboliser status apart from the susceptibility to FP-related toxicity. Complete absence of DPD enzyme activity occurs in only 1-2 per 1,000 of the Western European population and can be fatal upon exposure to an FP (Dean and Kane 2021 (Update)).

---

[2] CPIC Guidelines for Fluoropyrimidines and DPYD (updated January 2024), available at: https://cpicpgx.org/guidelines/guideline-for-fluoropyrimidines-and-dpyd/

The prevalence of *DPYD* gene variant-related DPD enzyme deficiency varies with ancestry, although most published data were based on European populations (Meulendijks et al. 2015; Henricks et al. 2018; White et al. 2021). There was a paucity of data describing the prevalence of *DPYD* variant carriers in Australian and non-Caucasian populations, and no data describing *DPYD* variants in Aboriginal and Torres Strait Islander peoples. Furthermore, while the four aforementioned *DPYD* genetic variants indicate a significantly increased risk of FP-related toxicity in Caucasian populations, their impact on non-Caucasian populations is less known (White et al. 2021). Other variants have also been shown to impact DPD enzyme activity in non-Caucasian populations. For example, variant c.1905+1G>A is shown to increase the odds of toxicity in the Caucasian population by a factor of 5.4, but no data is available on non-Caucasians. On the other hand, a variant c.85T>C did not affect DPD enzyme activity in Caucasians but increased the risk of haematological toxicity in South Indian carriers (White et al. 2021). In Italy, the variant rs1801160 (c.2194G>A, p.Val732Ile, *DPYD**6), considered a 'normal functioning allele' by the CPIC was included in the *DPYD* genotyping panel because it increased the overall risk of toxicity by a factor of 1.73 (Kim et al. 2022). This genetic variant was also the most commonly found in a recent observational study in Greece (Ragia et al. 2023).

According to the applicant, the clinical characteristics (type of cancer etc) are not associated with having specific *DPYD* variants. Germline genotype for autosomal genes is also not expected to differ by demographic characteristics (e.g., age, sex). However, there are differences in the frequency of specific variants between different ancestries. For example, the frequency of the four *DPYD* variants identified by the CPIC as being of specific relevance in patients receiving 5-FU varies from less than 0.1% (South East Asia, Sub-Saharan Africa) to 3.6% (European)[3]. The difficulty is that self-declared ethnicity can be a poor guide to the genetic ancestry of a patient.

Observational data suggest that 40-50% of the patients will develop FP-related toxicity despite pre-treatment *DPYD* genotyping-guided FP dose reduction. This residual toxicity potentially results from other genetic variants that could potentially reduce DPD enzyme activity (Palles et al. 2021; Tsiachristas et al. 2022). Published evidence suggests that pre-treatment *DPYD* genotyping for the four variants recommended by the CPIC has very low sensitivity to predict FP-related toxicity (approximately 4% for any grade toxicity to 12% for grade ≥3 toxicity) but has very high specificity (97-100%) (Lee et al. 2014; Ragia et al. 2023). Expanding this genetic panel to include additional *DPYD* variants significantly increased the sensitivity of predicting grade 3-4 toxicity (Boige et al. 2016; Palles et al. 2021; Maslarinou et al. 2023).

FP-related toxicity and *DPYD* genotyping

Individuals who are *DPYD* intermediate or poor metabolisers cannot metabolise FPs at normal rates and are at risk of severe (grade 3-4), and sometimes potentially life-threatening toxicity typically developing within the first two cycles of standard-dose FP-based chemotherapy. Toxicity may present as bone marrow suppression, haematologic reactions (leukopenia, neutropenia, anaemia, thrombocytopenia), gastrointestinal symptoms (mucositis, diarrhoea, nausea, and vomiting), neurotoxicity and palmar-plantar erythrodysaesthesia (hand-foot syndrome). Generally, while approximately 30-40% of all patients on FPs experience severe toxicity (Van Cutsem et al. 2001), the risk of toxicity events is increased by 1.7- to 5-fold in *DPYD* variant carriers (Meulendijks et al. 2015).

---

[3] https://files.cpicpgx.org/data/report/current/frequency/DPYD_frequency_table.xlsx

Guidelines suggest orally administering an oral formulation of uridine triacetate to treat FP-related severe toxicity (eviQ 2020). The uridine triacetate is a pyrimidine analogue and acetylated pro-drug of uridine. After administration, metabolically, the uridine triacetate converts to uridine. This product is a compound that inhibits cell damage and cell death, thereby acting as an effective antidote to FP-related toxicity. Patients with severe toxicity may require hospitalisation or a follow-up visit with the specialist. Management of FP-related toxicity is usually based on clinical manifestations, for example, patients with FP-related cardiotoxicity presenting with chest pain associated with electrocardiogram (ECG) changes are treated with anti-anginal drugs (Saif et al. 2023). Depending on the severity and type of toxicity event, patients may require anti-diarrhoeal drugs, a visit to the emergency department, FP dose modification, treatment delay, or discontinuation of treatment (eviQ 2021; Ontario Health 2021; eviQ 2022a).

A complete absence of DPD enzyme activity can often be fatal with exposure to FP-based chemotherapy. A systematic review of 35 studies (25 observational and 10 trials) (N= 13,929 patients receiving standard doses of FP-based chemotherapy) found that 566 patients (4.1%) had at least one *DPYD* variant, with c.1129-5923C>G being the most common variant (prevalence 3.9%). The review reported 27 treatment-related deaths (crude treatment-related mortality rate 0.2%), 13 of which were in patients with a *DPYD* variant. The meta-analysis showed that people with variants had a 25.6 times increased risk of treatment-related death (Sharma et al. 2021).

DPYD genotyping before the first exposure to FP-based chemotherapy may help identify patients at risk of DPD enzyme deficiency and avoid severe toxicities in carriers of clinically significant *DPYD* variants.

International guidelines

The 2020 statements by the European Medicines Agency (EMA) and the National Health Service (NHS) in England prompted the development of several national, regional, and institutional guidelines, on pre-treatment DPD deficiency testing, in Europe and the UK (EMA, 2020; NHS England 2020). These statements and the resulting guidelines recommend pharmacogenomic testing for *DPYD* polymorphisms and testing for the lack of the DPD enzyme before commencing FP-based chemotherapies. The guidelines include those published by the Dutch Pharmacogenetics Working Group (DPWG) and the CPIC, and are among the most commonly used guidelines globally (Lunenburg et al. 2020; Wörmann et al. 2020) (Amstutz et al. 2018).

In Australia, the eviQ also follows the CPIC and DPWG guidelines in their recommendations (eviQ 2023b). The Therapeutic Goods Administration (TGA) Advisory Committee on Medicines (ACM) considered the adequacy of the communication in the current Product Information on the risk of 5-FU toxicity related to DPD deficiency at its December 2021 meeting (TGA 2021). The ACM noted that:
- While the laboratory tests for DPD enzyme deficiency (including phenotype and genotype testing) in Australia follow international methods, there are international differences in clinical practice regarding whether to test for DPD activity prior to the start of treatment or to monitor the patient's experience.
- There continues to be uncertainty on the thresholds defining complete and partial DPD deficiency. There are no large studies confirming the best threshold for this biomarker, although blood uracil level > 150 ng/mL is associated with severe enzyme deficiency.
- Factors other than DPD deficiency remain important to minimise toxicity e.g., dosing and monitoring.

Therefore, the ACM advised that:

- DPD testing can be a reasonable clinical choice but need not be mandated. The treating team would consider availability of the test (cost, potential delay to decision on drug and dosing) for the individual patient.
- If a partial DPD deficiency is detected, a reduced starting dose is supported by multiple studies. The wording about dosage used in Europe is appropriate for 5-FU and capecitabine in Australia.

The ACM did not support the inclusion of detailed information on genotypic and phenotypic characterisation in the Product Information (TGA 2021).

The current TGA-approved Product Information documents for 5-FU and capecitabine contain a contraindication for patients with known complete DPD deficiency. There was also a warning about the potential for "severe and potentially life-threatening toxicity in patients with a total or partial DPD deficiency". These documents recommend a reduced starting FP dose to limit FP-related toxicity, and include DPD deficiency as a parameter for dose reduction, in conjunction with other routine measures (Sandoz Pty Ltd 2022; Pfizer Australia Pty Ltd 2023).

In the USA, neither the US National Comprehensive Cancer Network (NCCN) nor the American Society of Clinical Oncology (ASCO) endorse routine pre-treatment *DPYD* genotyping because of sub-optimal prediction of DPD activity in non-European ethnicities (Cascorbi 2023). The US FDA does not recommend *DPYD* genotyping but rather includes a warning in the medicine labels regarding a potential deficiency in DPD activity among people with 'certain' homozygous or compound heterozygous variants (Cascorbi 2023).

Commonly used international guidelines e.g., CPIC and DPWG, recommend pre-treatment *DPYD* genotyping to guide FP dosing, without restricting the eligible population by age (Amstutz et al. 2018; Lunenburg et al. 2020).

Patient characteristics

The prevalence of the known *DPYD* variants is very low in the Western European population, and varies widely across the world. As described above, the presence of certain *DPYD* variant(s) may significantly lead to DPD deficiency, and result in FP-related toxicity. In addition to the presence of *DPYD* variant(s), the prevalence of DPD deficiency, and therefore FP-related toxicity, varies with patient characteristics. These other factors include:

- Age: Observational studies have indicated an association between old age (≥65 years) and developing FP-related toxicity. But data is limited for other age groups including children (Knikman et al. 2021).
- Sex: Observational evidence suggests that capecitabine is catabolised slower in women than in men, so women are at a higher risk than men to develop FP-related toxicity. However, the studies did not adjust for confounders such as body size, warranting the need for further research (Knikman et al. 2021).
- Body composition: Decreased lean body mass could be associated with a higher risk of developing severe FP-related toxicity, but the data are limited with observational studies reporting conflicting findings (Knikman et al. 2021).

- Renal function: Evidence suggests an association of renal impairment and decreased creatinine clearance with a high risk of FP-related toxicity. Some countries, including Australia have incorporated renal insufficiency as a contraindication in the product information leaflets of FPs (EMA 2008; Sandoz Pty Ltd 2022; Pfizer Australia Pty Ltd 2023).
- The type and stage of cancer.
- Concurrent exposure to other anticancer drugs such as platinum-based drugs: FPs are often used in combination with other anticancer drugs that are associated with toxicity (e.g., platinum-based chemotherapies), and can increase the overall risk of toxicity (Ontario Health 2021; White et al. 2021). Future research should account for the confounding effect of chemotherapies on toxicity.

Because several factors potentially influence the risk of developing FP-related toxicity, evidence suggests combining patient characteristics with genotyping strategies to better manage FP dosing (Knikman et al. 2021).

As for age, the application did not explicitly specify the age of the proposed MBS population for pre-treatment *DPYD* genotyping, and did not propose an age restriction on this testing, although there may be differences in clinical utility by age (different cancers, different adverse events, different utility and cost impacts of those adverse events). Clinical experts at the pre-PASC teleconference (12 October 2023) advised that most of the patients qualifying for *DPYD* genotyping were expected to be adults although some would be children. Some children with congenital DPD deficiency have unusual development and medical issues, compared with those without DPD deficiency. The cause of the unusual development in these children is unclear. On the other hand, some children with solid tumours (e.g., colorectal cancer) require FP-based chemotherapy and the treating paediatric oncologists would refer/order *DPYD* genotyping before initiating chemotherapy. Paediatric testing for *DPYD* variants is also included in the NHS test directory (NHS England 2023). The CPIC guidelines also recommend genotyping children given that the FP-related pharmacokinetics are similar in children and adults. The CPIC guidelines further recommend performing early phenotypic (e.g., urine screening of uracil and its degradation products) and/or genetic testing (pre- or postnatal) of offspring of *DPYD* no function variant carriers (Amstutz et al. 2018).

The population for the genotyping test will include adults and the paediatric population.

PASC noted the applicant had proposed this testing without any age restriction, and that the applicant expert (a medical oncologist) reported that there was no reason why children should metabolise 5-FU differently from adults and that the CPIC guidelines did not restrict using DPYD genotyping by age group, nor describe different dose adjustment. PASC considered there may be a potential impact of dose adjustments (post-test) on cancer outcomes in children but noted the applicant clinical expert advice that in general and irrespective of age, the starting dose adjustment recommended is 50% of the target dose and up-titrated if tolerant, as per eviQ guidelines[4]. PASC advised the proposed MBS population for pre-treatment DPYD genotyping should not be restricted by age and would therefore include adults and the paediatric population.

---

[4] https://www.eviq.org.au/clinical-resources/side-effect-and-toxicity-management/prophylaxis-and-treatment/1744-dihydropyrimidine-dehydrogenase-dpd-enzyme

Number of eligible patients

The application estimated the number of eligible patients for *DPYD* genotyping under the proposed MBS listing based on the total number of cancers detected in the respective year, and the proportion of patients who currently receive FP-based chemotherapy.

Direct sources to collect this information are the Australian population-based cancer registries. But the development of these registries is still in its infancy, because they do not yet collect data on cancer treatments. In the future, national administrative healthcare data linkages will provide such information including the stage of cancer at diagnosis, treatment, and recurrence of cancer (Cancer Australia 2023). One of the indirect sources for such information on FP-based chemotherapy use is to review the TGA's Database of Adverse Event Notifications. The database is a voluntary reporting mechanism, that includes reports on adverse reactions to FPs. These notification records are incomplete making it difficult to estimate toxicity rates or patient characteristics. Therefore, there is lack of robust Australian data describing the overall incidence of severe toxicity related to the administration of FPs, and rates of toxicity-associated morbidity and mortality (White et al. 2021).

Utilisation of *DPYD* genotyping will be based on the number of people receiving FP-based chemotherapy in Australia. This data on FP use may be extracted from the national pharmaceutical dispensing dataset (e.g., PBS data).

Another method to estimate the utilisation of *DPYD* genotyping is by calculating the incidence of cancers that require FP-based chemotherapy. Table 2 presents the incidence of common cancers requiring FP-based chemotherapy based on the Australian Institute of Health Welfare (AIHW)'s projections of cancer incidence over the next 10 years (AIHW, 2023) (list of cancer type derived from the application (Attachment with the application 'Estimated Utilisation').

Table 2   Projected incidence of common cancers potentially receiving FP-based chemotherapy, over the next eight years and first six years of the proposed MBS listing

Cancer type[a]	2024	2025	2026[b]	2027	2028	2029	2030	2031
Breast	20,771	21,186	21,599	22,015	22,434	22,854	23,278	23,709
Colorectal	16,693	16,921	17,138	17,351	17,677	17,988	18,285	18,576
Head and neck	5,401	5,498	5,600	5,701	5,800	5,900	6,000	6,100
Oesophageal	1,794	1,832	1,871	1,910	1,953	1,996	2,038	2,080
Pancreas	4,752	4,930	5,110	5,295	5,468	5,643	5,818	5,995
Stomach	2,693	2,750	2,803	2,858	2,916	2,978	3,031	3,087
Total cancers	52,104	53,117	54,121	55,130	56,248	57,359	58,450	59,547

Source: *Table compiled during PICO development using the same method as in the application but updated with the most recently available data (AIHW Cancer data in Australia, 2023 (AIHW, 2023)) rather than the AIHW 2022 data used in the application* (Table 5, p8 of the application attachment titled "*Estimated utilisation*" submitted by the applicant).
AIHW = Australian Institute of Health and Welfare; FP = Fluoropyrimidine; MBS = Medicare Benefits Schedule.
Head and neck cancers excluded lip cancers.
[a] The list of cancers does not include numbers for extrahepatic bile duct cancers (type described in the application) because the most recent AIHW update did not provide numbers for these cancers and the application only provided projections until 2026.
[b] The first year of the proposed MBS listing of *DPYD* genotyping may be 2026 assuming MSAC advice in November 2024.

As described in the application, only a proportion of the cancers will be treated by FP-based chemotherapy (Sheet 1 of the attachment "*DPYD* Genotyping estimates"). Table 3 presents the estimated number of patients eligible for *DPYD* genotyping in the first year of the proposed MBS listing — approximately 22,000 patients— based on the assumptions described in the application. The applicant indicated that the assumptions used in the application were based on clinical experience (pre-PASC teleconference, 12 October 2023).

Table 3 Estimated number of patients eligible for *DPYD* genotyping in 2024 in the application (based on the proportion of people with cancer receiving FP-based chemotherapy)

Cancer	Stage	Annual incidence (N) [A][f]	% by stage [B]	Incidence in 1st year (N) [C=AxB]	% on FP [D]	% relapse [E]	% on FP after relapse [F]	Sub-total [G=CxD+ CxExF]	TOTAL
Colorectal[a]	All	16,693							
	1		25%	4,173	–	–	–		
	2		25%	4,173	10%	20%	80%	1,085	
	3		25%	4,173	80%	32%	25.6%	3,680	
	4		25%	4,173	80%	–	–	3,339	
									8,104
Breast[b]	All	20,771							
	2		50%	10,386	–	40%	60%	2,493	
	3		10%	2,077	20%	60%	60%	1,163	
	4		20%	4,154	30%	–	–	1,246	
									4,902
Upper GI	All	9,239							
Oesophagus[c]		1,794	100%	1,794	50%	–	–	897	
Stomach[d]		2,693	100%	2,693	80%	–	–	2,154	
Pancreas[e]		4,752	100%	4,752	80%[g]	–	–	3,802	
									6,853
Head & Neck		5,401		5,401	40%	–	–		2,160
Total									**22,019**[h]

Source: *Table compiled during PICO development based on Sheet 1 of attachment "DPYD genotyping estimates" but updated with AIHW (2023) incidence data (application used AIHW 2022 data).* The applicant reported that the proportions of incident cancers receiving FP therapy were based on clinical expert opinion. *Note that there were a few inconsistencies between the text description and the cell values in tab "Sheet 1" of Excel Workbook titled "DPYD genotyping estimates" of the application.*

5-FU = 5-Fluorouracil; AIHW = Australian Institute of Health and Welfare; FP = fluoropyrimidine; GI = gastrointestinal.

[a] Each stage of colorectal cancer represents about 25% of total cases. 10% of stage 2 colorectal cancers have adjuvant FP, 20% of all stage 2 relapses 80% of which have FP. 80% of stage 3 colorectal cancers have adjuvant FP (20% refuse), and 40% of these relapses of which 80% have FP. 80% of stage 4 colorectal cancers have FP (20% too ill or palliative care only).

[b] Most initial breast cancers are stage 1 (20%) or stage 2 (50%), with 10% stage 3 and 20% metastatic at first diagnosis. No stage 2 breast cancers have primary chemotherapy including FP. 40% of stage 2 relapse and 60% of these have FP at some time. 20% of stage 3 (locally advanced) breast cancers have primary FP, 60% of stage 3 relapse 60% of which have FP at some time. About 30% of all metastatic breast cancers have capecitabine or 5-FU at some time.

[c] For the oesophagus, about 50% of cases have either neoadjuvant chemotherapy or chemotherapy at relapse.

[d] For gastric (stomach) cancer, about 80% of cases have neoadjuvant FP-based chemotherapy. All those who relapse have FP.

[e] For pancreatic cancer, FOLFIRINOX (chemotherapy combination of leucovorin calcium (folinic acid), fluorouracil, irinotecan hydrochloride, and oxaliplatin) or gemcitabine/paclitaxel are the preferred regimens used in about 80% of all cases.

[f] Annual incidence is based on (AIHW 2023) projections for the year 2024.

[g] There were a few inconsistencies between the text description and the cell values in tab "Sheet 1" of Excel Workbook titled "*DPYD* genotyping estimates" of the application. For example, the text description reported that 80% of all cases of pancreatic cancer used FP-based chemotherapy whereas 40% was used in the calculation.

[h] The application estimated that a total of 16,776 cancer patients would require FP-based chemotherapy, based on the AIHW 2022 projections. The total estimate in Table 3 was higher (22,019) for a few reasons, e.g., Table 3 used AIHW 2023 incidence projections and 80% rather than 40% for pancreatic cancer patients using FP-based chemotherapy.

Intervention

The intervention proposed by the applicant was *DPYD* genotyping before the commencement of FP-based chemotherapy (to identify patients at risk of severe FP-related toxicity).

The goal of pre-treatment *DPYD* genotyping is to reduce the risk of severe toxicity by identifying patients at risk of DPD enzyme deficiency, to change patient management by reducing FP dose or changing the treatment (Innocenti et al. 2020). Lowering the FP dose in patients with DPD deficiency will help maintain therapeutic plasma concentrations of 5-FU and its metabolites, thereby decreasing the risk of severe toxicity while maintaining treatment efficacy (Ontario Health 2021).

The applicant reported that genotyping of *DPYD* variants is typically conducted using a polymerase chain reaction (PCR)-based method on DNA extracted from peripheral blood cells (4 ml Ethylenediaminetetraacetic acid (EDTA) sample) (p6 in the applicant's supporting PICO document titled "Adults undergoing fluoropyrimidine-based treatment for cancer" (MSAC 2023), (Ontario Health 2021))(Keen et al. 2022), or buccal swab or saliva samples (Mayo Clinic Laboratories 2023). The applicant reported that there are several commercially available kits for testing all the variants described in this application. The kits include the VeriDose *DPYD* panel (Agena Biosciences), the Elucigene® *DPYD* kit (Yourgene), and the TaqMan® OpenArray® Pharmacogenomics (PGx) Panel (Thermo Fisher Scientific). The applicant also indicated that these kits do not need to be TGA-registered and would be validated by pathology providers in accordance with the National Pathology Accreditation Advisory Council (NPAAC) standards as in-house In Vitro Diagnostic. All these commercially available kits use amplicon (PCR)-based technology (source: applicant's document titled "Questions from the Assessment Group Final" supplied to the Department for the pre-PASC teleconference, 12 October 2023).

PASC noted consultation comments had raised concerns that testing should not be restricted to blood samples. PASC considered that no restriction on sample types was proposed, and that testing could therefore use any appropriate sample type without requiring adjustment to the proposal.

Most European countries and the United Kingdom (UK) publicly reimburse routine pre-treatment *DPYD* genotyping (SSCPT 2020; Wörmann et al. 2020; de With et al. 2023). A few others (e.g., France, Denmark, and the Netherlands) provide additional reimbursement for *DPYD* phenotype testing (de With et al. 2023). In Canada, pre-treatment *DPYD* genotyping is publicly funded in Quebec for the four common variants (Ontario Health 2021), but the funding status is unclear in other provinces. In Australia, *DPYD* genotyping is currently not publicly funded on the MBS but is available in some medical genetics laboratories (e.g., MyDNA), hospitals (e.g., the Peter MacCallum Cancer Centre), and local area health networks (e.g., the Southern Adelaide Local Health Network. Genotyping is requested by medical oncologists as in-patient services (i.e., state-funded) or as private individuals (out-of-pocket expenses) (pre-PASC teleconference, 12 October 2023). However, the extent of this funding or the number of patients covered is unknown. Regarding the current practice around genotyping in Australia, the applicant stated during the pre-PASC teleconference that it is performed in patients who have already initiated FP-based chemotherapy. The applicant also indicated that pathology laboratories are generally not informed of the specific clinical context of the *DPYD* genotyping requested. *DPYD* genotyping available through some commercial

providers costs approximately $140- $160, with costs varying between the laboratories offering the test.[5,6,7,8]

The proposed *DPYD* genotyping will be performed before initiating FP-based chemotherapy, administered orally or intravenously. The proposed listing includes genotyping for *at least* the four *DPYD* variants that are recommended by international guidelines including by the CPIC (Amstutz et al. 2018), the DPWG (Lunenburg et al. 2020), European consensus guidelines (Wörmann et al. 2020), and the EMA (EMA 2020), and adopted in several European countries:

- c.1905+1G>A
- c.1679T>G
- c.2846A>T
- c.1129-5923C>G

Note that the guidelines published before 2024 describe c.1236G>A. as a proxy for c.1129-5923C>G. However new evidence was published in 2024 that the proxy SNP is no longer considered appropriate. *PASC noted (out-of-session) that the item descriptor's practice note had been updated to reflect the new evidence that these SNPs are not in complete linkage disequilibrium (Turner et al., 2024).*

PASC further considered (out-of-session) that at least some of the relevant studies likely used the proxy SNP rather than c.1129-5923C>G itself. However PASC considered that the applicability of evidence using the proxy SNP was not necessarily greatly reduced for the HTA, because Turner et al. reported unlinked genotypes between the SNP and its proxy to be very rare ("0.223% of subjects with c.1236G>A lack c.1129-5923G>A"). PASC therefore advised that literature using the proxy SNP still be considered for inclusion in the DCAR's assessment, including an assessment of the applicability of the identified evidence as per usual.

The proposed MBS item descriptor, by stating "genetic testing for four or more variants …. ", leaves the potential to expand the panel to other variants as more data emerge on genotypes that can increase the risk of FP-related toxicity, or are found to be more common in Australia (see "Proposal for public funding").

PASC considered that more research may be needed to determine relevant alleles in non-Caucasian populations, such as Aboriginal and Torres Strait Islanders and other minorities. PASC considered that the testing being for "four or more variants" would include the proposed variants and also permit any other variants with relevant clinical utility (including those in minority populations, and those discovered in the future) to be included as necessary.

The turnaround time for *DPYD* genotyping is approximately 5-6 days (p5 of the application), with some delays in rural and regional areas (eviQ 2023b).

A second tier of testing (using e.g., gene sequencing) after genotyping was not proposed in this application, although there was some evidence on two-tiered testing in the literature. Some observational studies conducted standard PCR testing followed by Sanger sequencing of the entire coding region of the *DPYD* gene in a subset of patients to "confirm the genotype or detect rare variants" (Coenen et al. 2019). The Scottish regulatory authority—the Healthcare Improvement Scotland (SHTG)— also recommends Sanger

---

[5] Australian Clinical Labs: https://www.clinicallabs.com.au/cancer-services/pharmacogenetics/
[6] Sonic Genetics: https://www.sonicgenetics.com.au/our-tests/all-our-tests/dpyd-screen/
[7] Austin pathology: https://www.austinpathology.org.au/test-directory/2034
[8] MyDNA: https://www.mydna.life/dpydtesting/

single gene sequencing following PCR amplification (SHTG 2020). However, a follow-up investigation like NGS may not be required to validate the PCR results because the accuracy of the PCR testing in detecting the four variants is well studied, and the literature suggests a robust functional significance of the proposed variants (Amstutz et al. 2018; EMA 2020; Hamzic et al. 2020; Lunenburg et al. 2020). The applicant also confirmed that *DPYD* sequencing is not currently conducted in Australia following a PCR-based test.

DPYD genotyping will be conducted in a National Association of Testing Authorities (NATA) accredited diagnostic laboratory in accordance with the NPAAC guidelines (ACSQHC 2022).

Gene sequencing

An alternative approach is sequencing the gene or exons, for example using Next Generation Sequencing (NGS). This has the potential to reveal additional rare pathogenic or likely pathogenic genetic variants, although would likely also be more costly than testing of the four SNPs. For example, a recent study explored genotype-phenotype correlations by conducting exon sequencing of the *DPYD* gene in a group of 94 patients. The study found eight common and 23 rare variants, collectively accounting for 42.5% of all DPD deficiencies observed in the study population (De Luca et al. 2022).

PASC noted that the applicant had proposed genotyping of four SNPs for the intervention, but that Sanger sequencing, next generation sequencing (NGS) and single nucleotide polymorphism (SNP) arrays were other methods that could be used to identify these and potentially other DPYD variants. PASC considered that non-targeted sequencing methods were likely to identify variants of unknown significance (VUS) or variants with no demonstrated effect on DPD enzyme reduction, and potentially at greater testing cost. PASC also considered the applicant's advice that genotyping is the 'gold standard' method for identifying the four proposed genetic variants. Overall, PASC advised the intervention should be genotyping of at least the four proposed SNPs, and gene sequencing should not be added to the HTA as a scenario of the intervention.

PASC noted that the method-agnostic genotyping item descriptor would not exclude laboratories from choosing to use non-targeted methods, and in that case use of virtual panels could ensure only the variants described in line with current guidelines were reported thereby preventing implications from the detection of VUSs.

Phenotypic testing

Some phenotypic methods can identify decreased DPD enzyme activity and are endorsed by some international guidelines (Lunenburg et al. 2020) and publicly subsidised in some European countries (de With et al. 2023). However, the applicant did not propose phenotypic methods as interventions based on their drawbacks (Knikman et al. 2021) (p8 of the [application](#)).

Phenotypic methods and their drawbacks are:
- **Determining pre-treatment dihydrouracil to uracil ratio in blood:** The test is based on DPD enzyme converting its endogenous substrate uracil (U) into dihydrouracil (DHU). Testing U or DHU concentrations in blood plasma entails ultra-performance liquid chromatography–tandem mass spectrometry (UPLC–MS/MS) assay using the patient's blood sample (Jacobs et al. 2016). The pre-treatment ratio of serum concentrations of DHU to U – the DHU/U ratio – has been investigated as a phenotypic measure of systemic DPD activity (Meulendijks et al. 2017). A low ratio would

indicate low DPD enzyme activity and predict high 5-FU plasma levels. But there is no consensus on the exact association of U and DHU levels with FP-related toxicity (de With et al. 2022; Paulsen et al. 2022). The test is recommended by some international guidelines that were based on one single centre observational study published in 2017. This study showed a significant association of plasma U and DHU levels above 16 ng/ml with FP-related toxicity (Meulendijks et al. 2017; European Medicines Agency (EMA) 2020). Newer retrospective data have shown its correlation with the prediction of severe toxicity, but prospective data are lacking. Also, this method requires expensive laboratory facilities. Studies have shown conflicting results of association of pre-treatment DHU/U ratio with FP-related toxicity:

- o A Dutch prospective observational study found no correlation between phenotypic testing using pre-treatment U levels and DHU/U ratio, and FP-related toxicity. The study also found no significant relationship between U levels and DHU/U ratio and DPD enzyme activity in peripheral blood mononuclear cells. This study indicated plausible between-centre differences in the plasma uracil levels, underlining that its measurements could be sensitive to pre-analytical errors and may be affected by circadian rhythm and food intake (de With et al. 2022).
- o One French non-randomised cohort study found a significantly lower incidence of ≥3 toxicity in patients who received FP doses based on both *DPYD* genotyping and phenotypic testing using plasma U and DHU levels), compared to those who received standard doses. However, the study did not provide the thresholds/cut-offs of plasma U and DHU levels used to alter the dose. Further, the study population comprised a higher number of participants with partial (CPIC's intermediate metaboliser) deficiency in the intervention arm compared with the standard dose arm, biasing the results in favour of the combined genotyping-phenotypic approach (Boisdron-Celle et al. 2017).
- o Another French study found that *DPYD* genotyping poorly correlated with plasma U and DHU levels. But the study did not test the predictive value of either method in estimating the risk of severe FP-related toxicity (Pallet et al. 2020).
- o Given the uncertainty around the standardisation of plasma U and DHU measurement and the lack of agreed clinical cut-offs for the phenotypic tests, comparing this phenotypic testing method with no testing may be difficult in this assessment. Based on evidence that pre-treatment U and DHU levels can accurately predict DPD enzyme activity, it is publicly subsidised in two European countries including France and the Netherlands (Laures et al. 2022). But other countries like Italy do not support measuring plasma U and DHU levels to predict FP-related toxicity. This reluctance is primarily due to a lack of standardisation of testing processes (Bignucolo et al. 2023).
- **Uracil breath test:** This method primarily evaluates pyrimidine catabolism. The method measures the amount of modified compound of carbon dioxide (CO_2)— $^{13}CO_2$ —in exhaled breath after ingestion of a defined amount of ^{13}C-uracil. Since CO_2 is one of the final metabolites in pyrimidine catabolism, its availability in breath would reflect the rate of uracil catabolism. This method does not differentiate between deficiency in DPD enzymes or of other enzymes involved in the conversion of an uracil intermediate compound 1C to carbon dioxide. While it differentiates between DPD-deficient and non-deficient patients, it does not predict toxicity (Knikman et al. 2021). It is also difficult to employ in a large-scale DPD screening implementation.

- **DPD enzyme activity in peripheral blood mononuclear cells (PBMCs):** This ex-vivo test uses radioenzymatic assays to measure the levels of DPD enzyme activity directly (Chazal et al. 1996; Pluim et al. 2015). Evidence suggests the robustness of this test in predicting DPD enzyme activity. But there is no consensus on the threshold to determine DPD deficiency. Studies have also shown inter-patient variability of test results. These issues make it difficult to compare and interpret DPD activity in PBMCs. Further, the sample processing may be laborious, time-consuming, and expensive, and the technology might not be employed in all hospitals (Etienne-Grimaldi et al. 2023).
- **5-FU degradation rate in peripheral blood mononuclear cells:** This method lacks prospective validation, which makes it difficult to assess clinical utility. Further, it is not proven if it is correlated with severe toxicity. It also requires special equipment and may be laborious, time-consuming, and expensive (Knikman et al. 2021).

None of the phenotypic tests described above are listed on the MBS.

Given the drawbacks of phenotypic testing, the lack of evidence around their utility and analytical accuracy in predicting FP-related toxicity, phenotypic testing was not considered as a scenario of the intervention.

PASC noted DPD deficiency has a highly variable disease penetrance, and genotyping has low sensitivity (4-12%) for the four proposed variants, therefore genotyping may not detect many patients at risk of toxicity. PASC noted the applicant did not propose phenotypic methods, and the applicant's clinical expert informed not being aware of phenotypic testing being routinely done in clinical practice. PASC further noted the pros and cons of genotyping and phenotypic methods in the target population to predict FP-related toxicity in Paulsen et al (2022)[9]. PASC acknowledged the applicant's reasons for not adopting phenotypic tests in standard clinical practice, which included: phenotypic methods cannot be reliably reproduced, phenotypic method results are affected by other factors such as kidney diseases, systemic DPD activity is hard to monitor using phenotypic tests e.g., blood plasma levels of uracil, and that there is a lack of consensus for a threshold to define metaboliser status based on phenotypic tests. PASC noted the applicant also stated that offering phenotypic testing in Australia could be challenging because of geographical access limitations (samples required for phenotypic testing may be unstable, and the logistics involved may not be feasible). Overall, PASC considered that the analytical accuracy of proposed phenotypic testing methods was not known, and therefore advised that phenotypic testing should not be added as a scenario of the intervention.

Comparator

The comparator will be no pre-treatment *DPYD* genotyping.

The application specified that currently no toxicity-related testing is publicly funded for patients who will initiate systemic FP-based chemotherapy for solid tumours (p10 of the application). All patients receive standard-dose systemic FP-based chemotherapy unless they experienced a previous episode of toxicity or are deemed unfit to receive full-dose chemotherapy following medical assessment by an oncologist.

---

[9]Paulsen NH et. al. (2022). *DPYD* genotyping and dihydropyrimidine dehydrogenase (DPD) phenotyping in clinical oncology. A clinically focused minireview. *Basic & Clinical Pharmacology & Toxicology* 131(5): 325-346.

The applicant also confirmed that the current demand for phenotypic testing nationally is not available as there is no national "clearing house" of demand, outcomes, and indications for testing. Furthermore, a pathology laboratory would usually not be informed of the clinical context in which the test is requested. The applicant also stipulated that the MSAC may wish to seek input from current requestors of the test. (p2 of the document received in confidence from the applicant titled "Questions from the assessment group_Final").

PASC agreed that the comparator should be no pre-FP-based-treatment DPYD genotyping.

Reference standard (for investigative technologies only)

- Clinical utility standard: Ability to predict FP-related toxicity.

The application did not specify any non-clinical, clinical reference or clinical utility standard for comparative analytical performance of the proposed *DPYD* genotyping. The accuracy of PCR testing in detecting the four variants is well studied, and the literature suggests a robust functional significance of the proposed variants (EMA Amstutz et al. 2018; 2020; Hamzic et al. 2020; Lunenburg et al. 2020).

The sensitivity of genotyping the four variants in predicting FP-related toxicity is low. To predict the response to treatment in *DPYD* variant carriers, a clinical utility standard is necessary (MSAC guidelines for preparing assessments TG 11.2, p103-105). The proposed clinical utility standard is the ability of *DPYD* genotyping to predict FP-related toxicity.

PASC agreed that the clinical utility standard should be the ability of DPYD genotyping to predict FP-related toxicity.

PASC advised that an assessment of the comparative analytical performance of genotyping against a non-clinical reference standard was not required. PASC agreed with the applicant that a comparison between genotyping and non-targeted methods like Sanger sequencing will not be useful because the latter is known to be less sensitive than PCR, which is the most commonly used genotyping method. PASC, therefore, advised that because genotyping was considered the 'gold standard,' comparison against a non-clinical reference standard was not required.

Outcomes

Safety outcomes:

- Adverse events (AEs) related to *DPYD* genotyping.
- AEs (or avoided AEs) from any change in patient management (e.g., treatment modifications, monitoring).

Test performance:

- Prognostic accuracy: Sensitivity, specificity, positive predictive value, and negative predictive value of *DPYD* genotyping to predict toxicity outcomes (e.g., AEs).
  The application did not include prognostic accuracy of *DPYD* genotyping as an outcome. Published evidence suggests *DPYD* genotyping has very low sensitivity but high specificity to predict FP-

related toxicity (Lee et al. 2014; Ragia et al. 2023). Low sensitivity would result in a high number of false negative results, potentially missing patients at high risk of developing FP-related toxicity. On developing FP-related toxicity these patients will require treatment, and thereafter dose titration or therapy switching based on the type of cancer.

- Any differences in prognostic accuracy by patient characteristics (e.g., age, ancestry), and cancer characteristics (e.g., type, stage).

Change in management:

- Change in patient management (e.g., treatment modifications, monitoring).
- Any differences in patient management by patient characteristics (e.g., age, ancestry), and cancer characteristics (e.g., type, stage).

Clinical effectiveness outcomes:

- Direct: Change in patient-relevant health outcomes (e.g., AEs, the effectiveness of cancer treatment, mortality, morbidity, quality of life) comparing patients who received pre-treatment *DPYD* genotyping versus those who did not receive pre-treatment *DPYD* genotyping.
  Large prospective observational data found that *DPYD* genotyping-guided dose adjustment significantly reduced the prevalence of severe toxicity in *DPYD* variant carriers compared with historical controls (Henricks et al. 2018).
- Indirect: Change in patient-relevant health outcomes (e.g., AEs, the effectiveness of cancer treatment, mortality, morbidity, quality of life) in patients who experienced severe FP-related toxicity and received treatment modifications compared to receiving the standard dose.
- Any harm from *DPYD* genotyping (e.g., false negatives, test turn-around time (TAT) resulting in potential delay in commencing treatment noting that the test turnaround time is approximately 5-10 days, with delays in rural and regional areas (eviQ 2023b)).
- Any differential clinical effectiveness outcomes by patient characteristics (e.g., age, ethnicity), and cancer characteristics (e.g., type, stage).

Cost-effectiveness outcomes:

- Cost per patient with a *DPYD* variant identified — c.1905+1G>A, c.1679T>G, c.2846A>T, or c.1129–5923C>G.
- Cost per patient experiencing severe (≥Grade 3) FP-related toxicity avoided.
- Cost per quality-adjusted life year (QALY) gained.
- Any differential results by patient characteristics (e.g., age, sex, ancestry), and cancer characteristics (e.g., location, stage).

Health system resources:

- Cost of *DPYD* genotyping.
- Change in the costs associated with the investigation, monitoring, and management of FP-related toxicity (e.g., drugs, hospitalisation) and other AEs if applicable.
- Change in the cost of treatment because of a change in clinical management (e.g., dose modification, alternative non-FP-based treatment).
- Total Australian Government healthcare costs.

The PASC agreed with the list of outcomes described in the PICO.

Assessment framework (for investigative technologies)

Figure 1 Generic assessment framework showing the links from the test population to health outcomes
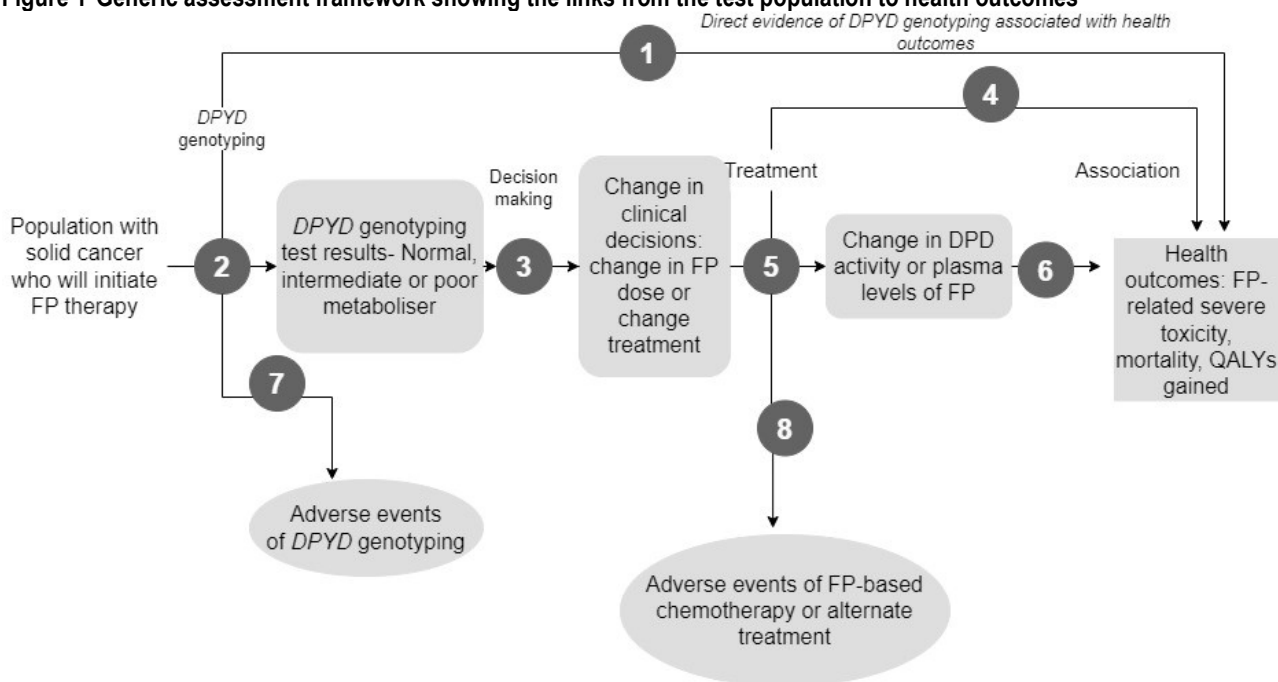
DPD = dihydropyrimidine dehydrogenase enzyme; FP = fluoropyrimidine; QALYs = Quality adjusted life years.
Figure notes: 1: direct from test (pre-treatment *DPYD* genotyping) to health outcomes evidence; 2: test accuracy; 3: change in treatment/management: including dose adjustment of FP-based chemotherapy or switching to alternate treatment; 4: influence of the change in management on health outcomes; 5: influence of the change in management on intermediate outcomes, for example increase in DPD activity or therapeutic drug monitoring results indicating optimal plasma levels of FP; 6: association of intermediate outcomes with health outcomes such as decreased incidence of severe toxicity events and decreased mortality; 7: adverse events due to *DPYD* genotyping; 8: adverse events due to FP-based chemotherapy.

Assessment questions mapped to the assessment framework:

1. What is the safety and effectiveness of pre-treatment *DPYD* genotyping versus no pre-treatment *DPYD* genotyping in patients with solid tumours including, but not limited to colorectal, upper gastrointestinal, head and neck, breast, and pancreatic cancers? (Direct evidence)
2. What is the diagnostic yield of *DPYD* genotyping in patients with solid tumours? Do the results of *DPYD* genotyping predict a treatment effect modification with fluoropyrimidines?
3. What proportion of patients with a *DPYD* variant receive a change in treatment/management, such as dose adjustment of FP-based chemotherapy or switching to alternate treatment?
4. What is the effectiveness of the FP dose adjustment/ change in therapy vs no FP dose adjustment/change in therapy for health outcomes such as mortality, severe FP-related toxicity, and quality of life?
5. What is the effectiveness of FP dose modifications/change in therapy vs no FP dose modification/change in therapy for intermediate health outcomes such as DPD enzyme activity in

the blood, and therapeutic levels of FP in the blood (ascertained through therapeutic dose monitoring)?
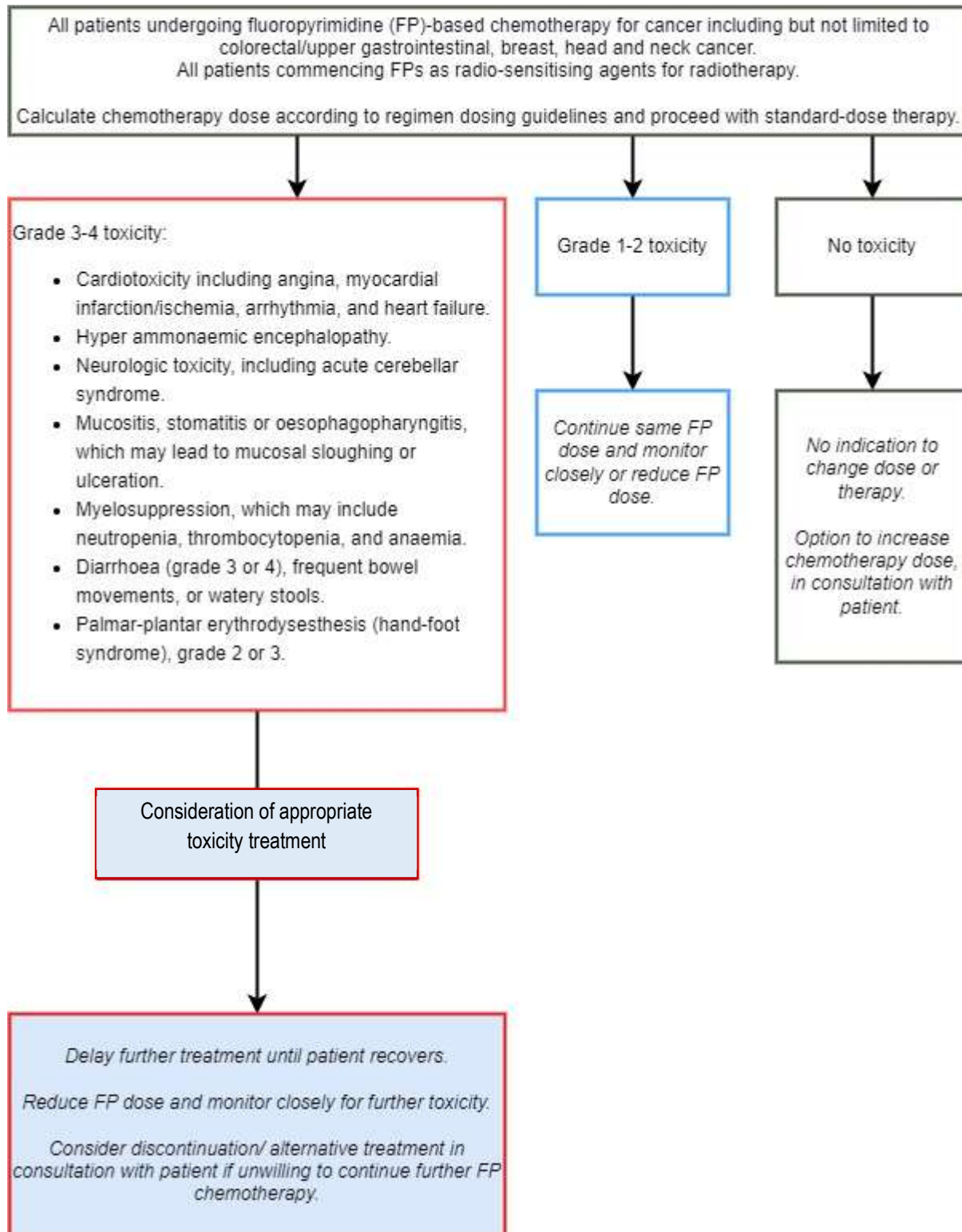
6. How strong is the association between intermediate health outcomes such as DPD activity, and incidence of severe toxicity events and mortality?

7. What is the safety of pre-treatment *DPYD* genotyping vs no *DPYD* genotyping testing for false negatives and test turn-around time (TAT) resulting in potential delay in commencing treatment?

8. What is the safety of dose adjustments to FP-based chemotherapy or changed treatment for mortality, morbidity, quality of life?

The PASC agreed with the assessment framework described in the PICO.

Clinical management algorithms

Figure 2 presents the current clinical algorithm in the application.

Figure 2 Current clinical algorithm (no routine pre-treatment *DPYD* genotyping)

At present there is no routine pre-treatment *DPYD* genotyping or any phenotypic testing. Changes to FP-based chemotherapy doses are based on FP-related toxicity events as and when encountered. These toxicity events may lead to hospitalisation, dose reduction, treatment delay, or treatment discontinuation/switch (Ontario Health 2021). Other treatments for toxicity include:

- Mucositis, stomatitis or oesophagopharyngitis: Saline, benzydamine or anaesthetic mouthwashes, systemic analgesics, topical, systemic or intralesional steroids (eviQ 2023d).
- Myelosuppression (e.g., neutropenia, thrombocytopenia, anaemia): Antibiotics, platelet or blood transfusions (eviQ 2023c; eviQ 2023a).
- Diarrhoea: Loperamide, diphenoxylate and atropine, octreotide (eviQ 2022b).
- Palma-plantar erythrodysesthesia: topical application of an emollient containing urea 10%, topical wound care, cold compresses, moisturisers, topical corticosteroids and systemic analgesics (eviQ 2021).

An oral formulation of uridine triacetate is used as an antidote for patients who develop FP-related toxicity. The exogenous uridine in uridine triacetate would compete with 5-FU for incorporation into RNA, diluting the toxic effects of high 5-FU levels (Dean and Kane 2021 (Update)). Currently, while it is approved by the US FDA, there are no registered antidotes for FP overdose or overexposure in Australia. Uridine triacetate can only be obtained through the Special Access Scheme from pharmaceutical companies (eviQ 2020). As of October 2023, the antidotes for FP-related toxicity (such as uridine triacetate) are not listed on the Australian Pharmaceutical Benefits Scheme. Management of FP-related toxicities depends on the type and severity of the toxicity, as presented in the FP-related toxicity and *DPYD* genotyping section. However, uridine triacetate is rarely used and is difficult and expensive to obtain.

PASC also noted that treating toxicity with uridine triacetate was rarely used, difficult to source and expensive. The clinical experts present at the pre-PASC teleconference informed that clinicians frequently discuss the benefits of *DPYD* genotyping with prospective patients and recipients of FP-based chemotherapy. This practice is in line with the Australian eviQ guidelines for DPD deficiency, which recommends clinicians to "discuss *DPYD* gene testing with all patients who are going to start fluoropyrimidines, with the decision to conduct testing made between the clinician and the patient" (eviQ 2023b).

Figure 3 presents the proposed clinical management algorithm using *DPYD* genotyping in guiding the dose of FPs or discontinuing FPs. The dose guidance in the proposed algorithm reflects the recommendations from international authorities including the CPIC and DPWG. FP dose adjustments are based on gene activity scoring, which is similar across the guidelines except the DPWG recommends phenotype testing to guide treatment decisions for poor metabolisers, while the CPIC recommends avoiding FP-based chemotherapy in these cases (Table 4). In Australia, FP dose recommendations following *DPYD* genotyping were also based on these two international guidelines (eviQ 2023b).

On the current clinical management algorithm, PASC considered that "adults" should be changed to "all patients." PASC advised to include patients requiring radio sensitisation and exposure to FP agents.

PASC noted that a subgroup of patients will receive alternative treatment based on the level of DPD enzyme activity. Therefore, PASC requested adding a box for alternative therapy into the proposed and current clinical management algorithm for patients who have no enzyme activity (poor metabolisers), or reduced enzyme activity (intermediate metabolisers).

PASC noted that uridine triacetate was included in the proposed algorithm but omitted from the current algorithm, which incorrectly implied it was part of the proposed intervention. PASC requested this be clarified by either removing it from the proposed algorithm or adding it to the current algorithm. PASC noted (out-of-session) the applicant's comments that uridine triacetate is not mandated because it requires clinical consideration regarding timing of administration and drug availability on a case-by-case basis. PASC therefore considered it was appropriate to update "Treat toxicity with uridine triacetate" to "Consideration of appropriate toxicity treatment". PASC also noted (out-of-session) the applicant's comments that alternative therapy options were tumour specific, hence in the clinical algorithm "alternative therapy" was changed to "alternative therapy (if available).

Figure 3 Proposed clinical management algorithm after introducing pre-treatment *DPYD* genotyping.
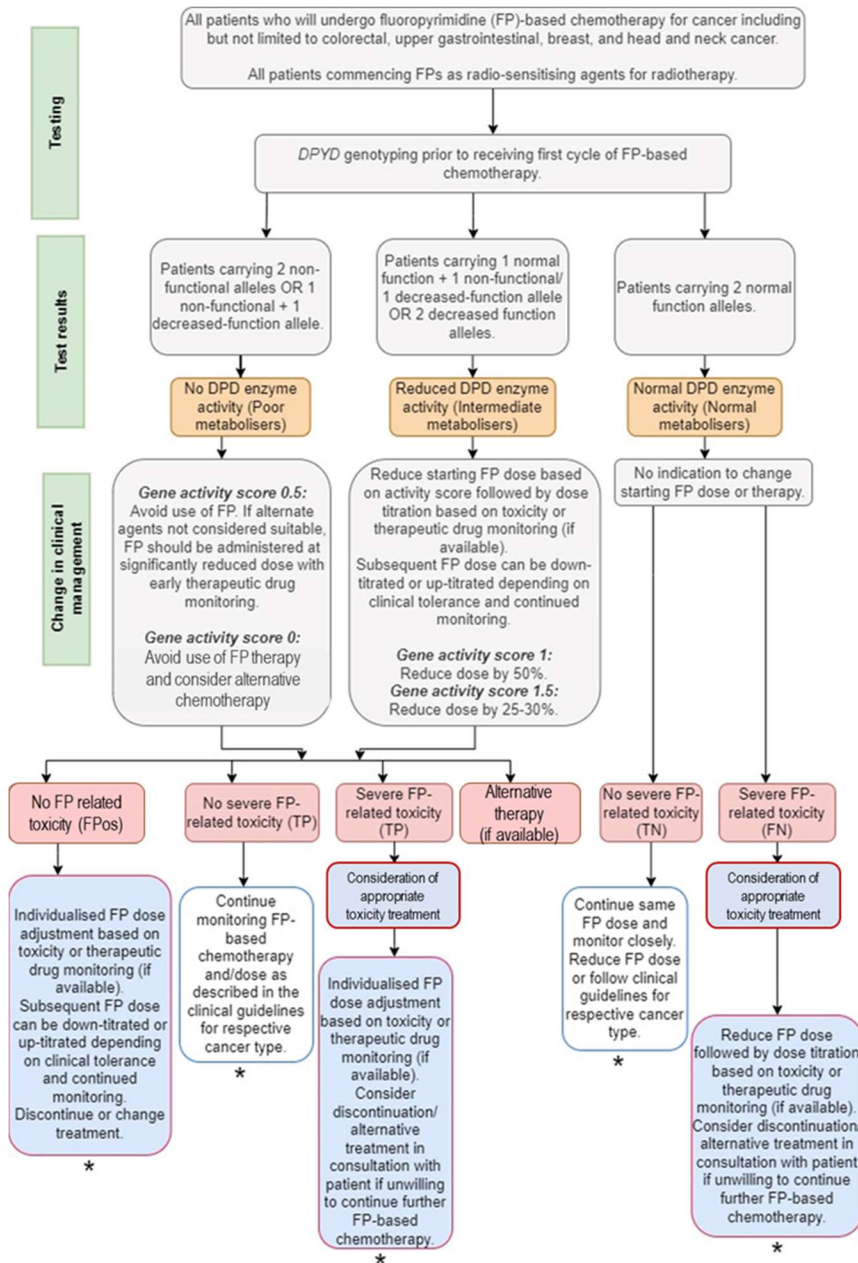
Source: adapted from Figure 2, (p15) of the PICO set document submitted by the applicant and additional changes suggested by PASC in the PASC meeting held on 7 December 2023. The algorithm in this document contains downstream effects of encountering true positives, false positives, true negatives, and false negatives—features that were not included in the application.

Changes suggested by the PASC and added to the proposed algorithm: Population referred to as 'patients' as opposed to 'individuals'. Population included patients commencing FPs as radio-sensitising agents. A process step was added for alternative therapy for patients who have no enzyme activity (poor metabolisers), or reduced enzyme activity (intermediate metabolisers).

DPD = dihydropyrimidine dehydrogenase enzyme; FN = False negative; FP = fluoropyrimidine; FPos = False positive; TN = True negative; TP = True positive

*Patients will be followed up based on the current clinical algorithm when they experience FP-related toxicity.

Table 4 presents the CPIC guideline for FP dose adjustments following *DPYD* genotyping (Amstutz et al. 2018).

Table 4 Description of *DPYD* genotype and DPD activity and FP dose recommendations

CPIC gene activity score	Phenotype: *DPYD* metaboliser type	Example *DPYD* genotype (c. variants are for NM_000110.4(DPYD))	Approximate DPD enzyme activity	Diplotype	DPD deficiency classification	CPIC recommended starting FP dose
2.0	Normal	wildtype /wildtype	100%	Homozygous wildtype	DPD sufficient	Label-recommended
1.5	Intermediate	wildtype/ c.2846A>T	75%	Heterozygous variant	Partial DPD deficiency	Reduce dose by 50%
1.0	Intermediate	wildtype/ c.1905+1 G>A, or c.2846A>T / c.2846A>T, or c.2846A>T /*wild type /c.1905+1 G>A, or c.2846A>T / c.2846A>T, or c.2846A>T / c.1129–5923C>G	50%	Heterozygous, or homozygous variant, or compound heterozygous	Partial DPD deficiency	Reduce dose by 50%
0.5	Poor	c.1905+1 G>A / c.2846A>T	25%	Compound heterozygous	Partial DPD deficiency	Avoid use or strongly reduce dose by 75%
0.0	Poor	c.1905+1 G>A/ c.1905+1 G>A, or c.1905+1 G>A /c.1679 T>G	0%	Homozygous variant, or compound heterozygous	Complete DPD deficiency	Avoid use[a]

Source: Table compiled during PICO development adapted from the CPIC guidelines by Amstutz et al. (2018).
CPIC = Clinical Pharmacogenetics Implementation Consortium; DPD = dihydropyrimidine dehydrogenase; FP = fluoropyrimidine.
The CPIC activity score is calculated as the sum of the two lowest individual variant activity scores.
'Wildtype' refers only to the genotype at the 4 positions described in the relevant guidelines.
[a] Known complete DPD deficiency is a contradiction in the TGA-approved Product Information for 5-FU and capecitabine.

Under the proposed clinical management, the findings of *DPYD* genotyping and subsequent gene activity scoring will inform FP dosing (Amstutz et al. 2018). However, irrespective of the genotype results, all patients receiving systemic FP-based chemotherapy should be closely monitored for potential adverse events and toxicity. This is because even when all DPYD genotyping results are accurate, some patients identified as 'normal' metabolisers based on genotype may still develop toxicity (i.e., false negatives in terms of ability of the DPYD test result activity score to predict FP-related toxicity that may be related to other genetic/ epigenetic factors). On the other hand, it is possible that patients identified as 'intermediate' or 'poor' metabolisers may not develop the predicted extent of toxicity (i.e., false positives in terms of ability of the DPYD test result activity score to predict FP-related toxicity). Such patients will require up-titration of the FP dose to achieve a desirable and tolerable therapeutic effect. The clinical care for patients switching to another chemotherapy regimen would follow the clinical guidelines for the specific cancer type.

Proposed economic evaluation

The application claimed that pre-treatment *DPYD* genotyping was superior to the comparator (no pre-treatment *DPYD* genotyping). Therefore, the economic evaluation will be a cost-effectiveness analysis (cost per patient with a *DPYD* variant identified and cost per patient avoiding severe (≥grade 3) FP-related toxicity, and cost-utility analysis (cost per QALY gained) (Table 5).

Modelling cost per QALY only considering adverse events is anticipated to be simpler and more reliable than estimating cost per QALY also taking into account changes in clinical management and health outcomes (progression-free survival and overall survival). Including the change in management and health outcomes would be indication-specific and require evidence on the efficacy of using fewer lines of therapy, as well as likely requiring more assumptions and therefore potentially producing highly uncertain results. However, restricting the model to only the more certain impacts on utilities (i.e., due to adverse events) would require evidence that reducing the dose of FP-based chemotherapy for patients with *DPYD* variants did not impact other health outcomes (e.g. overall survival). Therefore, a stepped presentation of the economic results (i.e. including all QALY types) will be used.

PASC noted the Assessment Group recommended limiting the scope of the economic model to the impact on adverse events, including death and disutilities associated with adverse events. PASC considered the economic model should be developed following the evidence as per usual. PASC acknowledged that modelling the impact of changing dosing and chemotherapy regimens on survival outcomes would be less certain but considered that restriction to only the more certain impacts on utilities (i.e., due to adverse events) would require evidence that reducing the dose of FP-based chemotherapy for patients with DPYD variants did not impact other health outcomes (e.g. overall survival). PASC advised that in the absence of robust evidence that there was no consequence of dose reduction on cancer outcomes, excluding the impact on cancer outcomes would also produce highly uncertain results. PASC considered that stepped presentation of the economic results (i.e. including all QALY types but with less attention to the less certain ones) may sufficiently clarify the uncertainty to be informative for decision-making.

While the economic model would aim to use data from a heterogeneous combination of cancer types, an exemplar cancer—specifically colorectal cancer— could be used for modelling if sufficient evidence was not available for heterogeneous cancers to inform the model. An exemplar cancer could be used noting that the economic model would still require modelling the complete range of chemotherapies indicated, the sequence of delivery of these therapies, the adverse events from each therapy, the changes in management from *DPYD* genotyping (dose changes, chemotherapy changes and sequence changes) and the subsequent impact on health outcomes. These data might be sourced from heterogeneous cancer types to maximise the inputs required for the model. The rationale for using exemplars is primarily because the clinical management is dependent on the type of cancer. The clinical experts indicated that the most common indications for receiving FP-based chemotherapy in Australia are colorectal and breast cancers (pre-PASC teleconference on 12 October 2023). Therefore, the majority of patients who will utilise *DPYD* genotyping will comprise patients with these two cancers. There is a paucity of data on whether some *DPYD* variants are found in certain types of cancer but not others. It is unlikely that the predisposing *DPYD* genetic variants found in colorectal and breast cancers are not found in other cancers treated with FPs like head and neck cancer. Published systematic reviews of the effectiveness of *DPYD* genotyping in predicting FP-based toxicity have included studies with patients on FP-based chemotherapy mostly for

colorectal and breast cancers, but also include other cancer types such as head and neck, upper gastrointestinal, pancreatic, and oesophageal cancers (Meulendijks et al. 2015; Glewis et al. 2022). Furthermore, published meta-analyses provide pooled estimates based on the type of *DPYD* variant as opposed to cancer type, assuming that the health outcomes are similar across the cancer types (Meulendijks et al. 2015).

PASC noted that the pre-PASC PICO also sought advice on whether an exemplar cancer could be used for the economic model. PASC considered that if an exemplar approach were acceptable colorectal cancer appeared a reasonable choice, because it was the most common cancer that required an FP-based chemotherapy regimen as the main therapy, as opposed to other types of cancers in which FPs are used intermittently. PASC noted that the published economic evaluation by Henricks et al. (2018)[10] included a heterogeneous population with cancers, so it would not be an appropriate guide for an assessment based on an exemplar. PASC noted the Assessment Group's advice that modelling multiple cancers would be complex, primarily because DPYD genotyping would be used by patients with heterogeneous cancer types that have different types and sequences of chemotherapies. However, focusing on one cancer type would still require modelling the complete range of chemotherapies indicated, the sequence of delivery of these therapies, the adverse events from each therapy, the changes in management from DPYD genotyping (dose changes, chemotherapy changes and sequence changes) and the subsequent impact on health outcomes. PASC noted that these complexities in the treatment algorithms will therefore introduce significant uncertainty in the economic model, whether an exemplar is used or not. PASC considered that data may be scarce to inform such a large model, and advised that a heterogeneous combination of cancer types may be used rather than an exemplar, as this would allow the most data to inform the model. The financial analysis would still examine the whole patient population.

Table 5   Classification of comparative effectiveness and safety of the pre-treatment *DPYD* genotyping, compared with no pre-treatment *DPYD* genotyping, and guide to the suitable type of economic evaluation.

Comparative safety	Comparative effectiveness			
	Inferior	**Uncertain[a]**	**Noninferior[b]**	**Superior**
Inferior	Health forgone: need other supportive factors	Health forgone possible: need other supportive factors	Health forgone: need other supportive factors	? Likely CUA
Uncertain[a]	Health forgone possible: need other supportive factors	?	?	? Likely CEA/CUA
Noninferior[b]	Health forgone: need other supportive factors	?	CMA	CEA/CUA
Superior	? Likely CUA	? Likely CEA/CUA	CEA/CUA	CEA/CUA

CEA = cost-effectiveness analysis; CMA = cost-minimisation analysis; CUA = cost-utility analysis.
? = reflect uncertainties and any identified health trade-offs in the economic evaluation, as a minimum in a cost-consequences analysis.
[a] 'Uncertainty' covers concepts such as inadequate minimisation of important sources of bias, lack of statistical significance in an underpowered trial, detecting clinically unimportant therapeutic differences, inconsistent results across trials, and trade-offs within the comparative effectiveness and/or the comparative safety considerations.
[b] An adequate assessment of 'noninferiority' is the preferred basis for demonstrating equivalence.

---

[10] Henricks LM, 2018, *Annals of Oncology*, "*DPYD* genotype-guided dose individualization of fluoropyrimidine therapy: A prospective safety and cost-analysis on *DPYD* variants."

Proposal for public funding

The applicant proposed the public funding of *DPYD* genotyping targeting at least four *DPYD* gene variants on the MBS.

Searches conducted during the PICO development stage did not identify any relevant in-progress applications with the Medical Services Advisory Committee or the Pharmaceutical Benefits Advisory Committee.

Table 6 presents the proposed MBS item descriptor from the application form *with modifications to reflect PASC's advice*. The proposed MBS fee for *DPYD* genotyping was $188. The applicant advised that the proposed fee included a commercially available kit, specimen collection and transportation, sample processing and consumables, technician labour, genomic analysis, interpretation and report generation, and pre-analytical steps required such as DNA extraction (pre-PASC teleconference, 12 October 2023).

Table 6   Proposed MBS item descriptor incorporating PASC's advice

Category 6 – PATHOLOGY SERVICES Genetics	Group P7 –

MBS item AAAAA
 	
Genetic testing for four or more variants in the *DPYD* gene to predict fluoropyrimidine-induced toxicity in a patient, where:
 	
   a. the service is requested by a specialist or consultant physician; and
   b. the service is conducted prior to the initiation of chemotherapy, or radio-sensitisation, with a fluoropyrimidine, administered systemically; and
   c. genotyping is conducted to detect at least four *DPYD* variants that can lead to reduced or completely absent dihydropyrimidine dehydrogenase (DPD) activity.
 	
Once per lifetime.	

Fee: $188.00 Benefit: 75%=$141.00 85%=$159.80	

Explanatory note:
 	
  - The variants analysed should be selected in line with current guidelines, and must include at least:
      • NM_000110.4(DPYD):c.1905+1G>A
      • NM_000110.4(DPYD):c.1679T>G
      • NM_000110.4(DPYD):c.2846A>T
      • NM_000110.4(DPYD):c.1129-5923C>G
 	
      Published evidence for the association of presence of these four variants with severe fluoropyrimidine-related toxicity, is based on people with Caucasian ancestry only.	

Source: p10 of the application , with changes suggested by PASC in the PASC meeting on 7 December 2023.
DPD = dihydropyrimidine dehydrogenase enzyme; *DPYD* = dihydropyrimidine dehydrogenase gene; MBS = Medicare Benefits Schedule.

PASC considered the item descriptor should include the reason for DPYD genotyping, and that this was to predict fluoropyrimidine-induced toxicity.

PASC noted the descriptor was proposed to state the FP was "administered either orally or intravenously". PASC considered that simpler wording would be "administered systemically".

PASC considered the MBS item descriptor should be method agnostic, to futureproof the item and permit laboratories to choose an appropriate method. Therefore, "using Polymerase Chain Reaction" should be removed from the MBS item descriptor.

PASC considered the policy proposal that the MBS item specifies the item can be requested by a specialist or a consultant physician was reasonable. This should, therefore, be added to the item descriptor.

To reflect PASC's advice on the patient population as described above, the MBS item should not be restricted by age, and should include patients receiving FPs prior to radio-sensitisation.

PASC considered that the variants recommended for testing may change over time and that the inclusion of the four proposed variants (based on current guidelines) in the testing will be ensured through the external quality assessment (EQA) process. PASC considered that specifying the variants in the item descriptor was therefore not necessary, and doing so would make the item less future-proofed. PASC advised the four currently specified variants should be moved to an explanatory note, and the item descriptor should state "at least four DPYD variants that can lead to reduced or completely absent dihydropyrimidine dehydrogenase (DPD) activity". PASC noted (out-of-session) that the variants in the practice note had been updated based on new clinical evidence that c.1236G>A was no longer accepted as a suitable proxy for c.1129-5923C>G (Turner et al., 2024).

PASC considered the item descriptor was phrased adequately to allow testing for newly identified gene variants in future as evidence emerged on new genetic variants associated with severe FP-related toxicity and in population groups with ancestry other than Caucasian. PASC considered the explanatory note should further state that these variants are based on studies in Caucasian populations only, to convey to laboratories that additional variants may also be appropriate in patients of ancestries with other relevant variants.

PASC noted queries as to the potential need for a second test in children later in life if they develop a second cancer that also requires treatment with FP-based chemotherapy. PASC considered that in such cases the person's experience during their first FP therapy would already reveal whether or not they would develop toxicity with FP exposure, and germline genotype does not change in a person's lifetime. PASC therefore agreed the test should be once-per-lifetime, as proposed.

PASC noted the proposed fee for the MBS item was $188. PASC considered that this fee was similar to the range of current fees for this testing in private laboratories in Australia ($95-$160, as described by eviQ). PASC noted the fee was also similar to that for MBS item 73397 (Fee $200) for characterisation of variants in the CALR and MPL genes, although much higher than MBS item 73317 ($36) for detecting genetic mutations for haemochromatosis. On balance, PASC considered the proposed fee of $188 appeared reasonable.

Summary of public consultation input

PASC noted and welcomed consultation input from 9 organisations and three individuals, one of whom were consumer and two were health professionals. The 9 organisations that submitted input were:

- Therapeutic Goods administration (TGA)
- National Pathology Accreditation Advisory Council (NPAAC)
- The Royal College of Pathologists of Australasia (RCPA)
- Australasian Society of Clinical and Experimental Pharmacologists and Toxicologists (ASCEPT)
- Australian Pathology

- PathWest Nedlands Clinical Biochemistry Clinical Pharmacology and Toxicology
- Society of Hospital Pharmacists of Australia (SHPA)
- Consumer Representatives from Melbourne Genomics Health Alliance
- Australian Genomics

The consultation feedback received was supportive. The consultation feedback generally considered *DPYD* genotyping as providing significant physical and mental benefits through this robust genotyping methodology. Australian genomics asserted that there is no benefit of cascade testing to family members.

The consultation feedback raised a few concerns, predominately in relation to the population not including paediatric patients. Another concern was raised in relation to the intervention that the samples should not be limited to blood but also include cheek cell samples for easy accessibility for the regional population.

Australian Genomics consider the risk of lack of uptake of the proposed service due to a lack of awareness and suitable educational programs.

Clinical need and public health significance

The main benefits of public funding received in the consultation feedback included the ability to provide the appropriate dose of fluoropyrimidine and prevent the occurrence of severe and sometimes life-threatening adverse effects from the toxicity of the drugs. *DPYD* testing benefits the individual as they can have confidence in the treatment being personalised to their physiology (including lower doses of medication) and there is less stress and concern regarding treatment. Australian Genomics stated that "reimbursement may act as a catalyst for the adoption of *DPYD* testing as best practice". Other benefits included fewer hospitalisations and lower health costs due to fewer adverse events.

The SHPA pointed at the lack of availability of Uridine Triacetate (VISTOGRAD) in Australia which is used in FP overdose or overexposure. They stated that in a time-critical scenario such as a severe FP toxicity, sourcing uridine triacetate currently presents financial and logistical challenges. Therefore, they advised that public funding of *DPYD* testing to determine DPD enzyme deficiency prior to FP treatment initiation was essential in reducing harm for individuals initiating FP-based treatment.

The main disadvantages included that the absence of four *DPYD* variants characterised in the proposed service does not eliminate the risk of toxicity and patients can still experience the severe adverse side effects. Short delays in the treatment commencement while waiting for the results of the proposed test.

Indication(s) for the proposed medical service and clinical claim

The consultation feedback ranged from strongly agreeing to disagreeing with the proposed population. The individual advocated to include paediatric population along with adults in the proposed population. They considered that fluoropyrimidines may have a role in paediatric solid tumours such as hepatoblastoma, nasopharyngeal carcinoma and ependymoma. The Society of Hospital Pharmacists of Australia (SHPA) indicated to include all adults undergoing FP treatment for malignancy and not limit this test to solid tumours only.

The consultation feedback ranged from strongly agreeing to disagreeing with the proposed comparator. One individual commented that phenotypic testing was not satisfactory in predicting patients at risk for fluoropyrimidine-induced toxicity.

Pathwest Laboratory Medicine disagreed with the proposed comparator - "no *DPYD* genotyping" and indicated including an established phenotypic test as a comparator. Pathwest Laboratory Medicine

disagreed with the service descriptor and advocated for combined genotype and phenotype testing for improved sensitivity and specificity of the proposed service.

Australian Genomics acknowledged the technicalities involved with phenotypic testing and cannot be easily implemented as a routine test.

The consultation feedback ranged from agreeing to strongly agreeing with the clinical claim.

Cost information for the proposed medical service

The consultation feedback ranged from disagreeing to strongly agreeing with the proposed service descriptor. However, one individual supported the need of a provision in the descriptor to include additional *DPYD* variants as they become known in Aboriginal and Torres Strait Islander and minority populations in future.

The consultation feedback ranged from agreeing to strongly agreeing with the proposed service fee. One individual indicated that the proposed fee would be extensively offset, as fewer patients would have severe side effects and associated health costs. Australian pathology indicated the proposed fee for the proposed medical service was deemed adequate at the current time.

Additional comments

One respondent highlighted the risk of litigation claims against the treating clinician because death occurred as an adverse reaction to therapy when a test to predict these adverse reactions was available.

Consumer Feedback

Consumer Representatives from Melbourne Genomics Health Alliance indicated additional testing and treatment costs because testing can be an issue and cost to the consumer.

Next steps

PASC advised that, upon ratification of the post-PASC PICO, the application can proceed to the Evaluation Sub-Committee (ESC) stage of the MSAC process.

PASC noted the applicant has elected to progress its application as a DCAR (Department Contracted Assessment Report).

Applicant Comments on Ratified PICO

Note that the statement describing Decreased function variants (page 5) is incorrect and should be deleted based on updated information provided by Turner et al (2024): "c.1129–5923C>G (also known as rs75017182, HapB3). The variant c.1236G>A is in complete linkage disequilibrium with the HapB3 variant and can be used as a suitable proxy (Dean and Kane 2021 (Update))."

Variant c.1236G>A is **not** in complete linkage disequilibrium, and therefore cannot be used as a suitable proxy. The c.1129-5923C>G variant is the variant causally linked to decreased DPD function.

Turner AJ, Haidar CE, Yang W et al. *Updated DPYD HapB3 haplotype structure and implications for pharmacogenomic testing*. Clin Transl Sci. 2024 Jan;17(1):e13699. doi: 10.1111/cts.13699. PMID: 38129972; PMCID: PMC10777430.

Reference to the variant c.1236G>A on page 15 of this document should be deleted based on updated information provided by Turner et al (2024). The College agrees that "testing could use any appropriate sample type"; however, it is important that the performance of other specimen types has validated by the testing laboratory in accordance with NPAAC standards. In addition, cancer biopsy samples should be excluded as appropriate specimens for DNA extraction due to the potential of poor test performance and the risk of false negatives.

The College would like to stress that the MBS item descriptor should be method agnostic, to futureproof the item and to permit laboratories to choose an appropriate method. The statements by PASC on page 16 and 30 of this document appear to contradict each other and require clarification.

"Overall, PASC advised the intervention should be genotyping of at least the four proposed SNPs, and gene sequencing should not be added as a scenario of the intervention."

PASC concerns regarding variants of uncertain significance can be overcome with the use of virtual panels that ensure only the variants described are identified, and that the variants analysed are selected in line with current guidelines.

Regarding Figures 2 and 3 – Clinical Algorithms - clinician response: Alternative therapy options are tumour stream specific and should not be explicitly listed in this document. The decision for marked FP dose reduction/ careful monitoring versus alternative therapy needs careful clinical consideration and discussion with patients. Likewise, the decision to manage severe toxicity with uridine triacetate should not be mandated as this requires clinical consideration regarding timing of administration and drug availability on a case-by-case basis. Treatment with uridine triacetate should be removed from the clinical algorithms.

The College agrees with PASC that the item number should remain methodology agnostic and should not restrict the testing population by age.

The College notes the generally supportive consultation feedback received.

References

Alsanosi SMM, Skiffington C and Padmanabhan S (2014). Chapter 17 - Pharmacokinetic Pharmacogenomics. Handbook of Pharmacogenomics and Stratified Medicine. S. Padmanabhan. San Diego, Academic Press**:** 341-364.

Amstutz U, Froehlich TK and Largiadèr CR (2011). "Dihydropyrimidine dehydrogenase gene as a major predictor of severe 5-fluorouracil toxicity." Pharmacogenomics **12**(9): 1321-1336.

Amstutz U, Henricks LM, Offer SM, Barbarino J, Schellens JHM, Swen JJ, Klein TE, McLeod HL, Caudle KE, Diasio RB and Schwab M (2018). "Clinical Pharmacogenetics Implementation Consortium (CPIC) Guideline for Dihydropyrimidine Dehydrogenase Genotype and Fluoropyrimidine Dosing: 2017 Update." Clin Pharmacol Ther **103**(2): 210-216.

Australian Commission on Safety and Quality in Health Care (ACSQHC). (2022). "Requirements for medical testing for human genetic variation- Third Edition." Third. Retrieved 16 Oct, 2023, from https://www.safetyandquality.gov.au/sites/default/files/2023-03/requirements_for_medical_testing_for_human_genetic_variation_third_edition_2022.pdf.

Australian Institute of Health and Welfare (AIHW) (2023). Cancer Data in Australia: Book 1e – Long-term incidence projections for selected cancers. Australian Institute of Health and Welfare (AIHW). Canberra.

Bignucolo A, De Mattia E, Roncato R, Peruzzi E, Scarabel L, D'Andrea M, Sartor F, Toffoli G and Cecchin E (2023). "Ten-year experience with pharmacogenetic testing for DPYD in a national cancer center in Italy: Lessons learned on the path to implementation." Front Pharmacol **14**: 1199462.

Boige V, Vincent M, Alexandre P, Tejpar S, Landolfi S, Le Malicot K, Greil R, Cuyle PJ, Yilmaz M, Faroux R, Matzdorff A, Salazar R, Lepage C, Taieb J and Laurent-Puig P (2016). "DPYD Genotyping to Predict Adverse Events Following Treatment With Fluorouracil-Based Adjuvant Chemotherapy in Patients With Stage III Colon Cancer: A Secondary Analysis of the PETACC-8 Randomized Clinical Trial." JAMA Oncology **2**(5): 655-662.

Boisdron-Celle M, Capitain O, Faroux R, Borg C, Metges JP, Galais MP, Kaassis M, Bennouna J, Bouhier-Leporrier K, Francois E, Baumgaertner I, Guerin-Meyer V, Cojocarasu O, Roemer-Becuwe C, Stampfli C, Rosenfeld L, Lecompte T, Berger V, Morel A and Gamelin E (2017). "Prevention of 5-fluorouracil-induced early severe toxicity by pre-therapeutic dihydropyrimidine dehydrogenase deficiency screening: Assessment of a multiparametric approach." Semin Oncol **44**(1): 13-23.

Calais G, Alfonsi M, Bardet E, Sire C, Germain T, Bergerot P, Rhein Ba, Tortochaux J, Oudinot P and Bertrand P (1999). "Randomized Trial of Radiation Therapy Versus Concomitant Chemotherapy and Radiation Therapy for Advanced-Stage Oropharynx Carcinoma." JNCI: Journal of the National Cancer Institute **91**(24): 2081-2086.

Cancer Australia. (2023). "Cancer Research in Australia." Retrieved 9 Oct, 2023, from https://www.canceraustralia.gov.au/research/data-and-statistics/cancer-data/improving-cancer-data.

Cascorbi I (2023). "Why is There Still Debate About Recommending DPYD-Testing Before Fluoropyrimidine Treatment?" Clinical Pharmacology & Therapeutics **114**(4): 733-737.

Chazal M, Etienne MC, Renée N, Bourgeon A, Richelme H and Milano G (1996). "Link between dihydropyrimidine dehydrogenase activity in peripheral blood mononuclear cells and liver." Clinical Cancer Research **2**(3): 507-510.

Coenen MJH, Paulussen ADC, Breuer M, Lindhout M, Tserpelis DCJ, Steyls A, Bierau J and van den Bosch BJC (2019). "Evolution of Dihydropyrimidine Dehydrogenase Diagnostic Testing in a Single Center during an 8-Year Period of Time." Curr Ther Res Clin Exp **90**: 1-7.

De Luca O, Salerno G, De Bernardini D, Torre MS, Simmaco M, Lionetto L, Gentile G and Borro M (2022). "Predicting Dihydropyrimidine Dehydrogenase Deficiency and Related 5-Fluorouracil Toxicity: Opportunities and Challenges of DPYD Exon Sequencing and the Role of Phenotyping Assays." Int J Mol Sci **23**(22).

de With M, Knikman J, de Man FM, Lunenburg CATC, Henricks LM, van Kuilenburg ABP, Maring JG, van Staveren MC, de Vries N, Rosing H, Beijnen JH, Pluim D, Modak A, Imholz ALT, van Schaik RHN, Schellens JHM, Gelderblom H, Cats A, Guchelaar H-J, Mathijssen RHJ, Swen JJ and Meulendijks D (2022). "Dihydropyrimidine Dehydrogenase Phenotyping Using Pretreatment Uracil: A Note of Caution Based on a Large Prospective Clinical Study." Clinical Pharmacology & Therapeutics **112**(1): 62-68.

de With M, Sadlon A, Cecchin E, Haufroid V, Thomas F, Joerger M, van Schaik RHN, Mathijssen RHJ and Largiadèr CR (2023). "Implementation of dihydropyrimidine dehydrogenase deficiency testing in Europe." ESMO Open **8**(2): 101197.

Dean L and Kane M (2021 (Update)). Fluorouracil Therapy and DPYD Genotype. Medical Genetics Summaries. V. M. Pratt, S. A. Scott, M. Pirmohamed et al. Bethesda (MD), National Center for Biotechnology Information (US)**:** 291-308.

Denis F, Garaud P, Bardet E, Alfonsi M, Sire C, Germain T, Bergerot P, Rhein B, Tortochaux J, Oudinot P and Calais G (2003). "Late toxicity results of the GORTEC 94-01 randomized trial comparing radiotherapy with concomitant radiochemotherapy for advanced-stage oropharynx carcinoma: comparison of LENT/SOMA, RTOG/EORTC, and NCI-CTC scoring systems." International Journal of Radiation Oncology*Biology*Physics **55**(1): 93-98.

Desilets A, McCarvill W, Aubin F, Bahig H, Ballivy O, Charpentier D, Filion É, Jamal R, Lambert L, Nguyen-Tan PF, Vadnais C, Weng X and Soulières D (2022). "Upfront DPYD Genotyping and Toxicity Associated with Fluoropyrimidine-Based Concurrent Chemoradiotherapy for Oropharyngeal Carcinomas: A Work in Progress." Current Oncology **29**(2): 497-509.

Etienne-Grimaldi M-C, Pallet N, Boige V, Ciccolini J, Chouchana L, Barin-Le Guellec C, Zaanan A, Narjoz C, Taieb J, Thomas F and Loriot M-A (2023). "Current diagnostic and clinical issues of screening for dihydropyrimidine dehydrogenase deficiency." European Journal of Cancer **181**: 3-17.

European Medicines Agency (EMA). (2008, 26 Oct 2022). "Xeloda - EMEA/H/C/000316 - IAIN/0099/G Product information- Summary of product characteristics."   Retrieved 25 Oct, 2023, from

https://www.ema.europa.eu/en/documents/product-information/xeloda-epar-product-information_en.pdf.

European Medicines Agency (EMA). (2020). "Fluorouracil and fluorouracil related substances (capecitabine, tegafur and flucytosine) containing medicinal products." Retrieved 19th April, 2023, from https://www.ema.europa.eu/en/medicines/human/referrals/fluorouracil-fluorouracil-related-substances-capecitabine-tegafur-flucytosine-containing-medicinal.

eviQ. (2020). "Management of fluoropyrimidine overdose or overexposure." Retrieved 17 Oct, 2023, from https://www.eviq.org.au/clinical-resources/side-effect-and-toxicity-management/prophylaxis-and-treatment/738-management-of-fluoropyrimidine-overdose-or-ove#management.

eviQ. (2021). "Hand-foot syndrome (HFS) - palmar-plantar erythrodysaesthesia (PPE) associated with chemotherapeutic agents." Retrieved 30 Oct, 2023, from https://www.eviq.org.au/clinical-resources/side-effect-and-toxicity-management/hair-skin-and-nails/1416-hand-foot-syndrome-associated-with-chemothera#management.

eviQ. (2022a). "Severe enteropathy associated with fluoropyrimidine chemotherapy in colorectal cancer." Retrieved 30 Oct, 2023, from https://www.eviq.org.au/clinical-resources/side-effect-and-toxicity-management/prophylaxis-and-treatment/1739-severe-enteropathy-associated-with-fluoropyri#management.

eviQ. (2022b). "Treatment induced diarrhoea." Clincial resources- Side effect and toxicity management- Gastrointestinal Version 5. Retrieved 31 October, 2023, from https://www.eviq.org.au/clinical-resources/side-effect-and-toxicity-management/gastrointestinal/779-treatment-induced-diarrhoea#management.

eviQ. (2023a). "Anaemia." Version 4. Retrieved 31 October, 2023, from https://www.eviq.org.au/clinical-resources/side-effect-and-toxicity-management/haematological/1754-anaemia#history.

eviQ. (2023b). "Dihydropyrimidine dehydrogenase (DPD) enzyme deficiency." Retrieved 9 Oct, 2023, from https://www.eviq.org.au/clinical-resources/side-effect-and-toxicity-management/prophylaxis-and-treatment/1744-dihydropyrimidine-dehydrogenase-dpd-enzyme#history.

eviQ. (2023c, 5 October 2022). "Immediate management of neutropenic fever." Version 6. Retrieved 31 October, 2022, from https://www.eviq.org.au/clinical-resources/oncological-emergencies/123-immediate-management-of-neutropenic-fever#management.

eviQ. (2023d, 9 March 2023). "Oral mucositis and stomatitis." Version 5. Retrieved 31 October, 2023, from https://www.eviq.org.au/clinical-resources/side-effect-and-toxicity-management/oropharyngeal/210-oral-mucositis-and-stomatitis#history.

Gentile G, Botticelli A, Lionetto L, Mazzuca F, Simmaco M, Marchetti P and Borro M (2016). "Genotype–phenotype correlations in 5-fluorouracil metabolism: a candidate DPYD haplotype to improve toxicity prediction." The Pharmacogenomics Journal 16(4): 320-325.

Glewis S, Alexander M, Khabib MNH, Brennan A, Lazarakis S, Martin J, Tie J, Lingaratnam S and Michael M (2022). "A systematic review and meta-analysis of toxicity and treatment outcomes with pharmacogenetic-guided dosing compared to standard of care BSA-based fluoropyrimidine dosing." Br J Cancer **127**(1): 126-136.

Hamzic S, Aebi S, Joerger M, Montemurro M, Ansari M, Amstutz U and Largiadèr C (2020). "Fluoropyrimidine chemotherapy: recommendations for DPYD genotyping and therapeutic drug monitoring of the Swiss Group of Pharmacogenomics and Personalised Therapy." Swiss Med Wkly **150**: w20375.

Healthcare Improvement Scotland (SHTG). (2020). "Pre-treatment *DPYD* genetic testing for patients who are prescribed chemotherapy involving fluoropyrimidines." SHTG Assessment Number 04. Retrieved 22 October, 2023, from https://shtg.scot/media/1873/pre-treatment-dpyd-genetic-testing-for-pts-prescribed-chemotherapy-involving-fluoropyrimidines-shtg-assess-04-med-onco-and-clin-genetics.pdf.

Henricks LM, Lunenburg CATC, de Man FM, Meulendijks D, Frederix GWJ, Kienhuis E, Creemers G-J, Baars A, Dezentjé VO, Imholz ALT, Jeurissen FJF, Portielje JEA, Jansen RLH, Hamberg P, ten Tije AJ, Droogendijk HJ, Koopman M, Nieboer P, van de Poel MHW, Mandigers CMPW, Rosing H, Beijnen JH, Werkhoven Ev, van Kuilenburg ABP, van Schaik RHN, Mathijssen RHJ, Swen JJ, Gelderblom H, Cats A, Guchelaar H-J and Schellens JHM (2018). "DPYD genotype-guided dose individualisation of fluoropyrimidine therapy in patients with cancer: a prospective safety analysis." The Lancet Oncology **19**(11): 1459-1467.

Innocenti F, Mills SC, Sanoff H, Ciccolini J, Lenz HJ and Milano G (2020). "All You Need to Know About DPYD Genetic Testing for Patients Treated With Fluorouracil and Capecitabine: A Practitioner-Friendly Guide." JCO Oncol Pract **16**(12): 793-798.

Jacobs BAW, Rosing H, de Vries N, Meulendijks D, Henricks LM, Schellens JHM and Beijnen JH (2016). "Development and validation of a rapid and sensitive UPLC–MS/MS method for determination of uracil and dihydrouracil in human plasma." Journal of Pharmaceutical and Biomedical Analysis **126**: 75-82.

Jurczyk M, Król M, Midro A, Kurnik-Łucka M, Poniatowski A and Gil K (2021). "Cardiotoxicity of Fluoropyrimidines: Epidemiology, Mechanisms, Diagnosis, and Management." Journal of Clinical Medicine **10**(19): 4426.

Keen J, Groves E, Shaunak N and Newman W (2022). "DPYD genetic testing and the future of pharmacogenomic testing in routine care." The Pharmaceutical Journal **308**(7962).

Kim W, Cho YA, Kim DC and Lee KE (2022). "Elevated Risk of Fluoropyrimidine-Associated Toxicity in European Patients with DPYD Genetic Polymorphism: A Systematic Review and Meta-Analysis." J Pers Med **12**(2).

Knikman JE, Gelderblom H, Beijnen JH, Cats A, Guchelaar HJ and Henricks LM (2021). "Individualized Dosing of Fluoropyrimidine-Based Chemotherapy to Prevent Severe Fluoropyrimidine-Related Toxicity: What Are the Options?" Clin Pharmacol Ther **109**(3): 591-604.

Laures N, Konecki C, Brugel M, Giffard AL, Abdelli N, Botsen D, Carlier C, Gozalo C, Feliu C, Slimano F, Djerada Z and Bouché O (2022). "Impact of Guidelines Regarding Dihydropyrimidine Dehydrogenase (DPD)

Deficiency Screening Using Uracil-Based Phenotyping on the Reduction of Severe Side Effect of 5-Fluorouracil-Based Chemotherapy: A Propension Score Analysis." Pharmaceutics **14**(10).

Lee AM, Shi Q, Pavey E, Alberts SR, Sargent DJ, Sinicrope FA, Berenberg JL, Goldberg RM and Diasio RB (2014). "DPYD variants as predictors of 5-fluorouracil toxicity in adjuvant colon cancer treatment (NCCTG N0147)." J Natl Cancer Inst **106**(12).

Longley DB, Harkin DP and Johnston PG (2003). "5-Fluorouracil: mechanisms of action and clinical strategies." Nature Reviews Cancer **3**(5): 330-338.

Lunenburg C, van der Wouden CH, Nijenhuis M, Crommentuijn-van Rhenen MH, de Boer-Veger NJ, Buunk AM, Houwink EJF, Mulder H, Rongen GA, van Schaik RHN, van der Weide J, Wilffert B, Deneer VHM, Swen JJ and Guchelaar HJ (2020). "Dutch Pharmacogenetics Working Group (DPWG) guideline for the gene-drug interaction of DPYD and fluoropyrimidines." Eur J Hum Genet **28**(4): 508-517.

Maslarinou A, Manolopoulos VG and Ragia G (2023). "Pharmacogenomic-guided dosing of fluoropyrimidines beyond DPYD: time for a polygenic algorithm?" Frontiers in Pharmacology **14**.

Mattison LK, Soong R and Diasio RB (2002). "Implications of dihydropyrimidine dehydrogenase on 5-fluorouracil pharmacogenetics and pharmacogenomics." Pharmacogenomics **3**(4): 485-492.

Mayo Clinic Laboratories. (2023). "Dihydropyrimidine Dehydrogenase, DPYD Full Gene Sequencing." Retrieved 8 Nov, 2023, from https://www.mayocliniclabs.com/test-catalog/overview/610051#Specimen.

Medical Services Advisory Committee (MSAC). (2023). "MSAC Application 1760 - DPYD genotyping to predict fluoropyrimidine-induced toxicity." Department of Health and Aged Care Retrieved 29 Sep 2023, from http://www.msac.gov.au/internet/msac/publishing.nsf/Content/1760-public.

Meulendijks D, Henricks LM, Jacobs BAW, Aliev A, Deenen MJ, de Vries N, Rosing H, van Werkhoven E, de Boer A, Beijnen JH, Mandigers CMPW, Soesan M, Cats A and Schellens JHM (2017). "Pretreatment serum uracil concentration as a predictor of severe and fatal fluoropyrimidine-associated toxicity." British Journal of Cancer **116**(11): 1415-1424.

Meulendijks D, Henricks LM, Sonke GS, Deenen MJ, Froehlich TK, Amstutz U, Largiadèr CR, Jennings BA, Marinaki AM, Sanderson JD, Kleibl Z, Kleiblova P, Schwab M, Zanger UM, Palles C, Tomlinson I, Gross E, van Kuilenburg ABP, Punt CJA, Koopman M, Beijnen JH, Cats A and Schellens JHM (2015). "Clinical relevance of DPYD variants c.1679T>G, c.1236G>A/HapB3, and c.1601G>A as predictors of severe fluoropyrimidine-associated toxicity: a systematic review and meta-analysis of individual patient data." The Lancet Oncology **16**(16): 1639-1650.

NHS England (2020). Clinical Commissioning Urgent Policy Statement: Pharmacogenomic testing for DPYD polymorphisms with fluoropyrimidine therapies. United Kingdom, National Health Service England.

NHS England. (2023). "National genomic test directory for cancer."   Retrieved 8 Nov, 2023, from https://www.england.nhs.uk/publication/national-genomic-test-directories/.

Oncology (2001). FDA Approves Capecitabine, First Oral Chemotherapy for the Treatment of Metastatic Colorectal Cancer. New Jersey, USA, MJH Life Sciences. **15**.

Ontario Health (2021). DPYD genotyping in patients who have planned cancer treatment with fluoropyrimidines: a health technology assessment. Ontario Health Technology Assessment Series. Ontario, Ontario Health. **21:** 1–186.

Palles C, Fotheringham S, Chegwidden L, Lucas M, Kerr R, Mozolowski G, Rosmarin D, Taylor JC, Tomlinson I and Kerr D (2021). "An Evaluation of the Diagnostic Accuracy of a Panel of Variants in DPYD and a Single Variant in ENOSF1 for Predicting Common Capecitabine Related Toxicities." Cancers (Basel) **13**(7).

Pallet N, Hamdane S, Garinet S, Blons H, Zaanan A, Paillaud E, Taieb J, Laprevote O, Loriot MA and Narjoz C (2020). "A comprehensive population-based study comparing the phenotype and genotype in a pretherapeutic screen of dihydropyrimidine dehydrogenase deficiency." Br J Cancer **123**(5): 811-818.

Paulsen NH, Vojdeman F, Andersen SE, Bergmann TK, Ewertz M, Plomgaard P, Hansen MR, Esbech PS, Pfeiffer P, Qvortrup C and Damkier P (2022). "DPYD genotyping and dihydropyrimidine dehydrogenase (DPD) phenotyping in clinical oncology. A clinically focused minireview." Basic & Clinical Pharmacology & Toxicology **131**(5): 325-346.

Pfizer Australia Pty Ltd. (2023). "Product Information- Fluorouracil Injection BP (Fluorouracil)." Version: pfpfluoi10723, Last updated 5 July 2023. Retrieved 30 Oct, 2023, from https://www.ebs.tga.gov.au/ebs/picmi/picmirepository.nsf/pdf?OpenAgent&id=CP-2011-PI-02952-3&d=20231030172310101.

Pluim D, Jacobs BA, Deenen MJ, Ruijter AE, van Geel RM, Burylo AM, Meulendijks D, Beijnen JH and Schellens JH (2015). "Improved pharmacodynamic assay for dihydropyrimidine dehydrogenase activity in peripheral blood mononuclear cells." Bioanalysis **7**(5): 519-529.

Ragia G, Maslarinou A, Atzemian N, Biziota E, Koukaki T, Ioannou C, Balgkouranidou I, Kolios G, Kakolyris S, Xenidis N, Amarantidis K and Manolopoulos VG (2023). "Implementing pharmacogenetic testing in fluoropyrimidine-treated cancer patients: DPYD genotyping to guide chemotherapy dosing in Greece." Frontiers in Pharmacology **14**.

Saif WM, Banchs J and Kohne C-H (2023). Fluoropyrimidine-associated cardiotoxicity: Incidence, clinical manifestations, mechanisms, and management. UpToDate: Evidence-based Clinical Decision Support. R. M. Goldberg, J. C. Kaski and S. M. Shah, Wolters Kluwer.

Sandoz Pty Ltd. (2022). "Product Information- Capacitabine tablets." Last updated 5 July 2022. Retrieved 30 Oct, 2023, from https://www.ebs.tga.gov.au/ebs/picmi/picmirepository.nsf/pdf?OpenAgent&id=CP-2014-PI-01318-1.

Sharma BB, Rai K, Blunt H, Zhao W, Tosteson TD and Brooks GA (2021). "Pathogenic DPYD Variants and Treatment-Related Mortality in Patients Receiving Fluoropyrimidine Chemotherapy: A Systematic Review and Meta-Analysis." Oncologist **26**(12): 1008-1016.

Swiss Society of Clinical Pharmacology and Toxicology (SSCPT). (2020). "Pharmacogenetic testing information." Retrieved 29 Sep, 2023, from https://clinpharm.ch/en/pharmacogenetics_spt/swiss_group_of_personalised_therapy_and_pharmacogenomics/pharmacogenetic_testing_information.

Therapeutic Goods Administration (TGA). (2021). "Advisory Committee on Medicines Meeting Statement-Meeting 30, Thursday 2 and Friday 3 December 2021. Section B: Post-market item referred for advice. Fluorouracil toxicity and dihydropyrimidine dehydrogenase (DPD) deficiency." Retrieved 30 October, 2023, from https://www.tga.gov.au/sites/default/files/acm-meeting-statement-meeting-30-2-3-december-2021.pdf.

Tsiachristas A, Vallance G, Koleva-Kolarova R, Taylor H, Solomons L, Rizzo G, Chaytor C, Miah J, Wordsworth S and Hassan AB (2022). "Can upfront DPYD extended variant testing reduce toxicity and associated hospital costs of fluoropyrimidine chemotherapy? A propensity score matched analysis of 2022 UK patients." BMC Cancer **22**(1): 458.

Turner AJ, Haidar CE, Yang W, Boone EC, Offer SM, Empey PE, Haddad A, Tahir S, Scharer G, Broeckel U, Gaedigk A (2024). "Updated DPYD HapB3 haplotype structure and implications for pharmacogenomic testing." Clin Transl Sci. 17(1):e13699. doi: 10.1111/cts.13699.

US FDA. (2022). "FDA approves updated drug labeling including new indications and dosing regimens for capecitabine tablets under Project Renewal." Retrieved 25 Oct, 2023, from https://bit.ly/3Ysotk8.

Van Cutsem E, Twelves C, Cassidy J, Allman D, Bajetta E, Boyer M, Bugat R, Findlay M, Frings S, Jahn M, McKendrick J, Osterwalder B, Perez-Manga G, Rosso R, Rougier P, Schmiegel WH, Seitz JF, Thompson P, Vieitez JM, Weitzel C and Harper P (2001). "Oral capecitabine compared with intravenous fluorouracil plus leucovorin in patients with metastatic colorectal cancer: results of a large phase III study." J Clin Oncol **19**(21): 4097-4106.

White C, Scott RJ, Paul C, Ziolkowski A, Mossman D and Ackland S (2021). "Ethnic Diversity of DPD Activity and the DPYD Gene: Review of the Literature." Pharmgenomics Pers Med **14**: 1603-1617.

Wörmann B, Bokemeyer C, Burmeister T, Köhne CH, Schwab M, Arnold D, Blohmer JU, Borner M, Brucker S, Cascorbi I, Decker T, de Wit M, Dietz A, Einsele H, Eisterer W, Folprecht G, Hilbe W, Hoffmann J, Knauf W, Kunzmann V, Largiadèr CR, Lorenzen S, Lüftner D, Moehler M, Nöthen MM, Pox C, Reinacher-Schick A, Scharl A, Schlegelberger B, Seufferlein T, Sinn M, Stroth M, Tamm I, Trümper L, Wilhelm M, Wöll E and Hofheinz RD (2020). "Dihydropyrimidine Dehydrogenase Testing prior to Treatment with 5-Fluorouracil, Capecitabine, and Tegafur: A Consensus Paper." Oncol Res Treat **43**(11): 628-636.