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MEDGENOME

DNA TEST REPORT – MEDGENOME LABORATORIES

Full Name/ Ref No:	**Mrs. P.W.B. Renuka Vithana**	**Order ID/ Sample ID:**	**1128577/8840972**
Date of Birth / Age:	59 years	**Gender:**	Female
Referring Clinician:	Dr. Mahendra Perera, Aegle Omics Private Limited	**Specimen Type:**	Blood in EDTA tube
		Date of Sample Collection:	17-09-2024
Date of Order:	04-12-2024	**Date of Sample Receipt:**	04-12-2024
Test Requested:	DPYD mutation analysis (MGM340)	**Date of Report:**	18-12-2024

CLINICAL DIAGNOSIS / SYMPTOMS / HISTORY

The subject has been tested for variation in the *Dihydropyrimidine dehydrogenase* (*DPYD*) gene for Fluorouracil (5-FU) drug sensitivity.

RESULTS

c.85T>C (*9A) (ONE COPY) and c.1905+1G>A (*2A) (ONE COPY) IDENTIFIED IN THE *DPYD* GENE

INTERMEDIATE METABOLIZER

Gene	Allele	Zygosity	Allele function[#]
DPYD	c.85T>C (*9A)	Heterozygous	Normal function
DPYD	c.1905+1G>A (*2A)	Heterozygous	No function

[#] **- Allele function is according to The Clinical Pharmacogenetics Implementation Consortium (CPIC).**

VARIANT INTERPRETATION AND RECOMMENDATION

Variants c.85T>C (*9A) one copy (chr1:97883329A>G; Total read depth: 74X; Alternate allele read depth:44X) and c.1905+1G>A (*2A) one copy (chr1:97450058C>T; Total read depth: 76X; Alternate allele read depth:38X) were detected in the *DPYD* gene for the sample. Fluoropyrimidines (5-fluorouracil and capecitabine) are antimetabolite drugs widely used in the treatment of cancer including colorectal and breast cancer and cancers of the aerodigestive tract. Individuals with *9A/*2A genotype carrying one normal function allele and one no function allele, having an activity score of 1 are intermediate metabolizers of fluoropyrimidines. These individuals have decreased DPD activity (leukocyte DPD activity at 30% to 70% that of the normal population) and increased risk for severe or even fatal drug toxicity when treated with fluoropyrimidine drugs. CPIC recommends to reduce starting dose by 50% followed by titration of dose based on toxicity or therapeutic drug monitoring (if available). [1] Other genetic and clinical factors may also influence response to fluoropyrimidine-based chemotherapy. All changes in medications must be done under clinical supervision.

BACKGROUND INFORMATION

DPYD (OMIM*612779) gene encodes Dihydropyrimidine dehydrogenase (DPD), a rate limiting enzyme in pyrimidine catabolism. In the liver, DPD is known to degrade 85% of most frequently prescribed anticancer drug 5-FU, thus limiting the amount of drug available for conversion into active metabolites. Various clinical studies across the globe, have established that approximately 40-60% of cancer patients who are DPD deficient develop severe life threatening 5-FU toxicities. However, 5-FU is generally well tolerated at standard doses. The *DPYD* variants can lead to a decreased enzyme activity and hence increase the risk for 5-FU toxicity. [1, 2] Till date, more than 30 *DPYD* variations have been reported, of which some of them are clearly linked to 5-FU toxicity e.g., IVS14+1G>A point variation. Genetic screening of cancer patients for the presence of *DPYD* variations before the administration of 5-FU is appropriate in order to prevent lethal 5-FU-related toxicity.

METHODOLOGY

Targeted gene sequencing: Genomic DNA was used to perform targeted gene capture using a custom capture kit. The libraries were sequenced to mean >80- 100X coverage on Illumina sequencing platform. We follow the GATK best practices framework for identification of variants in the sample using Sentieon (v201808.07). [3] The sequences obtained are aligned to human reference genome (GRCh38.p13) using Sentieon aligner [3, 4] and analyzed using Sentieon for removing duplicates, recalibration and re-alignment of indels. [3] Sentieon haplotype caller has been used to identify variants which are relevant to the clinical indication. Gene annotation of the variants is performed using VEP program [5] against the Ensembl release 99 human gene model. [6]

GUIDELINES FOR CLINICAL INTERPRETATION

The Clinical Pharmacogenetics Implementation Consortium (CPIC) [7] is an international consortium that facilitates clinical implementation of pharmacogenetic tests through published gene/drug clinical practice guidelines. CPIC clinical guidelines are endorsed by American Society of Health-System Pharmacists (ASHP) and American Society for Clinical Pharmacology and Therapeutics (ASCPT). This Pharmacogenomic test has been assessed based on recommendations of the Clinical Pharmacogenetics Implementation Consortium, CPIC, as updated on December 2022. Gene and disease coverage will change as per recommendations and research advancements/reports.

LIMITATIONS

- Only CPIC recommended alleles in *DPYD* are assayed in this test.
- Genetic testing is highly accurate. Rarely, inaccurate results may occur for various reasons. These reasons include, but are not limited to: mislabeled samples, inaccurate reporting of clinical/medical information, rare technical errors or unusual circumstances such as bone marrow transplantation, blood transfusion; or the presence of change(s) in such a small percentage of cells that may not be detectable by the test (mosaicism).

DISCLAIMER

- Interpretation of variants in this report is performed to the best knowledge of the laboratory based on the information available at the time of reporting. The classification of variants can change over time and MedGenome cannot be held responsible for this. Please feel free to contact MedGenome Labs (techsupport@medgenome.com) in the future to determine if there have

been any changes in the classification of any variations. Re-analysis of variants in previously issued reports in light of new evidence is not routinely performed but may be available upon request.

- Very rarely in spite of having normal report, absence of toxicity cannot be guaranteed because of other rare genetic variations present in DPYD gene and which are not recommended by CPIC. Uncertain function/unknown function alleles are considered as normal function alleles for reporting.
- The variations have not been validated/confirmed by Sanger sequencing.
- The report shall be generated within agreed turnaround time (TAT), however, such TAT may vary depending upon the complexity of test(s) requested. MedGenome under no circumstances will be liable for any delay beyond afore mentioned TAT.
- It is hereby clarified that the report(s) generated from the test(s) do not provide any diagnosis or opinion or recommends any cure in any manner. MedGenome hereby recommends the patient and/or the guardians of the patients, as the case may be, to take assistance of the clinician or a certified physician or doctor, to interpret the report(s) thus generated. MedGenome hereby disclaims all liability arising in connection with the report(s).
- In a very few cases genetic test may not show the correct results, e.g. because of the quality of the material provided to MedGenome. In case where any test provided by MedGenome fails for unforeseeable or unknown reasons that cannot be influenced by MedGenome in advance, MedGenome shall not be responsible for the incomplete, potentially misleading or even wrong result of any testing if such could not be recognised by MedGenome in advance.
- This is a laboratory developed test and the development and the performance characteristics of this test was determined by MedGenome.

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2. Diasio RB, *et al*., (1988). Familial deficiency of dihydropyrimidine dehydrogenase. Biochemical basis for familial pyrimidinemia and severe 5- fluorouracil-induced toxicity. *J Clin Invest*., 81:47-51.
3. Freed D, *et al*., (2017). The Sentieon Genomics Tools-A fast and accurate solution to variant calling from next-generation sequence data. *BioRxiv*., 115717.
4. Li H, *et al*., (2010). Fast and accurate long-read alignment with Burrows-Wheeler transform. *Bioinformatics.*, 26:589-595.
5. McLaren W, *et al*., (2010). Deriving the consequences of genomic variants with the Ensembl API and SNP Effect Predictor. *Bioinformatics*., 26:2069-2070.
6. Zerbino D, R. *et al*., (2018). Ensembl 2018. *Nucleic Acids Res*. 46:D754-D761.
7. CPIC: https://cpicpgx.org/

APPENDIX: LIST OF *DPYD* ALLELES COVERED IN THE PANEL

Sl. No	ALLELES COVERED	ALLELE FUNCTION	Sl. No	ALLELES COVERED	ALLELE FUNCTION
1	*10	No function	22	c.1278G>T	Normal function
2	*11	Normal function	23	c.1294G>A	Normal function
3	*12	No function	24	c.1314T>G	Decreased function
4	*13	No function	25	c.1349C>T	Normal function
5	*2A	No function	26	c.1358C>G	Normal function
6	*3	No function	27	c.1371C>T	Normal function
7	*4	Normal function	28	c.1403C>A	Normal function
8	*5	Normal function	29	c.1475C>T	No function
9	*6	Normal function	30	c.1484A>G	No function
10	*7	No function	31	c.1519G>A	Normal function
11	*8	No function	32	c.1543G>A	Normal function
12	*9A	Normal function	33	c.1577C>G	Normal function
13	*9B	Normal function	34	c.1615G>A	Normal function
14	c.1024G>A	No function	35	c.1682G>T	Normal function
15	c.1057C>T	No function	36	c.1774C>T	No function
16	c.1108A>G	Normal function	37	c.1775G>A	No function
17	c.1129-5923C>G	Decreased function	38	c.1777G>A	No function
18	c.1180C>T	Normal function	39	c.1796T>C	Normal function
19	c.1181G>T	Normal function	40	c.1896T>C	Normal function
20	c.1218G>A	Normal function	41	c.1905C>G	Normal function
21	c.1260T>A	Normal function	42	c.1906A>C	Normal function

Sl. No	ALLELES COVERED	ALLELE FUNCTION	Sl. No	ALLELES COVERED	ALLELE FUNCTION
43	c.1990G>T	Normal function	63	c.3049G>A	Normal function
44	c.2021G>A	No function	64	c.3061G>C	Normal function
45	c.2161G>A	Normal function	65	c.3067C>A	Normal function
46	c.2186C>T	Normal function	66	c.313G>A	Normal function
47	c.2195T>G	Normal function	67	c.343A>G	Normal function
48	c.2279C>T	Decreased function	68	c.451A>G	Normal function
49	c.2303C>A	Normal function	69	c.46C>G	Normal function
50	c.2336C>A	Normal function	70	c.496A>G	Normal function
51	c.2482G>A	Normal function	71	c.498G>A	Normal function
52	c.2582A>G	Normal function	72	c.525G>A	Normal function
53	c.2623A>C	Normal function	73	c.557A>G	Decreased function
54	c.2639G>T	No function	74	c.601A>C	No function
55	c.2656C>T	Normal function	75	c.61C>T	No function
56	c.2846A>T	Decreased function	76	c.62G>A	Normal function
57	c.2872A>G	No function	77	c.632A>G	No function
58	c.2915A>G	Normal function	78	c.868A>G	Decreased function
59	c.2921A>T	Normal function	79	c.929T>C	Normal function
60	c.2933A>G	No function	80	c.934C>T	Normal function
61	c.2977C>T	Normal function	81	c.967G>A	Normal function
62	c.2978T>G	Normal function			

----------End of report----------