Table S1. Folding free energy, rSASA and Polyphen predictions for the mutations in DHCR7 protein. ΔΔGs are shown in kcal/mol and average ΔΔG are also calculated using the results from multiple webservers. Mutations A206T and H390T are located on the loop, not present in the template, thus the corresponding rSASA is highlighted in red to indicate low confidence for these two mutations.

Pathogenic missense mutations									
Mutation	**rSASA_mem**	**CV score**	**SAFFEC**	**mCSM**	**SDM**	**DUET**	**FOLDX**	**ΔΔG_ave**	**Polyphen**
T93M	0.21	0.62	0.53	0.10	0.30	-0.03	-0.52	0.08	Probably damaging
G147D	0.01	1.00	3.30	-1.53	-0.35	-1.39	7.40	1.49	Probably damaging
T154R	0.01	0.97	-3.55	-0.59	-1.45	-0.60	1.62	-0.91	Probably damaging
S169L	0.31	0.85	1.39	-0.25	0.69	-0.25	0.62	0.44	Probably damaging
R242H	0.02	1.00	0.17	-2.38	-0.05	-2.59	-0.20	-1.01	Probably damaging
R242C	0.02	1.00	0.40	-1.96	0.39	-2.05	-3.95	-1.43	Probably damaging
G244R	0.01	1.00	0.96	-1.39	-0.10	-1.20	4.72	0.60	Probably damaging
V281M	0.02	0.91	-0.19	-0.28	-0.08	-0.30	0.40	-0.09	Probably damaging
E288K	0.10	1.00	-15.77	-0.27	-1.17	-0.24	0.78	-3.33	Probably damaging
T289I	0.43	0.29	-0.60	-0.09	1.26	0.18	-0.70	0.01	Possibly damaging
G303R	0.04	1.00	-1.48	-1.15	-2.73	-1.22	21.73	3.03	Probably damaging
V326L	0.00	0.94	0.24	-1.08	0.20	-1.00	1.01	-0.13	Benign
R352W	0.06	0.94	0.00	-0.37	2.57	-0.55	0.12	0.35	Probably damaging
R352Q	0.06	0.94	-0.91	-0.81	-0.63	-0.81	0.60	-0.51	Probably damaging
R404C	0.04	0.97	-0.05	-1.97	0.11	-2.18	3.00	-0.22	Probably damaging
G410S	0.01	0.91	-2.71	-1.81	-0.55	-1.69	11.05	0.86	Probably damaging
Missense mutations with unknown effects									
Mutation	**rSASA_mem**	**CV score**	**SAFFEC**	**mCSM**	**SDM**	**DUET**	**FOLDX**	**Average**	**SD**
A41V	0.05	0.44	0.03	-0.28	-0.29	0.01	-0.30	-0.17	Benign
I44T	0.18	0.76	0.69	-1.29	-2.79	-1.19	1.40	-0.64	Benign
A67T	0.00	0.18	2.48	-1.59	-0.79	-1.66	5.65	0.82	Possibly damaging
I75F	0.53	0.24	-0.47	-0.64	0.12	-0.61	-0.56	-0.43	Benign
R81W	0.51	0.21	-0.55	-0.49	1.18	-0.58	-0.64	-0.22	Probably damaging
A97T	0.24	0.80	0.18	-1.28	-2.29	-1.19	1.64	-0.59	Possibly damaging
V126I	0.12	0.76	0.60	-0.78	0.74	-0.72	0.04	-0.02	Probably damaging
V134L	0.28	0.32	1.36	-0.63	1.13	-0.43	-1.24	0.04	Benign
A162V	0.13	0.76	1.18	0.00	0.68	0.19	-1.50	0.11	Possibly damaging
R228Q	0.18	0.97	1.27	-1.01	-0.84	-1.13	-0.23	-0.39	Probably damaging
V330M	0.58	0.85	-2.47	-0.52	-0.51	-0.45	-0.57	-0.90	Probably damaging
V338M	0.25	0.41	-0.92	-0.38	0.80	-0.19	-0.70	-0.28	Benign
F361L	0.01	0.91	-0.16	-2.72	-0.17	-2.80	-0.90	-1.35	Probably damaging
T364M	0.25	0.85	-0.63	-0.03	0.30	-0.32	-0.60	-0.26	Probably damaging
R367C	0.56	0.32	4.01	-0.37	0.83	-0.30	0.30	0.89	Probably damaging
G424S	0.08	0.35	-0.48	-0.48	2.60	-0.02	-0.03	0.32	Probably damaging
G425S	0.12	0.56	-0.95	-0.68	2.54	-0.28	-0.10	0.11	Probably damaging
R461C	0.54	0.71	3.02	-0.39	0.16	-0.31	1.25	0.75	Probably damaging
Non-pathogenic missense mutations									
Mutation	**rSASA_mem**	**Conser**	**SAFFEC**	**mCSM**	**SDM**	**DUET**	**FOLDX**	**Average**	**SD**
V43I	0.16	0.74	0.02	-0.32	0.59	-0.10	0.61	0.16	Benign
G70S	0.00	0.12	0.67	-1.68	-2.79	-1.95	2.51	-0.65	Benign
V76I	0.43	0.06	-1.00	-0.27	0.01	-0.19	0.75	-0.14	Benign
A137S	0.68	0.97	0.39	-0.81	-1.42	-0.66	-0.19	-0.54	Possibly damaging
V140M	0.37	0.53	-0.75	-0.49	-0.74	-0.59	-0.81	-0.67	Benign
A162V	0.13	0.76	0.94	0.00	0.68	0.19	-1.47	0.07	Possibly damaging
V191I	0.31	0.82	0.43	-0.45	0.59	-0.20	-1.11	-0.15	Possibly damaging
A195T	0.03	0.85	0.37	-0.96	-0.79	-0.96	4.06	0.35	Probably damaging
M196V	0.03	0.41	-0.37	-0.97	-1.11	-1.08	4.19	0.13	Benign
A206T	0.29	0.59	2.01	-1.05	-1.72	-0.99	-0.16	-0.38	Probably damaging
M220L	0.08	0.85	0.27	0.16	0.40	0.47	0.14	0.29	Benign
R260Q	0.25	0.15	-1.37	-0.49	-0.26	-0.28	0.23	-0.43	Benign
I295V	0.09	0.88	0.55	-1.04	-0.59	-0.97	0.50	-0.31	Possibly damaging
P335R	0.32	0.38	-3.49	0.15	1.18	0.40	1.85	0.02	Possibly damaging
R363C	0.54	0.88	2.53	-0.17	0.16	-0.10	0.29	0.54	Benign
R363H	0.54	0.88	2.00	-0.98	-0.41	-0.99	0.29	-0.02	Probably damaging
T364M	0.25	0.85	0.59	-0.03	0.30	-0.32	-0.62	-0.02	Probably damaging
R367C	0.56	0.32	3.65	-0.37	0.83	-0.30	0.28	0.82	Probably damaging
H390T	0.36	0.85	0.49	1.55	1.47	1.48	-0.34	0.93	Benign
G425S	0.12	0.56	1.13	-0.68	2.54	-0.28	-0.10	0.52	Benign
A452T	0.32	0.32	1.32	-1.28	-2.72	-1.18	0.41	-0.69	Possibly damaging
G456S	0.33	0.94	-0.43	-1.30	-2.55	-1.21	4.05	-0.29	Probably damaging
R461C	0.54	0.71	3.71	-0.39	0.16	-0.31	1.25	0.88	Probably damaging

Table S2. KNN classifications using different properties and K values. True positive (TP), true negative (TN) and accuracy are calculated for each K value.

Using rSASA, EC score, PD and ΔΔG				Using only rSASA, EC score, PD			
K	TP	TN	Accuracy	K	TP	TN	Accuracy
1	4	2	0.6	1	5	4	0.9
2	4	2	0.6	2	5	4	0.9
3	5	2	0.7	3	6	4	1
4	4	2	0.6	4	6	3	0.9
5	5	2	0.7	5	6	4	1
6	5	2	0.7	6	6	4	1
7	5	2	0.7	7	6	4	1
8	5	3	0.8	8	6	4	1
9	5	2	0.7	9	6	4	1
10	5	0	0.5	10	5	4	0.9

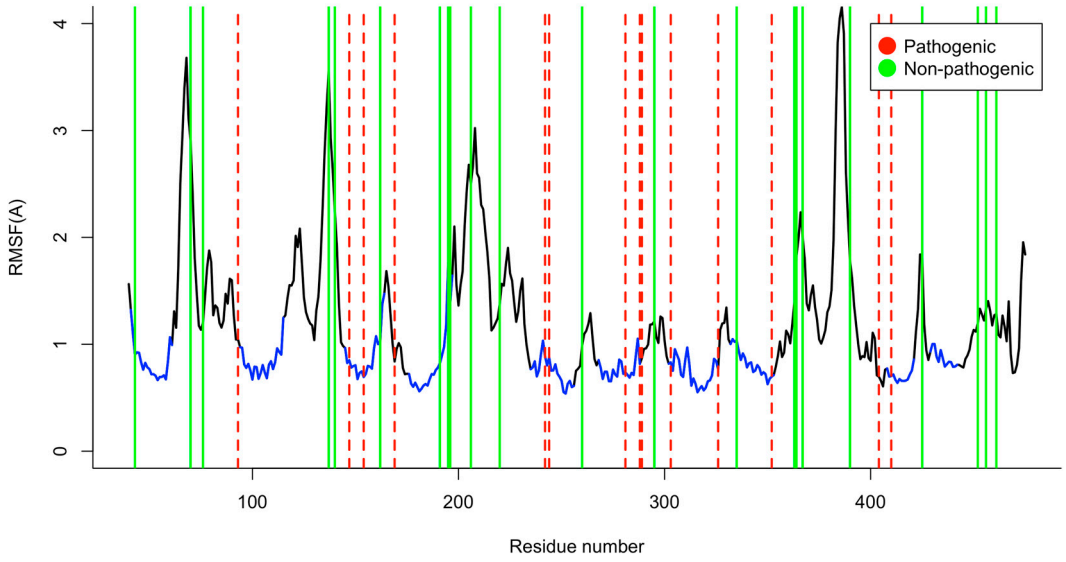

Figure 1. The pathogenic and non-pathogenic mutation occurring sites mapping on the average RMSF of the wild type DHCR7 protein. Pathogenic and non-pathogenic mutation sites are marked with red and green lines. The RMSF of transmembrane region are shown in blue.

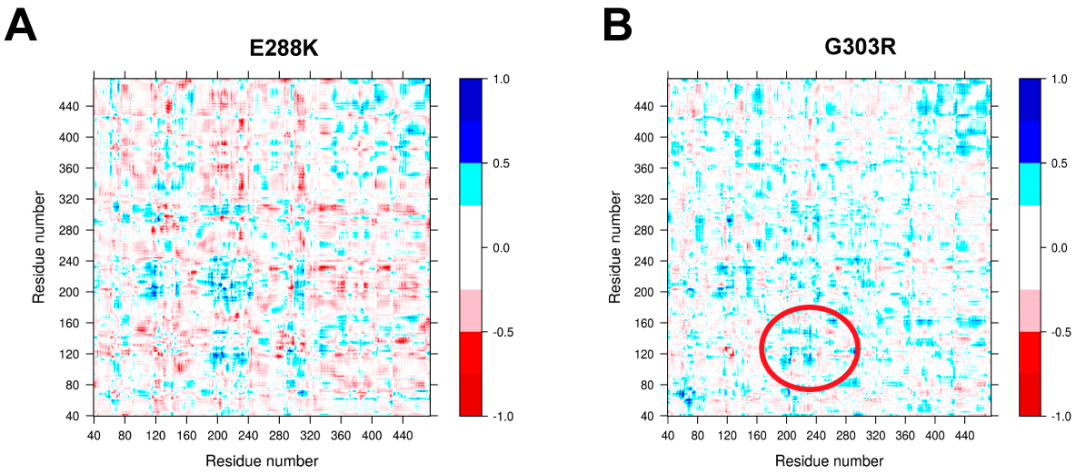

Figure S2. The changes in residue cross-correlation for mutations E288K and G303R.

```
DHCR7          MAAKSQPNIPKAKSLDGVTNDRTASQGQWGRAWEVDWFSLASVIFLLLFAPFIVYYFIMACDQYSCALTGPVVDIVTGHA
4QUV           MSEQE--------SRDNAAVDAVRQKYGFGFSWLV----------LMIALPPLVYYLWICVTYYQGELV-----FTSDAA
               *: :.        * *..: * . .:  :* :* *          *::  * :***: :.   *.  *.     :.:. *

DHCR7          RLSDIWAKTPPITRKAAQLYTLWVTFQVLLYTSLPDFCHKFLPGYVGGIQEGAVTPAGVVNKYQINGLQAWLLTHLLWFA
4QUV           AWRRFWSHVAPPTWHAAGLYAAWFLGQAALQVWAP-----------GPTVQGMKLPDGSRLDYRMNGIFSFLFTLAVVFG
                   :*::..* * :** **: *.  *. * .  *           *   :*   * *   .*::**: ::*:*  : *.

DHCR7          NAHLLSWFSPTIIFDNWIPLLWCANILGYAVSTFAMVKGYFFPTSARDC-KFTGNFFYNYMMGIEFNPRIGKWFDFKLFF
4QUV           LV-TMGWLDATVLYDQLGPLLTVVNIFTFVFAGFL----YFWGLNGKQWERPTGRPFYDYFMGTALNPRIGS-LDLKLFC
                .  :.*:..*:::*:  ***  .**: :..: *     **:  ..::  : **. **:*:**  :*****. :*:***

DHCR7          NGRPGIVAWTLINLSFAAKQRELHSHVTNAMVLVNVLQAIYVIDFFWNETWYLKTIDICHDHFGWYLGWGDCVWLPYLYT
4QUV           EARPGMIFWLLMNLSMAAKQYELHGTVTVPMLLVVGFQSFYLIDYFIHEEAVLTTWDIKHEKFGWMLCWGDLVWLPFTYT
               :.***:: * *:***:**** ***. ** .*:**  :*::*:**:* :*   *.* ** *::*** * *** ****: **

DHCR7          LQGLYLVYHPVQLSTPHAVGVLLLGLVGYYIFRVANHQKDLFRRTDGRCLIWGRKPKVIECSYTSADGQRHHSKLLVSGF
4QUV           LQAQYLVHHTHDLPVWGIIAIVALNLAGYAIFRGANIQKHHFRRDP-NRIVWGKPAKYIKT--------KQGSLLLTSGW
               **. ***:*. :*..   :.:: *.*.** *** ** **. ***   . ::**: .* *:         :: * **.**:

DHCR7          WGVARHFNYVGDLMGSLAYCLACGGGHLLPYFYIIYMAILLTHRCLRDEHRCASKYGRDWERYTAAVPYRLLPGI-
4QUV           WGIARHMNYFGDLMIALSWCLPAAFGSPIPYFHIVYFTILLLHREKRDDAMCLAKYGEDWLQYRKKVPWRIVPKIY
               **:***:**.**** :*::**... *  :***:*:*::*** **  **:  * :***.** :*   **:*::* *
```

Figure S3. Sequence alignment between DHCR7 and template 4QUV.