SUPPORTING INFORMATION

In the supplementary section we present the figures and tables that are necessary to understand with more detail the studied mechanism. It contains the coordinates for full model used, and the structure of the stationary points discussed in the paper; Relevant interatomic distances for the optimized structures with the B3LYP/6-31G(d):FF99SB level of theory, changes involved in the charge distribution along the reaction path for the DFT layer (B3LYP/6-311+G(2d,2p):FF99SB level theory).

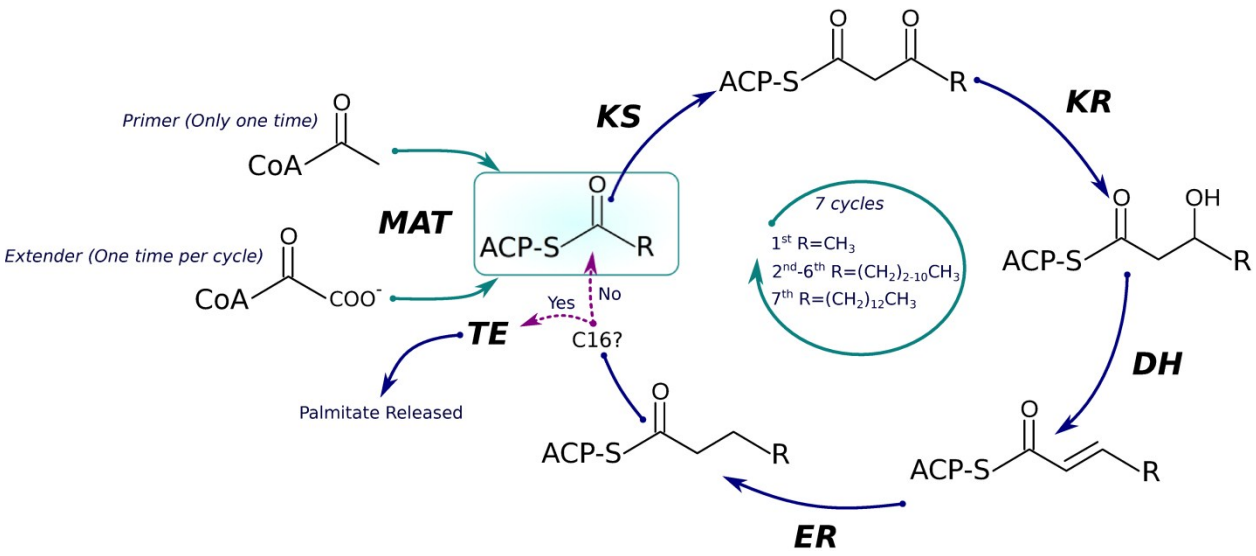

Figure S1. General scheme of the set of reactions that occur inside human FAS (see introduction section for details).

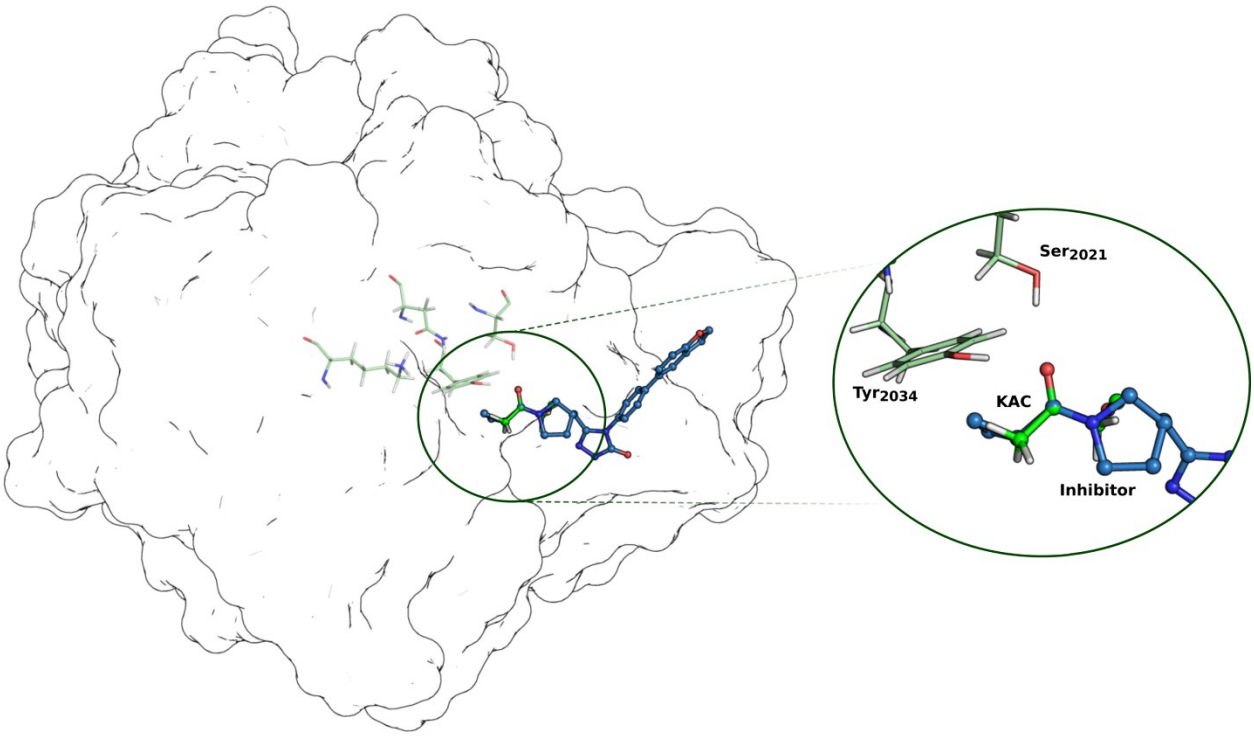

Figure S2. (Left) Surface representation of the KR human domain with catalytic residues (light green sticks representation), KAC substrate (green stick and ball representation) and GSK2194069 inhibitor[10] (blue stick and ball representation). (Right) KAC alignment (green) with the inhibitor (blue) (Ser and Tyr light green color).

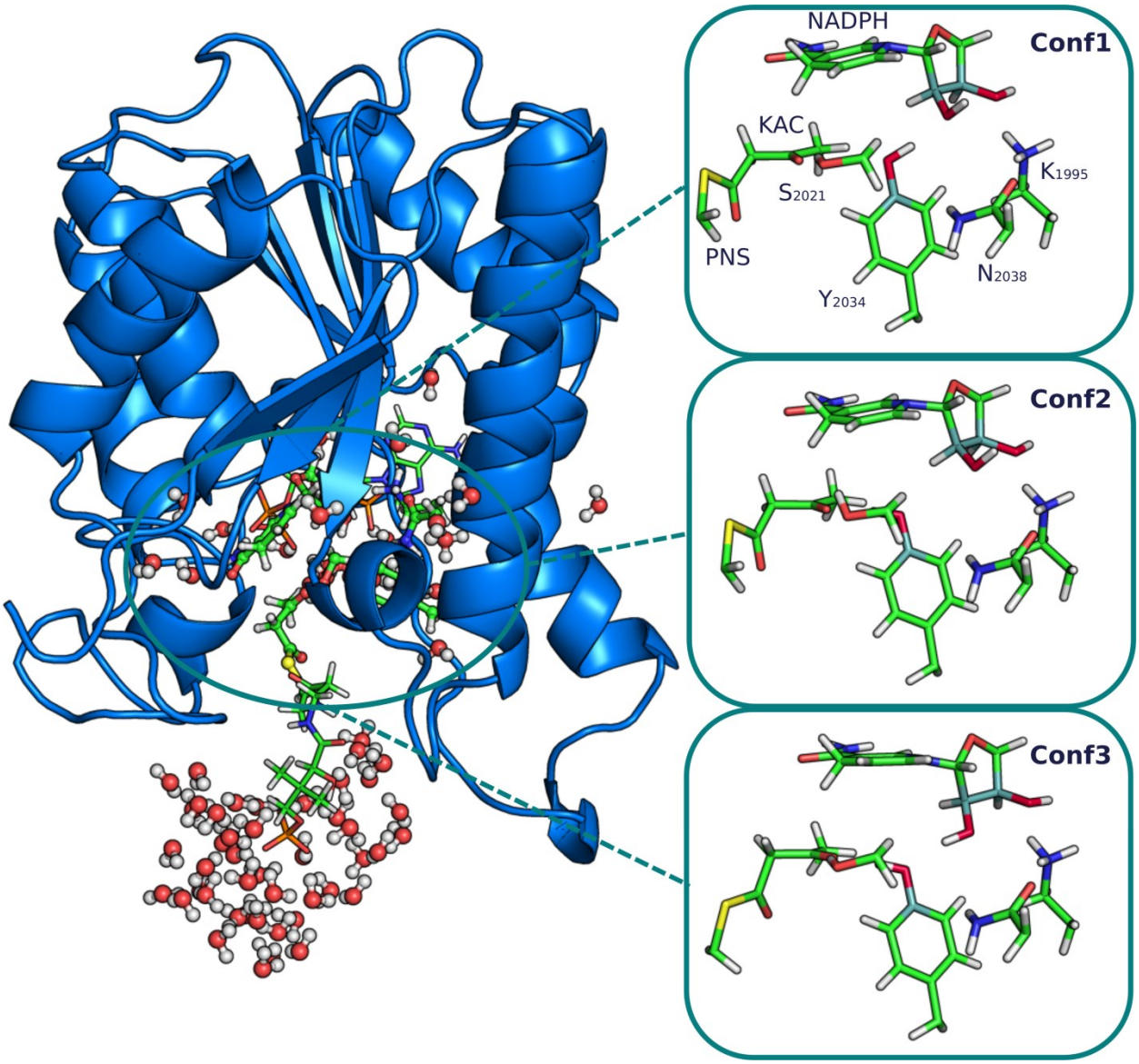

Figure S3. (Left) Full model used for the ONIOM calculations, the image includes: hKR (cartoon representation), water molecules within 6 Å around of catalytic domains, NADPH, PNS and KAC (spheres and sticks representations). (Right) The three conformations in the QM layer.

Table S1. Comparison of the activation electronic energies with the water nearby Lys1995 included either in the QM layer or the MM layer, in kcal·mol^{-1}.

	TS1		I	
	H_2O in MM	**H_2O in QM**	**H_2O in MM**	**H_2O in QM**
Conf-1	27.7	31.3	27.3	22.8
Conf-2	16.6	13.9	3.2	-1.1
Conf-3	14.8	14.0	-7.7	-7.8

Table S2. Single point energies (kcal·mol^{-1}) determined for the stationary structures with five different DFT functional and the 6-311+G(2d,2p) basis set.

	B3LYP	**M06-2X**	**ωB97XD**	**MPW1K**	**MPWB1K**
React	0.0	0.0	0.0	0.0	0.0
TS1	16.0	15.2	15.8	18.2	18.6
I	-5.8	-8.1	-7.2	-4.9	-5.7
TS2	-4.4	-6.4	-4.1	-2.6	-3.1
P	-12.8	-14.3	-12.0	-12.0	-13.4

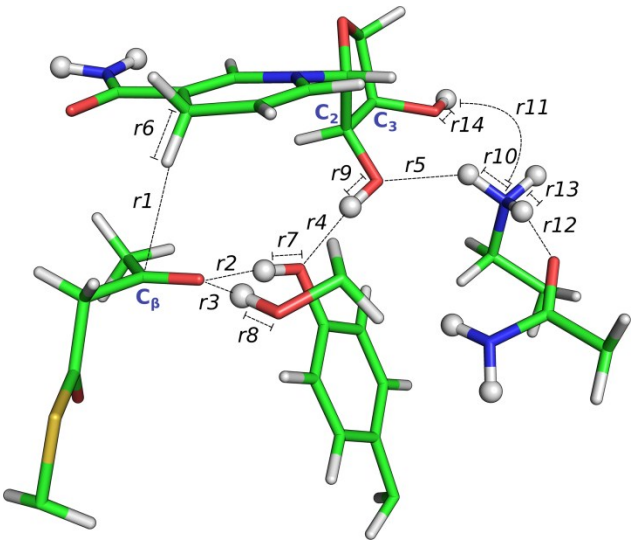

Figure S4. Representation of the most relevant distances in the QM-layer.

Table S3. NBO bond orders for the QM layer in **React**, **TS1** and **I** states. The electronic population values are determined at B3LYP/6-311+G(2d,2p) level.

	React		**TS1**		**I**	
	H_{Cre}	$H_{Tyr2034\text{-}OH}$	H_{Cre}	$H_{Tyr2034\text{-}OH}$	H_{Cre}	$H_{Tyr2034\text{-}OH}$
C_{re}	0.2249	0.0000	**0.1104**	0.0000	0.0007	0.0000
C_{β}	0.0003	0.0000	**0.1101**	0.0000	0.2282	0.0000
C_{β}-oxygen	0.0000	0.0180	0.0000	**0.0485**	0.0000	0.1612
Tyr_{2034}-oxygen	0.0000	0.1590	0.0000	**0.1308**	0.0000	0.0194

Table S4. Continuation of the **Table 1**. Distances (Å) for the optimized stationary points in the PESs (B3LYP/6-31G(d):FF99SB).

Coordinate	**React**	**TS1**	**I**	**TS2**	**P**
***r*11**	3.08	3.08	3.16	2.83	2.13
***r*12**	1.85	1.83	1.80	1.90	2.09
***r*13**	1.03	1.04	1.04	1.03	1.02
***r*14**	0.98	0.98	0.98	0.97	0.98

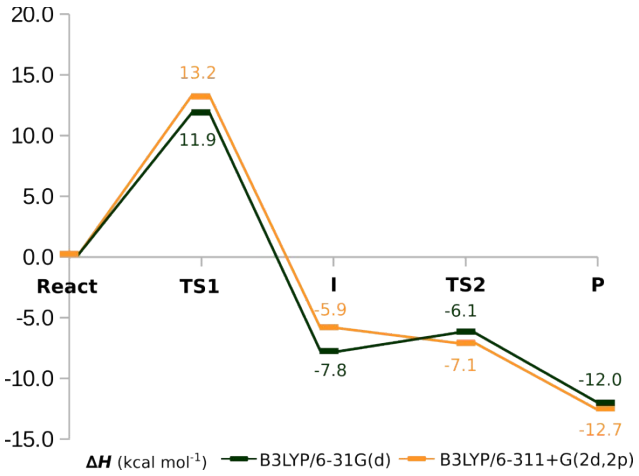

Figure S5. Electronic energy profile for the two stages observed in the reduction of KAC to HAC: (orange) electronic energy calculated at B3LYP/6-311+G(2d,2p) level theory and (gray) electronic energy at B3LYP/6-31G(d) level theory.

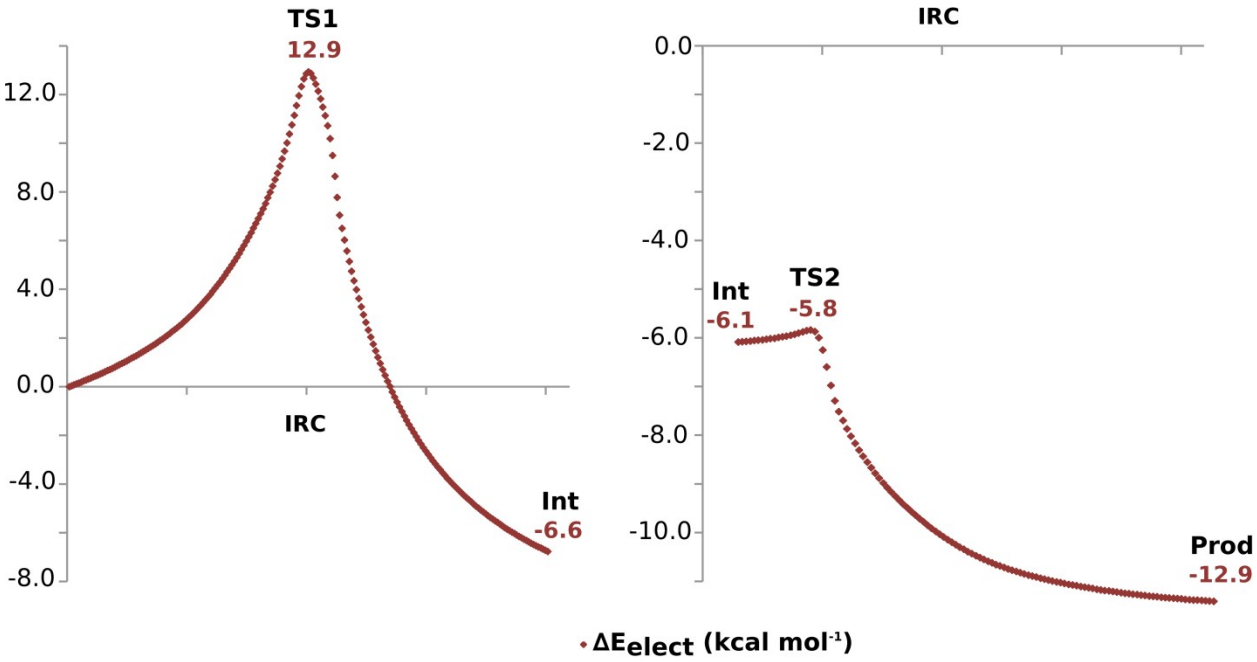

Figure S6. (Left) IRC obtained for the TS1. (Right) IRC obtained for the TS2. Both calculations were performed at the B3LYP/6-31G(d):FF99SB level.

The reaction paths followed by IRC calculations, show that both the stationary states and the energy barrier are very similar to those obtained in the study of the PESs for the catalytic mechanism (ΔE ~2 kcal mol^{-1} relatively to the PES results). However, the **I** states coming from **TS1** and the **TS2** have different energy values (which differ in 0.5 kcal mol^{-1}). We explored the differences between these: the IRC of the **TS1** leads to the formation of a **I** structure with a negatively charged Tyr2034 (**Figure S7**); the IRC of the**TS2** results in a **I** state where the Tyr2034 is already protonated by the C_2–hydroxyl of ribose (**Figure S7**). We studied the PES that interchanges the proton of the C_2–hydroxyl with Tyr2034 for both states. The results indicate that the energy difference in the proton transfer for both PES is ~0.16 kcal mol^{-1}. Moreover, we confirmed through an rmsd analysis that the DFT layers only differ in the position of this proton either in Tyr2034 or the C_2 of ribose (0.11 Å). Thus, we confirm that these differences in the IRC results do not affect the overall mechanism proposed.

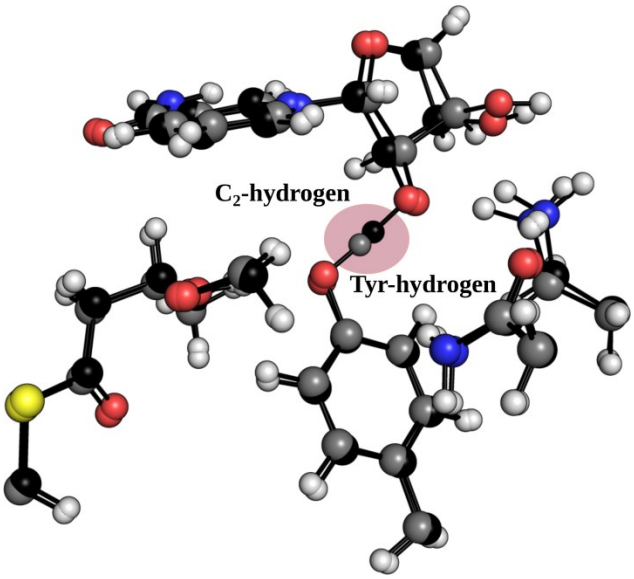

Figure S7. Aligned structures for the **I**. The black atoms represent the structure of the IRC from **TS1** and the gray atoms that of the **TS2**. The transparent red circle shows the hydrogen atoms that are interchanged in the **I** state.

XYZ coordinates of the QM layer from optimized structures

react.xyz				**ts1.xyz**			
C	5.155	6.115	-0.351	C	5.154	6.140	-0.372
H	5.963	6.673	-0.830	H	5.957	6.700	-0.859
H	5.252	6.236	0.732	H	5.262	6.262	0.710
C	5.263	4.641	-0.742	C	5.262	4.668	-0.763
H	4.935	4.484	-1.771	H	4.917	4.507	-1.787
H	6.289	4.288	-0.650	H	6.290	4.320	-0.686
N	4.438	3.742	0.125	N	4.456	3.769	0.122
H	3.442	4.025	0.140	H	3.453	4.025	0.131
H	4.407	2.764	-0.255	H	4.463	2.784	-0.227
H	4.813	3.727	1.089	H	4.836	3.787	1.085
C	9.523	0.178	3.053	C	9.528	0.347	3.213
H	8.804	-0.061	2.264	H	8.798	0.325	2.397
H	9.635	1.275	3.077	H	9.785	1.414	3.379
O	10.799	-0.401	2.805	O	10.679	-0.410	2.905
H	10.759	-0.919	1.982	H	10.647	-0.626	1.937
C	11.221	5.831	-0.746	C	11.257	6.012	-0.756
H	11.196	6.100	-1.809	H	11.234	6.309	-1.810
H	12.277	5.696	-0.505	H	12.311	5.861	-0.516
C	10.508	4.514	-0.553	C	10.526	4.704	-0.591
C	9.247	4.259	-1.113	C	9.272	4.476	-1.174
H	8.726	5.052	-1.648	H	8.779	5.278	-1.723
C	8.664	2.993	-1.045	C	8.661	3.224	-1.107
H	7.703	2.784	-1.500	H	7.710	3.035	-1.593
C	9.342	1.937	-0.421	C	9.298	2.150	-0.464
O	8.736	0.713	-0.422	O	8.675	0.948	-0.444
H	9.374	-0.018	-0.207	H	9.333	0.179	-0.141
C	10.586	2.172	0.175	C	10.550	2.362	0.134
H	11.107	1.366	0.682	H	11.060	1.533	0.610
C	11.145	3.449	0.100	C	11.135	3.625	0.066
H	12.125	3.616	0.539	H	12.116	3.770	0.512
C	7.192	5.541	4.044	C	7.223	5.591	4.013
H	8.238	5.827	4.204	H	8.268	5.878	4.177
H	6.878	4.969	4.920	H	6.908	5.009	4.883
C	6.992	4.617	2.856	C	7.023	4.680	2.815
O	5.859	4.282	2.508	O	5.889	4.319	2.490
N	8.127	4.115	2.300	N	8.155	4.227	2.222
H	8.052	3.617	1.419	H	8.090	3.733	1.338
H	8.939	4.733	2.345	H	8.964	4.847	2.282
C	6.391	-2.135	1.612	C	6.915	-1.834	1.314
H	5.868	-1.554	2.359	H	6.504	-1.200	2.085

C	7.478	-2.862	1.884	C	8.064	-2.539	1.476
H	7.855	-2.888	2.899	H	8.597	-2.490	2.414
C	8.170	-3.702	0.847	C	8.554	-3.376	0.407
H	9.221	-3.402	0.737	H	9.642	-2.726	-0.094
H	8.240	-4.739	1.202	H	9.142	-4.247	0.676
C	7.449	-3.659	-0.493	C	7.660	-3.566	-0.728
C	6.342	-2.904	-0.673	C	6.519	-2.823	-0.831
H	5.789	-2.893	-1.605	H	5.816	-2.914	-1.651
C	8.041	-4.544	-1.536	C	8.106	-4.622	-1.698
O	9.112	-5.133	-1.248	O	9.119	-5.281	-1.386
N	7.461	-4.715	-2.745	N	7.422	-4.834	-2.831
H	7.872	-5.500	-3.254	H	7.762	-5.661	-3.329
H	6.593	-4.315	-3.131	H	6.601	-4.347	-3.224
N	5.803	-2.111	0.340	N	6.159	-1.943	0.170
C	4.836	-1.092	0.032	C	5.049	-1.011	-0.029
H	4.596	-0.573	0.972	H	4.846	-0.544	0.945
C	5.315	-0.035	-0.996	C	5.379	0.096	-1.061
H	5.952	-0.531	-1.736	H	6.003	-0.344	-1.850
C	4.031	0.385	-1.694	C	4.013	0.411	-1.662
H	4.191	0.640	-2.740	H	4.082	0.676	-2.714
O	3.470	1.523	-1.017	O	3.421	1.499	-0.938
H	2.743	1.826	-1.595	H	2.706	1.829	-1.515
O	5.980	1.070	-0.410	O	5.984	1.234	-0.497
H	6.920	0.826	-0.297	H	6.958	1.077	-0.438
O	3.680	-1.689	-0.533	O	3.935	-1.708	-0.522
C	3.128	-0.838	-1.548	C	3.219	-0.881	-1.467
H	2.137	-0.495	-1.219	H	2.243	-0.625	-1.033
O	10.561	-1.221	0.107	O	10.299	-0.783	0.311
C	11.063	-1.959	-0.749	C	10.556	-1.838	-0.434
C	10.584	-2.006	-2.163	C	10.277	-1.714	-1.926
H	9.772	-1.299	-2.343	H	9.283	-1.293	-2.105
H	11.438	-1.794	-2.819	H	11.031	-1.048	-2.358
H	10.243	-3.028	-2.377	H	10.345	-2.687	-2.422
C	12.259	-2.809	-0.385	C	11.829	-2.606	-0.058
H	12.184	-3.127	0.658	H	11.791	-2.882	1.001
H	12.299	-3.697	-1.026	H	11.877	-3.535	-0.641
C	13.559	-1.992	-0.611	C	13.123	-1.810	-0.329
O	13.733	-1.305	-1.605	O	13.289	-1.086	-1.296
C	16.106	-1.181	0.052	C	15.757	-1.154	0.153
H	15.846	-0.123	0.094	H	15.541	-0.087	0.210
H	16.243	-1.470	-0.998	H	15.811	-1.441	-0.906
S	14.710	-2.175	0.704	S	14.373	-2.101	0.895

int.xyz				**ts2.xyz**			
C	5.147	6.163	-0.299	C	5.251	6.181	-0.272
H	5.949	6.728	-0.783	H	6.012	6.831	-0.716
H	5.252	6.286	0.783	H	5.327	6.274	0.817
C	5.272	4.693	-0.691	C	5.529	4.742	-0.709
H	4.923	4.525	-1.712	H	5.266	4.603	-1.761
H	6.308	4.366	-0.620	H	6.595	4.549	-0.611

168	N	4.491	3.778	0.201	N	4.846	3.678	0.077
169	H	3.475	3.973	0.177	H	3.825	3.732	-0.006
170	H	4.584	2.790	-0.116	H	5.234	2.571	-0.233
171	H	4.850	3.840	1.173	H	5.079	3.778	1.076
172	C	9.557	0.201	3.138	C	9.567	0.190	3.166
173	H	8.834	0.259	2.318	H	8.834	0.153	2.355
174	H	9.926	1.235	3.293	H	9.823	1.258	3.302
175	O	10.619	-0.693	2.852	O	10.728	-0.570	2.876
176	H	10.704	-0.795	1.876	H	10.727	-0.819	1.926
177	C	11.137	5.972	-0.685	C	11.285	5.863	-0.650
178	H	11.097	6.258	-1.743	H	11.316	6.148	-1.708
179	H	12.196	5.807	-0.467	H	12.323	5.697	-0.355
180	C	10.390	4.676	-0.479	C	10.513	4.575	-0.500
181	C	9.149	4.420	-1.076	C	9.285	4.375	-1.143
182	H	8.677	5.193	-1.683	H	8.866	5.166	-1.764
183	C	8.518	3.180	-0.950	C	8.596	3.167	-1.031
184	H	7.583	2.982	-1.464	H	7.655	3.005	-1.541
185	C	9.105	2.105	-0.230	C	9.114	2.118	-0.260
186	O	8.515	0.927	-0.132	O	8.429	0.945	-0.118
187	H	9.764	-0.263	-0.068	H	9.745	-0.489	-0.107
188	C	10.367	2.372	0.366	C	10.363	2.282	0.356
189	H	10.877	1.563	0.874	H	10.805	1.458	0.902
190	C	10.972	3.618	0.240	C	11.035	3.499	0.232
191	H	11.953	3.774	0.688	H	12.008	3.612	0.705
192	C	7.185	5.583	4.116	C	7.279	5.548	4.209
193	H	8.231	5.871	4.265	H	8.319	5.860	4.355
194	H	6.881	5.001	4.990	H	6.985	4.977	5.094
195	C	6.966	4.676	2.917	C	7.086	4.611	3.030
196	O	5.820	4.323	2.616	O	5.960	4.250	2.692
197	N	8.081	4.223	2.306	N	8.232	4.127	2.473
198	H	8.014	3.720	1.424	H	8.159	3.624	1.596
199	H	8.901	4.827	2.362	H	9.023	4.774	2.493
200	C	6.856	-1.858	1.376	C	6.569	-1.714	1.335
201	H	6.507	-1.131	2.092	H	6.172	-1.004	2.043
202	C	7.994	-2.618	1.552	C	7.591	-2.590	1.633
203	H	8.602	-2.507	2.439	H	8.035	-2.600	2.618
204	C	8.346	-3.515	0.553	C	8.007	-3.472	0.646
205	H	10.046	-2.413	-1.137	H	10.099	-2.734	-0.916
206	H	9.231	-4.129	0.642	H	8.808	-4.174	0.830
207	C	7.579	-3.645	-0.613	C	7.405	-3.485	-0.622
208	C	6.433	-2.874	-0.738	C	6.354	-2.603	-0.863
209	H	5.755	-2.914	-1.584	H	5.789	-2.572	-1.789
210	C	8.089	-4.662	-1.620	C	7.982	-4.525	-1.582
211	O	9.088	-5.319	-1.284	O	8.995	-5.134	-1.196
212	N	7.453	-4.820	-2.784	N	7.381	-4.764	-2.747
213	H	7.814	-5.626	-3.304	H	7.779	-5.572	-3.237
214	H	6.632	-4.333	-3.182	H	6.560	-4.296	-3.169
215	N	6.107	-2.004	0.258	N	5.976	-1.734	0.118
216	C	4.986	-1.046	0.060	C	4.843	-0.777	-0.082
217	H	4.744	-0.643	1.052	H	4.649	-0.348	0.908
218	C	5.416	0.109	-0.887	C	5.118	0.394	-1.072
219	H	6.115	-0.306	-1.626	H	5.578	-0.041	-1.977

C	4.101	0.428	-1.616	C	3.648	0.709	-1.428
H	4.283	0.653	-2.665	H	3.547	1.262	-2.361
O	3.475	1.544	-0.986	O	3.071	1.426	-0.323
H	2.775	1.830	-1.606	H	2.550	2.184	-0.641
O	5.963	1.196	-0.214	O	5.873	1.428	-0.556
H	6.992	1.096	-0.195	H	7.442	1.103	-0.334
O	3.923	-1.720	-0.524	O	3.761	-1.507	-0.564
C	3.254	-0.839	-1.468	C	3.006	-0.672	-1.498
H	2.285	-0.557	-1.037	H	1.994	-0.584	-1.093
O	10.548	-0.850	0.121	O	10.533	-1.010	0.149
C	10.840	-1.658	-1.025	C	10.849	-1.933	-0.913
C	10.860	-0.810	-2.297	C	10.808	-1.228	-2.267
H	9.874	-0.356	-2.449	H	9.799	-0.837	-2.455
H	11.606	-0.017	-2.232	H	11.524	-0.407	-2.307
H	11.092	-1.427	-3.172	H	11.048	-1.932	-3.072
C	12.120	-2.465	-0.737	C	12.174	-2.626	-0.566
H	11.991	-2.985	0.219	H	12.073	-3.098	0.419
H	12.200	-3.238	-1.513	H	12.310	-3.441	-1.290
C	13.448	-1.710	-0.754	C	13.453	-1.788	-0.612
O	13.791	-1.005	-1.691	O	13.718	-1.016	-1.521
C	15.987	-1.105	0.054	C	16.007	-1.137	0.132
H	15.786	-0.035	0.112	H	15.773	-0.076	0.215
H	16.121	-1.370	-1.002	H	16.141	-1.380	-0.930
S	14.510	-2.024	0.636	S	14.577	-2.120	0.720

prod.xyz

C	5.213	6.231	-0.298
H	5.999	6.845	-0.752
H	5.283	6.339	0.790
C	5.400	4.762	-0.696
H	5.231	4.653	-1.773
H	6.438	4.487	-0.503
N	4.552	3.781	0.012
H	3.562	4.042	-0.101
H	5.334	2.039	-0.248
H	4.750	3.840	1.014
C	9.597	0.138	3.114
H	8.867	0.094	2.300
H	9.879	1.202	3.221
O	10.733	-0.670	2.860
H	10.781	-0.886	1.904
C	11.308	5.899	-0.653
H	11.324	6.184	-1.712
H	12.351	5.749	-0.370
C	10.561	4.598	-0.501
C	9.282	4.413	-1.040
H	8.789	5.232	-1.560
C	8.630	3.183	-0.949
H	7.648	3.043	-1.386
C	9.248	2.103	-0.310
O	8.615	0.889	-0.198

272	H	9.883	-0.556	-0.163
273	C	10.535	2.253	0.214
274	H	11.034	1.403	0.666
275	C	11.167	3.493	0.117
276	H	12.171	3.601	0.518
277	C	7.272	5.552	4.193
278	H	8.309	5.868	4.352
279	H	6.971	4.977	5.074
280	C	7.099	4.616	3.008
281	O	5.991	4.229	2.656
282	N	8.267	4.156	2.458
283	H	8.190	3.696	1.557
284	H	9.027	4.840	2.474
285	C	6.597	-1.833	1.338
286	H	6.186	-1.140	2.055
287	C	7.675	-2.650	1.606
288	H	8.152	-2.628	2.576
289	C	8.106	-3.513	0.608
290	H	10.221	-2.821	-0.952
291	H	8.946	-4.173	0.769
292	C	7.466	-3.563	-0.640
293	C	6.365	-2.740	-0.855
294	H	5.770	-2.738	-1.763
295	C	8.048	-4.582	-1.614
296	O	9.061	-5.196	-1.238
297	N	7.444	-4.793	-2.783
298	H	7.830	-5.601	-3.283
299	H	6.617	-4.323	-3.189
300	N	5.974	-1.889	0.137
301	C	4.818	-0.967	-0.050
302	H	4.603	-0.555	0.943
303	C	5.079	0.207	-1.019
304	H	5.565	-0.169	-1.927
305	C	3.619	0.566	-1.329
306	H	3.502	1.171	-2.231
307	O	3.010	1.166	-0.191
308	H	3.238	2.114	-0.162
309	O	5.878	1.226	-0.456
310	H	7.648	1.015	-0.364
311	O	3.754	-1.691	-0.570
312	C	2.988	-0.811	-1.459
313	H	1.978	-0.747	-1.051
314	O	10.657	-1.090	0.104
315	C	10.969	-2.017	-0.958
316	C	10.915	-1.318	-2.314
317	H	9.905	-0.931	-2.500
318	H	11.630	-0.496	-2.364
319	H	11.151	-2.026	-3.117
320	C	12.301	-2.697	-0.617
321	H	12.204	-3.187	0.359
322	H	12.453	-3.496	-1.354
323	C	13.562	-1.831	-0.640

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O  13.812  -1.031  -1.528
C  16.096  -1.141   0.138
H  15.833  -0.088   0.233
H  16.250  -1.365  -0.925
S  14.682  -2.167   0.692
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TOC

We study the chemical reaction involved in the human β–ketoreductase domain. It was studied with the hybrid QM/MM methodology.