Supporting Information

for

Drawing the Retinal Out of its Comfort Zone: An ONIOM(QM/MM) Study of Mutant Squid Rhodopsin

Sivakumar Sekharan[†], Keiji Morokuma[†‡*]

[†]Cherry L. Emerson Center for Scientific Computation and Department of Chemistry, Emory University, Atlanta, Georgia 30322 and [¶]Fukui Institute for Fundamental Chemistry, Kyoto University, 34-4 Takano Nishihiraki-cho, Kyoto 606-8103, Japan

I. ONIOM (QM:MM) where QM=B3LYP/6-31G*; MM=AMBER; optimized cartesian coordinates (in Å)

1. Wildtype (WT) protonated Schiff base retinal optimized inside the protein environment.

51 atoms

N	9.796233	38.054782	57.858373
H	8.874230	38.327781	58.190451
H	9.949860	37.043264	57.784239
C	17.131212	44.828289	61.316098
C	18.532048	44.846576	60.654834
C	18.870817	46.259227	60.126053
H	19.906116	46.277020	59.772084
H	18.779472	47.006821	60.922821
H	18.237644	46.559165	59.284186
C	18.548568	43.860266	59.463264
H	18.399492	42.824057	59.788053
H	19.506550	43.917572	58.932764
H	17.755349	44.100632	58.751135
C	16.912775	44.359680	62.574668
C	15.608993	44.463847	63.332561
H	15.119096	43.483846	63.410627
H	14.902069	45.168229	62.891403
H	15.819203	44.788952	64.362000
C	19.631556	44.455889	61.667449
H	19.860961	45.321078	62.302797
H	20.550740	44.232709	61.115124
C	19.220124	43.289667	62.561279
H	18.961602	42.416279	61.948123
H	20.049083	42.987873	63.210366
C	18.004888	43.701138	63.388020
H	17.572340	42.835050	63.909033
H	18.305715	44.399838	64.183154
C	16.029066	45.291883	60.462118
H	16.203974	46.156939	59.824983
C	14.869973	44.599724	60.325323
H	14.771476	43.680386	60.893245
C	13.772753	44.876468	59.427957
C	13.645232	46.232245	58.791031
H	14.483039	46.425287	58.111967
H	13.676323	47.020597	59.551211
H	12.721317	46.331297	58.215712
C	12.925458	43.821423	59.175028
H	13.167594	42.897372	59.686577
C	11.761487	43.797790	58.360372
H	11.313150	44.752256	58.089777
C	11.061109	42.678445	57.961636
H	10.061518	42.853916	57.567039
C	11.469136	41.306496	57.959251
C	12.906167	40.878555	58.117623
H	13.087954	40.474542	59.121559
H	13.595416	41.701705	57.942621
H	13.152791	40.094901	57.395900
C	10.472504	40.349016	57.793664
H	9.437313	40.675394	57.796475
C	10.729282	38.979201	57.692339
H	11.721453	38.619755	57.440379

2. Demethyl (DM) protonated Schiff base retinal optimized inside the protein environment.

36 atoms

N	9.915745	38.080057	58.002919
H	9.020141	38.208349	58.488824
H	10.192285	37.099237	57.883151
C	17.186128	44.717314	61.187811
C	18.621500	44.930456	60.736518
H	18.737739	45.952250	60.349759
H	18.816066	44.273244	59.876336
C	16.882991	44.162850	62.383152
H	15.838511	44.087938	62.679825
C	19.650854	44.658283	61.844471
H	19.703863	45.530039	62.508804
H	20.645603	44.539774	61.402508
C	19.263020	43.423172	62.668098
H	19.178659	42.553239	62.001217
H	20.038917	43.184410	63.401498
C	17.909136	43.648094	63.353356
H	17.536802	42.724323	63.815716
H	18.012697	44.367390	64.182718
C	16.120213	45.111653	60.267981
H	16.258466	46.017334	59.675557
C	15.015524	44.344205	60.072249
H	14.976640	43.386970	60.585310
C	13.891377	44.638123	59.240499
H	13.780824	45.645314	58.837810
C	12.963336	43.672221	58.955792
H	13.165567	42.675341	59.340811
C	11.720235	43.869744	58.298236
H	11.409444	44.895116	58.108652
C	10.827360	42.858264	58.010018
H	9.799631	43.122391	57.770003
C	11.178349	41.482803	57.963395
H	12.234441	41.237911	57.871462
C	10.281601	40.437824	57.987568
H	9.232048	40.618786	58.192712
C	10.705225	39.113830	57.787550
H	11.704031	38.913537	57.399489

3. Deprotonated Schiff base retinal optimized in the presence of protonated Glu180 counterion inside the protein environment (SBR^{cip})

50 atoms

N	9.695676	38.104006	57.472643
H	9.896399	37.085265	57.492339
C	17.142197	44.840839	61.322339
C	18.537437	44.886512	60.649560
C	18.869412	46.320741	60.175676
H	19.898806	46.355960	59.803229
H	18.790147	47.035319	61.003600
H	18.218396	46.651346	59.359770
C	18.548482	43.957192	59.414113
H	18.410323	42.906373	59.695109
H	19.500288	44.047122	58.875884
H	17.742954	44.227186	58.727260
C	16.948504	44.344800	62.571324
C	15.655792	44.427810	63.351369
H	15.167137	43.446407	63.419088
H	14.943036	45.138064	62.928922
H	15.879222	44.735692	64.384314
C	19.652847	44.457969	61.629526
H	19.896111	45.299969	62.289661
H	20.563550	44.250361	61.055592
C	19.251242	43.266028	62.492279
H	18.980288	42.413786	61.854358
H	20.089923	42.940872	63.118354
C	18.049068	43.654809	63.349372
H	17.622275	42.770841	63.844687
H	18.366095	44.320872	64.165830
C	16.021802	45.309384	60.479890
H	16.177619	46.199772	59.873269
C	14.878477	44.602654	60.326615
H	14.792216	43.678847	60.890794
C	13.762184	44.866560	59.431252
C	13.655819	46.212230	58.762136
H	14.492785	46.385489	58.075885
H	13.697043	47.019562	59.503385
H	12.728659	46.310499	58.190294
C	12.901654	43.828159	59.210696
H	13.119538	42.922764	59.764682
C	11.733866	43.776529	58.364853

H	11.280078	44.727719	58.091937
C	11.080563	42.663374	57.921469
H	10.124490	42.837087	57.429238
C	11.451691	41.255954	57.982777
C	12.853410	40.839465	58.375143
H	12.924930	40.645042	59.450773
H	13.576298	41.614817	58.116429
H	13.157662	39.922247	57.867063
C	10.509448	40.330818	57.647610
H	9.515127	40.661127	57.362066
C	10.679438	38.891418	57.713503
H	11.679470	38.538137	57.997315

4. Deprotonated Schiff base retinal optimized in the presence of deprotonated Glu180 counterion inside the protein environment (SBR^{cidp})

50 atoms

N	9.643853	38.109596	57.571046
H	9.856519	37.092673	57.550323
C	17.150058	44.847725	61.320925
C	18.550389	44.894188	60.660561
C	18.881675	46.328078	60.184870
H	19.913036	46.365231	59.817857
H	18.795680	47.044823	61.010636
H	18.233364	46.654550	59.365209
C	18.574603	43.960646	59.428503
H	18.437786	42.910532	59.712100
H	19.530578	44.051690	58.897831
H	17.773638	44.223834	58.734024
C	16.945524	44.354835	62.569040
C	15.642558	44.432844	63.332854
H	15.155465	43.449993	63.392351
H	14.933579	45.140923	62.900436
H	15.850686	44.740994	64.369247
C	19.657695	44.471417	61.652232
H	19.892940	45.316169	62.311891
H	20.574564	44.263130	61.088135
C	19.249861	43.281478	62.515152
H	18.984363	42.428025	61.876925
H	20.083826	42.957934	63.148427
C	18.039647	43.670325	63.360896
H	17.610682	42.786374	63.854548
H	18.347664	44.339481	64.178975
C	16.033319	45.310586	60.469891
H	16.178104	46.213719	59.879503
C	14.904934	44.585004	60.298316
H	14.836255	43.648944	60.845188
C	13.779194	44.849069	59.415816
C	13.660918	46.200716	58.760169
H	14.492700	46.383826	58.070052
H	13.701575	47.003217	59.507403
H	12.730315	46.297322	58.193692
C	12.918350	43.810998	59.198773
H	13.150493	42.896889	59.733537
C	11.730271	43.767989	58.381548
H	11.273048	44.721760	58.123960
C	11.064292	42.657670	57.952668
H	10.095598	42.835468	57.486900
C	11.437956	41.249378	57.988567
C	12.858898	40.820301	58.292459
H	12.976869	40.540833	59.346188
H	13.566072	41.619359	58.067533
H	13.148280	39.955777	57.692369
C	10.471507	40.333794	57.702600
H	9.460932	40.670492	57.489006
C	10.649305	38.893780	57.720160
H	11.675342	38.540642	57.871286

II. Geometric parameters of Wildtype (**WT**) protonated Schiff base retinal, Demethyl (**DM**) protonated Schiff base retinal, deprotonated Schiff base retinal in the presence of a) protonated Glu180 counterion (**SBR**^{cip}) and b) deprotonated Glu180 counterion (**SBR**^{cidp}).

Bond lengths (Å)

Bonds	WT	DM	SBR^{cip}	SBR^{cidp}
C5–C6	1.361	1.352	1.358	1.357
C6–C7	1.469	1.462	1.478	1.478
C7–C8	1.357	1.359	1.353	1.352
C8–C9	1.444	1.429	1.455	1.455
C9–C10	1.377	1.369	1.367	1.366
C10–C11	1.421	1.420	1.443	1.443
C11–C12	1.379	1.380	1.365	1.364
C12–C13	1.431	1.420	1.457	1.457
C13–C14	1.391	1.377	1.362	1.362
C14–C15	1.397	1.405	1.451	1.451
C15–N16	1.324	1.318	1.283	1.284

Bond angles (°)

Bonds	WT	DM	SBR^{cip}	SBR^{cidp}
C5–C6–C7	121.748	120.220	122.142	121.949
C6–C7–C8	122.567	122.060	122.656	122.364
C7–C8–C9	127.903	127.375	128.557	128.704
C8–C9–C10	115.826	120.603	116.396	116.499
C9–C10–C11	128.475	126.254	129.157	129.176
C10–C11–C12	126.459	124.142	127.380	127.193
C11–C12–C13	129.361	123.845	130.720	130.820
C12–C13–C14	117.101	124.981	118.003	117.554
C13–C14–C15	123.500	121.089	125.557	125.294
C14–C15–N16	123.102	122.360	120.699	120.584

Dihedral angle (°)

Bonds	WT	DM	SBR^{cip}	SBR^{cidp}
C5–C6–C7–C8	-45.525	-40.891	-46.107	-47.181
C6–C7–C8–C9	-173.494	177.762	-172.954	-174.361
C7–C8–C9–C10	160.078	168.350	163.141	165.028
C8–C9–C10–C11	178.902	171.088	-178.694	-179.000
C9–C10–C11–C12	168.896	178.694	163.349	164.304
C10–C11–C12–C13	-17.328	-19.388	169.584	168.841
C11–C12–C13–C14	165.790	164.178	169.803	168.555
C12–C13–C14–C15	176.110	172.812	-176.787	-178.450
C13–C14–C15–N16	163.185	169.516	174.627	172.749

III. WS2 system corresponds to the inclusion of two internal water molecules (W1, W2) near the SB binding region. In this model, W1 forms an H-bond with SB nitrogen and Asn87, and W2 forms an H-bond with W1 and backbone oxygen atom of Tyr111 thus bringing the key distance between SB nitrogen and potential H-bonding residues (Asn87, Tyr111) within 0.5 Å deviation and also in good agreement with the X-ray data.