

Supporting Information

Oxyma-B, an Excellent Racemization Suppressor in Peptide Synthesis

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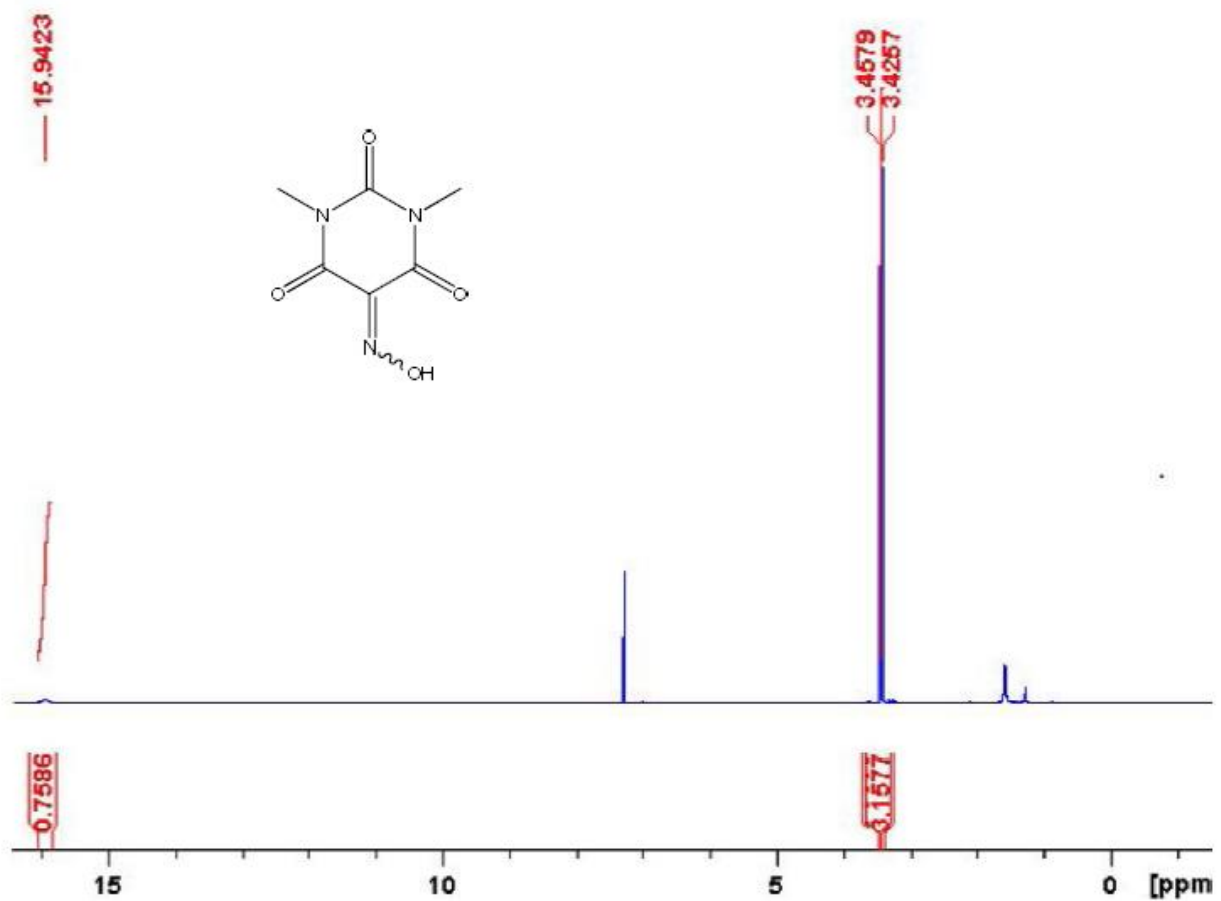
^d*Department of Chemistry, College of Science, King Saud University, P.O. Box 2455, Riyadh 11451, Saudi Arabia*

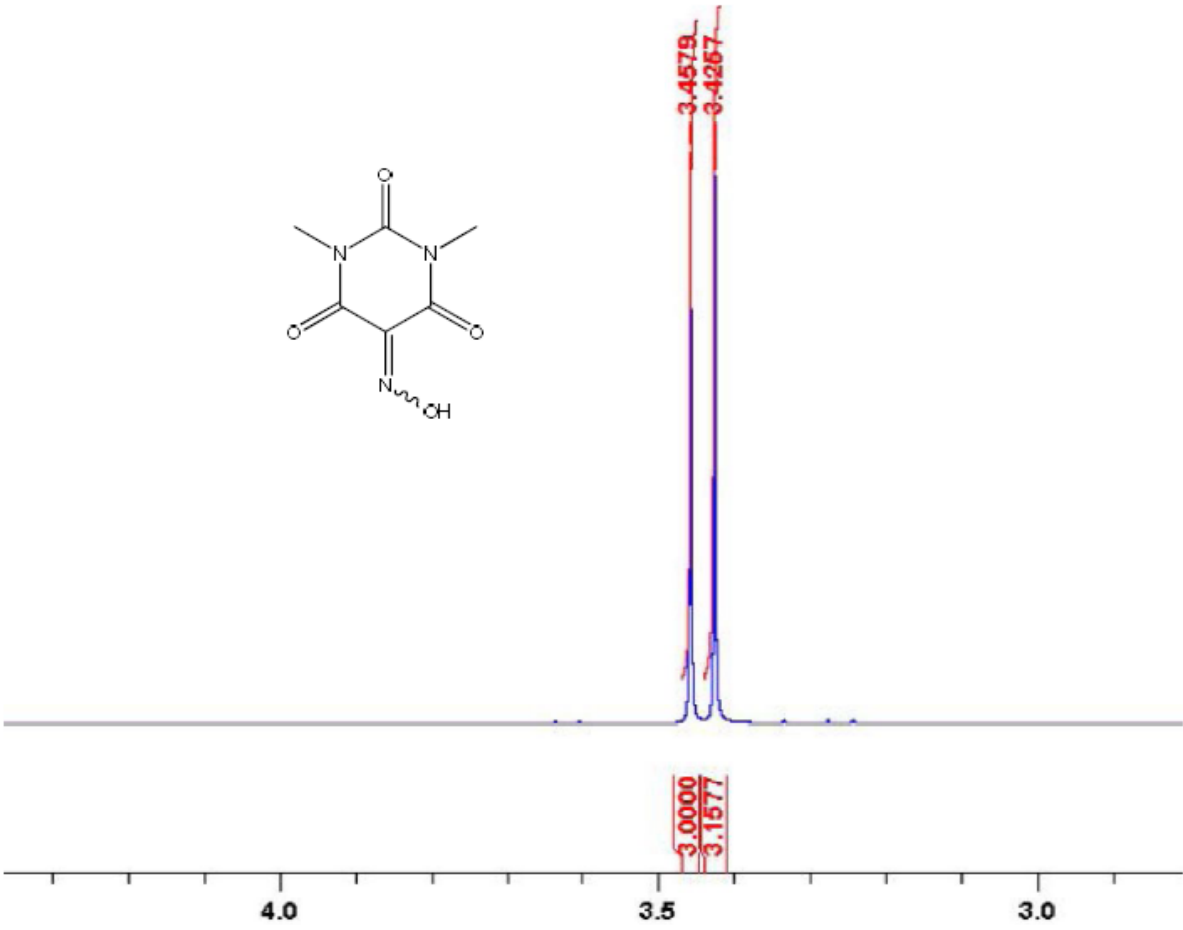
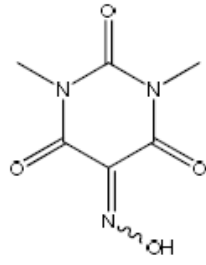
^e*School of Chemistry and Physics, University of KwaZulu-Natal, Durban 4001, South Africa*

^f*Institute for Research in Biomedicine and CIBER-BBN, Barcelona 08028, Spain*

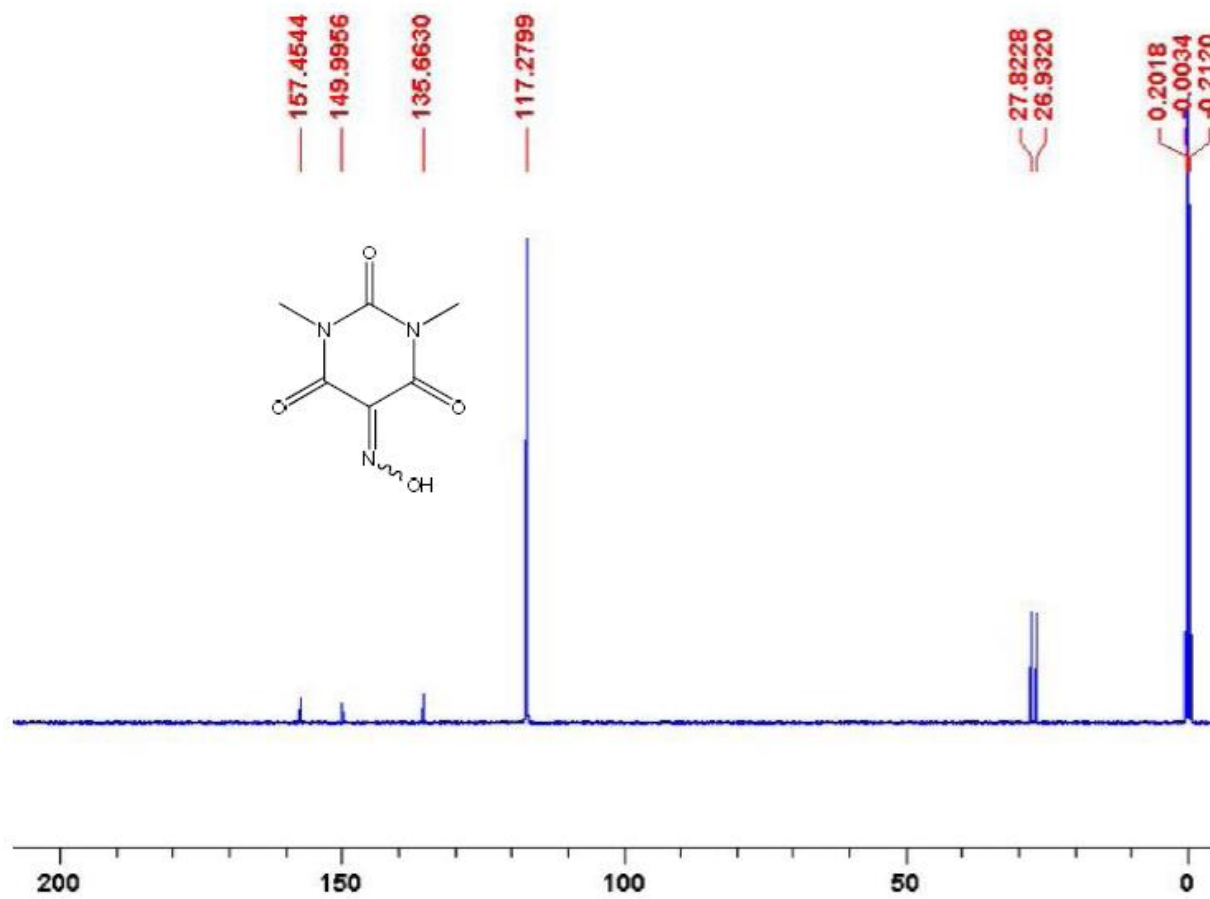
^g*Department of Organic Chemistry, University of Barcelona, Barcelona 08028, Spain*

^1H NMR of Oxyma-B in CDCl_3





^{13}C NMR of Oxyma-B in acetonitrile-d₄



Display Report

Analysis Info

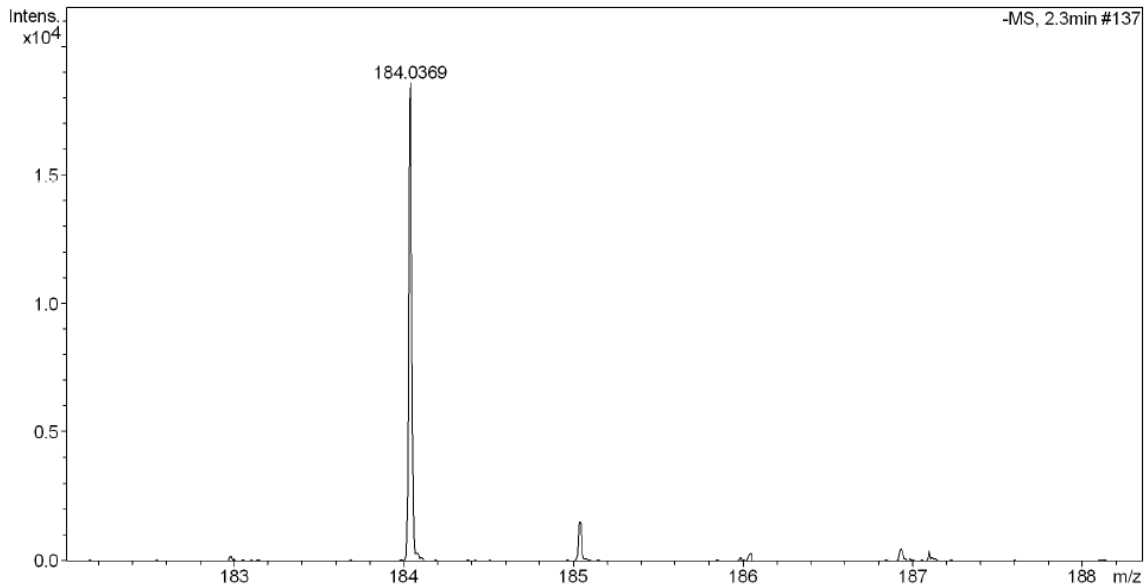
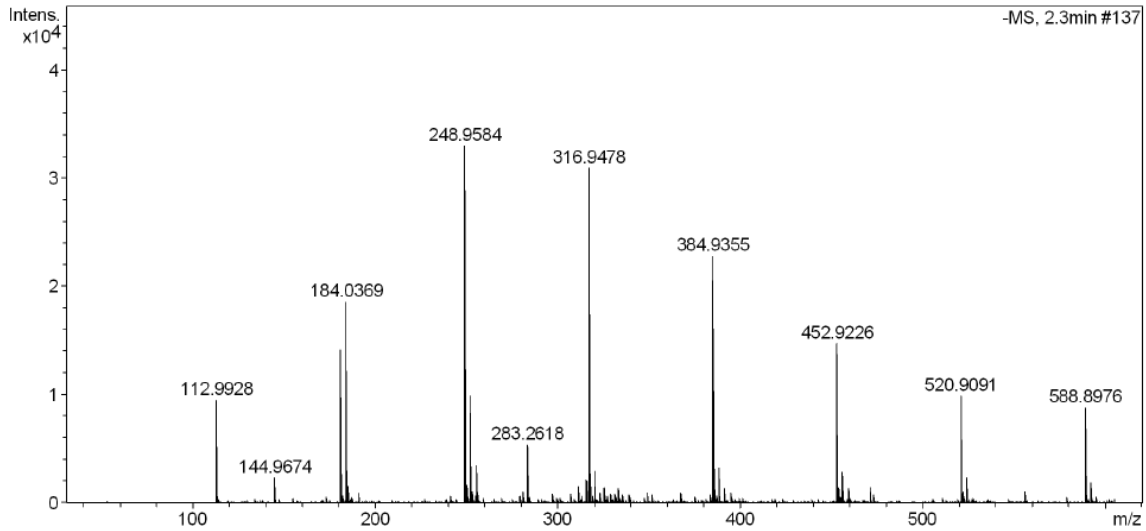
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Method tune_low.m
Sample Name OXYMAB
Comment

Acquisition Date 7/9/2014 11:58:47 AM

Operator BDAL@DE
Instrument micrOTOF-Q 10139

Acquisition Parameter

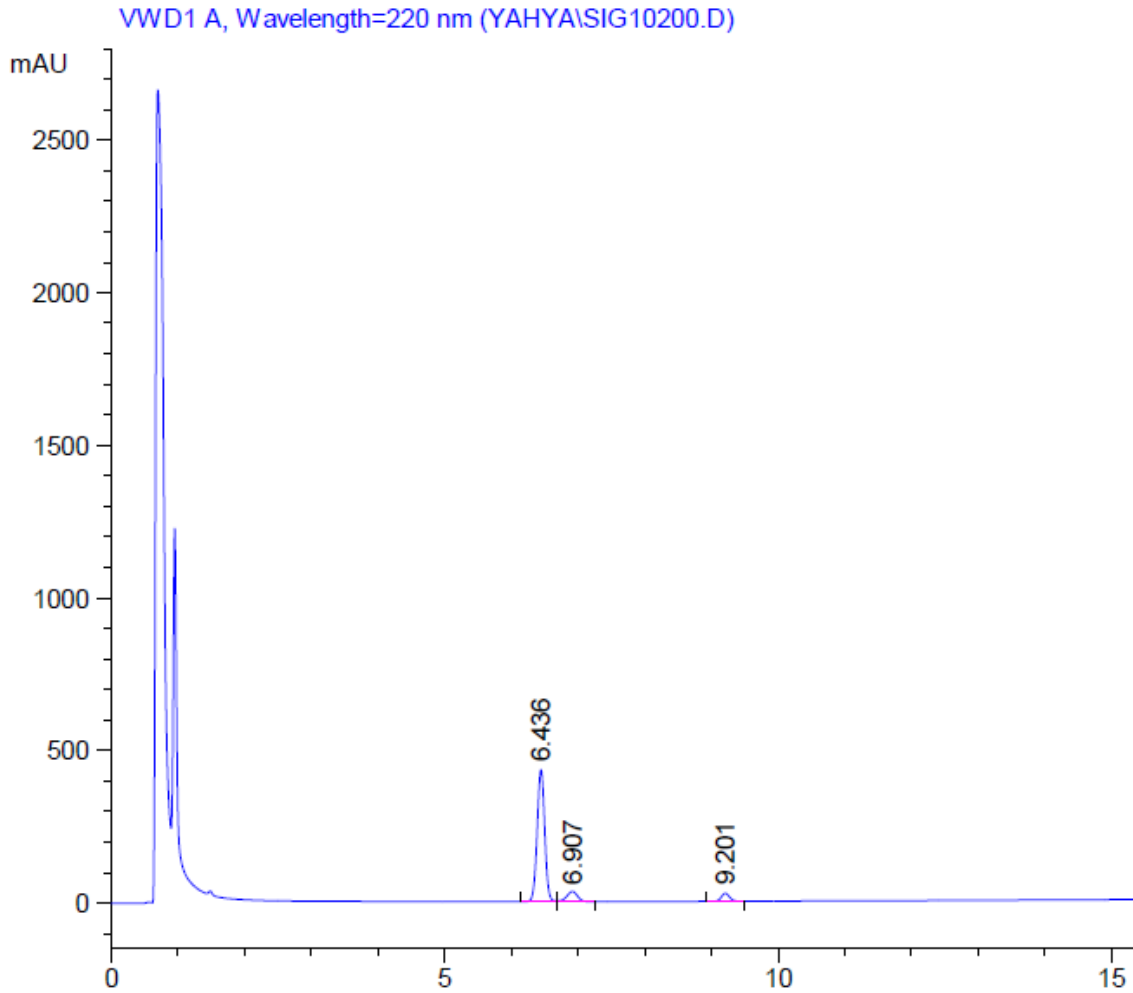
Source Type	ESI	Ion Polarity	Negative	Set Nebulizer	0.4 Bar
Focus	Active	Set Capillary	4000 V	Set Dry Heater	220 °C
Scan Begin	50 m/z	Set End Plate Offset	-500 V	Set Dry Gas	4.0 l/min
Scan End	600 m/z	Set Collision Cell RF	200.0 Vpp	Set Divert Valve	Source



HPLCs

1- Z-Phg-Pro-NH₂ Racemization test:

1.1- HOBt

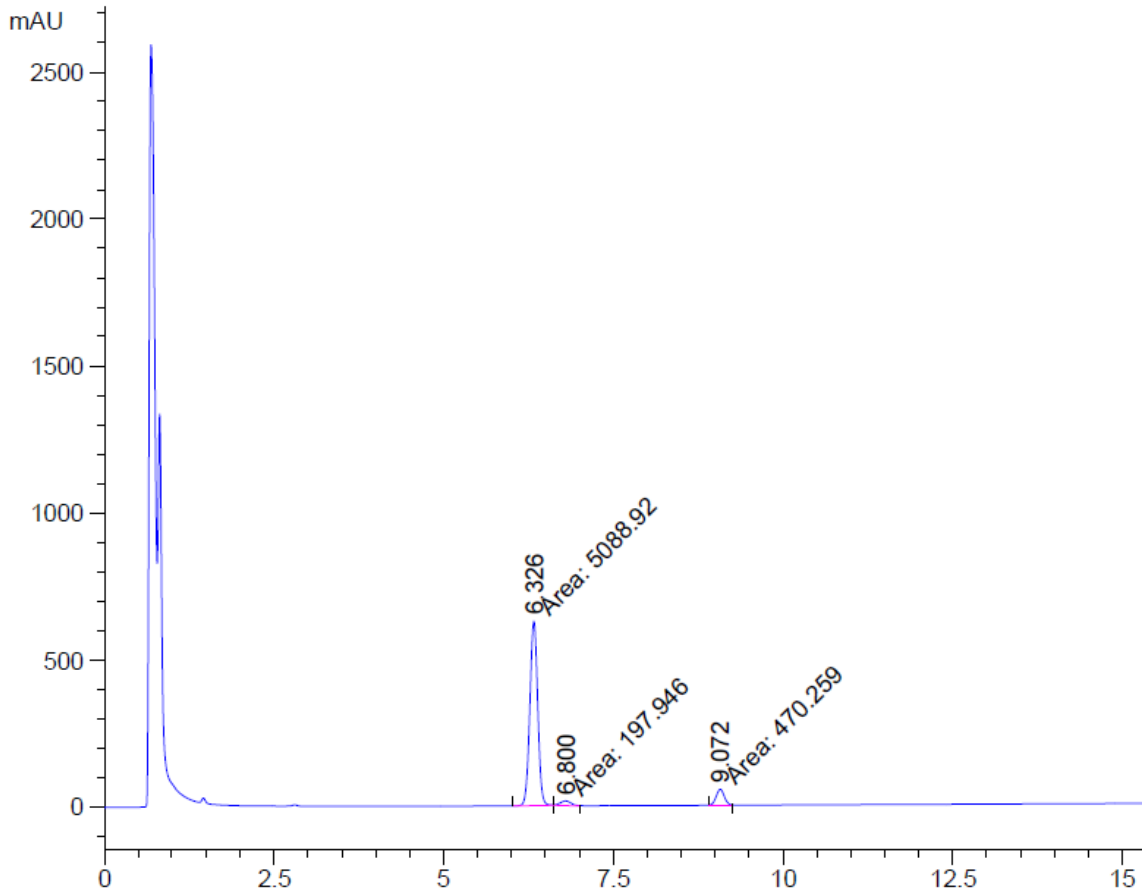


Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.436	PV	0.1242	3414.27417	432.31744	85.0454
2	6.907	VB	0.1751	375.11981	33.31265	9.3438
3	9.201	BB	0.1312	225.25343	26.50491	5.6108

Totals : 4014.64742 492.13500

1.2-HOAt

VWD1 A, Wavelength=220 nm (YAHYA\SIG10309.D)

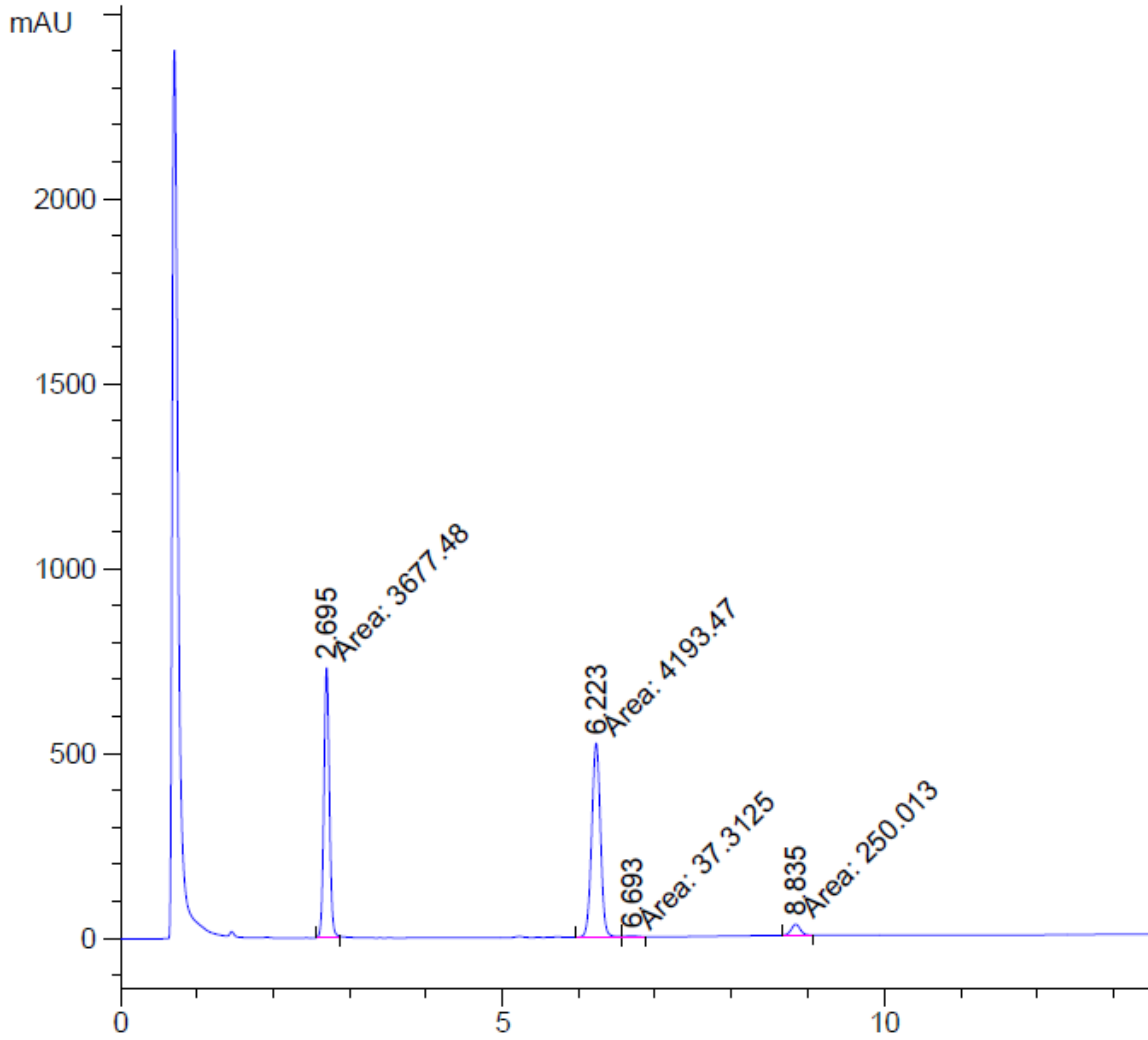


Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.326	MM	0.1349	5088.91650	628.60004	88.3934
2	6.800	MM	0.1974	197.94566	16.70938	3.4383
3	9.072	MM	0.1427	470.25909	54.91633	8.1683

Totals : 5757.12126 700.22575

1.3-OxymaPure

VWD1 A, Wavelength=220 nm (YAHYA\SIG10942.D)

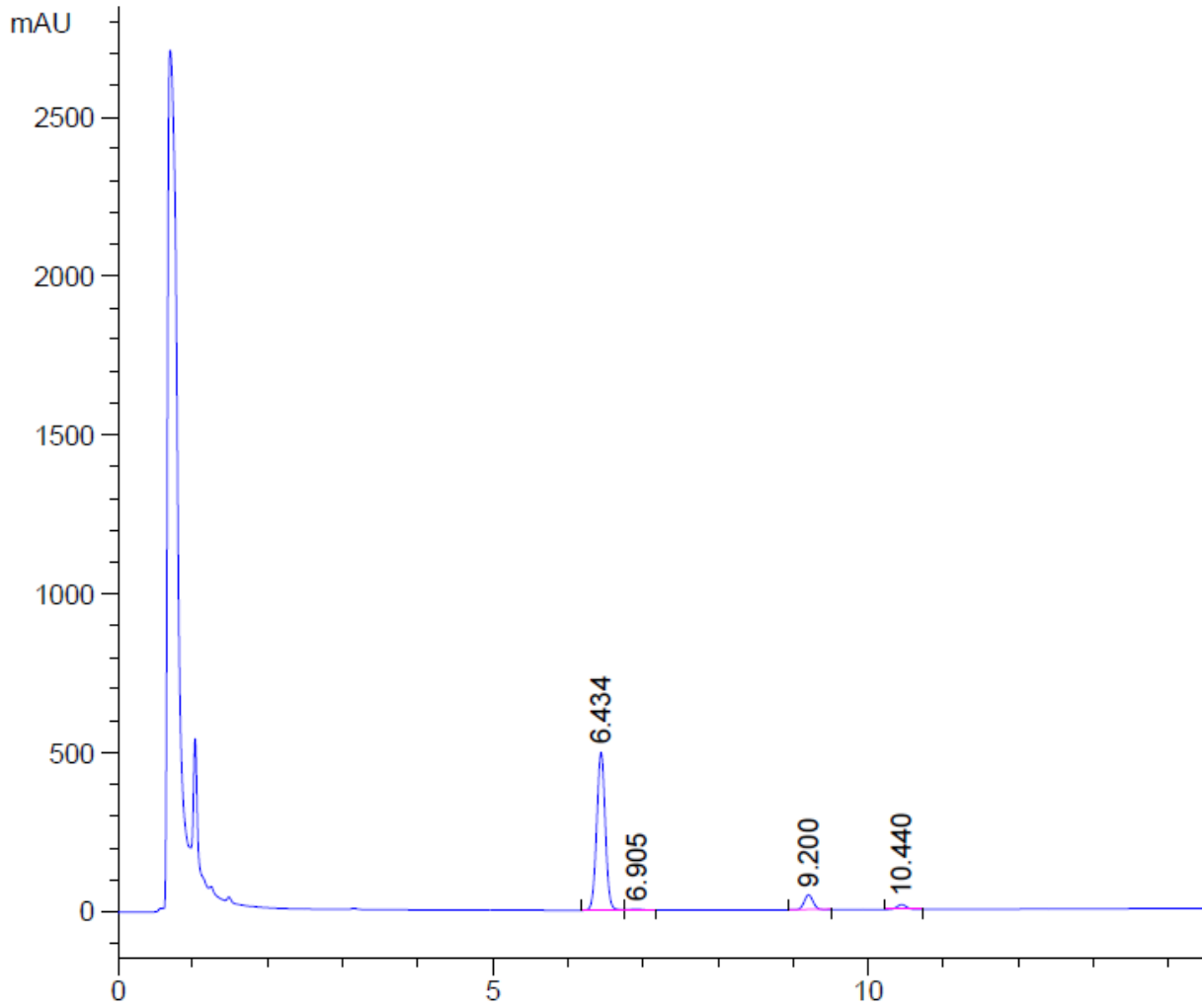


Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	2.695	MM	0.0834	3677.48315	735.30951	45.0767
2	6.223	MF	0.1329	4193.46582	525.72107	51.4014
3	6.693	FM	0.2116	37.31252	2.93868	0.4574
4	8.835	MM	0.1394	250.01295	29.89925	3.0645

Totals : 8158.27445 1293.86851

1.4-Oxyrna-B

VWD1 A, Wavelength=220 nm (YAHYA\SIG10198.D)

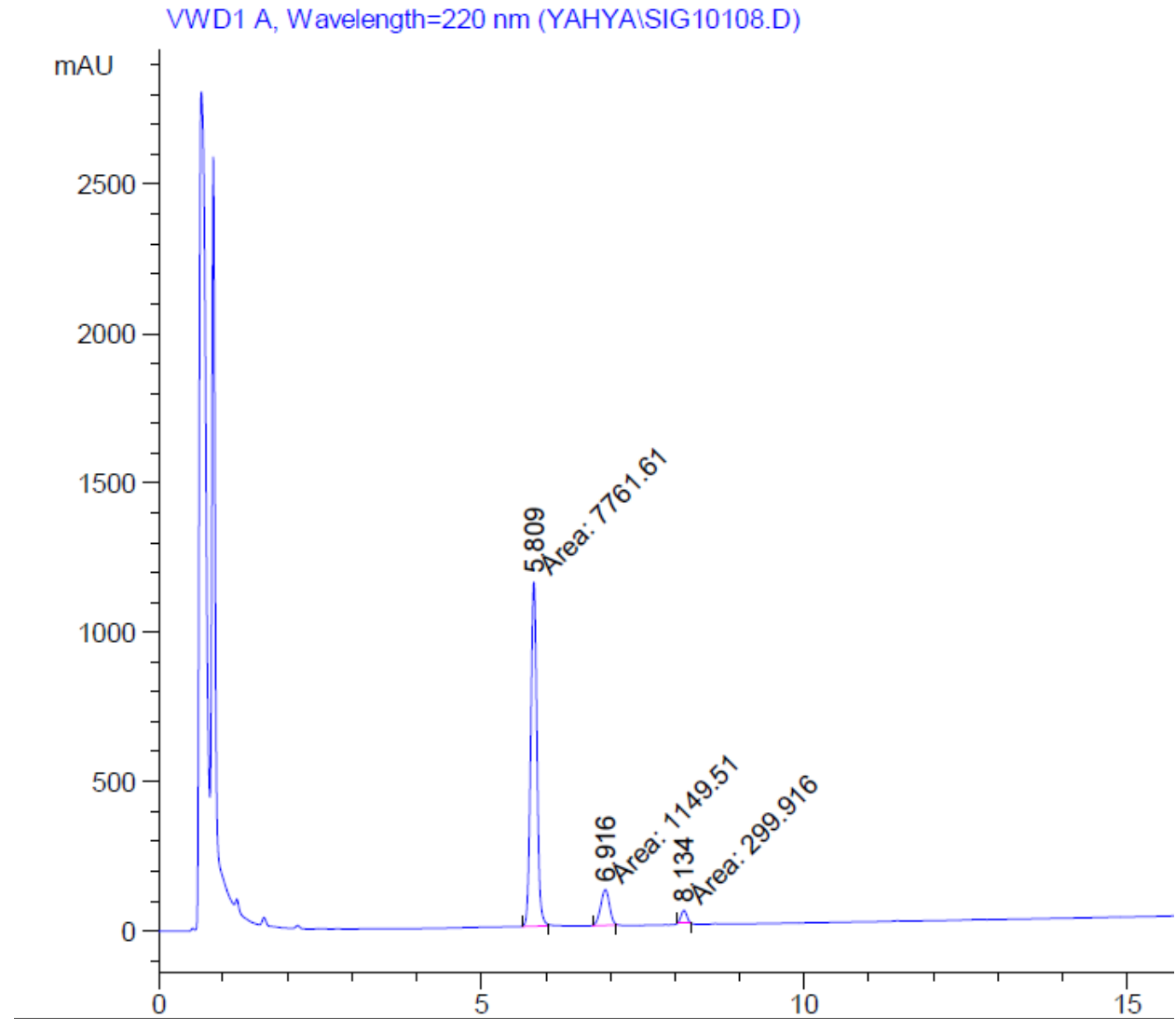


Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.434	PV	0.1222	3953.95679	495.56058	87.3388
2	6.905	VV	0.1890	40.04154	3.15152	0.8845
3	9.200	BB	0.1298	394.05420	47.02825	8.7042
4	10.440	BB	0.1418	139.09808	15.18406	3.0725

Totals : 4527.15061 560.92440

2-Z-Phe-Val-Pro-NH₂ Racemization test:

2.1- HOBt

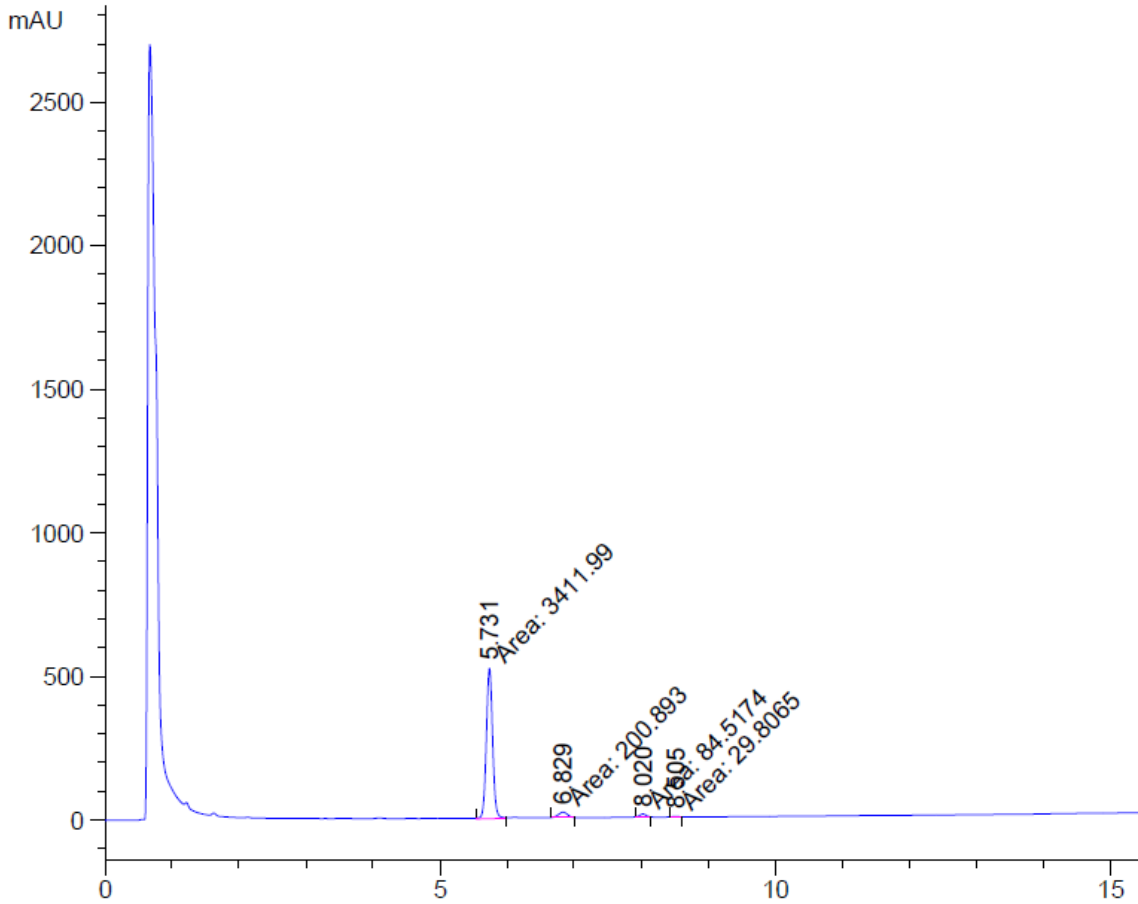


Peak #	RetTime [min]	Type	Width [min]	Area mAU	Area *s	Height [mAU]	Area %
1	5.809	MM	0.1118	7761.60791		1156.86853	84.2642
2	6.916	MM	0.1590	1149.51404		120.45636	12.4797
3	8.134	MM	0.1096	299.91574		45.59239	3.2560

Totals : 9211.03769 1322.91728

2.2-HOAt

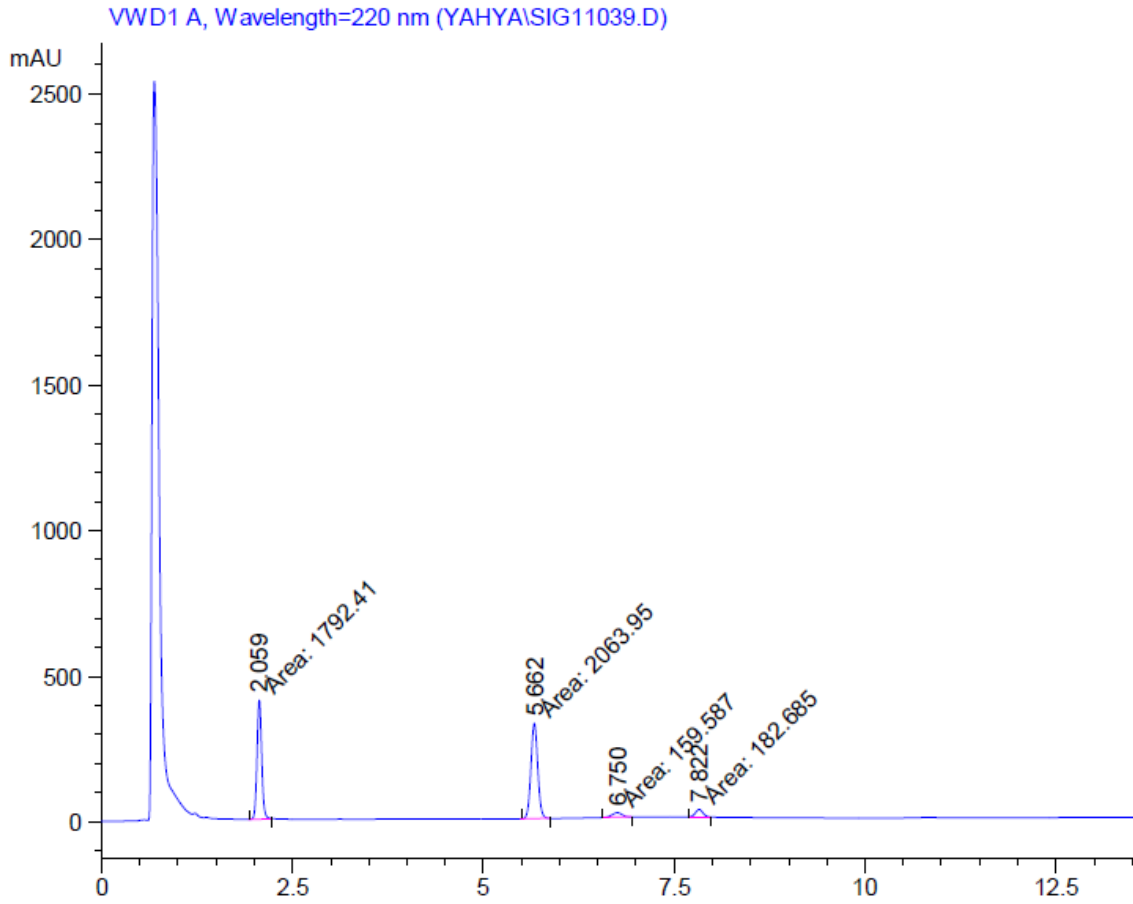
VWD1 A, Wavelength=220 nm (SUHAS\SIG10326.D)



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	5.731	MM	0.1082	3411.99072	525.58350	91.5428
2	6.829	MM	0.1680	200.89302	19.93414	5.3899
3	8.020	MM	0.1209	84.51738	11.64983	2.2676
4	8.505	MM	0.1416	29.80651	3.50751	0.7997

Totals : 3727.20763 560.67498

2.3-OxyrnaPure

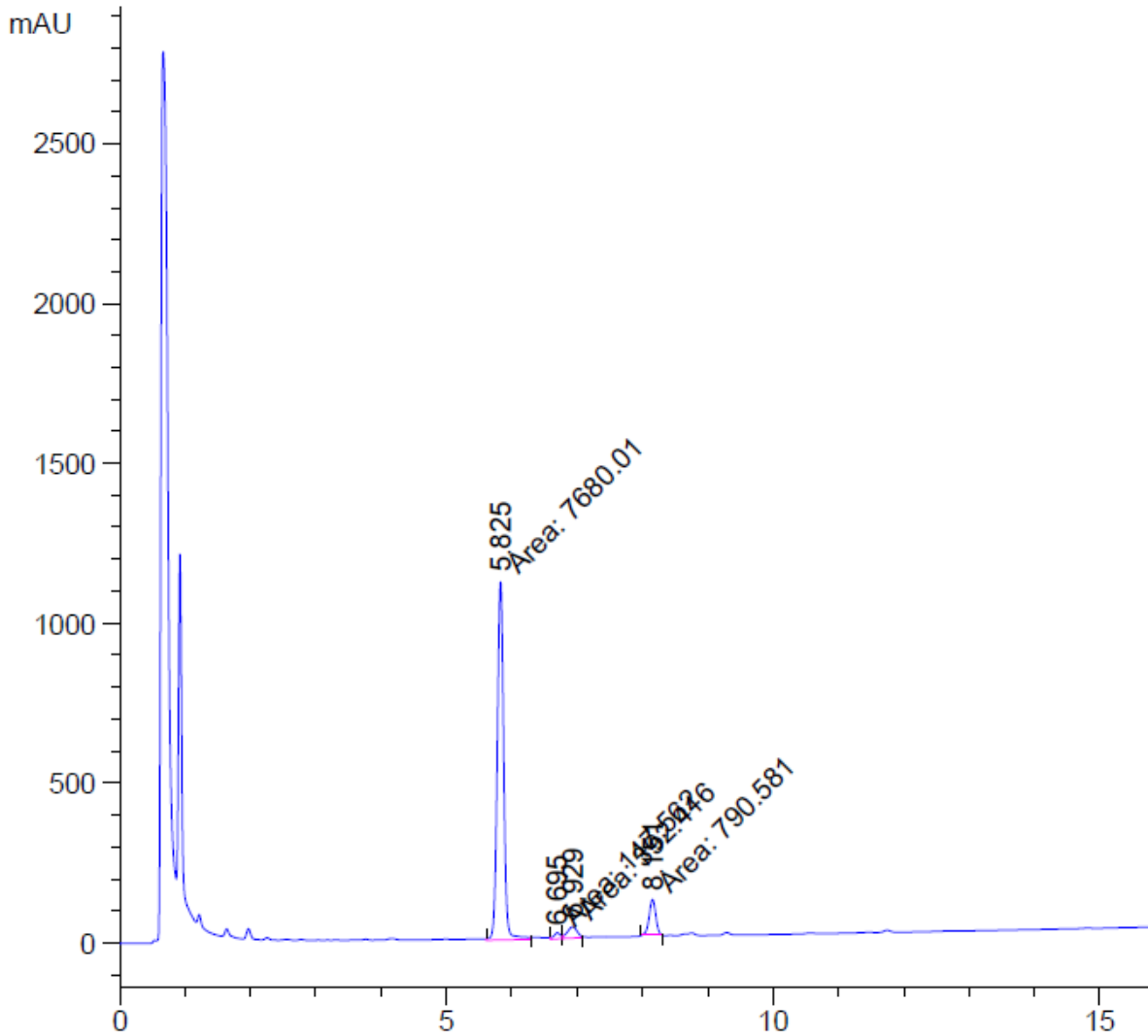


Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	2.059	MM	0.0720	1792.40674	414.90189	42.6902
2	5.662	MM	0.1045	2063.95459	329.12589	49.1578
3	6.750	MM	0.1629	159.58696	16.32829	3.8009
4	7.822	MM	0.1162	182.68468	26.21371	4.3511

Totals : 4198.63297 786.56978

2.4-Oxyrna-B

VWD1 A, Wavelength=220 nm (YAHYA\SIG10109.D)

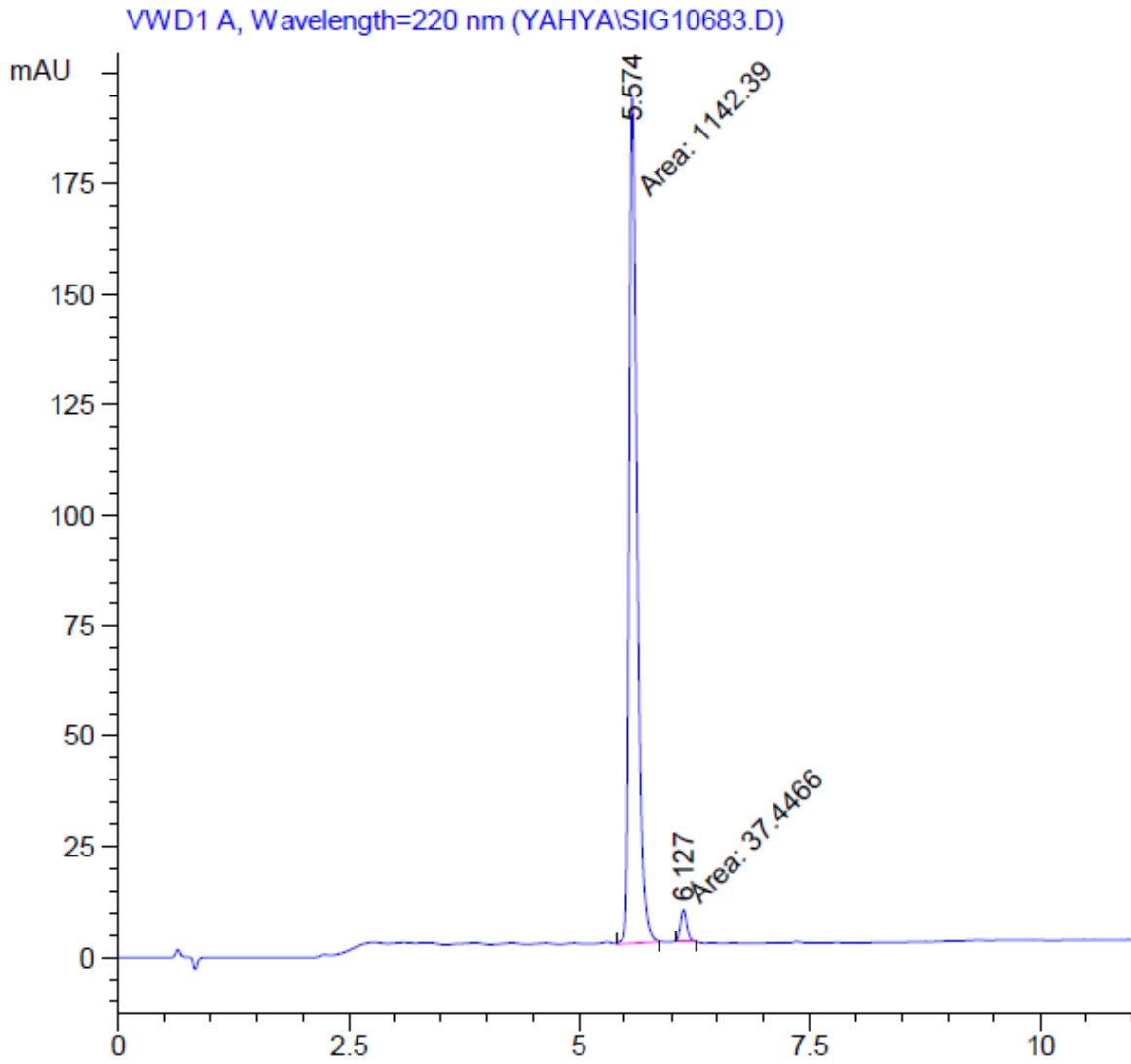


Peak #	RetTime [min]	Type	Width [min]	Area mAU	Area *s	Height [mAU]	Area %
1	5.825	MM	0.1138	7680.01074		1124.86694	85.2334
2	6.695	MF	0.1221	147.56154		20.13989	1.6376
3	6.929	FM	0.1768	392.41583		36.99476	4.3551
4	8.151	MM	0.1176	790.58063		112.03448	8.7739

Totals : 9010.56874 1294.03608

3- H-Gly-Ser-Phe-NH₂ racemization Experiments

3.1-HOBt

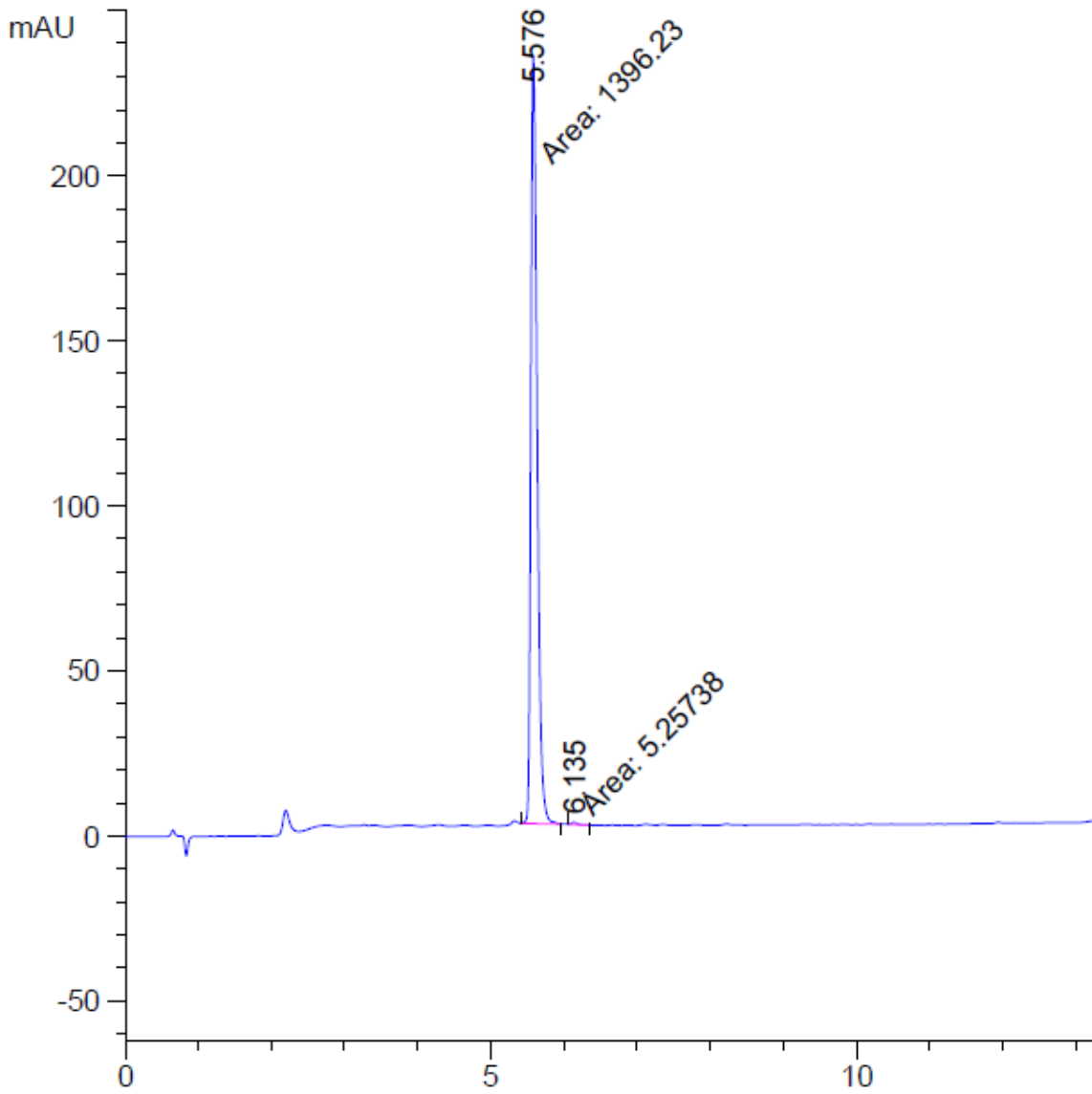


Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	5.574	MM	0.0991	1142.39124	192.13385	96.8261
2	6.127	MM	0.0834	37.44660	7.47892	3.1739

Totals : 1179.83784 199.61277

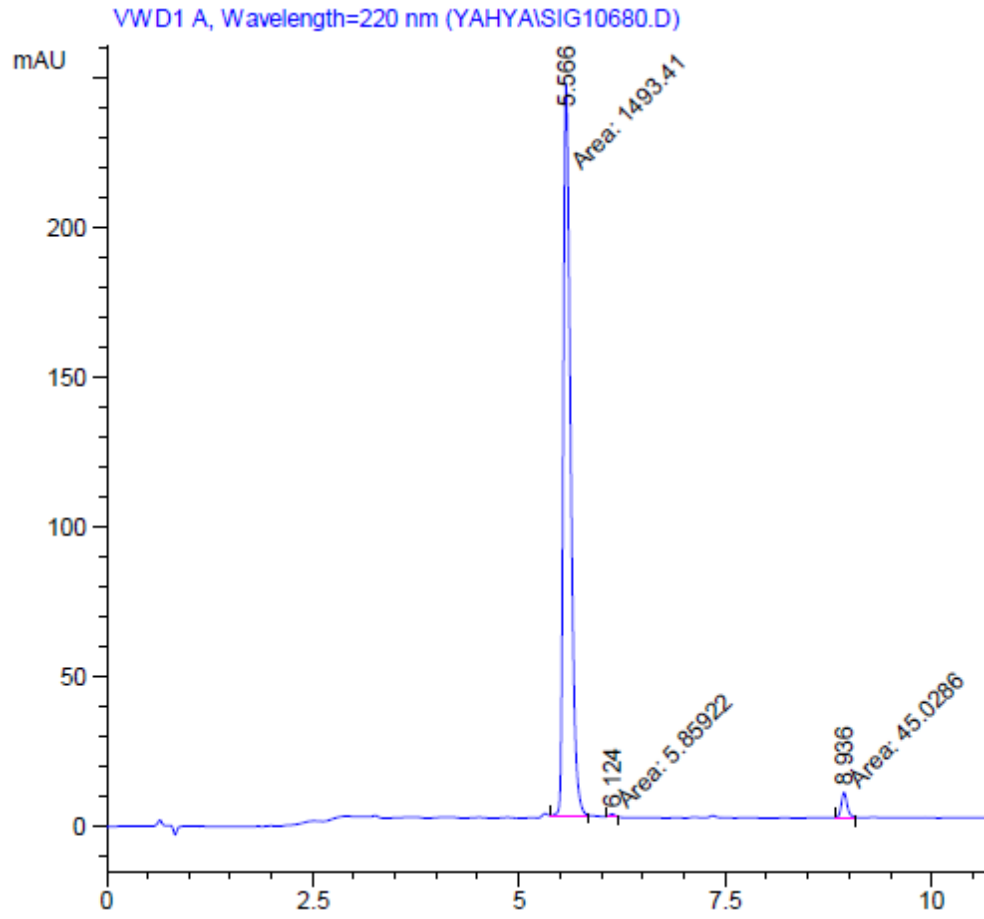
3.2-HOAt

VWD1 A, Wavelength=220 nm (YAHYA\SIG10682.D)



Peak #	RetTime [min]	Type	Width [min]	Area mAU	Area *s	Height [mAU]	Area %
1	5.576	MM	0.0996	1396.22803		233.66008	99.6249
2	6.135	MM	0.1179	5.25738		7.43010e-1	0.3751
Totals :				1401.48541		234.40309	

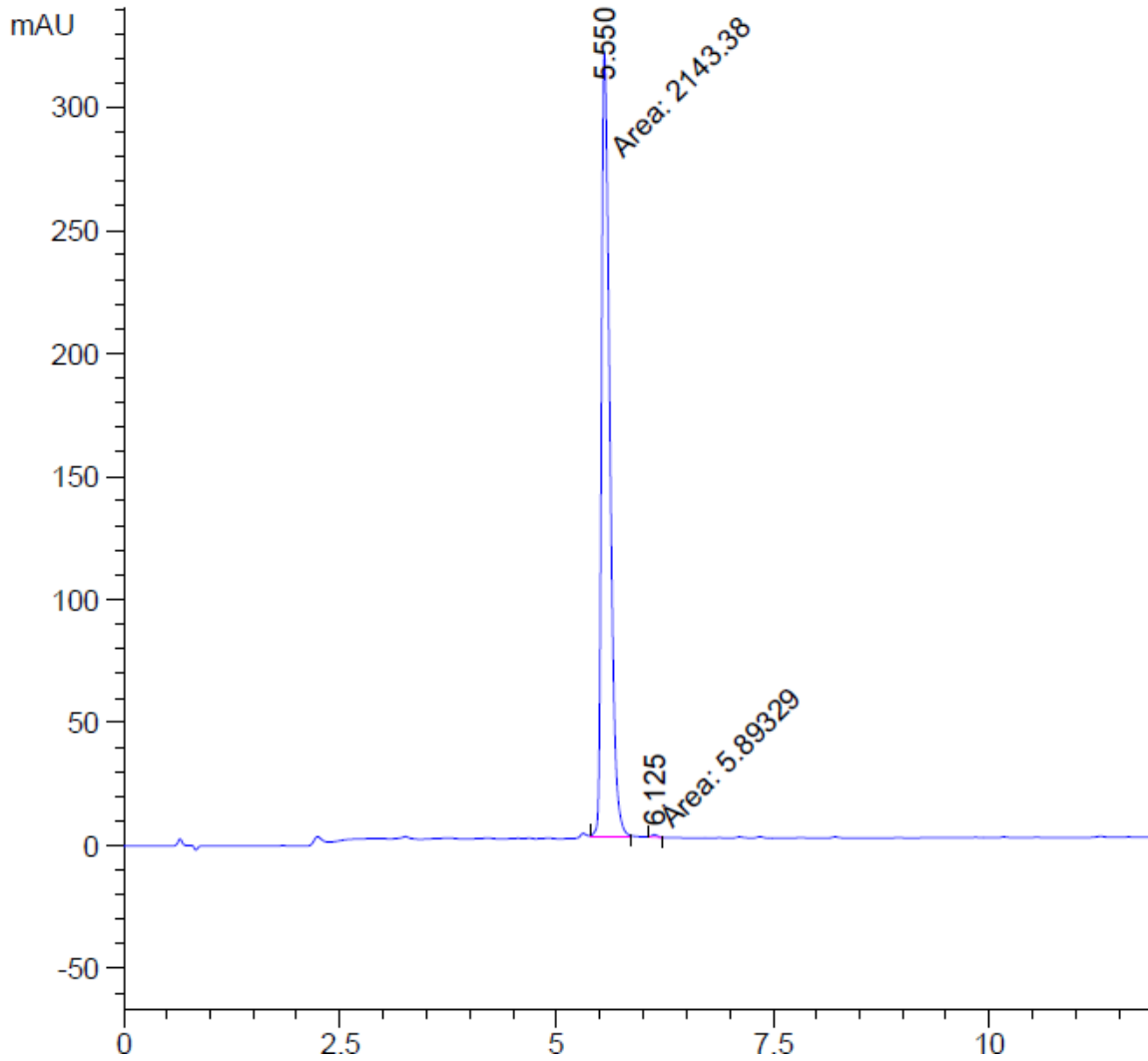
3.3-OxyrnaPure



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	5.566	MM	0.1013	1493.40833	245.60065	96.7048
2	6.124	MM	0.0862	5.85922	1.13334	0.3794
3	8.936	MM	0.0872	45.02864	8.60984	2.9158
Totals :				1544.29619	255.34382	

3.4-Oxyrna-B

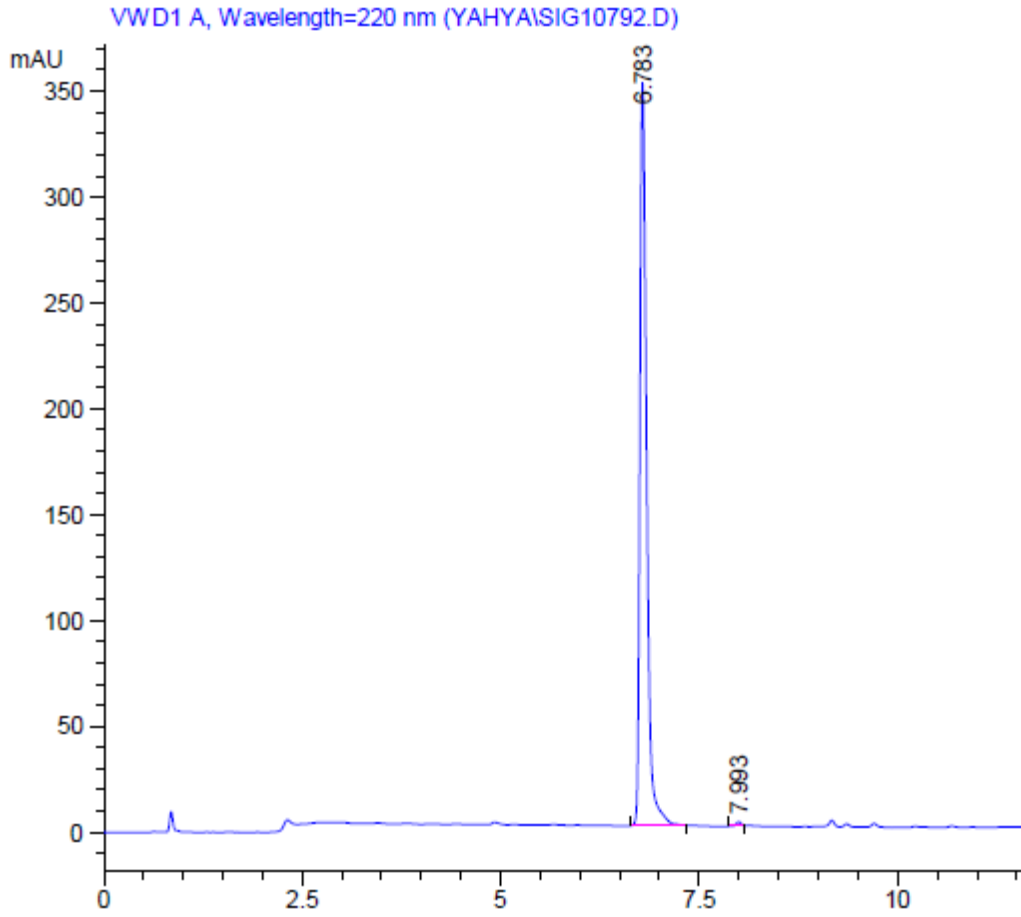
VWD1 A, Wavelength=220 nm (YAHYA\SIG10678.D)



Peak #	RetTime [min]	Type	Width [min]	Area mAU	Height [mAU]	Area %
1	5.550	MM	0.1119	2143.38477	319.10770	99.7258
2	6.125	MM	0.0873	5.89329	1.12541	0.2742
Totals :				2149.27806	320.23311	

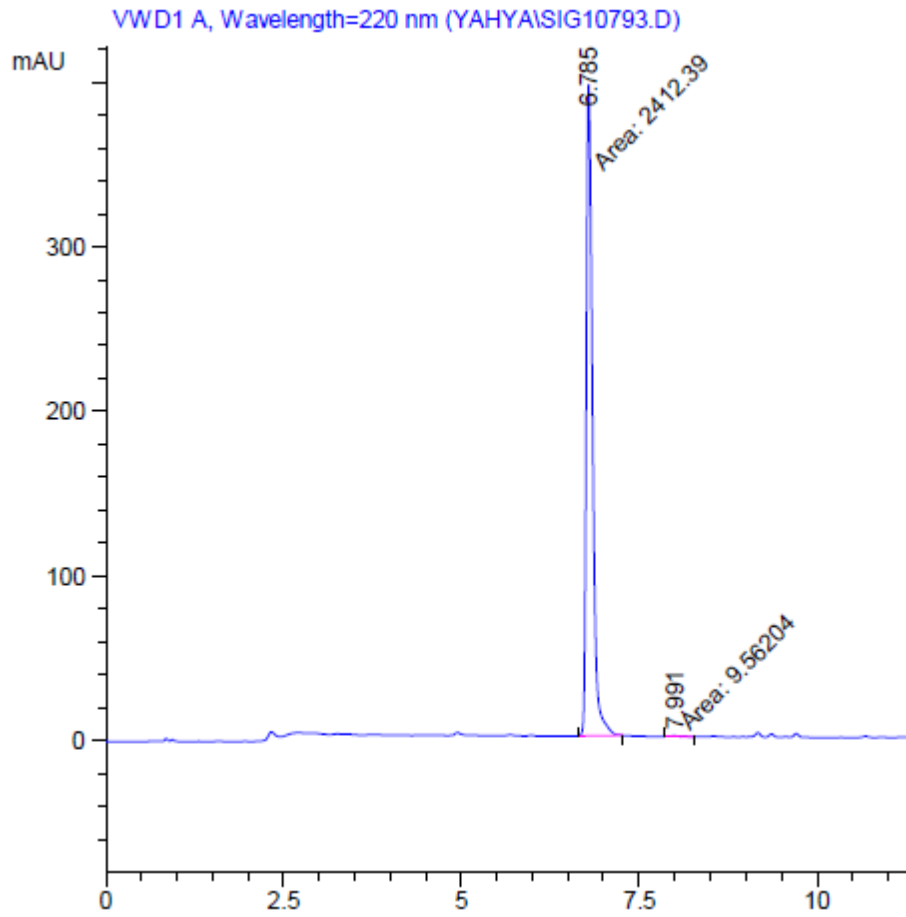
4- H-Gly-Cys-Phe-NH₂ racemization experiments

4.1-HOBt



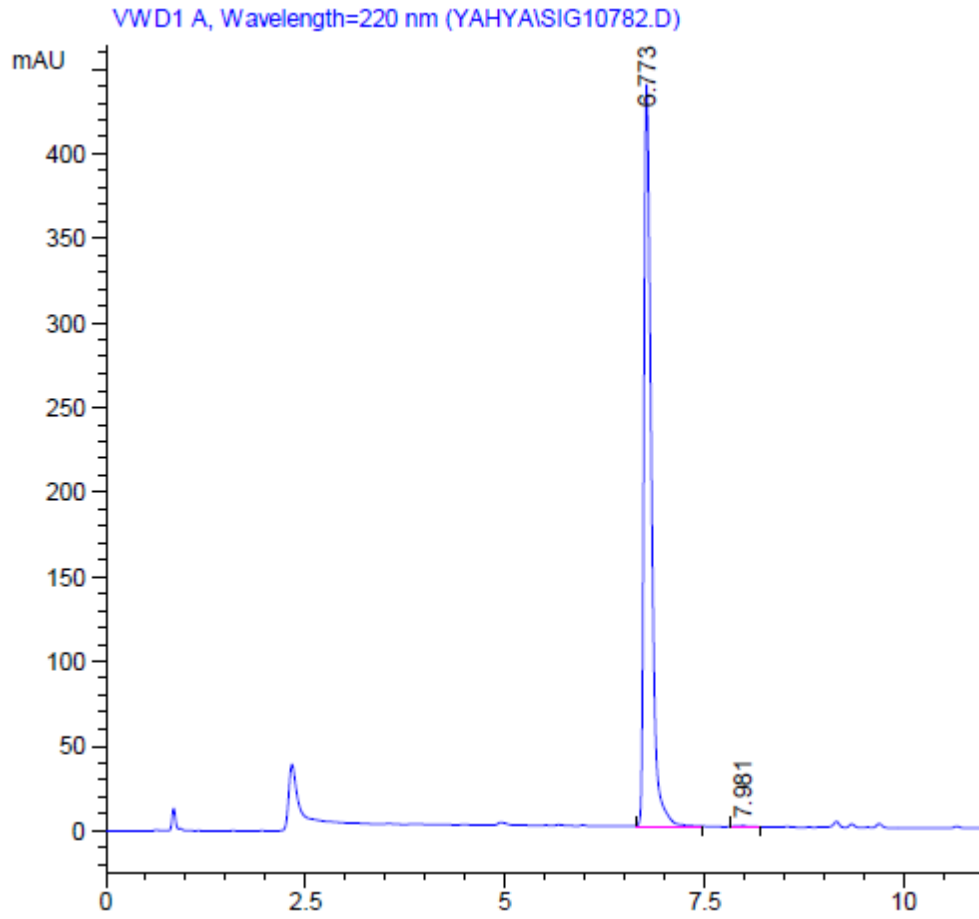
Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.783	BB	0.0929	2083.23022	349.22720	99.5156
2	7.993	PV	0.0744	10.13929	2.07863	0.4844
Totals :				2093.36952	351.30583	

4.2-HOAt



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.785	MM	0.1015	2412.39185	396.08578	99.6052
2	7.991	MM	0.2397	9.56204	6.64877e-1	0.3948
Totals :				2421.95389	396.75066	

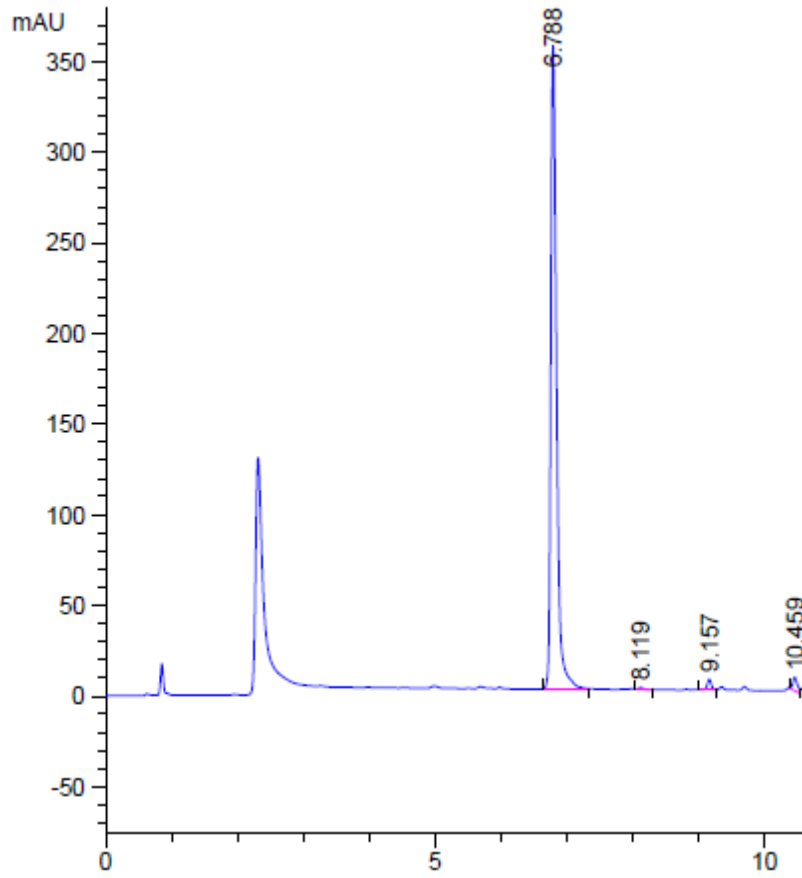
4.3-Oxympure



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.773	BB	0.1015	2812.95752	436.01724	99.6684
2	7.981	BB	0.1479	9.35891	8.37331e-1	0.3316
Totals :				2822.31643	436.85457	

4.4-Oxyrna-B

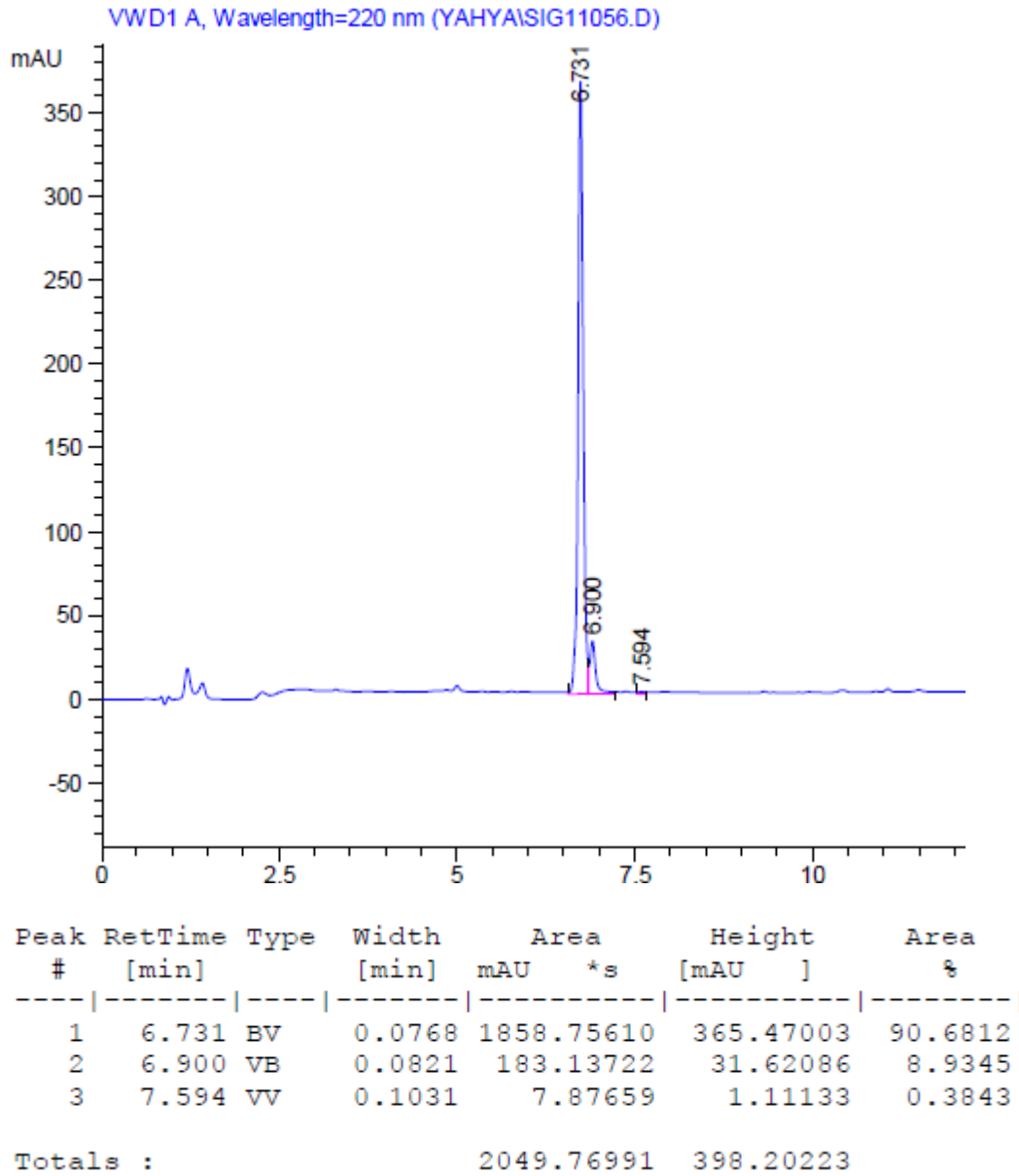
VWD1 A, Wavelength=220 nm (YAHYASIG10781.D)



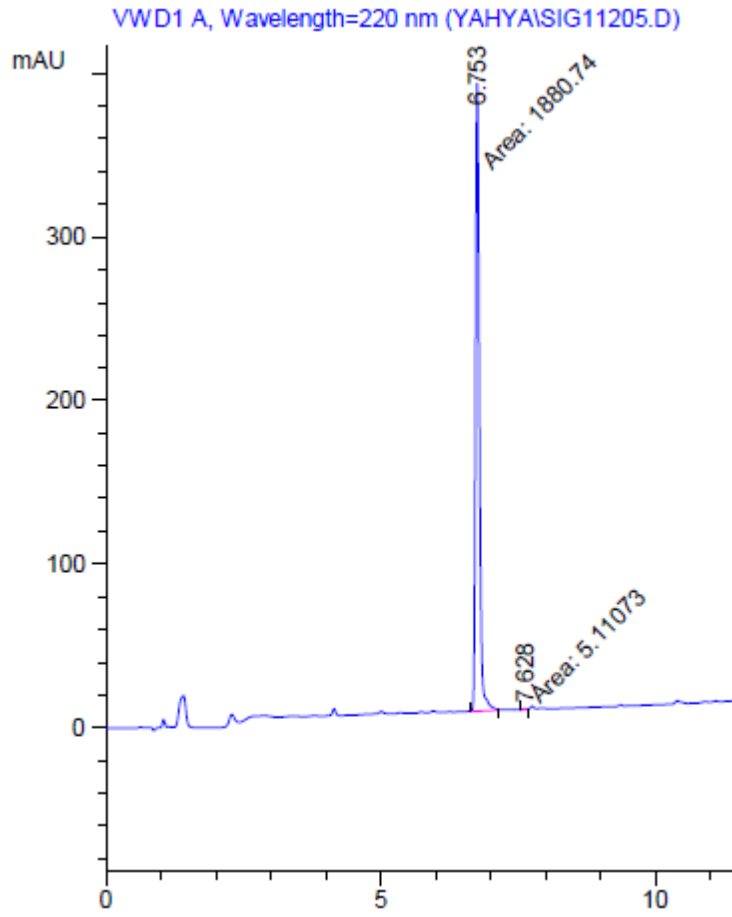
Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.788	BB	0.0939	2147.02563	354.76059	96.7380
2	8.119	VB	0.0845	6.22254	1.08330	0.2804
3	9.157	VV	0.0687	27.67116	5.96098	1.2468
4	10.459	MM	0.0859	38.50278	7.47316	1.7348
Totals :				2219.42212	369.27803	

5- H-Gly-Cys(Acm)-Phe-NH₂ racemization experiments

5.1-HOBt

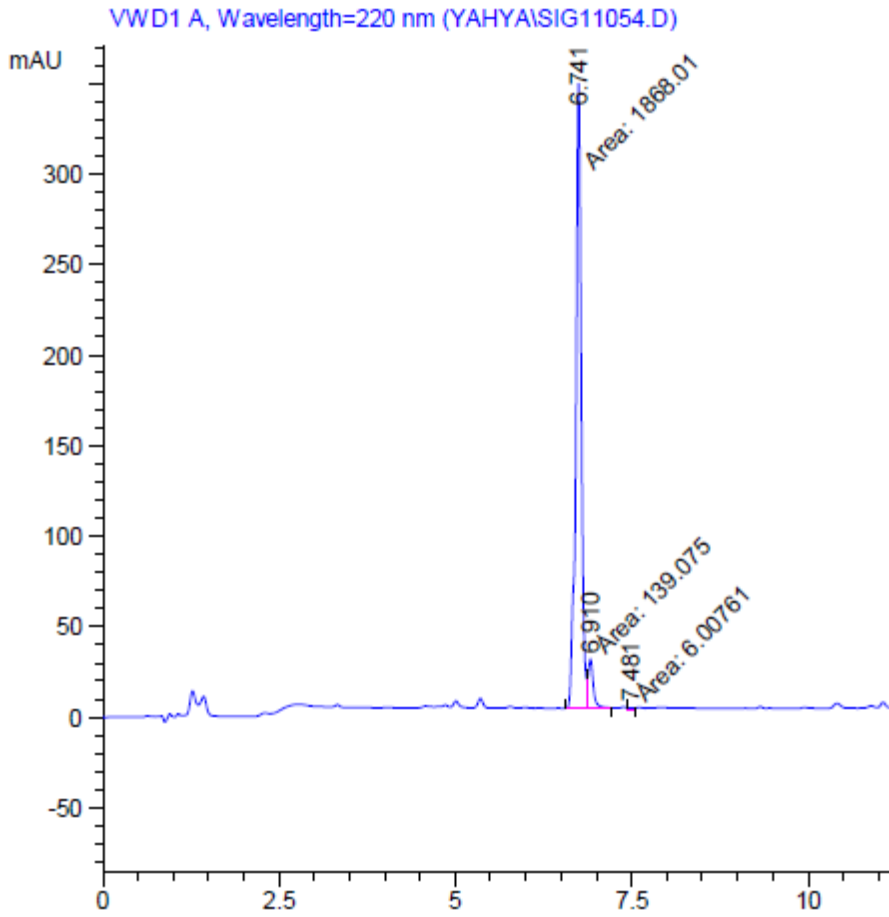


5.2-HOAt



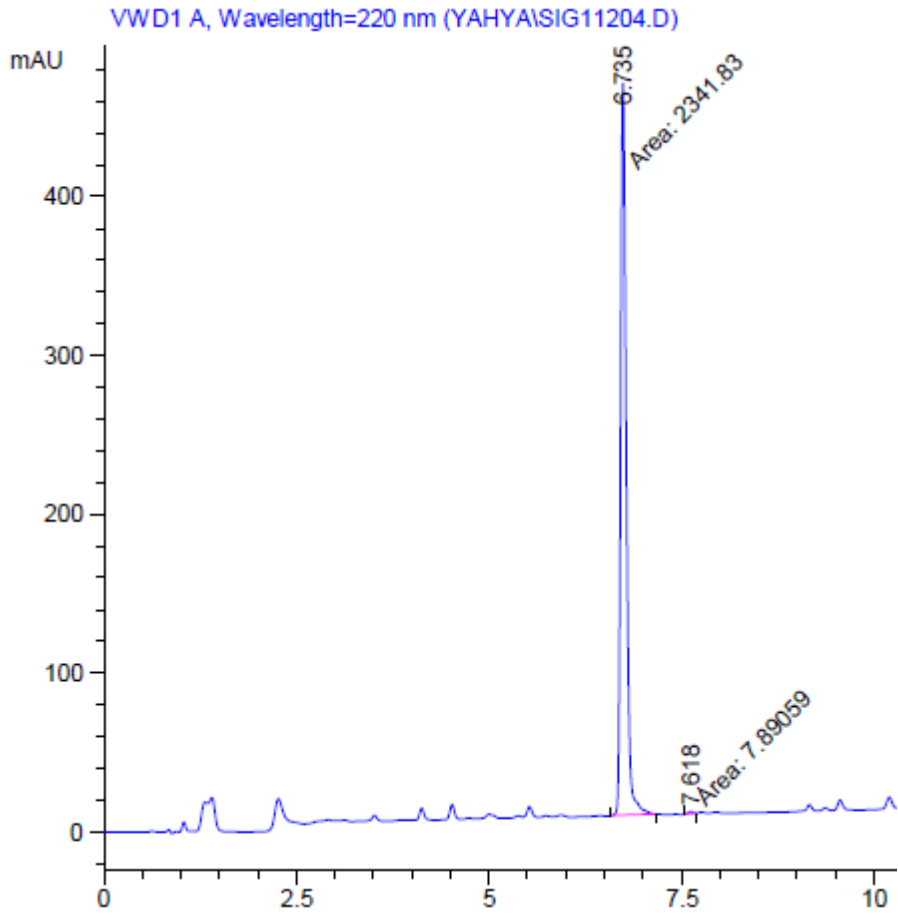
Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.753	MM	0.0812	1880.74109	385.99686	99.7290
2	7.628	MM	0.1056	5.11073	8.06673e-1	0.2710
Totals :				1885.85182	386.80353	

5.3-OxyrnaPure



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.741	MF	0.0900	1868.01404	345.85800	92.7931
2	6.910	FM	0.0853	139.07504	27.16454	6.9085
3	7.481	MM	0.0988	6.00761	1.01381	0.2984
Totals :				2013.09669	374.03636	

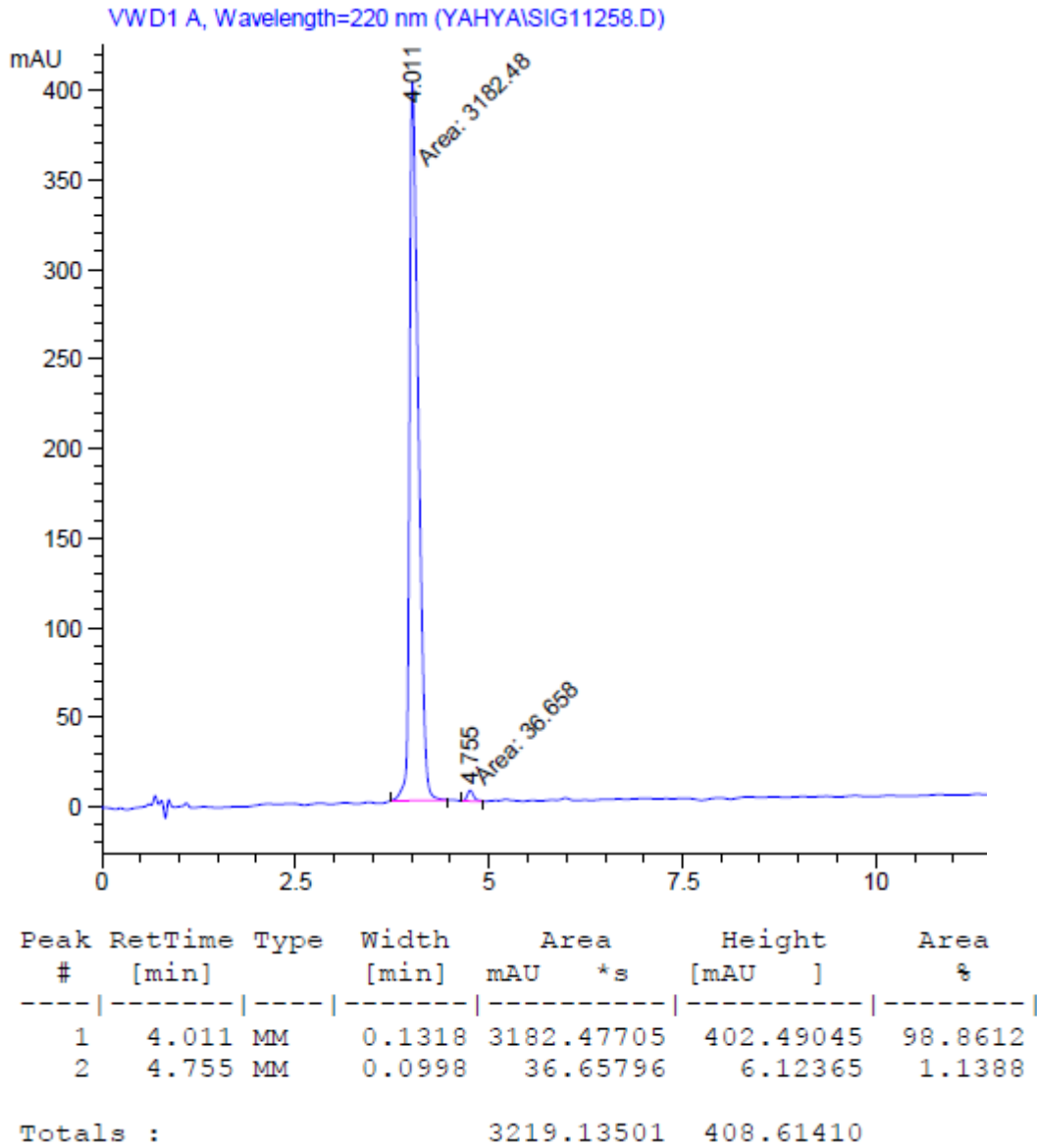
5.4-Oxyrna-B



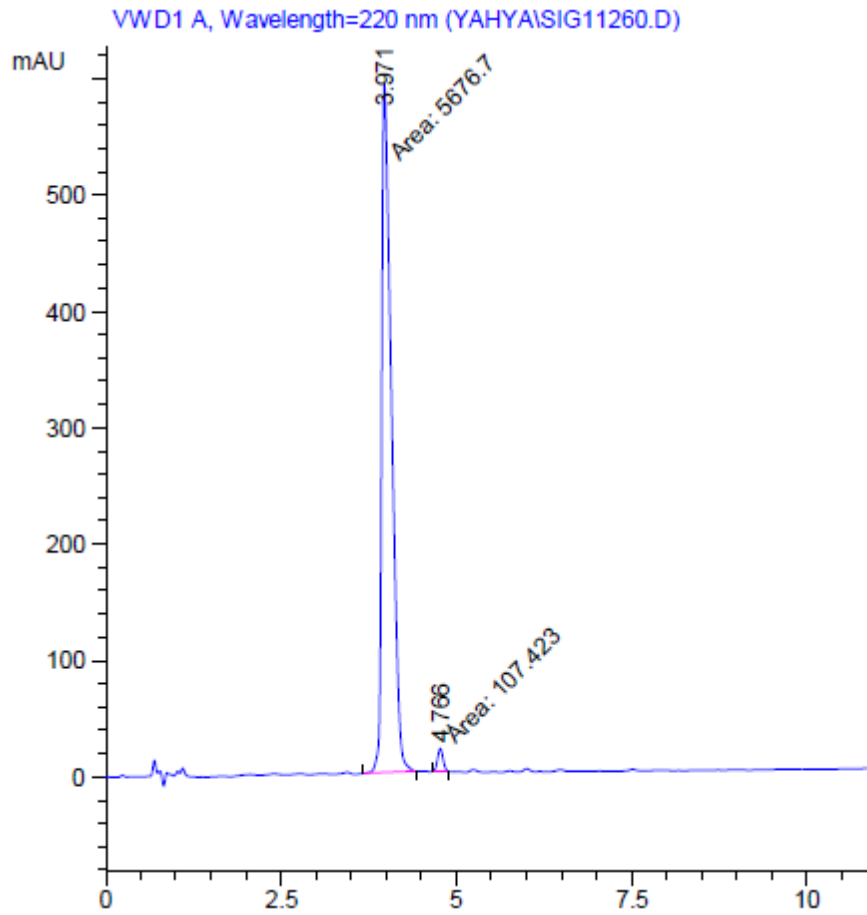
Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	6.735	MM	0.0841	2341.82642	464.22574	99.6642
2	7.618	MM	0.0804	7.89059	1.63655	0.3358
Totals :				2349.71700	465.86229	

6- H-Gly-His-Phe-NH₂ racemization experiments

6.1-HOBt



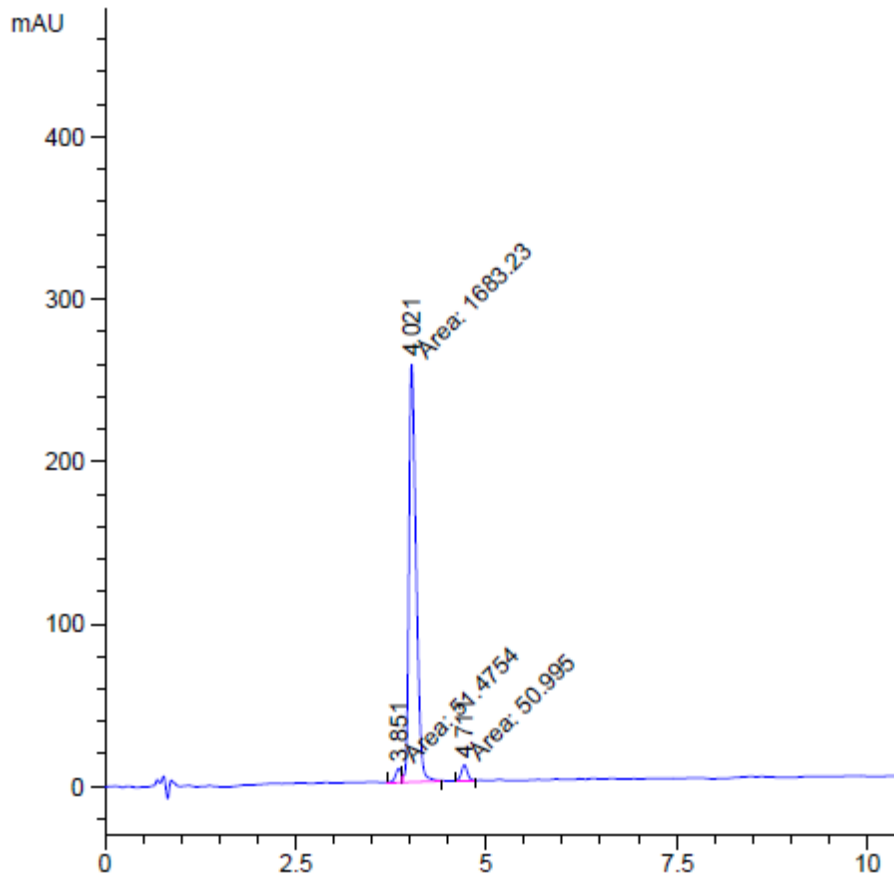
6.2-HOAt



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	3.971	MM	0.1595	5676.70312	593.04578	98.1428
2	4.766	MM	0.0910	107.42307	19.66497	1.8572
Totals :				5784.12619	612.71075	

6.3-OxyrnaPure

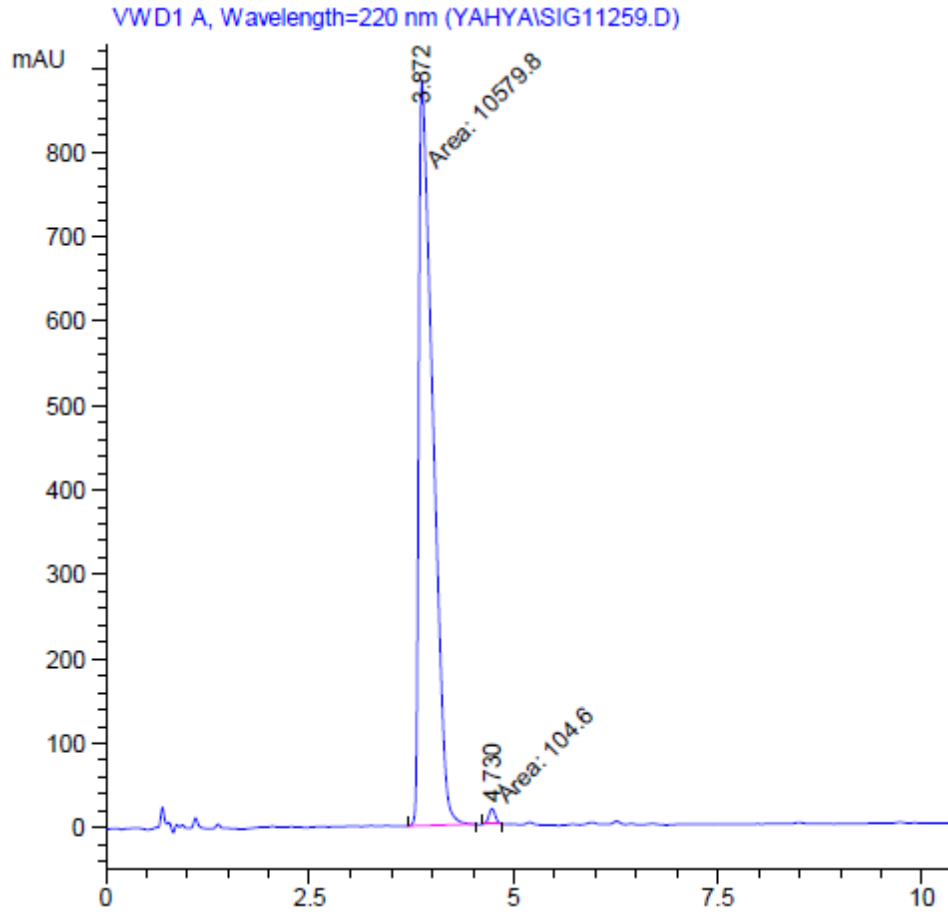
VWD1 A, Wavelength=220 nm (YAHYA\SIG11256.D)



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	3.851	MF	0.0959	51.47545	8.94173	2.8827
2	4.021	FM	0.1087	1683.22681	258.05417	94.2616
3	4.711	MM	0.0897	50.99501	9.47265	2.8557

Totals : 1785.69727 276.46854

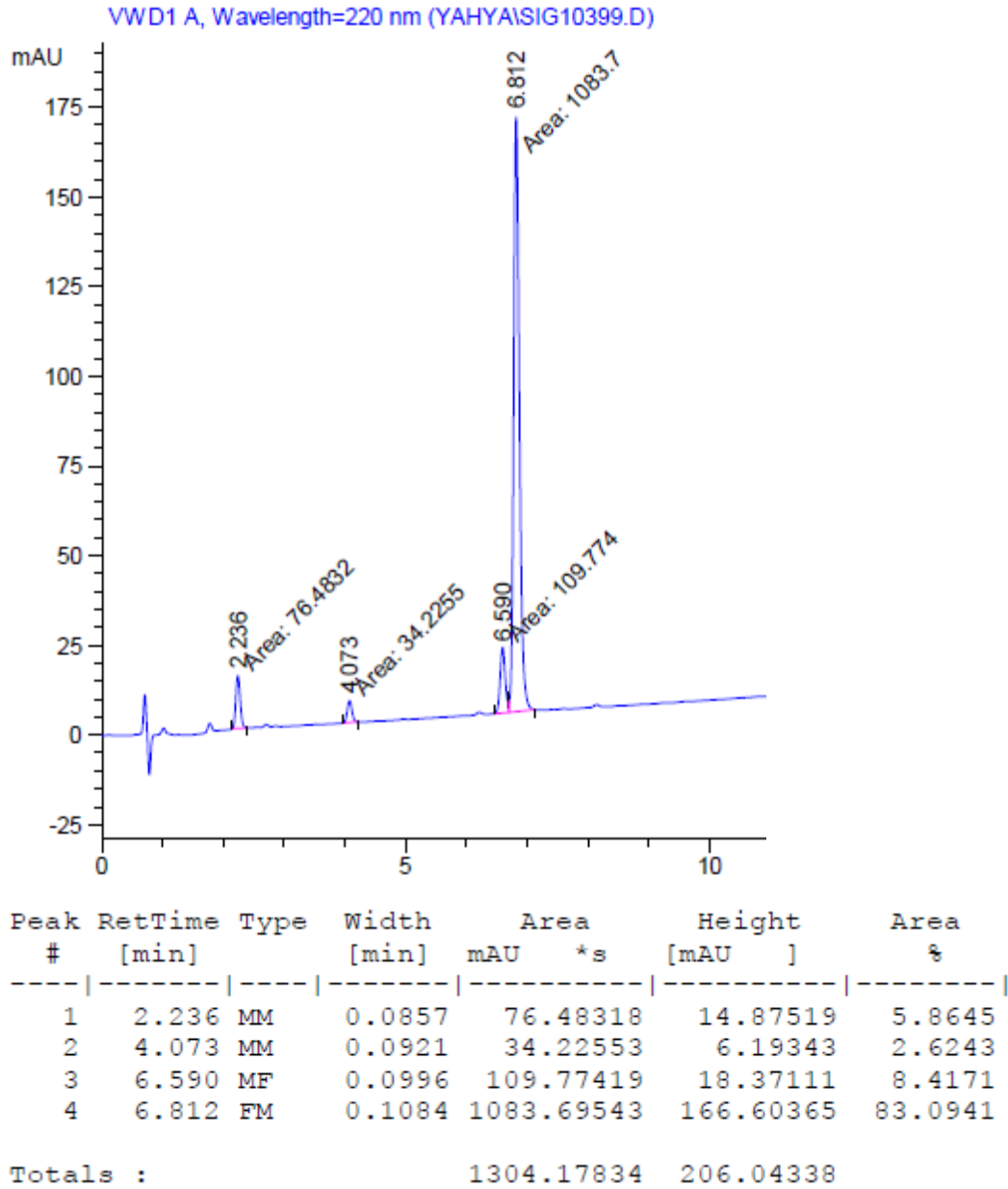
6.4-Oxyrna-B



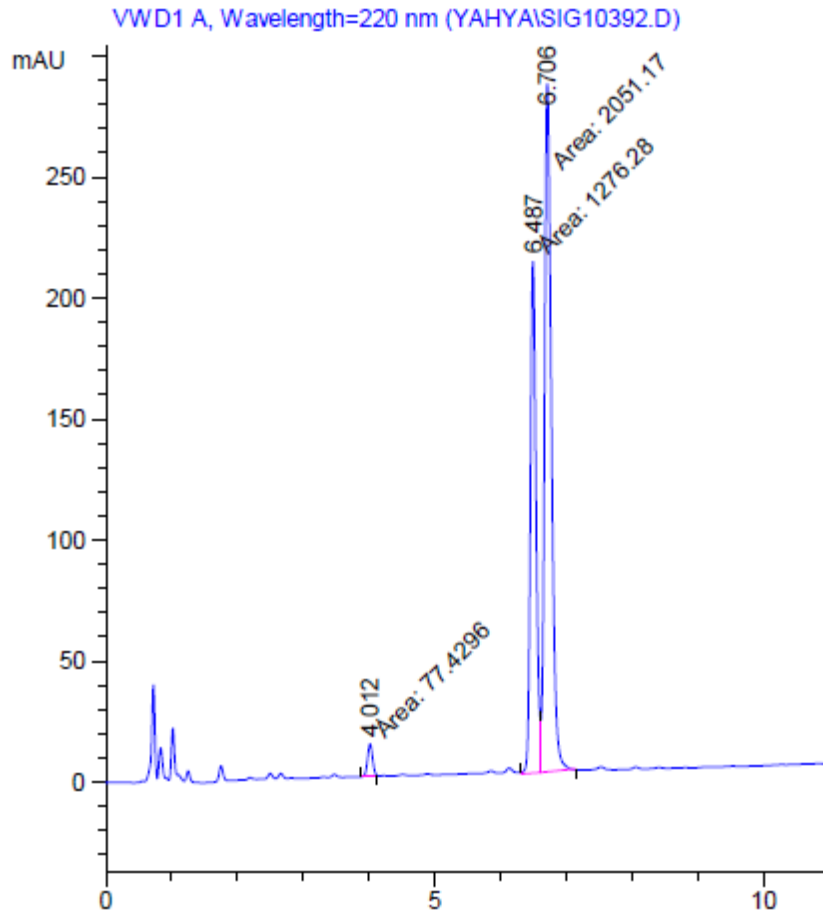
Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	3.872	MM	0.1996	1.05798e4	883.49829	99.0210
2	4.730	MM	0.0950	104.59971	18.34989	0.9790
Totals :				1.06844e4	901.84818	

7- Peptide synthesis experiments (Synthesis of H-Tyr-Aib-Aib-Phe-Leu-NH₂)

7.1- HOBt



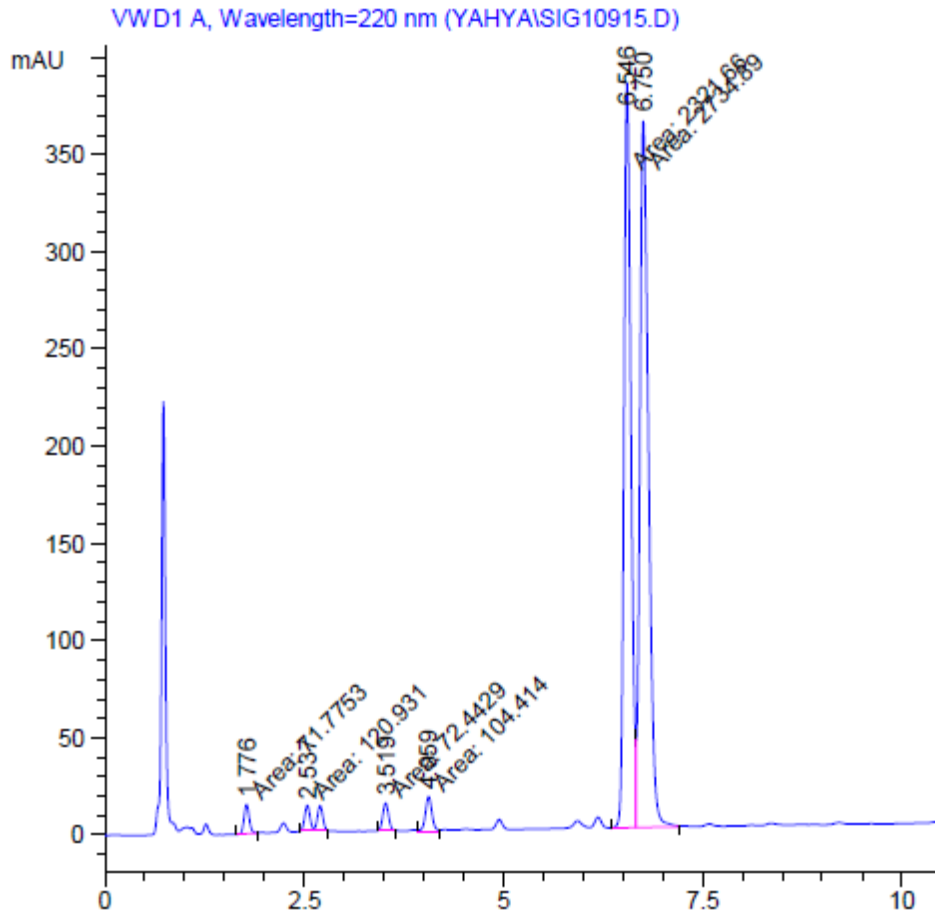
7.2- HOAt



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	4.012	MM	0.0934	77.42957	13.81854	2.2741
2	6.487	MF	0.1002	1276.28418	212.36047	37.4840
3	6.706	FM	0.1197	2051.16602	285.52606	60.2420

Totals : 3404.87976 511.70508

7.3- OxymaPure

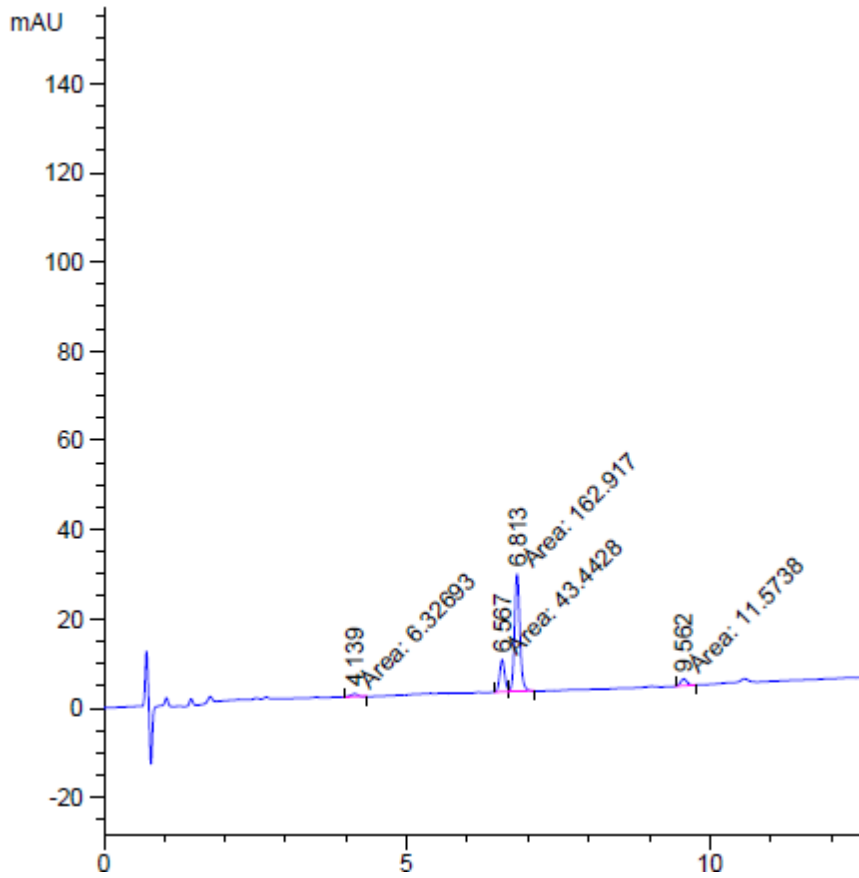


Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	1.776	MM	0.0783	71.77528	15.27117	1.3228
2	2.537	MM	0.1547	120.93087	13.02975	2.2287
3	3.519	MM	0.0846	72.44289	14.27463	1.3351
4	4.059	MM	0.0967	104.41430	18.00078	1.9243
5	6.546	MF	0.1006	2321.66406	384.56546	42.7868
6	6.750	FM	0.1250	2734.89282	364.77695	50.4024

Totals : 5426.12022 809.91874

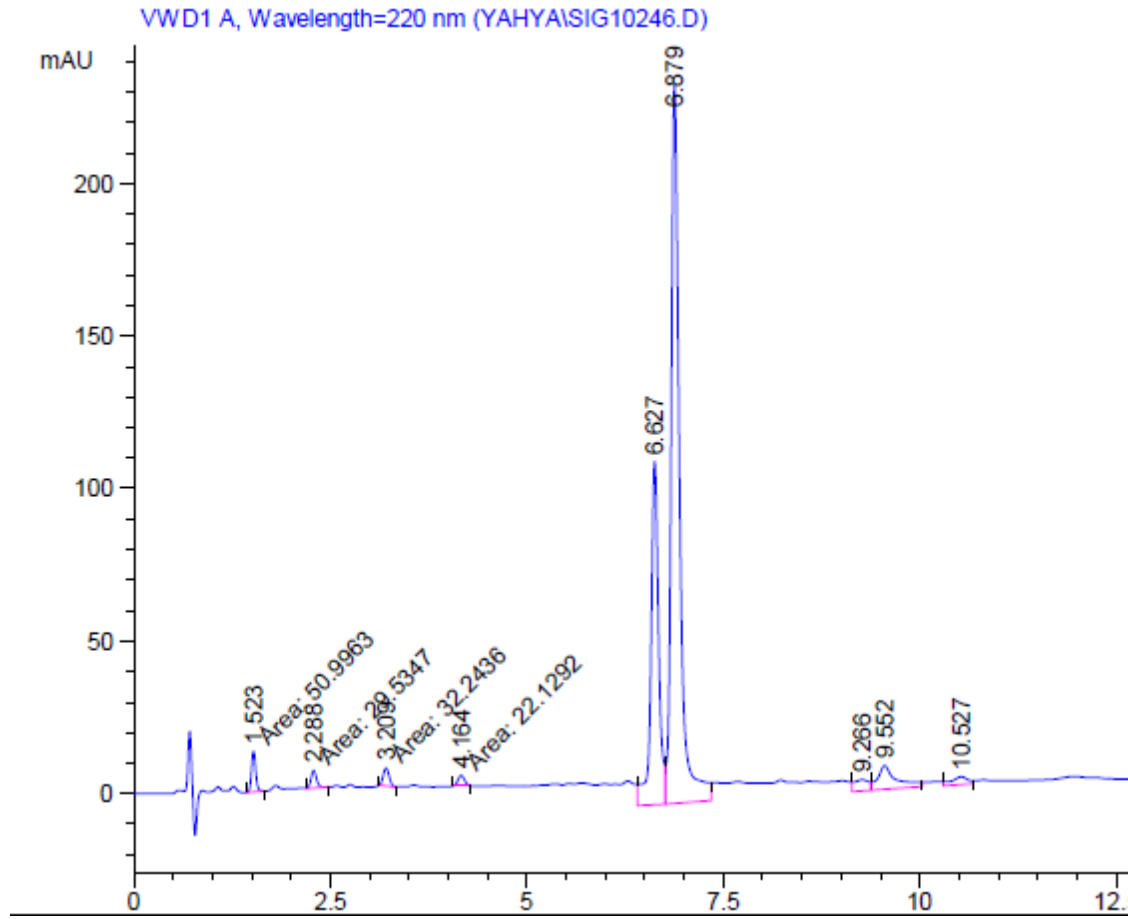
7.4-Oxyma-B (without preactivation)

VWD1 A, Wavelength=220 nm (YAHYASIG10300.D)



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	4.139	MM	0.1475	6.32693	7.14963e-1	2.8212
2	6.567	MF	0.0989	43.44279	7.32467	19.3715
3	6.813	FM	0.1024	162.91740	26.51380	72.6464
4	9.562	MM	0.1241	11.57382	1.55468	5.1609
Totals :				224.26094	36.10811	

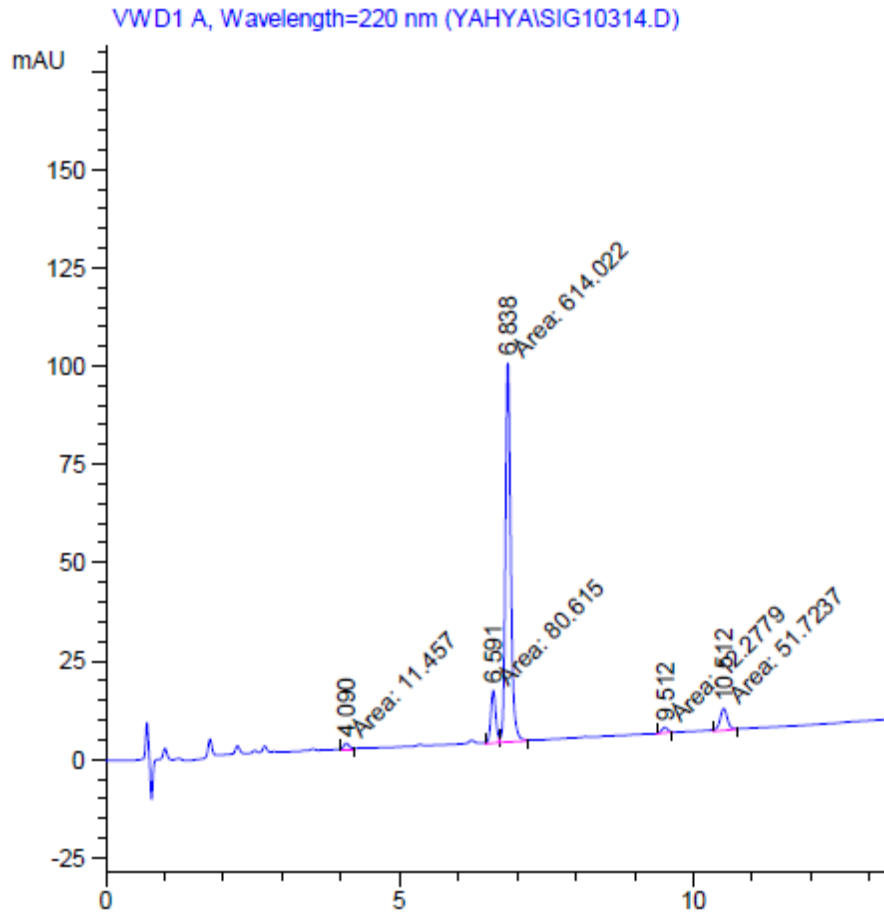
7.5-Oxyrna-B (3 min preactivation)



Peak #	RetTime [min]	Type	Width [min]	Area mAU	Area *s	Height [mAU]	Area %
1	1.523	MM	0.0625	50.99629		13.59133	1.7591
2	2.288	MM	0.0840	29.53468		5.85700	1.0188
3	3.209	MM	0.0904	32.24356		5.94303	1.1122
4	4.164	MM	0.1018	22.12924		3.62274	0.7633
5	6.627	VV	0.1027	766.14014		112.55873	26.4279
6	6.879	VB	0.1109	1771.56897		235.82402	61.1101
7	9.266	VV	0.1787	49.47144		3.78464	1.7065
8	9.552	VB	0.2309	137.40427		7.94631	4.7397
9	10.527	BV	0.2151	39.49155		2.59886	1.3623

Totals : 2898.98014 391.72666

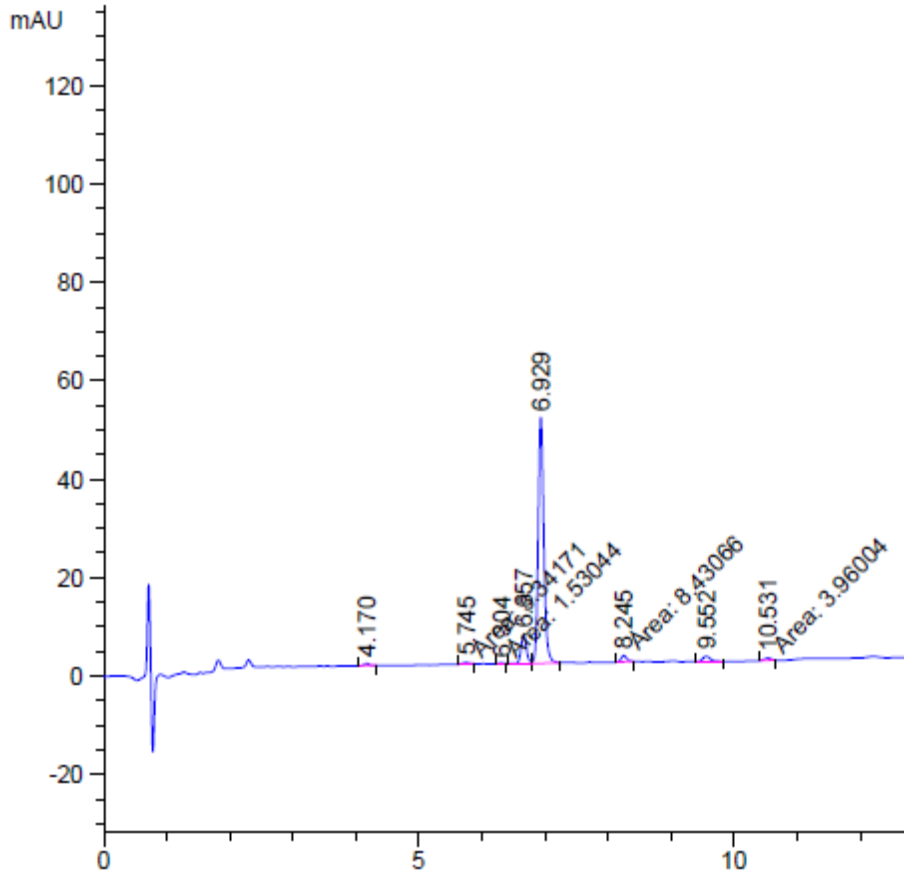
7.6-Oxyma-B (3 min preactivation with 0.1 eq. DIEA)



Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	4.090	MM	0.1207	11.45700	1.58226	1.4877
2	6.591	MF	0.0996	80.61497	13.49381	10.4682
3	6.838	FM	0.1058	614.02222	96.72003	79.7332
4	9.512	MM	0.1317	12.27792	1.55411	1.5943
5	10.512	MM	0.1524	51.72368	5.65723	6.7165
Totals :				770.09578	119.00743	

7.7-Oxyrna-B (7 min preactivation)

VWD1 A, Wavelength=220 nm (YAHYASIG10257.D)



Peak #	RetTime [min]	Type	Width [min]	Area mAU	Area *s	Height [mAU]	Area %
1	4.170	BP	0.0901	2.66901		4.66390e-1	0.7117
2	5.745	MM	0.1446	3.34171		3.85115e-1	0.8911
3	6.304	MM	0.1026	1.53044		2.48714e-1	0.4081
4	6.657	VV	0.0935	35.35608		5.87515	9.4282
5	6.929	VB	0.0958	310.49469		49.93639	82.7980
6	8.245	MM	0.1076	8.43066		1.30571	2.2482
7	9.552	PB	0.1312	9.21984		1.08481	2.4586
8	10.531	MM	0.1311	3.96004		5.03600e-1	1.0560

Totals : 375.00246 59.80588