Supporting Information

In-Silico approach towards lipase mediated chemoenzymatic synthesis of (*S*)-Ranolazine, an anti-anginal drug

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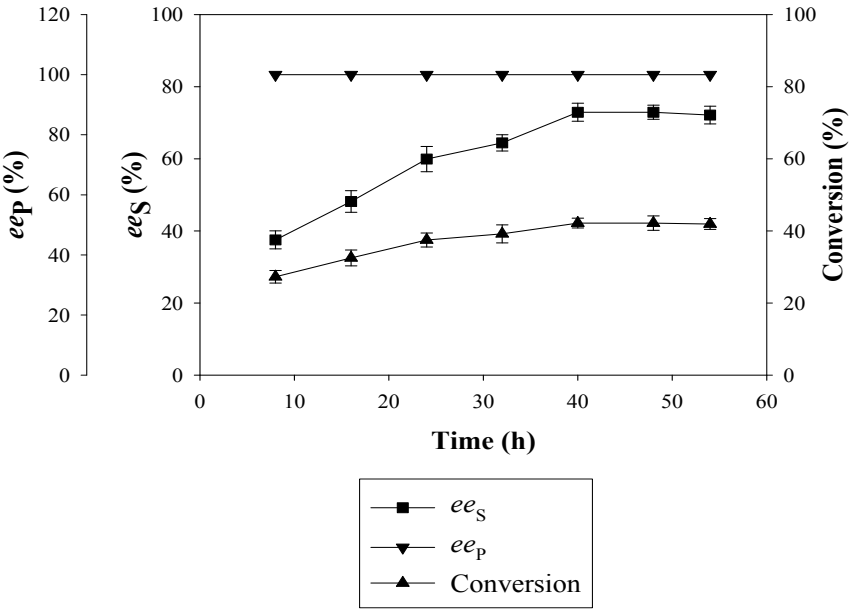
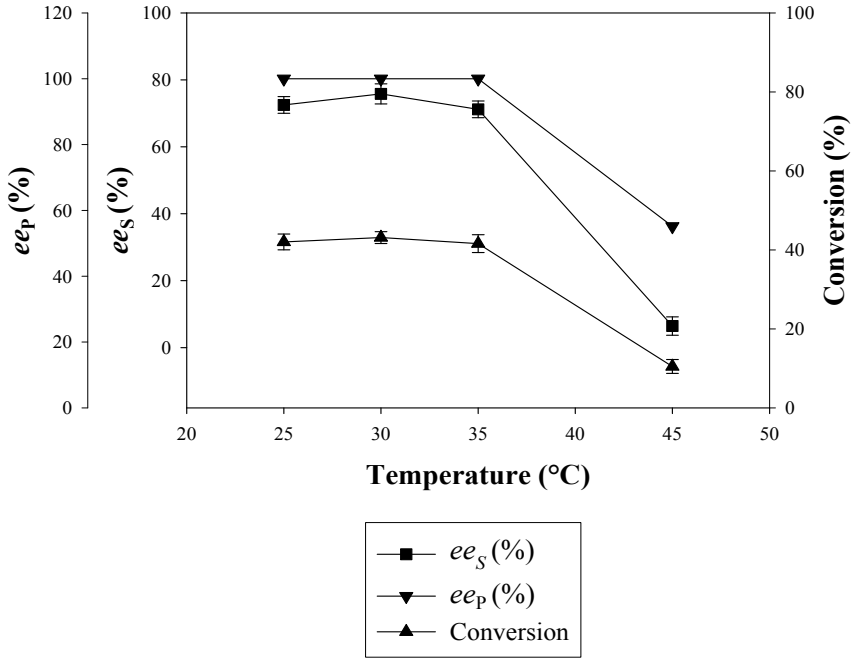
S1. Optimization of reaction parameters for the lipase mediated kinetic resolution of (*RS*)-5

Optimization of reaction parameters were carried out to obtain the ideal condition for the kinetic resolution of (*RS*)-**5**. For the optimization of reaction time, samples were collected at different time interval and the enzyme was removed by centrifugation at 7000 x g for five minutes. The supernatant collected was dried under vacuum and redissolved in 2-propanol for chiral HPLC analysis. The course of reaction was studied to determine the optimum reaction time. It was evident from Figure S1 (a) that both conversion and enantiomeric excess has increased with time. The maximum conversion of 42% and enantiomeric excess of substrate (ee_S) of 72% was achieved after 48 h (Figure S1 (a), see supporting information) and thereafter up to 54 h, no significant change in ee_S and conversion was observed. Hence, 48 h was selected for further optimization studies. To study the effect of temperature on enantioselectivity and conversion, reactions were carried out at different temperatures (25, 30, 35, 45 ˚C) for 48 h in toluene with vinyl acetate as acyl donor. After completion of the transesterification reaction, enantiomeric excess and conversion were determined by Chiral HPLC. It was evident from Figure S1 (b) that maximum ee_S (75.81%) and conversion (43.50%) was achieved at 30 °C, and further at 45 °C it was decreased to 6.45% and 10.48% respectively. Hence, 30 °C was selected as the optimum temperature for further reactions. To analyze the effect of enzyme concentration on CLEA catalyzed kinetic resolution of (*RS*)-**5**, various enzyme concentration of CLEA (5, 10, 15, 20, 25 and 30 mg/mL) in toluene at fixed substrate concentration (12.71 mg, 30 mM) were studied. It was evident from Figure S1 (c) that the enantiomeric excess and conversion showed an increasing trend with the increasing enzyme concentration and the maximum conversion was obtained with 30 mg/mL enzyme. Hence, further experiments were carried out with 30 mg/mL CLEA for the kinetic resolution of (*RS*)-**5**. To study the effect of substrate concentration on the transesterification, reactions were carried out using different concentrations of (*RS*)-**5** (5, 10, 20, 30, 40 mM) separately in toluene at 30 °C for 48 h with vinyl acetate as the acyl donor. It was evident from Figure S1 (d) that the best conversion and maximum *ee* was achieved with 5 mM substrate **5**. And with substrate concentration above 50 mM, there was non-selective acylation.

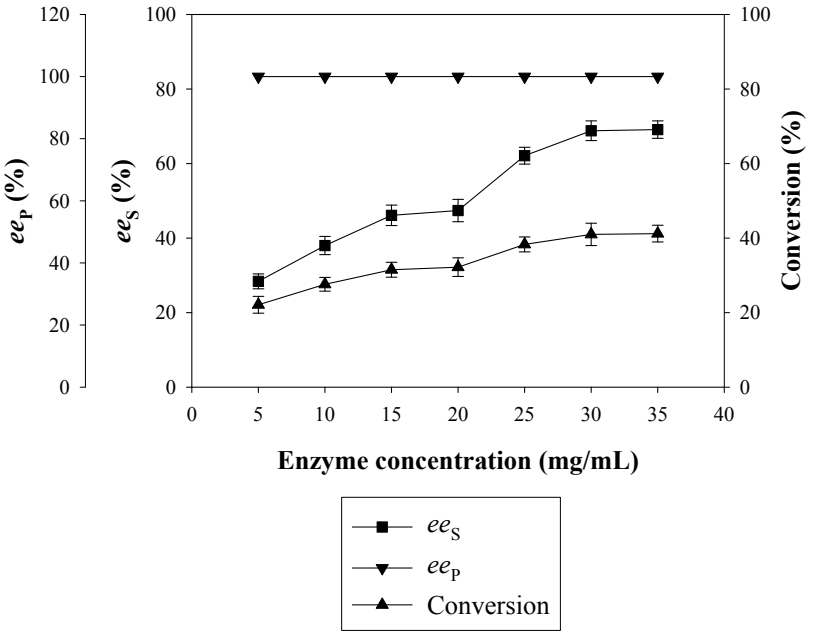


(a)

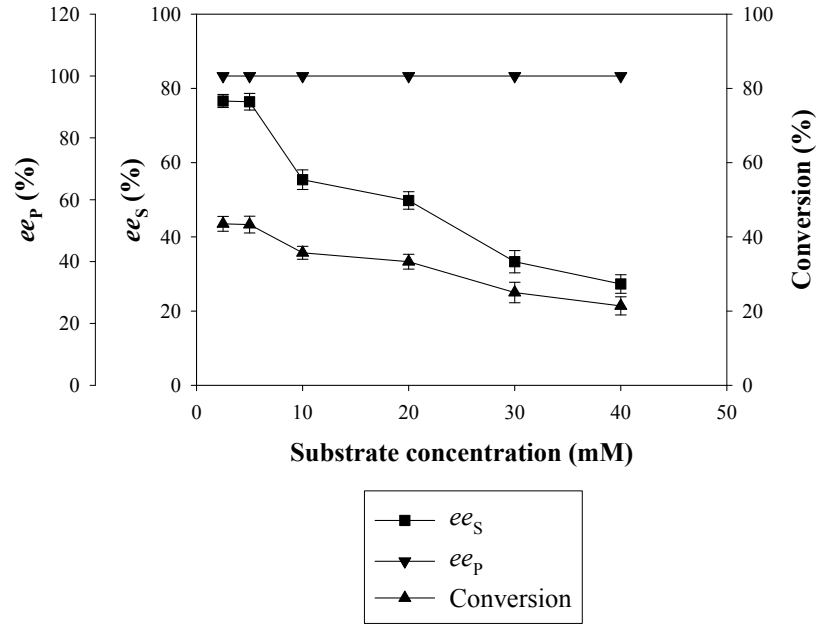


(b)

Figure. S1 (a) Course of reaction for the transesterification of (*RS*)-5, (b) Effect of temperature on transesterification of (*RS*)-5



(c)



(d)

Figure. S1 (c) Effect of enzyme concentration on transesterification of (*RS*)-5, (d) Effect of substrate concentration on transesterification of (*RS*)-5.

S2. Screening of organic solvent for the lipase mediated kinetic resolution of (*RS*)-5

(*RS*)-ranolazine is very slightly soluble in water having log P value 1.6. To study the effect of organic solvent on enantioselectivity and conversion, different polar and nonpolar solvents were selected and investigated for biocatalytic activity. Further the conversion was analyzed by using chiral HPLC. From the HPLC data, it is evident that the highest ee_S (89.7%) and conversion (47.8%) was obtained with diethyl ether (Table S1). Hence diethyl ether was selected as reaction medium for further optimization studies.

Table S1. Effect of solvent on CAL-A CLEA mediated kinetic resolution of (*RS*)-5[a]

Solvent	logP	ee_S (%)[b]	ee_P (%)[c]	Conversion (%)[d]	E[e]
Acetonitrile	-0.33	37 ± 2.3	93 ± 1.5	28 ± 2.4	37.0
Diethyl ether	0.85	90 ± 1.8	98 ± 1.3	48 ± 1.8	323
Dichloromethane	1.25	72 ± 0.8	99 ± 0.4	42 ± 2.3	205
t-Butyl methyl ether	1.35	74 ± 1.2	99 ± 0.3	43 ± 1.5	225
Benzene	2.0	28 ± 2.3	62 ± 2.3	31 ± 1.9	6.0
Toluene	2.5	32 ± 2.8	28 ± 1.9	54 ± 2.2	2.0

[a](*RS*)-5 (2.135 mg, 5 mM, 1 eq.) in respective solvent (800 µL) was treated with lipase (30 mg), vinyl acetate (200 µL) at 30 ˚C

[b]Enantiomeric excess of substrate was calculated by using formula = (*R*-*S*)/(*R*+*S*)*100

[c]Enantiomeric excess of product was calculated by using formula = (*S*-*R*)/(*S*+*R*)*100

[d]Conversions were calculated from enantiomeric excess of substrate (ee_S) and product (ee_P) using the formula: Conversion (C) = $ee_S/(ee_S + ee_P)$

[e]*E* values were calculated using the formula: $E = [\ln (1 - C (1 + ee_P)]/ [\ln (1 - C (1 - ee_P)]$.

S3. Screening of acyl donor for the lipase mediated kinetic resolution of (*RS*)-5

As acyl donor plays vital role in selectivity of CLEA catalyzed transesterification of (*RS*)-**5**, various acyl donors were investigated. For that, reactions were carried out with different acyl donors, using CLEA (30 mg/mL) in diethyl ether at 30 °C for 48 h. It was found that vinyl acetate is superior than the other acyl donors studied (Table S2).

Table S2. Effect of acyl donor on CAL-A CLEA mediated kinetic resolution of (*RS*)-5[a]

Acyl donor	ee_S **(%)[b]**	ee_P **(%)[c]**	**Conversion (%)[d]**	**E[e]**
Vinyl acetate	90 ± 0.8	99 ± 0.5	48 ± 0.9	588
Ethyl acetate	5.0 ± 1.7	35 ± 1.5	13 ± 1.2	2.20
Isopropenyl acetate	36 ± 2.0	90 ± 2.4	29 ± 2.8	25.6
Benzoyl acetate	36 ± 2..0	83 ± 1.8	26 ± 2.0	2.40
Acetic anhydride	No reaction	No reaction	No reaction	---

[a](*RS*)-5 (2.135 mg, 5 mM, 1 eq.) in diethyl ether (800 µL) was treated with lipase (30 mg/mL), acyl donor (200 µL) at 30 ˚C

[b]Enantiomeric excess of substrate was calculated by using formula = (*R*-*S*)/(*R*+*S*)*100

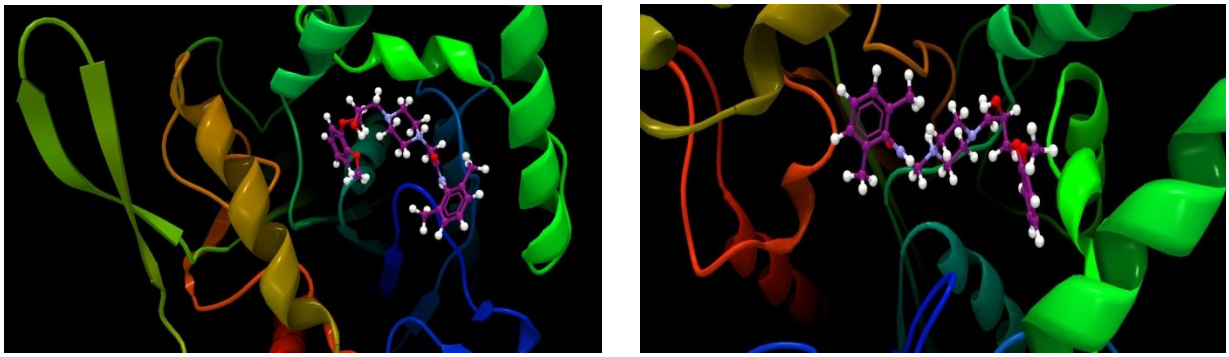
[c]Enantiomeric excess of product was calculated by using formula = (*S*-*R*)/(*S*+*R*)*100

[d]Conversions were calculated from enantiomeric excess of substrate (ee_S) and product (ee_P) using the formula: Conversion (C) = $ee_S/(ee_S + ee_P)$

[e]*E* values were calculated using the formula: $E = [\ln (1 - C (1 + ee_P)]/ [\ln (1 - C (1 - ee_P)]$.

S4. *In-Silico* study for the screening of different lipases

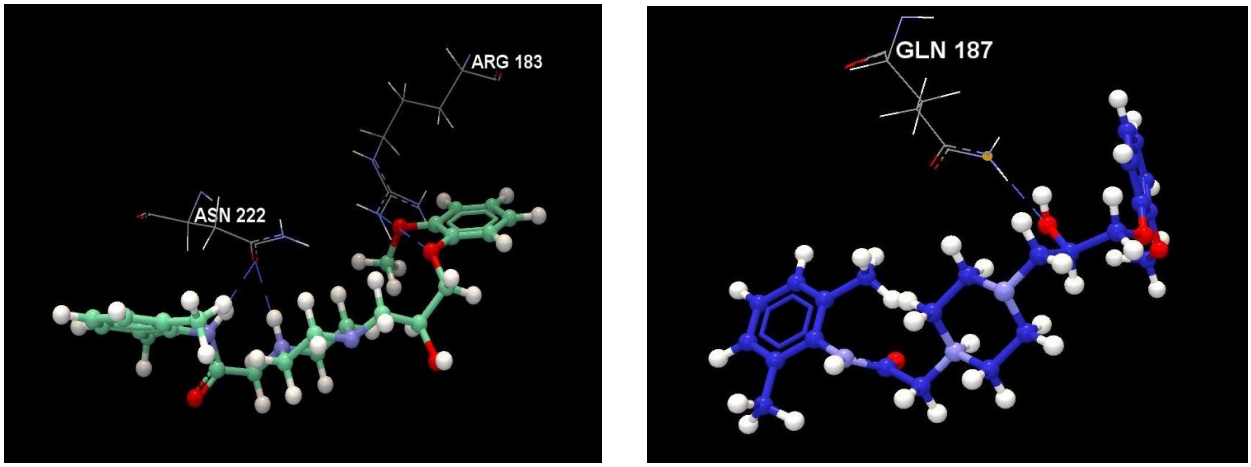
A. Amano 30 (2NW6)



(a) **(b)**

Figure. S4-1 (a) No interaction of Amano lipase with (*S*)-Ranolazine (b) No interaction of Amano lipase with (*R*)-Ranolazine

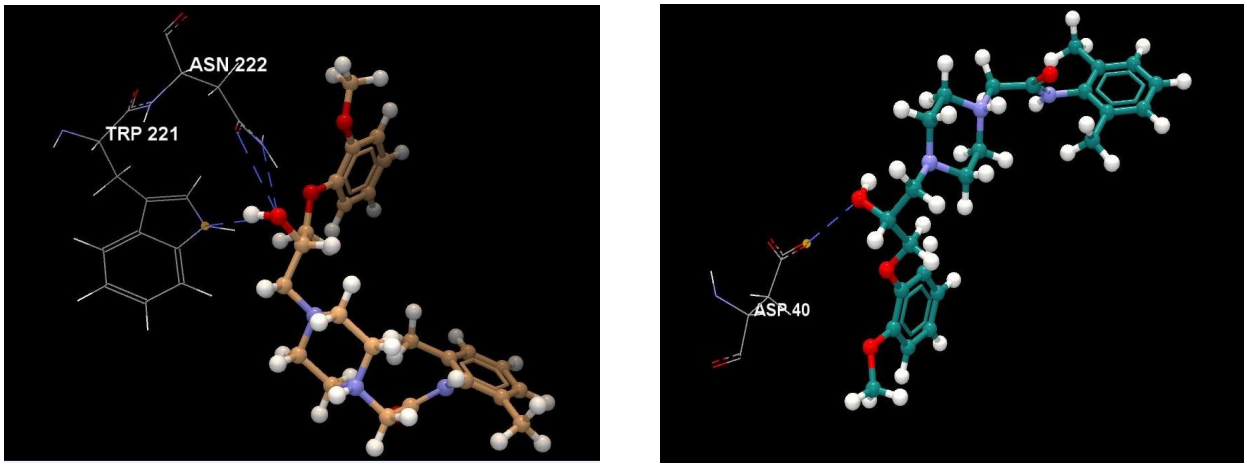
B. *C. rugosa* lipase 62316 (1LPN)



(a) **(b)**

Figure. S4-2 (a) Interaction of CRL62316 lipase with (*S*)-Ranolazine (b) Interaction of CRL62316 lipase with (*R*)-Ranolazine

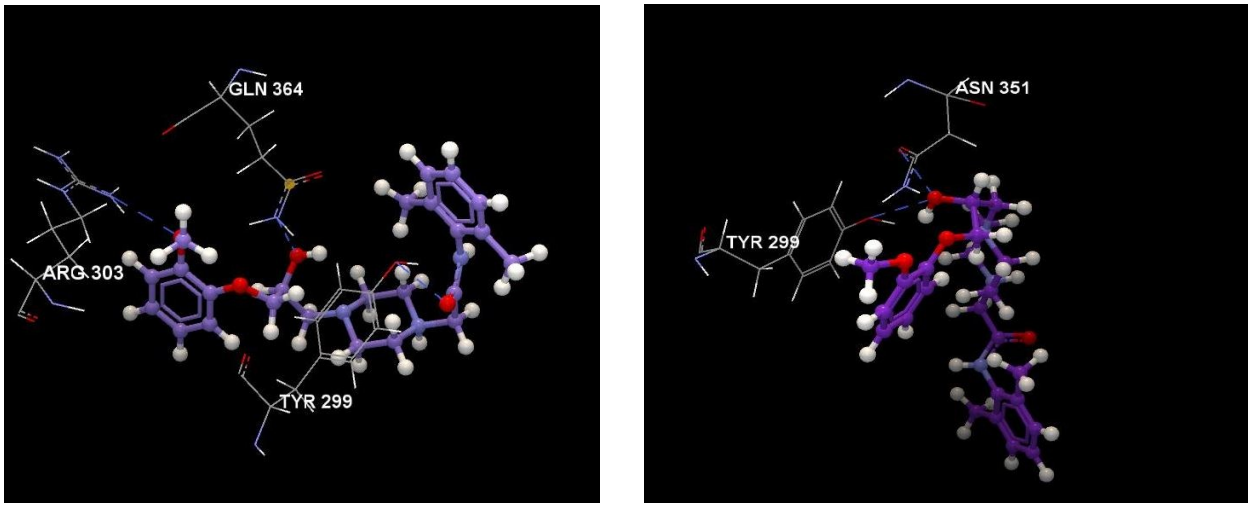
C. *C. rugosa* lipase 90860 (1LPO)



(a) **(b)**

Figure.S4-3 (a) Interaction of CRL 90860 lipase with (*S*)-Ranolazine (b) Interaction of CRL 90860 lipase with (*R*)-Ranolazine

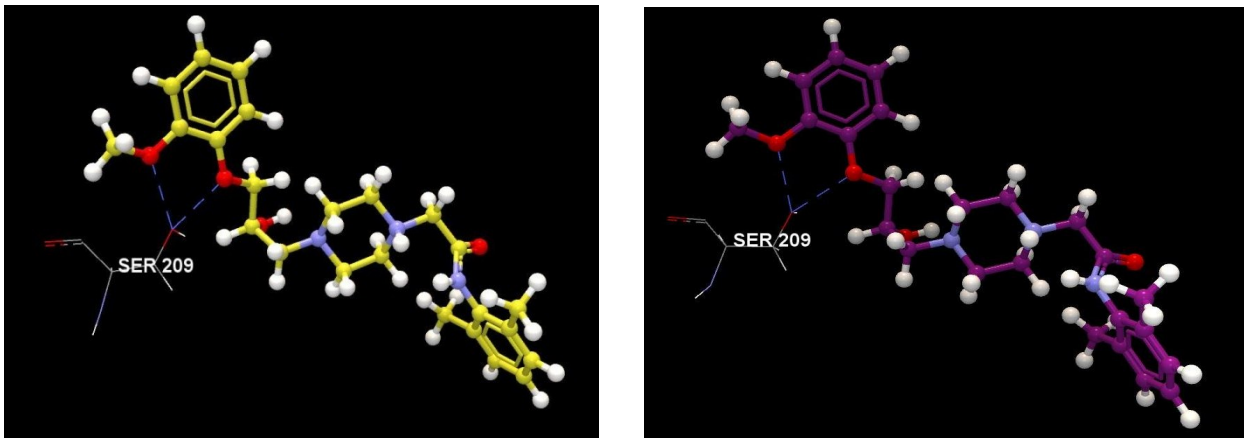
D. *C. rugosa* lipase L1754 (1GZ7)



(a) **(b)**

Figure. S4-4 (a) Interaction of CRL L1754 lipase with (*S*)-Ranolazine (b) Interaction of CRL L1754 lipase with (*R*)-Ranolazine

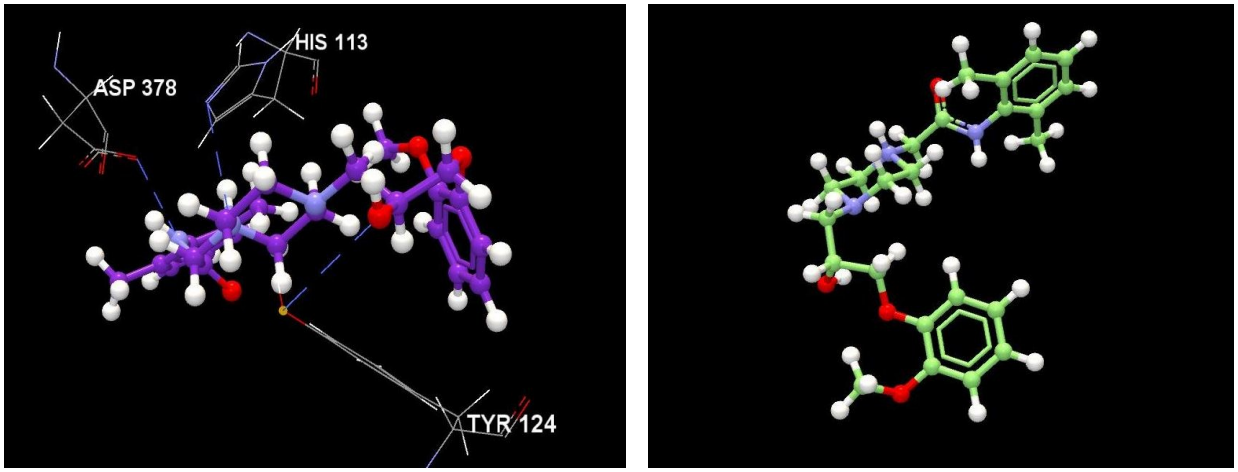
E. *C. cylindricea* lipase (1CLE)



(a) **(b)**

Figure. S4-5 (a) Interaction of *C. cylindricea* lipase with (*S*)-Ranolazine (b) Interaction of *C. cylindricea* lipase with (*R*)-Ranolazine.

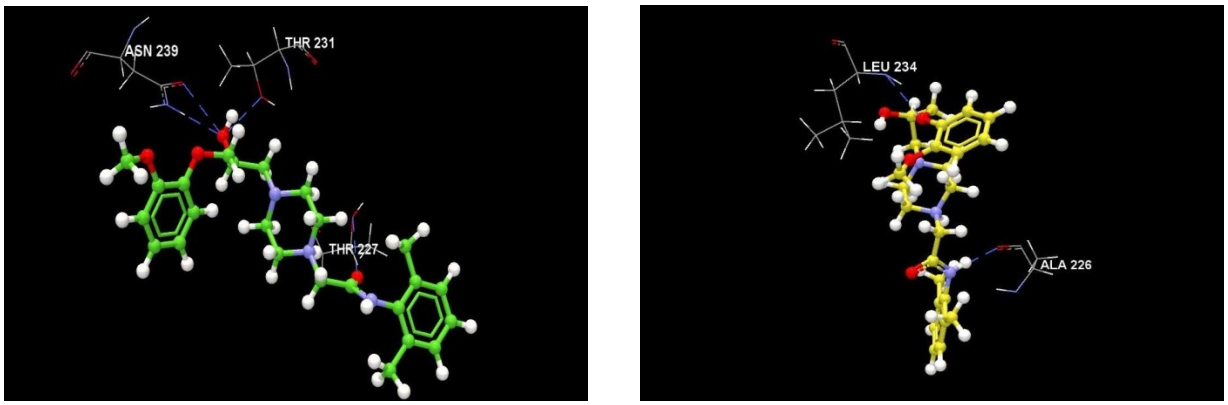
F. *Aspergillus niger* lipase (1UZA)



(a) **(b)**

Figure. S4-6 (a) Interaction of *Aspergillus niger* lipase with (*S*)-Ranolazine (b) No interaction of *Aspergillus niger* lipase with (*R*)-Ranolazine.

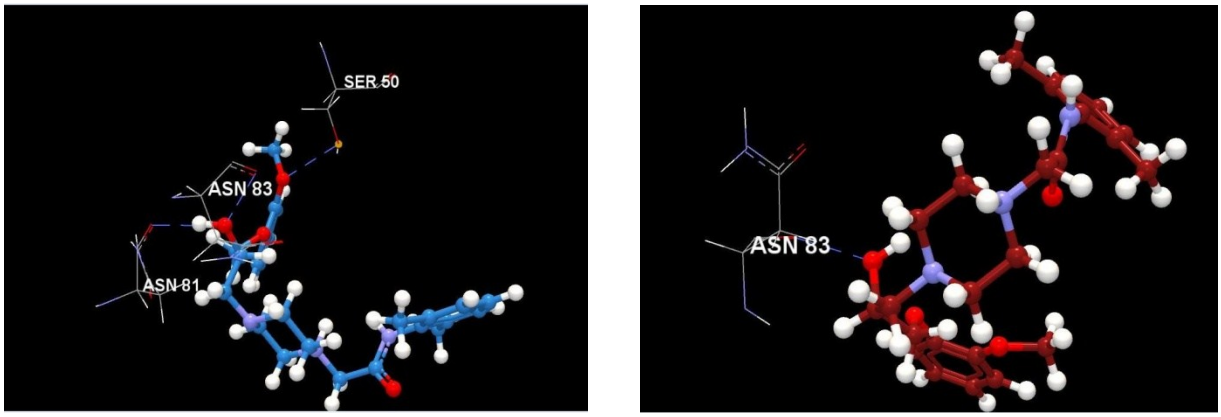
G. *Pseudomonas cepacia* lipase (3LIP)



(a) **(b)**

Figure. S4-7 (a) Interaction of *Pseudomonas cepacia* lipase with (*S*)-Ranolazine (b) Interaction of *Pseudomonas cepacia* lipase with (*R*)-Ranolazine.

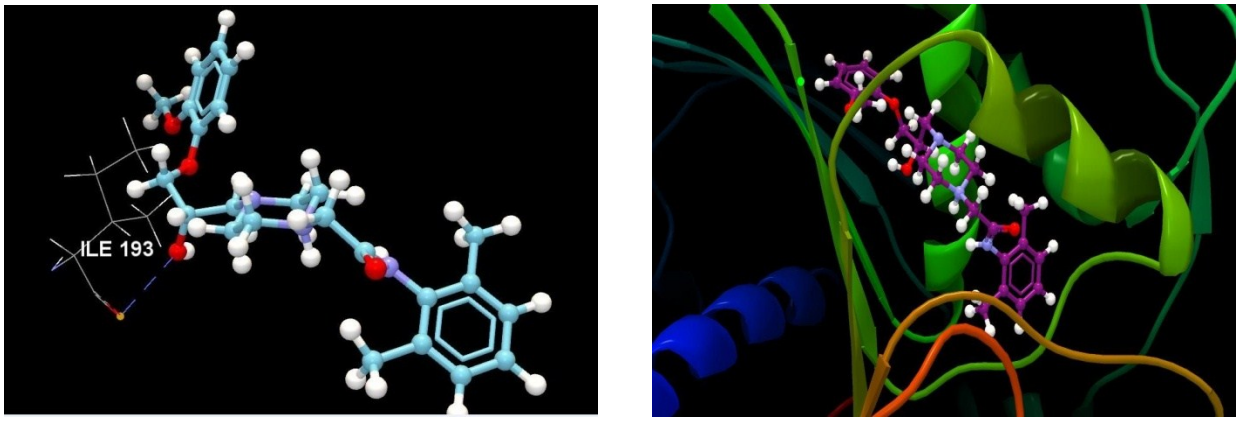
H. Porcine pancreatic lipase (1PCN)



(a) **(b)**

Figure. S4-8 (a) Interaction of Porcine pancreatic lipase with (*S*)-Ranolazine (b) Interaction of Porcine pancreatic lipase with (*R*)-Ranolazine.

I. *Mucor meihei* Lipase (3TGL)



(a) **(b)**

Figure. S4-9 (a) Interaction of *Mucor meihei* lipase with (*S*)-Ranolazine (b) No interaction of *Mucor meihei* lipase with (*R*)-Ranolazine.

J. *C. antartica* lipase in acrylic resin (3W9B)

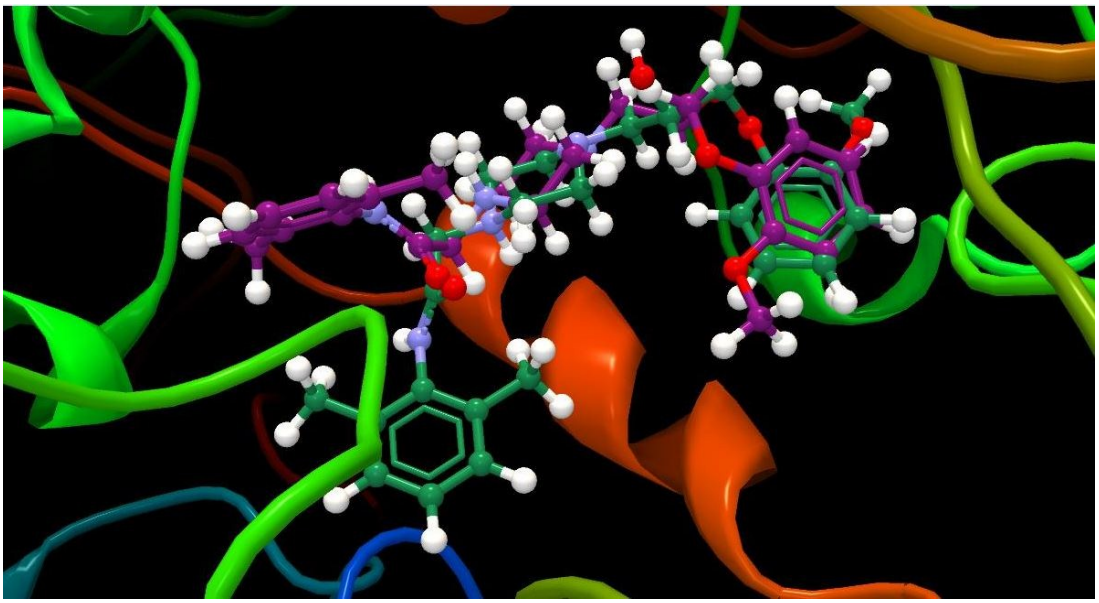
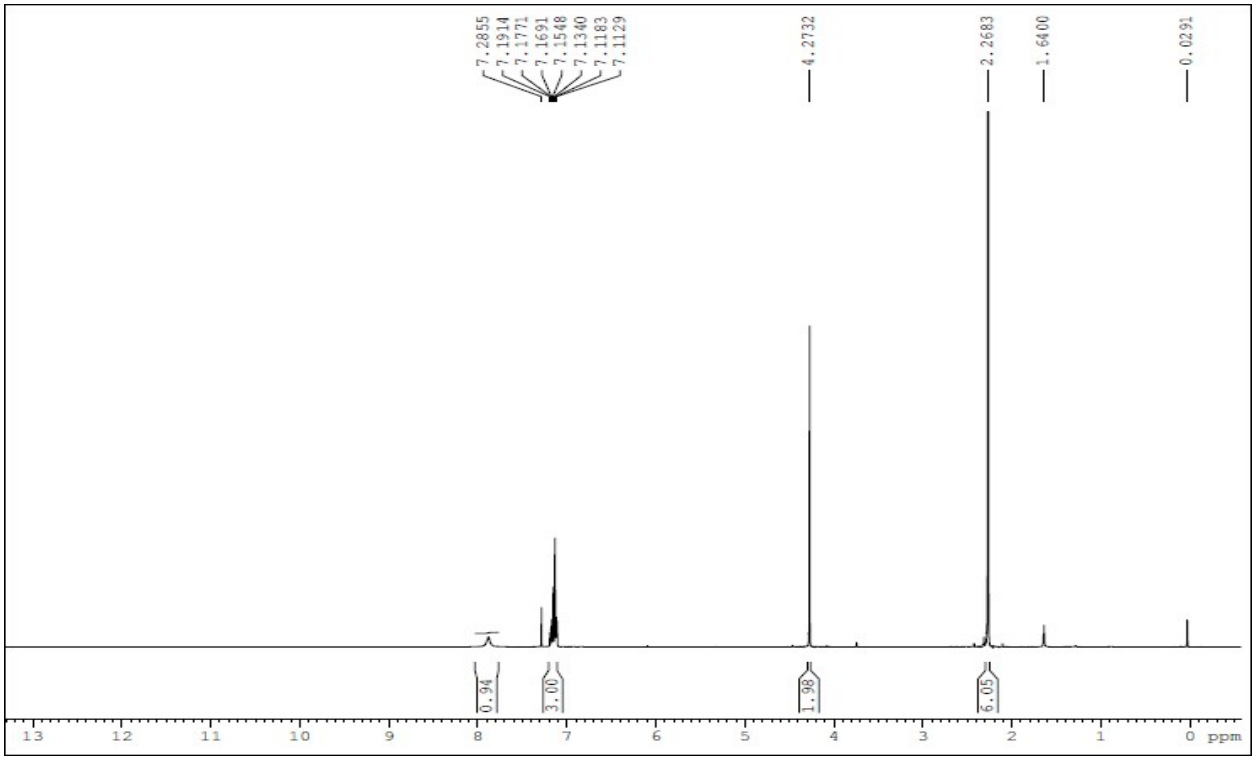
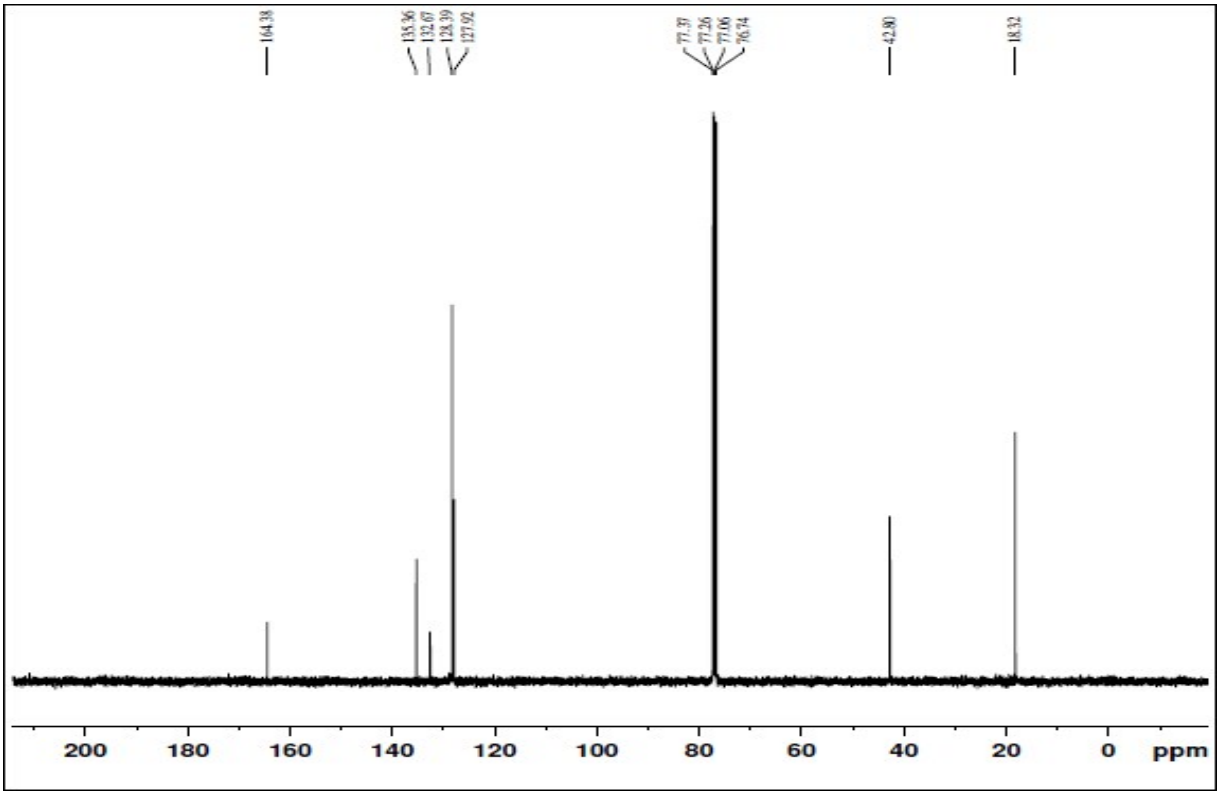


Figure. S4-10 No interaction of *C. antartica* lipase in acrylic resin with (*S*) and (*R*)-Ranolazine.

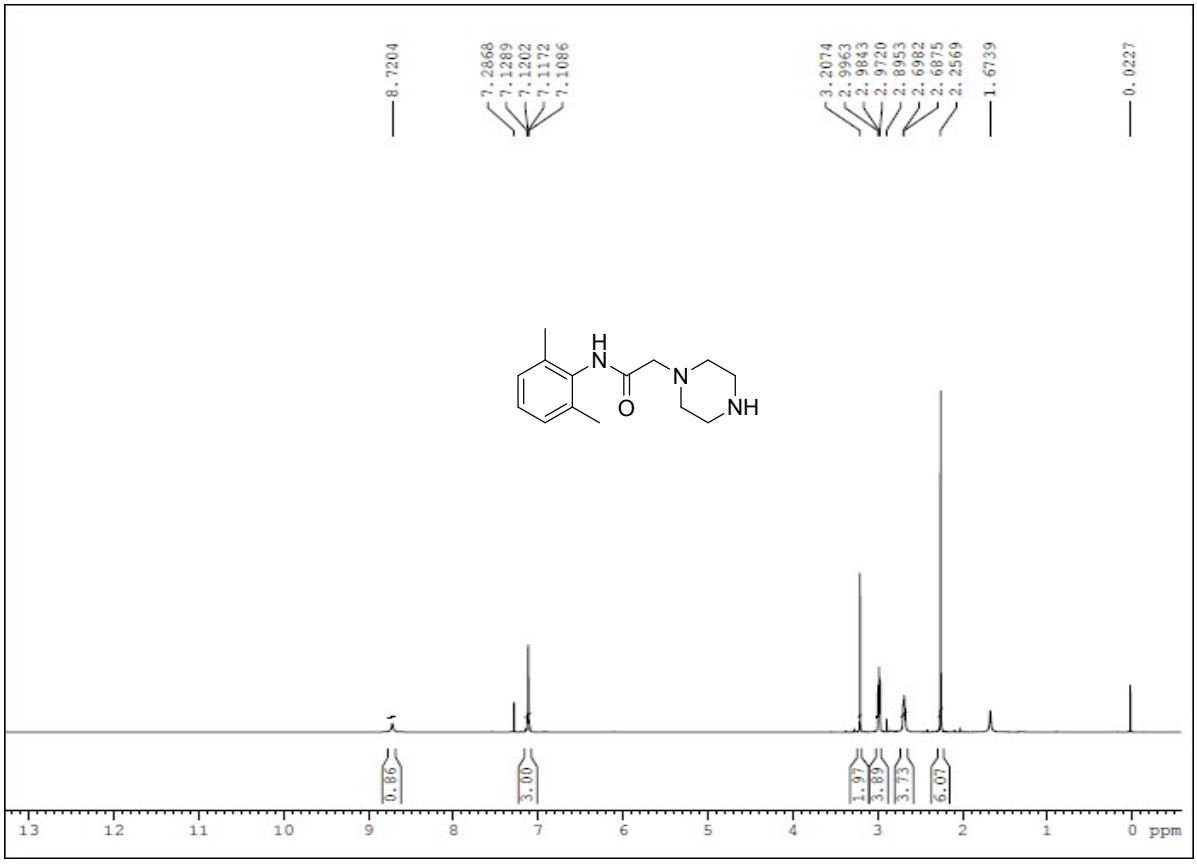
S5. ^{1}H NMR of 2-chloro-*N*-(2,6-dimethylphenyl)acetamide 2



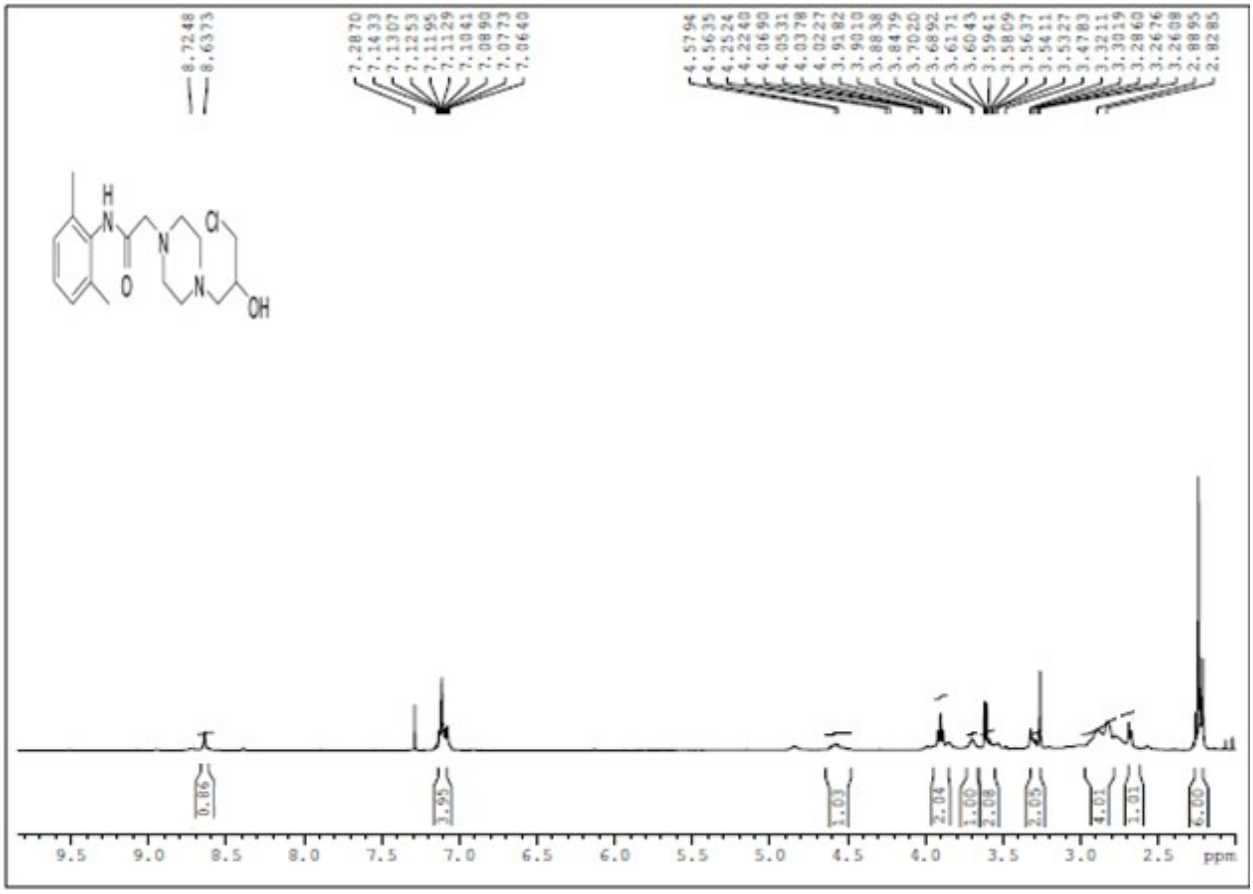
S6. ^{13}C NMR of 2-chloro-*N*-(2,6-dimethylphenyl)acetamide 2



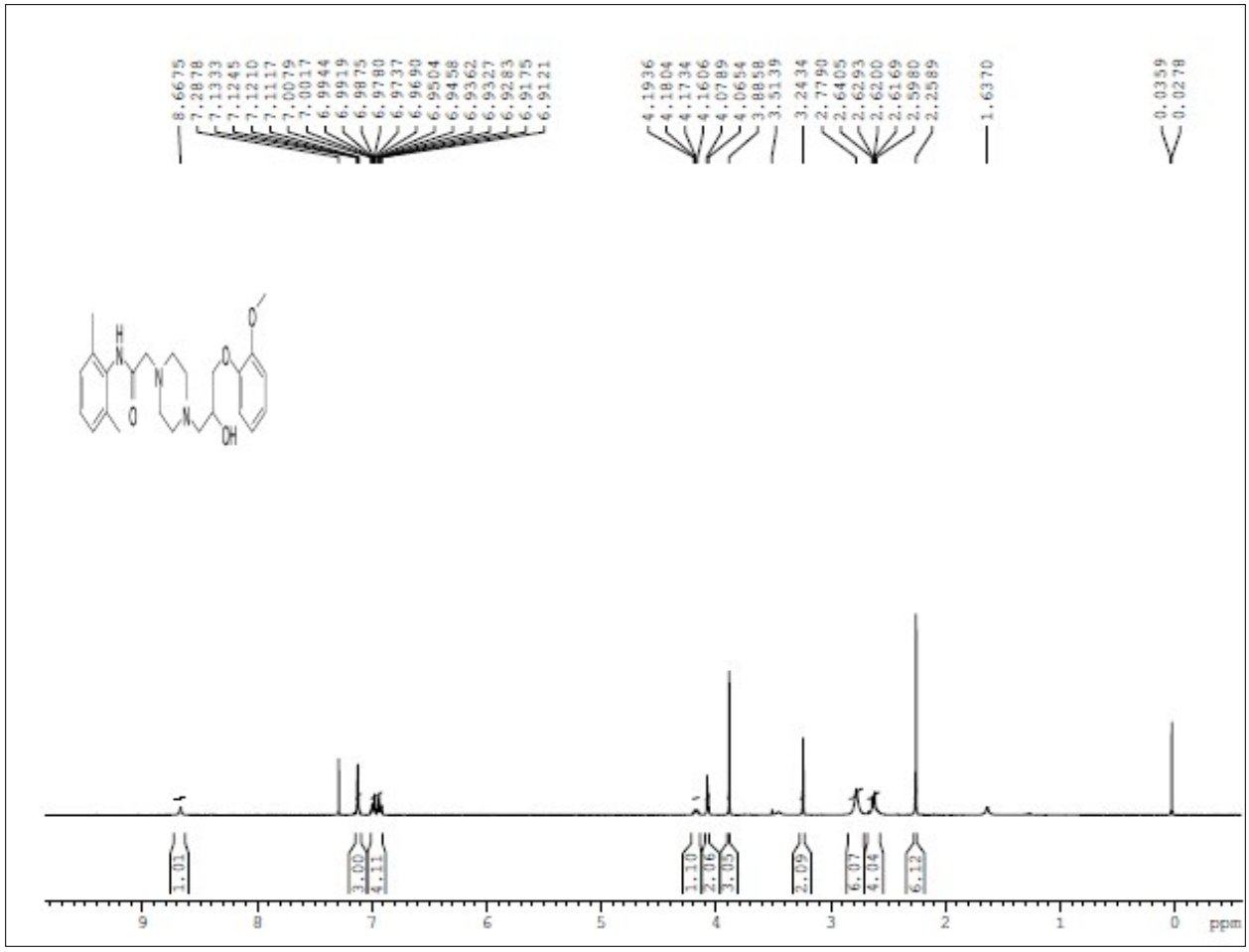
S7. Proton NMR of *N*-(2, 6 dimethylphenyl)-2-piperazin-1-yl)acetamide 3



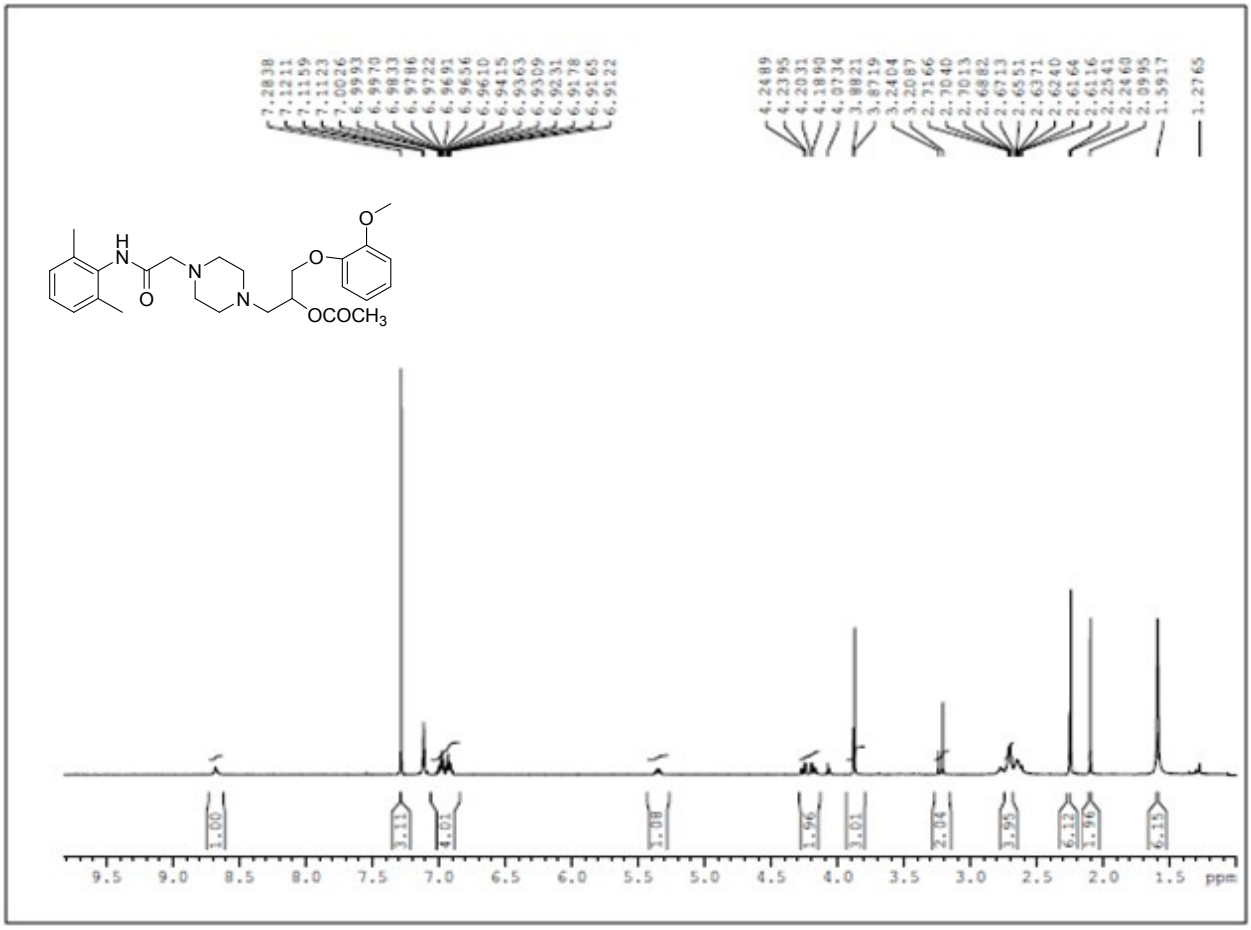
S8. Proton NMR of (*RS*)-4



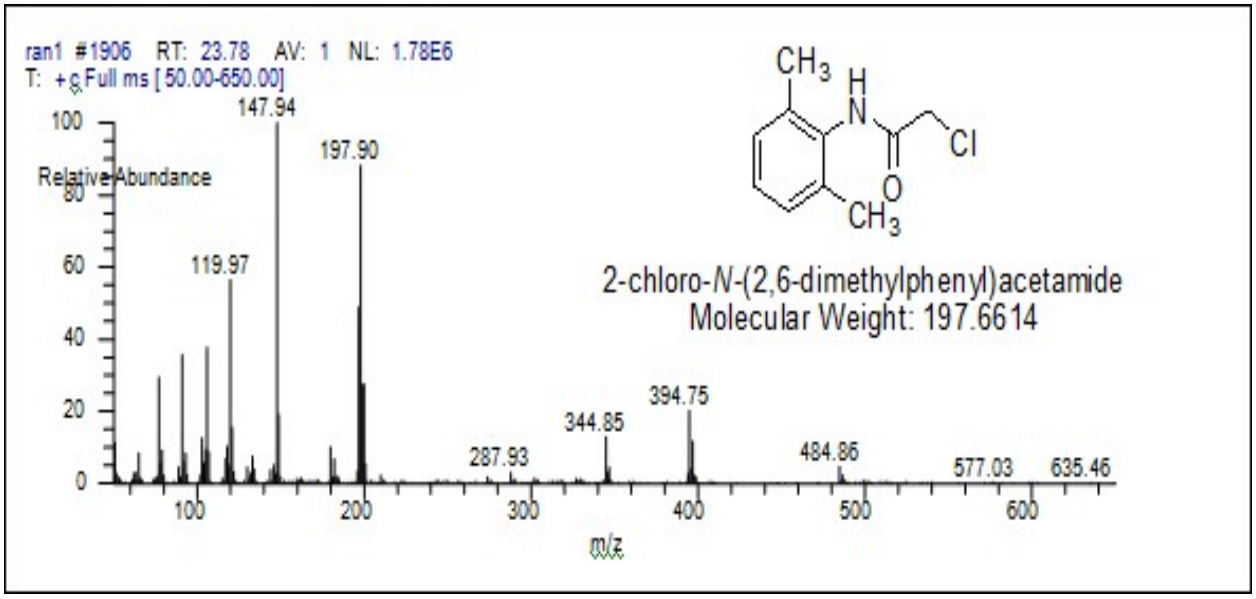
S9. Proton NMR of (*RS*)-Ranolazine 5



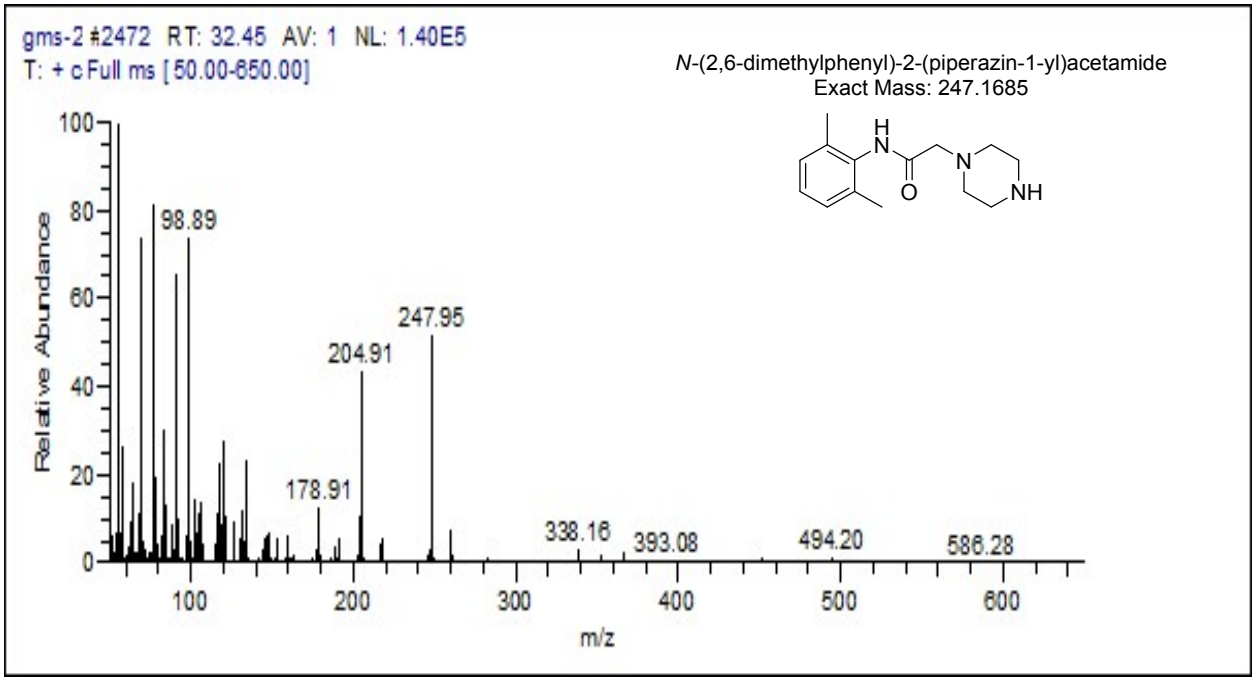
S10. Proton NMR of Acetylated (*RS*)-Ranolazine 6



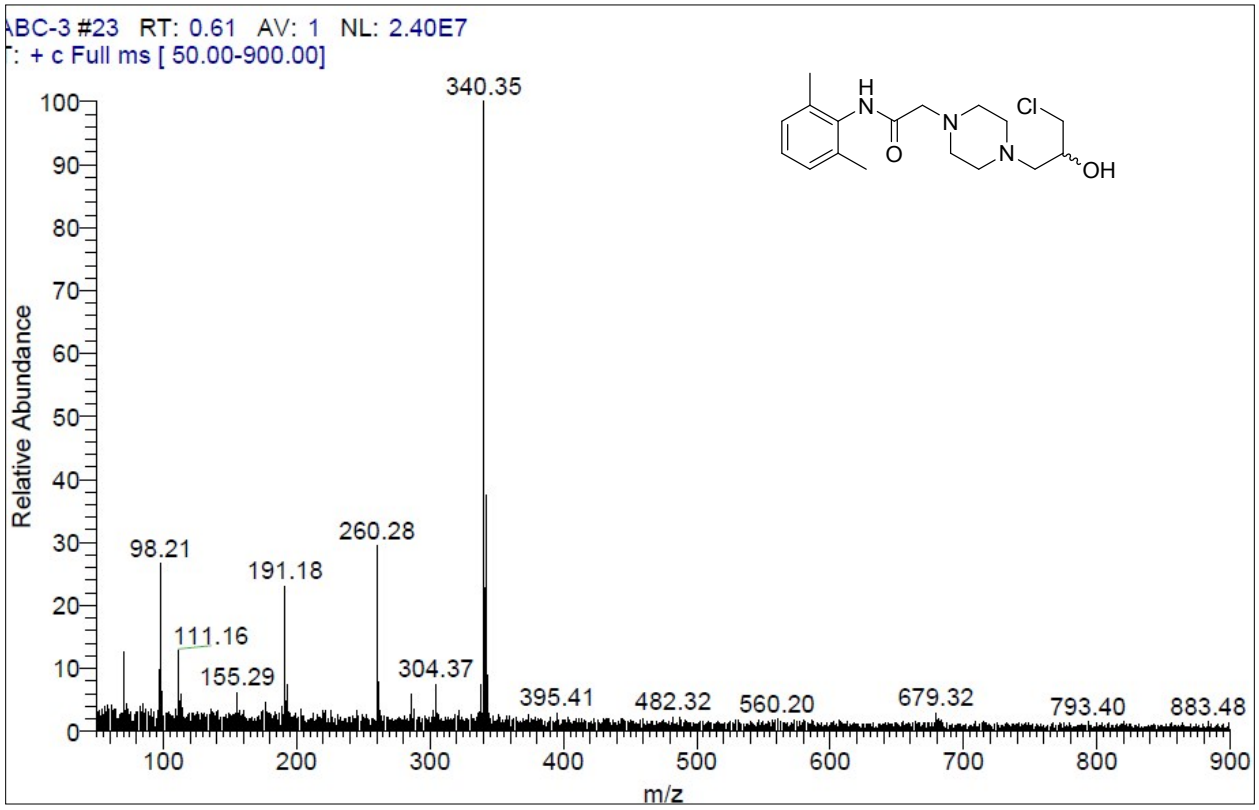
S11. GC-MS spectra of 2-chloro-*N*-(2,6-dimethylphenyl) acetamide 2



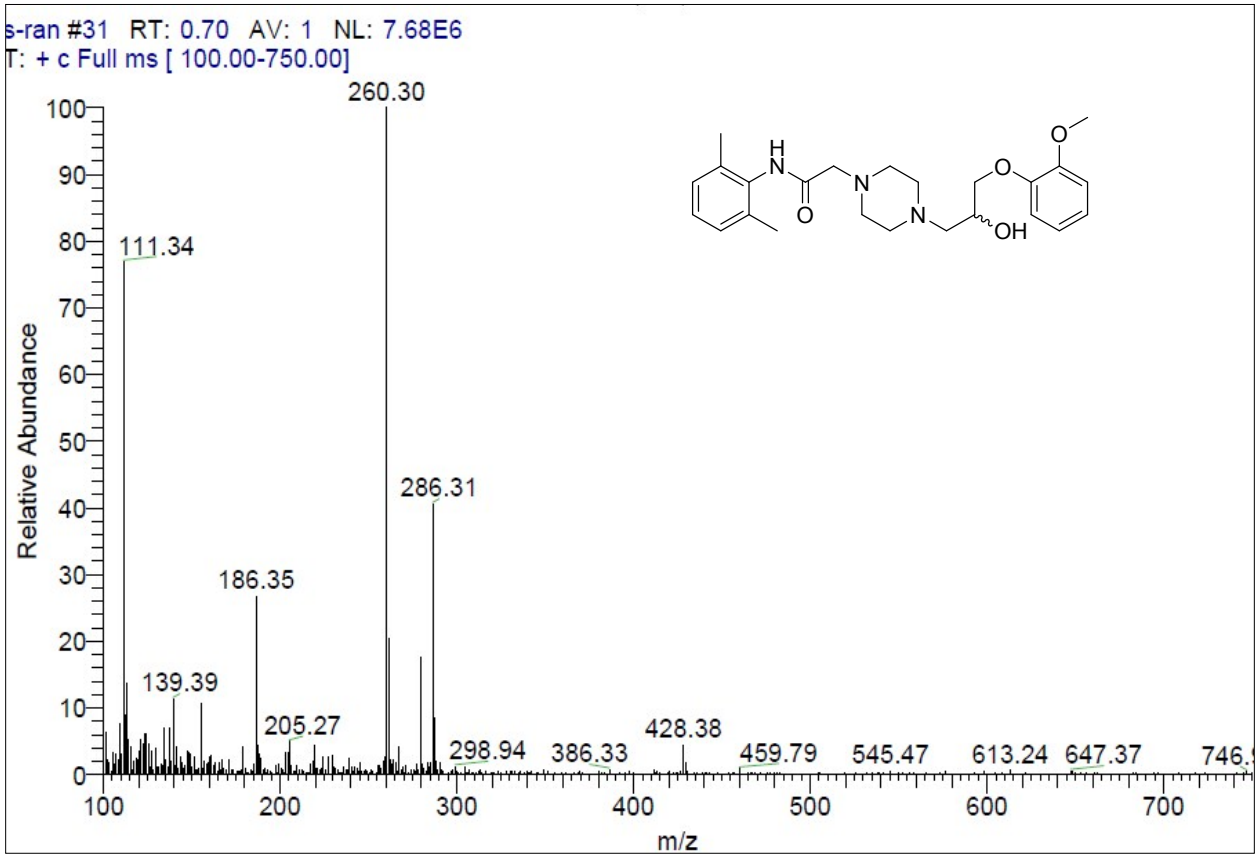
S12. GC-MS spectra of *N*-(2, 6 dimethylphenyl)-2-piperazin-1-yl)acetamide 3



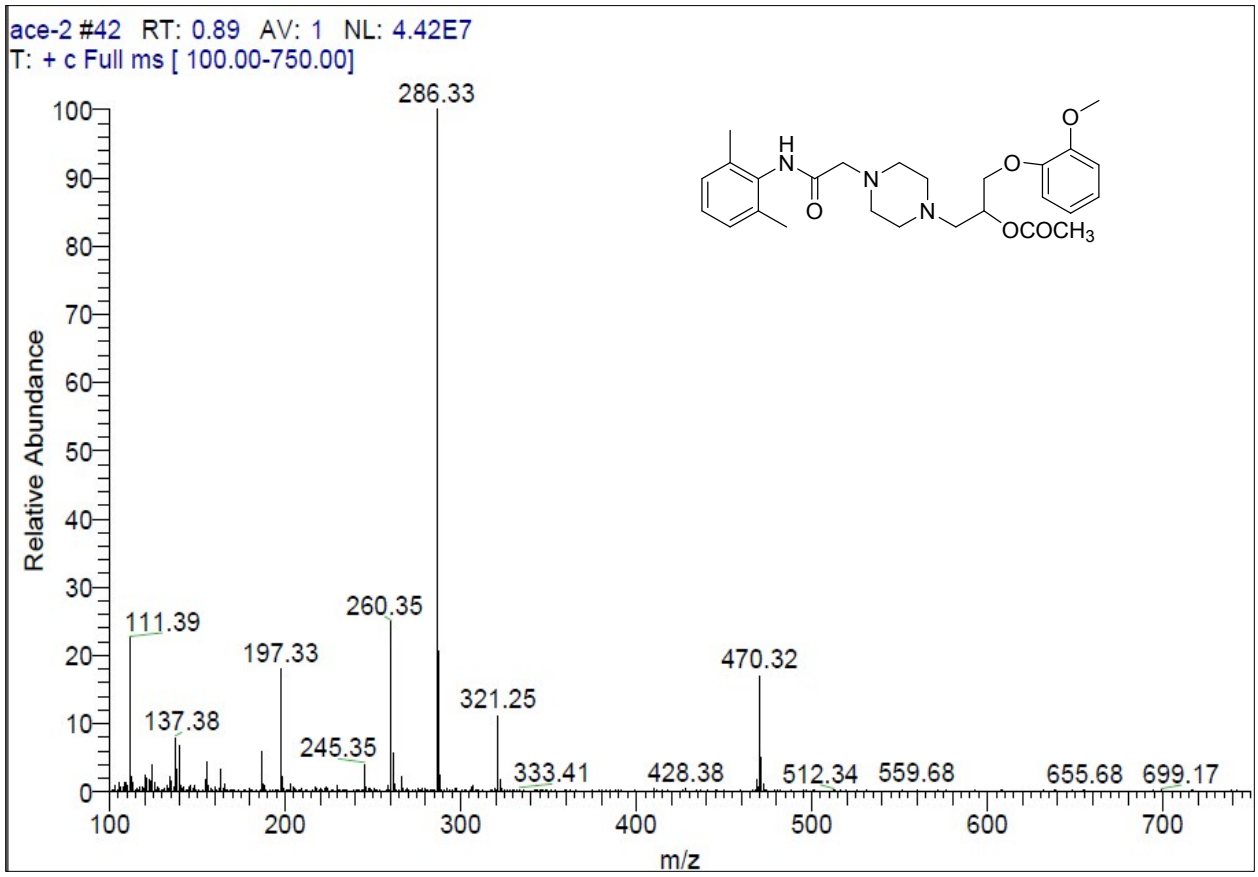
S13. GC-MS spectra of 2-(4-(3-chloro-2-hydroxyphenyl)piperazin-1-yl)-*N*-(2,6-dimethyl phenyl) acetamide 4



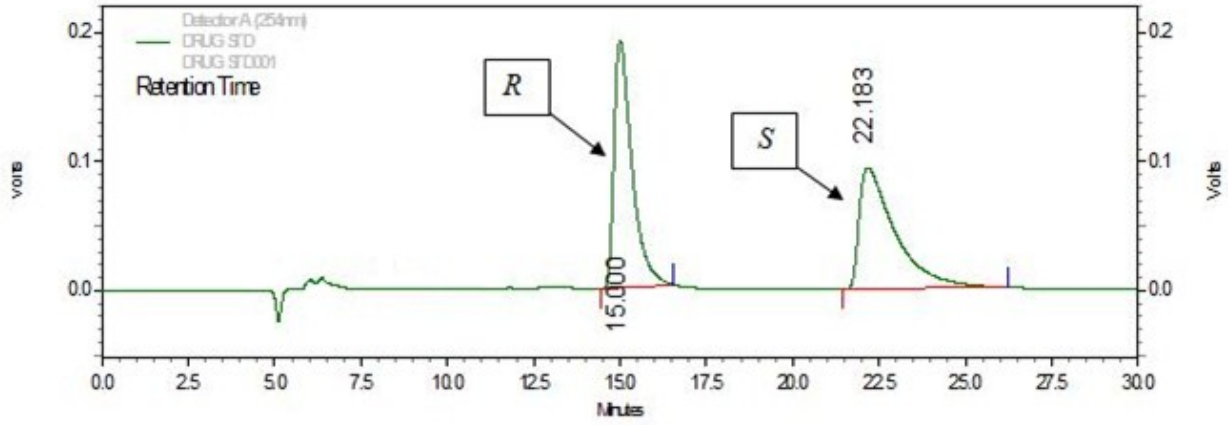
S14. GC-MS spectra of (*RS*)-Ranolazine 5



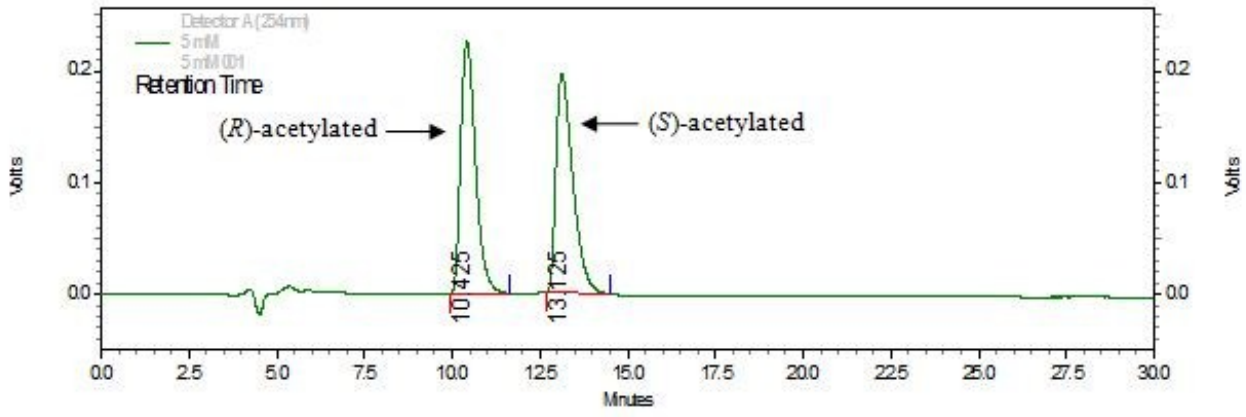
S15. GC-MS spectra of acetylated (*RS*)-Ranolazine 6



S16. HPLC chromatogram of (*RS*)-ranolazine



S17. HPLC chromatogram of acetylated (*RS*)-ranolazine 6



S18. HPLC chromatogram of *Candida antartica* lipase CLEA mediated kinetic resolution of (*RS*)-5 under optimized condition