Supporting Information

The identification and use of robust transaminases from a domestic drain metagenome

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Table of Contents

1. Taxonomic assignment of drain Class III TAms

Table S1. Protein length, predicted molecular weight (MW) (calculated using Expasy Protparam), protein annotation and organism and percentage identity to closest homologue in NCBI database. Data have been deposited in GenBank and the accession number of each enzyme coding sequence is listed below beside the pQR number.

pQR	Accession number	Length (aa)	Predicted MW (kDa)	Function	Taxonomic Assignment	% Ident. to NCBI
2188	MK121624	483	52.3	Aspartate aminotransferase family protein	*Pseudoxanthomonas sp.*	97
2189	MK121625	484	52.3	Aminotransferase	*Sphingopyxis sp.*	96
2190	MK121626	490	54.6	Adenosylmethionine-8-amino-7-oxononanoate transaminase	*Perlucidibaca sp.*	71
2191	MK121627	450	49.2	Glutamate-1-semialdehyde 2,1-aminomutase	*Novosphingobium aromaticivorans*	63
2192	MK121628	701	77.8	Glutamate-1-semialdehyde aminotransferase	*Herbaspirillum lusitanum*	77
2193	MK121629	417	44.4	Acetylornithine aminotransferase	*Azonexus hydrophilus*	85
2194	MK121630	456	48.1	Glutamate-1-semialdehyde aminotransferase	*Candidatus Propionivibrio aalborgensis*	83
2195	MK121631	478	51.5	Omega amino acid-pyruvate aminotransferase	*Acidovorax sp.*	99
2196	MK121632	469	51.1	Aspartate aminotransferase family protein	*Mesorhizobium*	90
2197	MK121633	414	44.5	Acetylornithine aminotransferase	*Alkanindiges illinoisensis*	78
2198	MK121634	454	47.7	Aspartate aminotransferase family protein	*Azospira oryzae*	99
2199	MK121635	468	49.8	Aspartate aminotransferase family protein	*Alkanindiges illinoisensis*	76
2200	MK121636	460	50.9	Aminotransferase	*Pseudoxanthomonas mexicana*	97
2201	MK121637	1494, 504	54.5	Lysine 6-aminotransferase	*Pseudoxanthomonas mexicana*	97
2202	MK121638	394	42.4	Acetylornithine aminotransferase	*Candidatus Accumulibacter phosphatis*	83
2203	MK121639	433	45.7	Aspartate aminotransferase family protein	*Azonexus hydrophilus*	96

2204	MK121640	401	42.8	Acetylornithine transaminase	*Sphingopyxis sp.*	98
2205	MK121641	447	48.4	Adenosylmethionine-8-amino-7-oxononanoate transaminase	*Roseateles depolymerans*	76
2206	MK121642	436	45.9	Glutamate-1-semialdehyde aminotransferase	*Pseudoxanthomonas sp.*	98
2207	MK121643	402	42.9	Acetylornithine transaminase	*Azospira oryzae*	98
2208	MK121644	1380, 466	50.1	Aspartate aminotransferase family protein	*Sphingomonas sp.*	83
2209	MK121645	404	42.4	Acetylornithine aminotransferase	*Curvibacter delicatus*	69
2210	MK121646	416	44.3	Acetylornithine aminotransferase	*Pseudoxanthomonas sp.*	94
2211	MK121647	412	44.2	Acetylornithine aminotransferase	*Pseudomonas lavalieres*	100
2212	MK121648	423	46.2	Denosylmethionine-8-amino-7-oxononanoate transaminase	*Sphingopyxis*	94
2213	MK121649	453	47.9	Aminotransferase	*Sphingopyxis*	87
2214	MK121650	459	49.5	Omega amino acid-pyruvate aminotransferase	*Aquabacterium parvum*	70
2215	MK121651	414	44.1	Acetylornithine aminotransferase	*Pseudoxanthomonas sp*	96
2216	MK121652	396	42.0	Acetylornithine aminotransferase	*Dechloromonas aromatica*	87

2. Percentage Identity Matrix

pQR	Cv	Vf	Mb	2188	2189	2190	2191	2192	2193	2194	2195	2196	2197	2198	2199	2200	2201	2202	2203	2204	2205	2206	2207	2208	2209	2210	2211	2212	2213	2214	2215	2216
Cv		38	13	33	48	27	19	17	26	20	34	34	24	20	18	54	17	25	19	28	22	18	27	48	26	28	30	26	18	31	28	26
Vf	38		12	30	36	28	21	18	27	18	30	32	22	19	19	36	16	26	18	30	25	20	24	34	26	25	26	22	16	31	26	26
Mb	13	12		19	15	13	13	16	18	18	17	16	18	19	20	13	16	17	18	17	16	17	15	13	16	18	16	18	20	18	17	19
2188	33	30	19		33	31	24	24	33	26	50	62	31	24	25	35	20	32	24	32	30	28	32	34	29	29	30	31	23	53	28	30
2189	48	36	15	33		26	20	20	26	20	29	31	23	19	19	49	20	28	19	26	26	21	27	69	27	25	26	25	17	31	25	27
2190	27	28	13	31	26		24	19	28	22	28	30	27	22	23	27	18	28	22	30	50	23	28	28	31	28	29	38	19	30	28	28
2191	19	21	13	24	20	24		30	22	31	20	19	21	30	29	19	16	22	31	22	21	31	21	20	24	23	21	19	23	19	23	21
2192	17	18	16	24	20	19	30		24	29	21	21	19	28	26	18	17	25	29	20	17	30	21	21	22	22	20	19	18	20	21	24
2193	26	27	18	33	26	28	22	24		27	28	32	54	26	25	27	23	71	24	41	27	22	39	27	40	41	42	25	21	29	42	81
2194	20	18	18	26	20	22	31	29	27		24	25	26	80	62	21	18	27	77	25	22	59	31	22	30	26	26	22	28	25	27	27
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2196	34	32	16	62	31	30	19	21	32	25	55		29	24	23	34	19	30	23	31	29	23	26	34	27	26	29	33	21	55	26	29
2197	24	22	18	31	23	27	21	19	54	26	27	29		24	25	25	24	54	23	42	27	24	38	24	39	38	38	25	23	28	39	54
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2210	28	25	18	29	25	28	23	22	41	26	24	26	38	26	24	27	26	42	24	41	27	23	39	27	38		50	25	25	28	93	41
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2215	28	26	17	28	25	28	23	21	42	27	24	26	39	27	25	28	26	42	24	40	28	24	39	28	39	93	51	25	26	27		41
2216	26	26	19	30	27	28	21	24	81	27	25	29	54	25	24	25	23	73	24	39	24	22	38	26	37	41	39	23	23	27	41	

0 50 100 %

Figure S1. Heat map of the percentage identity of 29 cloned drain TAms. Percentage identity matrix generated using Clustal Omega Multiple Sequence Alignment tool and visualised using Excel. Cv: *Chromobacterium violaceum* CV2025 accession number WP_011135573.1, Vf: *Vibrio fluvialis* JS17 accession number AEA39183.1, Mb: *Mycobacterium vanbaalenii* ABM15291.1.

3. Expression of 29 TAms

In the following SDS-Page gels, lane 1 is always ladder - NEB Broad Range protein ladder (10-250 kDa) (Figure S2-S4) or NEB color protein standard (Figure S5-S9), each protein is in two wells – the first is the cell free extract (CFE) and the second is the total protein (TP) fraction.

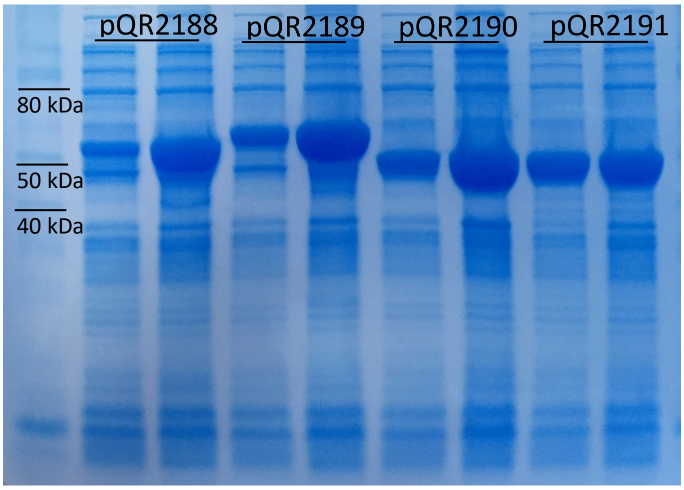

Figure S2. SDS-Page gel of pQR2188-2191.

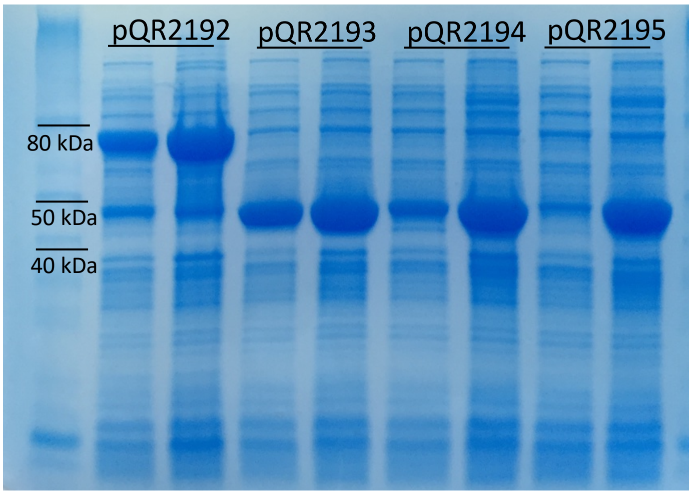

Figure S3. SDS-Page gel of pQR2192-2195.

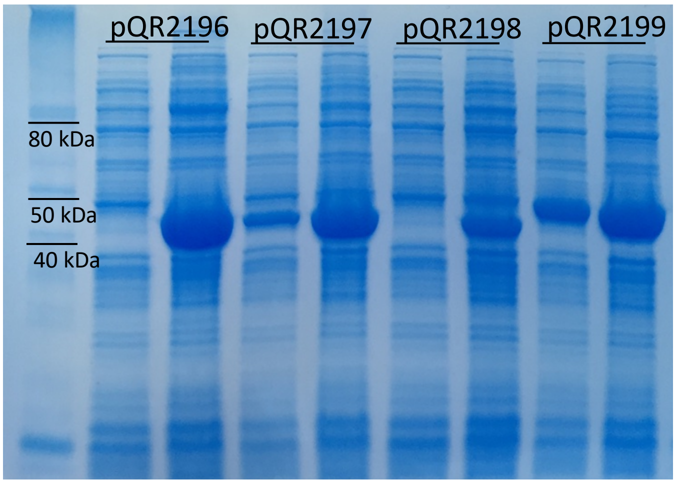

Figure S4. SDS-Page gel of pQR2196-2199.

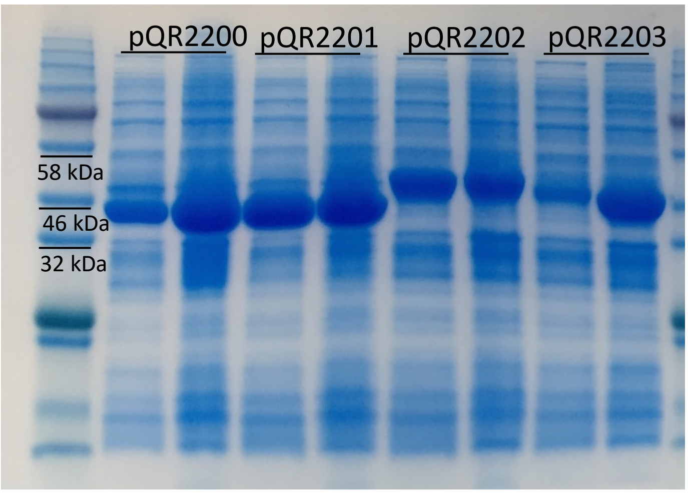

Figure S5. SDS-Page gel of pQR2200-2203.

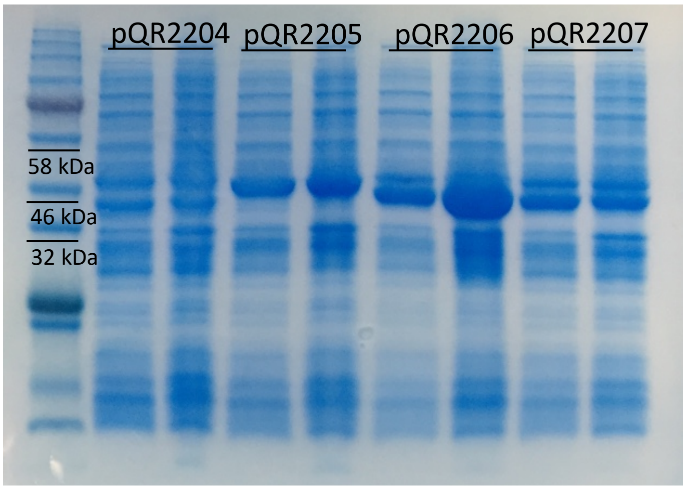

Figure S6. SDS-Page gel of pQR2204-2207.

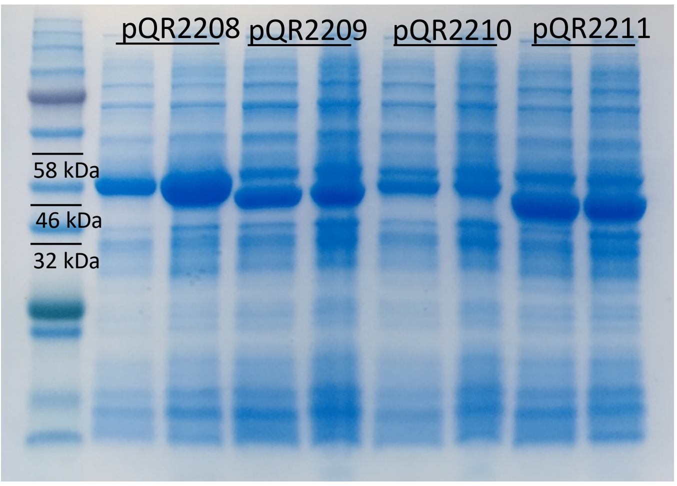

Figure S7. SDS-Page gel of pQR2208-2211.

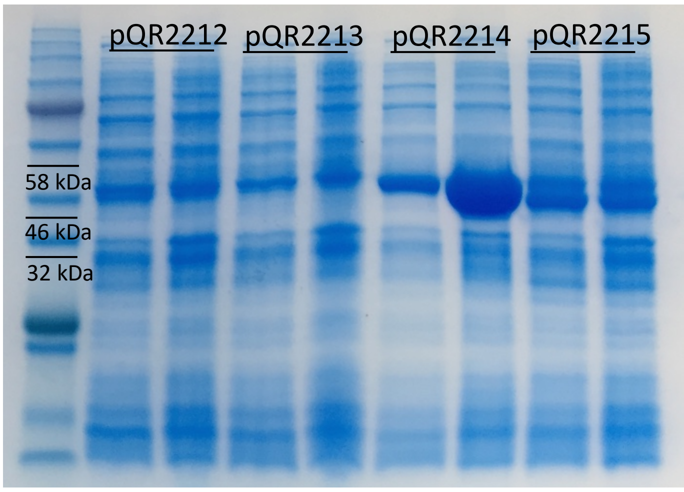

Figure S8. SDS-Page gel of pQR2212-2215.

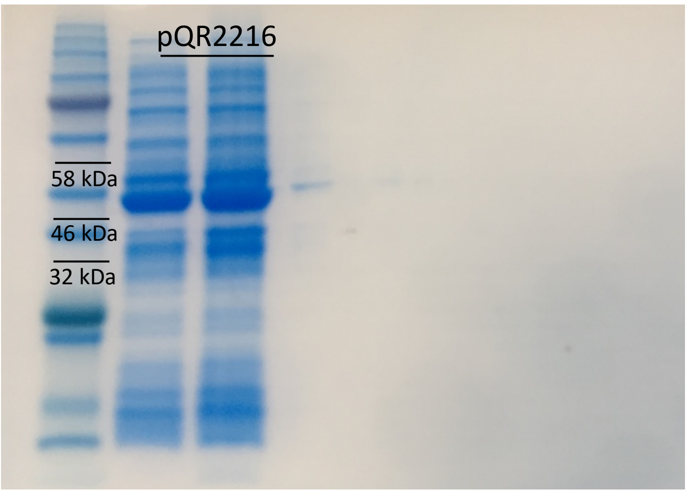

Figure S9. SDS-Page gel of pQR2216.

4. DNA Sequences

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> pQR2213
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> pQR2216
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5. Amino acid sequence

Protein sequences correspond to cloned genes; amino acids in **bold** correspond to the vector sequence.

> pQR2188
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> pQR2189
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ACIANGLMVRGIRDSLVMCPPLIISTEQIDEMVAIIRKSLDEVMPKLRAL**EHHHHHH**

> pQR2190
MGSSHHHHHHSSGLVPRGSHMNKNERLAQRDLRHVWHPCTQMQDHEQLPIVPIQRGQG
VWLEDFEGRRYLDAVSSWWVNLFGHANPRINNAVKEQLDTLEHVILAGFTHEPIVELSERL
VQLAPKGLTRCFYADNGSAATEIALKMSLHFWRNVGKAEKTRFICLENGYHGETLGSLSVT
DIPLFSATYAPLLKDHLRAPSPDCSRRDEGESWESFSRRQFAAMEALLEKHHAEVSAVILEP
LVQGAAGMKMYHPVYLTLLREACDRYGVHLIADEIAVGFGRTGTLFACEQAGITPDFLCLSK
GLTAGYLPMSVVMTTDTVYNAFYDSYESLKGFLHSHSYTGNALAARAALASLDIFASDNVLE
KNKLLAATMTDALRGLGDHQHVLEVRQTGMIAAVELVQDRRTRQPFDWRERRGLQIFQHA
LDKGVLLRPIGSVVYFIPPYVITPEEIRLMVDVAAAAIDVATAGTASRPGPGNIAL**EHHHHHH**

> pQR2191
MGSSHHHHHHSSGLVPRGSHMSGQRDQELRARAAKVMPSSAFGHVGTALLPANYPQFF
ERAEGAYVWDADGNRYLDYMCAFGPNLLGYRDPRVESAASAQAARGDVMTGPSPLAVEL
AEKFVEIVSHADWAFFCKNGTDATTIARTIARAQTGRRKILIAEGSYHGAAPWCNPFPAGTV
PEDRAHMLTFTFNDIASLEAAVAEAGDDLAGIIATPFKHEAFANQEFPTQDYARRCREICDA
SGAVLVVDDVRAGFRLAVDCSWATVGVKPDLSCWGKCFANGYSISAVMGSNRVKQGADS
IFATGSFWQSAISMAAALATLDIIRDGKVIEKTVRLGQRLRDGLDEVSRRHGFTLNQTGPVQ
MPQILFEGDPDFRVGFAWTSAMIDRGFYLHPWHNMFLCDAMTEEDIDQTIEAADSAFATVR
AALPTLQPHERVLALFSARA**EHHHHHH**

> pQR2192
MGSSHHHHHHSSGLVPRGSHMTRIVAIVQARMGSTRLPNKVMRPIAGIPMIEVLLKRLAQS
QRIDQICLATADDVRNQPLVAHVQQLGYAVYQGSEHDVLDRFYHAAEQMQADVVIRITGDC
PLIDAALVDLVIDRFLQGDVDYVSNAVPPTYPDGLDTEVFSMAALRQAWQQATSTFDHEHV
TPYLRDSGKFRLAVVSGEHDYSGERWTVDEPADFDVITQIFAHFAPRLDFSWTEVLALCHT
QPQLFAANQHLIRNEGAHMGTGQKLWKRAKNVIAGGNMLLSKRPEMFLPEQWPAYFSRA
QGCTVWDLDNQAYTDMIMGIGTNTLGYGHPEVDDAVRRTIDAGNMSTFNCPEEVYLAEKLI
ELHPWADMVRFARSGGEANAIAIRVARAATGKSKVAICGYHGWHDWYLAANLGDDKNLAG

HLLPGLEPNGVPESLRGTIYPFNYNNFAELEALVNSQDIGVIKMEVSRNHGPEDGFLHKVRE
LATARGIVLIFDECTSGFRQTFGGLHKLYGVEPDMAMFGKALGNGYAITATIGRREVMEAAQ
TTFISSTFWTERIGPTAALKTLEVMERERSWDTITQTGLAITERWKTLAARHGLSINTNGLPA
LTGFAFNSPNALAYKTLITQEMLGKGYLAGTSVYVCTAHTPEIVDGYFAALDPIFGVIRECED
GRDVMSLLKGPICHAGFKRL**EHHHHHH**

> pQR2193
MGSSHHHHHHSSGLVPRGSHMSHVMNTYARLPVAFSHGKGSRVTDTEGREYLDALSGIA
VNTLGHAHPRLVAAIAEQAGRLIHTSNLYGAVGQERLADRLCALSGMQEVFFGNSGAEANE
AAIKLARFYGHKKGIELPTVIVMEKSFHGRTMATLSATGNYKVQVGFEPLVAGFVRVPYGDL
DAIRAVAEQNPNIVAVMLEVIQGEGGIHLREPAYYQGVRQLCDAHDWLMICDEVQCGMGR
TGKWFGYQQVGVQPDIATLAKGLGSGVPIGACMAGGRAAGLFGPGNHGSTFGGNPLVCA
AALTTLDCIEEEGLLANAENIGKLIRQRLAAGLADARGVVDIRGHGLMIGIELDRPCGVLVTQ
GLAAGLLINVTGDTVVRLLPPLNFSERDASELVDRMIPLIKAFLAG**EHHHHHH**

> pQR2194
MGSSHHHHHHSSGLVPRGSHMSQGNQQLFERAQKHIPGGVNSPVRAFRSVGGTPRFFA
KGRGARVTDADGKTYLDYVGSWGPLILGHAHPEVVKAVQEAASDGLSFGAPTEREVEMAE
LLCAMLPSLDMVRLVSSGTEATMSAIRLARGHTGRDLLIKFEGCYHGHSDGLLVKAGSGLL
TFGNPSSGGVPADVAQHTMVLDYNDVGQLEAAFTEHGDRIAAVIVEPVAGNMNLIAPLPAF
LKTMRALCTQHGAVLIFDEVMTGFRVGPQCAQGFYGITPDLTTLGKVIGGGMPVGAFGGK
REIMEKIAPLGPVYQAGTLSGNPVAVAAGLATLRLIQAPGFYDALAASTRALCAGLTEAAKR
HGIAFSAQSVGGMFGIYFRASCPTSYAEVMECDKEAFNRFFHAMLDAGHYLAPSAFEAGF
VSATHSQADIAETVAAAGRWFASLQPS**EHHHHHH**

> pQR2195
MGSSHHHHHHSSGLVPRGSHMSFAVTDPTPTAPVRTDAAWLDAHWMPYTGNRQFKANP
RMIVEGSGAYYTDSEGRKIFDGLSGLWCAGLGHGRREIAEAISKQAMKLDYAPAFQFGHLL
SFELANRVKELTPAGLDYVFFTGSGSESADTSLKMARAYWRAKGQGTKTRLIGREKGYHG
VNYGGISVGGIVGNRKLFGQGVEADHLPHTQPPAGSFHKGMPPTGKELADRLLEVIGLHDA
SNIAAVIVEPFSGSAGVVIPPVGYLQRLREICTQNNILLIFDEVISGFGRSGAFTGAEAFGVTP
DILNFAKQVTNGAQPLGGVIASKEIYDTFMAAGGPEYMLEFPHGYTYSAHPVACAAGIAALD
ILQKEDMIGRVKALAPYFENAVHSLKGAKHVADIRNFGLAAGFTIAAVPGEPAKRPYEIAMKC
WEKGFYVRYGGDTIQLAPPFISTSAEIDRLVSALGDALQETA**EHHHHHH**

> pQR2196
MGSSHHHHHHSSGLVPRGSHMSNRLKVAPNDLSAFWMPFTSNRQFKQAPRMLAAAKDM
HYTTTDGRKILDGTAGLWCVNAGHCRPKITEAIQQQAGELDYAPAFQMGHPIVFELSNRLID
IAPAGMEHVFYTNSGSESVETALKIALAYHRAKGNGSRSRLIGRERGYHGVNFGGISVGGIV
NNRKMFGSLLTGVDHMPHTHNLAKNAFTKGEPEHGAELADELERIVTLHDASTIAAVIVEPV
AGSTGVLIPPKGYLKRLREICTKHGILLIFDEVITGFGRLGTPFAADYFDVQPDIITTAKGITNG
VIPMGAVFVTKEIHDAFMNGPEHVIEFFHGYTYSGNPIACAAALGTLDTYKEEGLLTRGAEL
APYFEEALHSLKGEPNVIDIRNIGMVGAIELEPIAGSPTKRAFQAFVKAYEKGCLIRTTGDIIAL
SPPLIITKGQINELVDHVRDVLRAVD**EHHHHHH**

> pQR2197
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HAHPTVAAAIADQAATLVHTSNLFEVPWQEAAGRLLCDVGGMQQCFFANSGAEANEAAIKL
ARMHGYKKDFQAPKIIVMEKSFHGRTLATLSATGNEKVQKGFYPLNDSFLRVPFGDVAAIEA
LAAQHSEIVAILVEPIQGEGGINTAPQGFVYLEQLRALCDQHDWLLMVDEIQTGNGRTGTYF
AYQHTSITPDVLTTAKGLGNGFPVGACLVSGKATQLFSAGNHGSTYGGTPLACRTVHTVIE
TLQTEQAMDNAARVGQWLKAQFTTQLAELGVEVRGFGMMIGIELPKACGALVARARDEQH
LILNVTADNVIRLLPPLNLSDVDAQDLVNRLVPLVKDFLAA**EHHHHHH**

> pQR2198
MGSSHHHHHHSSGLVPRGSHMSSRNQQLFDAAQRHIPGGVNSPVRAFRSVGGAPRFFTR
GEGPRVWDAEGKSYLDYVGSWGPLILGHAHAPTVKAVQEAAALGLSFGAPTEAEIEIADLL
CDILPSLDMVRLVSSGTEATMSAIRLARGHTGRDLLVKFEGCYHGHSDSLLVKAGSGLLTF
GNPSSGGVPADVAKHTLVLEYNNAEQLAEAFAKQGSEIAAVIVEPVAGNMNLIAPKPGFMQ
AMRELCSKHGAVLIFDEVMTGFRVGPQCAQGLFGITPDLTTLGKVIGGGMPVAAFGGKREI
MEKIAPLGPVYQAGTLSGNPVAVAAGLVTLKATRAPGFYDSLAARTKQLTDGLTAAAKKHG
VTFCAQSVGGMFGLYFSATPPTSFAEVMQCDKEAFNRFFHAMLEAGH
YLAPSAFEAGFVSAAHTEADIAATIAAAEAIFAKGV**EHHHHHH**

> pQR2199
MGSSHHHHHHSSGLVPRGSHMTDSIRPSSNADWFKAASQHIPGGVNSPVRAFKGVGGTP
VFVTKAQGAYLFDAEGKRYIDYIGSWGPMILGHAHPDVIKAVQDAAADGLSFGAPTPSEVT
VADWICQIMPSMDMVRMTSSGTEACMSAIRLARGYTRRDKIVKFEGCYHGHADSLLVKAG
SGMLTLGVPTSLGVPADLAQHTLTLPFNDIDAVKACFAQYGQQIACVIVEPVAGNMNLVLPI
QGFLETLRSECDQAGSVLIFDEVMTGFRVALGGAQAHYGVTPDLTTLGKIIGAGLPVGAFG
GKRAIMECIAPLGGVYQAGTLSGNPLAMRAGMAMLKLISEPHFYAMLSGKLAYLLGGLKAL
ADEIGIALQTQQAGGMFGIYFTQSTDLTSYEAMTHCDIAAFREFFHGMLKRGVYLAPSAFEA
GFISSAHSQTDLDETLDAARDTLLEMKAGIAQFEG**EHHHHHH**

>pQR2200
MTRLDTHTLQKLDAEHHLHPFNDNAALAKKGTRILTKGEGCYVWDADGNQLLDAFAGLWC
VNIGYGRKELGEVASKQMTQLAYYNSFFQCTTEPTIALAAKLAELAPGDLNHSFFVNSGSEA
NDTILRMVRHFWAVQDQPQKNIFIGRHDGYHGTTMAGASLGGMKGMHKQGGLPIPDIHHI
NPPFWFADGGDLSEDEYGLVAARRLEQKILELGPDRVAAFIGEPIMGAIGVYIPPKTYWPEIE
RICRQHDVLLVADEVICGFGRTGEWFGSQYFGFQPDIMPIAKGITSGYIPLGAAMFNDRVAK
VLKEQGGELAHGATYSGHPVCAAVALENIRILQDEKIVETAKNDIAPYLAQRWAELGEHRLV
GQARIAGMVGALELVPDKGKRAFFPERGTVGPRCRDHALKHGLILRATWDAMLLSPPLIITR
AQVDELFDKTWRALNDTATDLGM**HHHHHH**

> pQR2201
MALTDHLAPLRAHKGQRLTQGLDDATIERLAKGHPDLVAAIEAAAAEHARLQDEFAELLAM
DEAEQLRAVQAGYVNFYADDAINPYIALAARGPWVVTLNGAVLYDAGGYGMLGFGHTPAA
VLEAMARPQVMANIMTPSLSQLRFDRALRNEIGHTRGGCPFAKFLCLNSGSESVGLAARIA
DINSKLMTDPDGRHAGRTIKRIVVKGSFHGRTERPALYSDSSRKSYQQHLASYRGEDSVIAI
PPYDVDALKQAFADAEAKGWFVEAVFLEPVMGEGDPGRSVPPAFYAAARELTRSHGSLFL
VDSIQAGLRAHGVLSIIDYPGFEGLDAPDMETYSKALNAAQYPLSVLAVNERAAGLYRKGVY
GNTMTTNPRALDVACATLAQLTPQVRENIRKRGVEAVQKLQQLQGELGGLITNVQGTGLLF
SCELSPAFKCYGTGSTEEWLRQQGLNVIHGGANSLRFTPHFAMDGEELELLVGMVGRALR
EGPRISQAAAA**HHHHHH**

> pQR2202
MSHVMNTYARLPVAFSHGDGSWVTDTDGRIYLDALSGIAVSTLGHNHPELVAAIAAQAGRL
LHTSNLYRMPQQELLADKLTSLAGMDEVFFCNSGCEANEAAIKLARYYGHQQGVESPAIIV
MEKAFHGRTMATLSATGNRKTQAGFEPLVSGFVRVPYNDMAAIRAIAEHNKSVVAVMLEIV
QGEGGINIADLDYQRALRQLCDENGWLLICDEVQCGMGRTGTWFGFQHAGIRPDIVTLAK
GLGGGVPIGACLTAGKAACLFKPGNHGSTFGGNQLATTAALTTIDVVERDRLIANAESVGEL
IRKELAKALAGLTGVVDIRGQGLMIGIELDRPCGELVARALEAGLLINVTADKVVRLLPALTFS
MDEGRELVARLSLLIRNFLAS**HHHHHH**

> pQR2203
MTSRNEELFARAQKHIPGGVNSPVRAFRSVGGTPLFFQKGAGSQVQDTDGKWYTDYVGS
WGPMILGHAHPQVIAAVQAAVVDGLSFGAPTEREVEIADLLCDMVPSLDMVRLVSSGTEAT
MSAIRLARGFTGRDILVKFEGCYHGHADHLLVKAGSGLLTFGNPSSGGVPAGTAETTMVLT
YNDPQGLAEAFKTHGDKIAAVIVEPVVGNMNLIAPTPEFLKAMRDLTAQYGAVLIFDEVMTG

FRVGLKSAQGLFGITPDLSTFGKVVGGGMPMGAFGGRREIMEKIAPLGPVYQAGTLSGNPI
ATAAGLATLKLIQAPGFHETLTAKTKALCDGLVAAAQKHGVAFSAQNVGGMFGLYFAERCP
GSYDEVLACDKEAFNRFFHAMIEAGHYFAPSAFEAGFVSAAHSDAEIAATVAAADAWFATQ
R**HHHHHH**

> pQR2204
MTITPLMPVYPRCGVRPVRGEGAYLIGDRGERYLDFASGIAVNLLGHGHPHLTKAIQDQAA
TLMHVSNLYGSPQGEAYAARLVENTFADTVFLTNSGAEAVECSIKTARAYHSSAGNAEKHT
LITFNNAFHGRTLGTISATNQEKLRKGFDPLLPGFAYAPFDDINAALDLVDDNTAGFLVEPIQ
GEGGIRPASQPFLQALRDICDKRDLMLIFDEVQCGVARTGHLYAYEHFGVTPDIMASAKGIG
GGFPMGACLATEKAARGMVIGTHGSTYGGNPLACAAGQAVLDVVLEEGFLASVRTTGERL
RGALEQLIPNHDQLFDSVRGVGLMLGLKLSSDSRAFVAHLRDNHGLLTVAAGENVVRVLPP
LNIDDSHIAEFIEKLSAGAASYTPPEA**HHHHHH**

> pQR2205
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DGASAVEIALKQSFHSWRNLGQAQRREFVCLQNGYHGETIGALAVTDVAVFRDAYDPLLM
RAHTVESPDERRGNEAAALAAMRALLAERAEHIAAVIVEPLVQGAAGMVMHGPGYLRGLR
ALTREFGVHLIADEIAVGCGRTGTFFAWEQTEPTGPADWPDFILLSKGITAGTLPLSLVLSSE
AVYRAFWSEDVGRGFLHSHSYTGNALACAAANAVLDRFDAGQAERVRVQAACLATHCAP
LATHPRVRHWRQRGLILAFDVAEAGAGFSERFHLAARRHGLLIRPIGATVYLMPPYLIEDES
AAFLARAVAAALDDVTAKD**HHHHHH**

> pQR2206
MNHDQSHALFARAQQLLPGGVNSPVRAFKSVGGEPFFVQRADGAYLHDVDGNRYIDYVG
SWGPMIVGHNHPAVREAVQAAIQNGLSYGAPCPAEVTMAETITRLVPSCEMVRMVNSGTE
ATLSAIRLARGATGRNRIVKFEGCYHGHGDSFLVKAGSGMLTLGVPTSPGVPAGLSELTLTL
SYNDFEGATALFEQYGSEIACLIIEPVVGNANCLPPREGYLQHLRALCTQHGALLIFDEVMT
GFRVALGGAQAHYGITPDLTTFGKIIGGGMPVGAYGGRRALMQQIAPAGPIYQAGTLSGNP
VAMAAGLAMLELIQARGFHDGLAAATAALCEGMEAAARDAGVPLTTTRVGAMFGLFFTDQ
QVDTYAQAVACDTAAFNRFFHAMLERGVYLAPSAFEAGFMSSAHTPDVIDATISAARDAFK
VVAAG**HHHHHH**

> pQR2207
METPFAAAPAANALMWITQRPQLVFAEGRGSWLVDQQGKRYLDFVQGWAVNCLGHGHP
AIVEALASQAGKLINPSPAFYNEPSLKLAAGLAAHSCFDRVFFASTGAEANEGAIKLARKWG
QKHKGGAHEIITFAGGFHGRTLATMSASGKPGWDTLFAPQVPGFPKAQLNDLDSVAALINE
RTVAIMLEPIQGEGGVVPASVEFLQLLRQICDDRGLLLIVDEVQTGMGRTGKLFAHQHAGIE
PDIMTLGKGIGGGVPLSALLAKESVCCFEAGDQGGTYNGNPLMTAVGVAVLEVLTAPGFLD
EVAAKGEYLGAGLQRLSDRLGLRGERGEGLLRALLLADERGPAIVEAARERGPEGLLLNAP
RPHLLRFMPSLTVSREEIDQMLAWLEELLGA**HHHHHH**

> pQR2208
MTLRNYDMAELKRLDLAHHLPAQASYGLIRDLGGSRIITRAEGSTIWDAEGNAILDGMAGLW
CVDVGYGRAELAEVAREQMLELPYYNTFFRTATPPPVKLAAKIAGLLGGSLQHIFFNSSGSE
SNDTVFRLVRTYWALKGQPERTIFISRRNAYHGSTVAGVSLGGMAAMHAQGGLPIAGIEHV
MQPYAFGEGFGEDPEAFAARAAQEIEDRILAVGPEKVAAFIGEPVQGAGGVIIPPPGYWPR
VDAICRKYGILLVSDEVICGFGRLGEWFGFQKYGYTPDIVSMAKGLSSGYLPISATGVSSEIV
ETLRASGDDFVHGYTYSGHPVAAAVALRNLEIIKREGLVDRVRDDLAPYFAKALATLDDHPL
VGEARSVGLLGAVEIVSEKGTNHRFGGKEGTAGPVVRDHCIAGGLMVRAIRDSIVMCPPYV
ITHDEIDRMVAIIRSALDKAAVDLGGGA**HHHHHH**

> pQR2209
MNHPSALPSAASLMPIATRPDVLFVRGQGAWLFDAAGRRYLDWMQGWAVNCLGHSPQVI
VDAVAQQAATLLNPGPAFHNLPAMRLAEKLTAHSGFDHVFFASSGAEANEGAIKLARKWG
QLHKGGAHEIVTFVDGFHGRTLATMSASGKPGWDTLFAPQVAGFPKARLNDIATVDRLIGP
KTVAVMLEPIQGEAGVIPASGDFLRQLRQLCDERQLLLIVDEVQTGVGRTGHLFAHSAHGIQ
PDIMTLGKGLGGGLPISALLATRAASCFAPGDQGGTYCGNPLVCAAGLAVLDTLLADGFLA
ASRQRGEQLADALRTLSAELGLGAVRGEGFLLALELGADLGPAIATRARDLGLLVNAPRAH
CLRLMPALNTSAAEIAEGIALLRRAIADVQEAQA**HHHHHH**

> pQR2210
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PDLVAALTEQAGKLWHTSNVFYSEPPLRLAEELVTASRFAERVFLCNSGAEANEAAIKLVRK
WAASQGRAPDQRVIVTFRGSFHGRTLAAVTATAQPKYQEGYEPLPGGFRYVDFNDVTQLE
IAMSCGDVAAVMLEPVQGEGGVMPAASGFLRAVRELCDHHGALLVLDEIQAGMGRTGTLF
AHWQDGVVPDIVTLAKALGGGFPIGAMLAGPKVAQAMQFGAHGTTFGGNPLAAAVARVAL
RKLASPQIANNVARQSVALRKGLDAMNAELGLFSQVRGRGLMLGAVLDAKYAGRAGEVLD
LAAAKGLLMLQAGPDVLRFVPSLNITDEEVGEGLSRLHAALRAFAKPG**HHHHHH**

> pQR2211
MSVQHDPVQRADFDQYLVPNYAPAAFVPVRGLGSRVWDQSGRELIDFAGGIAVNALGHC
HPALVKALTEQANTLWHISNVFTNEPTLRLAHKLVDATFAERVFFCNSGAEANEAAFKLARR
VAHDRFGPEKYEIIAALNSFHGRTLFTVSVGGQPKYSDGFGPKIEGITHVPYNDLDALKAAIS
DKTCAVVLEPIQGEGGVLPADKAYLEGARALCDQHNALLVFDEVQSGMGRSGELFTYMHY
GVTPDILSSAKSLGGGFPIGAMLTTTELAKHLAVGTHGTTYGGNPLACAVAEAVLDIVNTPE
VLQGVKAKSEQFKQRLLAIGERYGMFAEVRGLGLLLGCVLNDAWKGKAKAVLDAAAAEGV
LVLQASPDVVRFAPSLVVEEADIVDGLDRFERAVAKLAQG**HHHHHH**

> pQR2212
MADTSPVWHPFTQHGLGDPIPLISHAKDAKLYAADGQSWIDAISSWWVTTHGHAHPRIMAA
IRAQTEKLDQLIFAGWTHEPAESLAAELIRITPAPLTRVFFSDSGSTSVEVALKMALGYWYNI
GEPRSRILVLEHSYHGDTIGTMSVGERGVYNRAWQPLLFDVDTIPFSYEGMEQATLDALEA
ACSAKPAAFIVEPLILGAGGMLIYPAWVLAEMRAICARHGVLFIADEVMTGWGRTGTRFACD
SAGVIPDIVCLSKGLTGGALPLAVTLCIEPIFEAHFSTDRSKTFYHSSSYTANPIACAAANANL
EIWREEPVQQRIDALAEAQAAHLSLLSHDPRVQNPRRLGTIAALDIVVADSGYLSNLAPRLIA
FYRDHGVLLRPLGNTLYVMPPYCITPDELAQVWSAITASLDAV**HHHHHH**

> pQR2213
MISPAAIARVAEREADRFRAANTRAFAHHAAATGWFQSVPFHWMKDWPSPVPIVAASAKD
AALTSIDGQTYDDFCLGDTASLFGHSPPALAAALARQAGEGLSYMLPTGRGAALSERLAVM
FALPQWQVTTTASEANRAVIRWCRGISGRPKILTFNGAYHGAVDDAFVDLKAGAPTMRASL
IGQAHDLCTTTAVIEFNDEDALANALRGGDVACVLAEPVMTNVGMVRDAPGFLATLRRLCD
ETGTLLVFDETHTISSGYGGHSVTHGPAPDLIVIGKSIGGGVPCAIYGFSAVVAERMAALNQ
SRPPGHSGIGTTLSANALAITAMDAMLGEVITSAAYDHMLRGAARLVAGLEQEIAHVGLDW
HVTQVGARVEFLTCPTPPRNGSEAKAAMHLELEAAMHLFLANRGILLAPFHNMMLVSPVTT
DDQIDRLVGAFADSVQALKE**HHHHHH**

> pQR2214
MNYPESIAAQVGTPQGLDNYWLPFTPNRYFREHPKLIAGAEGAYFILSDGRKLFDALSGLW
CCPLGHGNPKIVEALAKQAKALDYATAFQFANPVTLSLAERIATMAPEGLTRVFFANSGSES
VDTALKVAYGYQRLRGEGGRTRFIGREKGYHGVGFGGMSVGGMVANRKMFGPIMVPGVD
HLPHTYNLSQMAFSKGMPTWGAHLAEELERIVALHDASTIAAVIVEPMQGSVGVIAPPVGYL
QKLRDICTKHGILLIFDEVITGFGRMGTNFGSDFFGVTPDIICFAKGVTNGTVPMGGIIVREEI
YQAFMGVNAPEYAVELMHGYTYSGHPLAAAVGHVALDALVNDGLIQRAAELAPVLEDVIHG
LKGEPGVIDIRNVGLAAAVDLEGIPGKVGLRALRTFEAGIEEGLMLRFTADTIAMGPPFISTR
DEIEALGEKLRRAIRKAFSQT**HHHHHH**

> pQR2215
MSATTPDLLSNGQRYYLPVYRPREVILERGQGARVWDSEGREYLDLSAGIAVCGLGHNDP
DLVAALTEQAGKLWHTSNVFYSEPPLRLAEELVSASRFAERVFLCNSGAEANEAAIKLVRK
WATSQGRAPDQRVIVTFRGSFHGRTLAAVTATAQPKYQEGYEPLPAGFRYVDFNDLTQLEI
AMSCGDVAAVMLEPVQGEGGVMPAAPGFLSAVRALCDHHGALLVLDEIQAGMGRTGTLF
AHWQDGVVPDIVTLAKALGGGFPIGAMLAGPKVAEVMQFGAHGTTFGGNPLAAAVARVAL
RKLASPQIANNVARQSAALRKGLDAINAELGLFSQVRGRGLMLGAVLNAKYAGRAGEVLDL
AAAQGLLMLQAGPDVLRFVPSLNITDAEVAEGLKRLHTALKAFAAR**HHHHHH**

> pQR2216
MSHVMNTYARLPVTFSHGCGSRLFDVEGKEYLDALSGIAVSTLGHAHPKLVAALAAQAGR
MLHVSNLYRIAEQEQLADKLCSLSGMQEVFFGNSGAEANEAAIKLARFYGHKKGVELPTVIV
MEKAFHGRTMATLSATANRKAQAGFEPLVSGFVRVPYGDLDAIKAVAEHNKNIVAVMFEIIQ
GEGGIHLVDPAFYRGVRELCDRNEWLMMCDEVQCGMGRTGKWFGFQTAGVQPDVATLA
KGLGSGVPIGACLAGGKAAGLFGPGNHGSTFGGNPLVATAALTTIAVIEEEGLLDNAAKIGV
LIRQGFAEALAGVKGVVEIRGHGLMIGIELERPCGELVGQALAAGLLINVTADTVVRFLPPLN
FTENDARELVDRVAPLIKAFLAG**HHHHHH**

6. Other Substrates

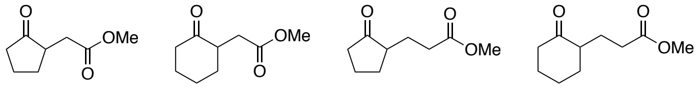

Figure S10. Substrates that showed no activity with any of the enzymes.

7. Analytical Methods

7.1 Achiral methods

Quantitative analysis of enzymatic reactions was analytical reverse phase HPLC analysis with respect of chemical standards using an Agilent 1260 Infinity or Dionex Ultimate 3000 with an Ace 5 C18 column 150 × 4.6 mm. Elution was carried out at 1 mL/min with a linear gradient of acetonitrile/0.1% TFA in water, injection volume 10 μL and column temperature 30 °C. Chromatograms showing retention times included below.

Method A:

Products were detected at 250 nm using a linear gradient 15 - 72% acetonitrile over 15 min. This method was used to detect acetophenone.

Method B:

Products were detected at 204 nm using a linear gradient 5 - 72% acetonitrile over 15 min. This method was used to detect all amine products **18b-25b**.

7.2 Chiral methods

Enantiomeric excess of **18b** and **20b** was determined by GC analysis using an Agilent 7820A GC System with a Supelco Beta Dex 225 capillary GC column 30 m × 250 μm × 0.25 μm with flame ionization detector at 300 °C, a temperature gradient and 1μL injection volume. Chromatograms showing retention times included below.

Method A:

Initial temperature 140 °C, 1 min hold, ramp 5 °C/min to 210 °C, 2 min hold. This method was used to detect **18b**.

Method B:

Initial temperature 150 °C, 1 min hold, ramp 10 °C/min to 210 °C, 3 min hold. This method was used to detect **TFA-20b**

8. HPLC and GC Traces

8.1. HPLC traces

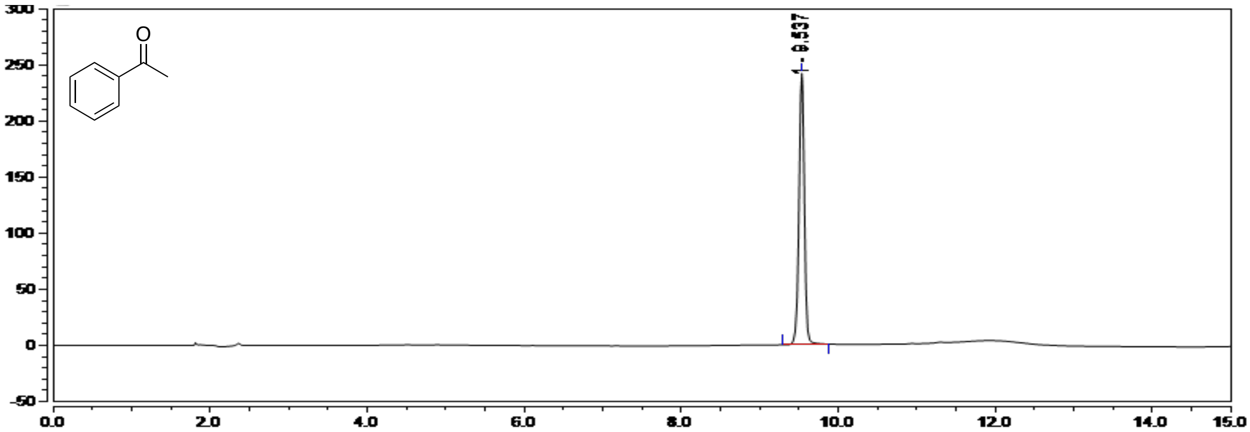

Figure S11. Chromatogram of acetophenone using HPLC method A. Retention time 9.5 min

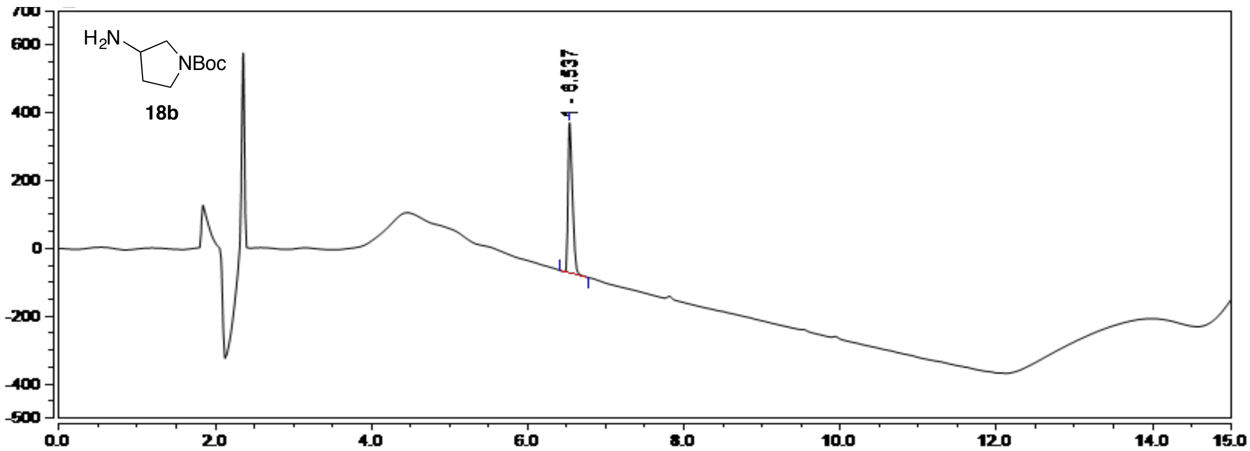

Figure S12. Chromatogram of amine **18b** using HPLC method B. Retention time 6.5 min.

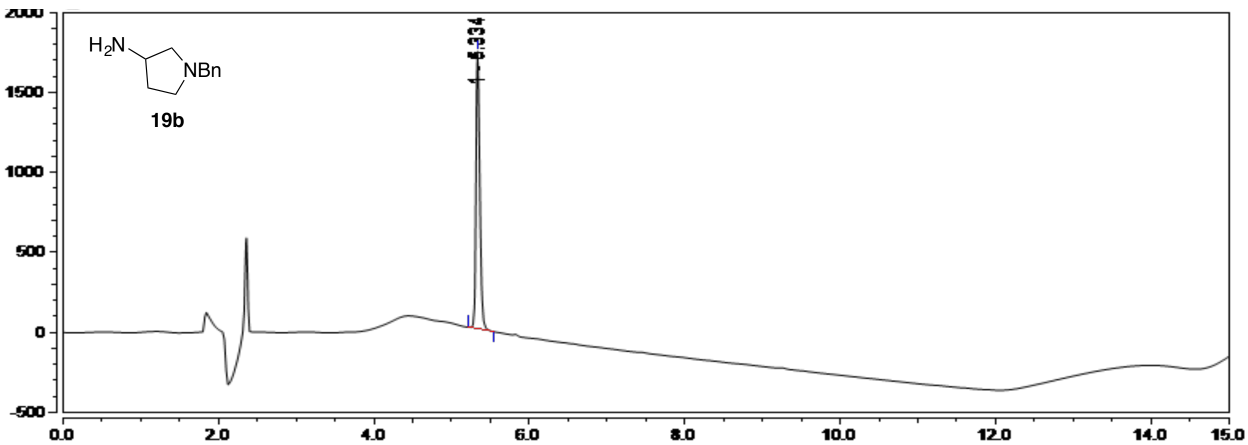

Figure S13. Chromatogram of amine **19b** using HPLC method B. Retention time 5.3 min.

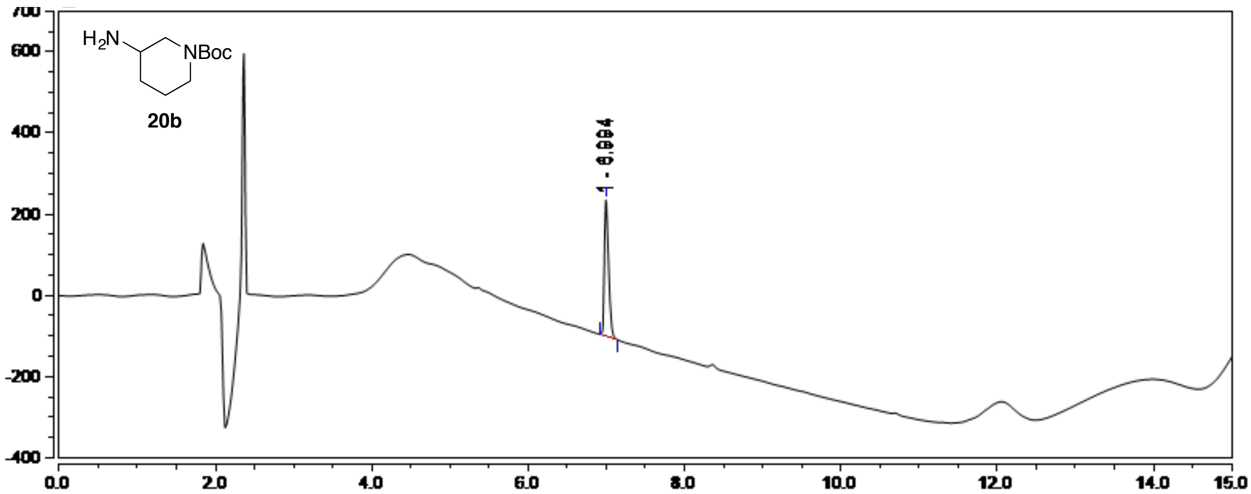

Figure S14. Chromatogram of amine **20b** using HPLC method B. Retention time 7.0 min.

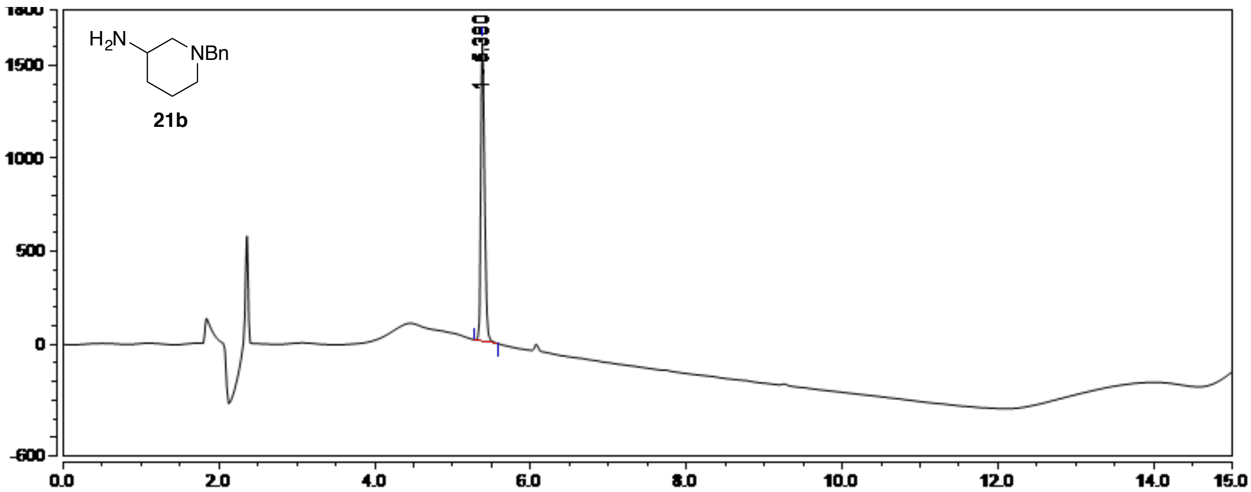

Figure S15. Chromatogram of amine **21b** using HPLC method B. Retention time 5.4 min.

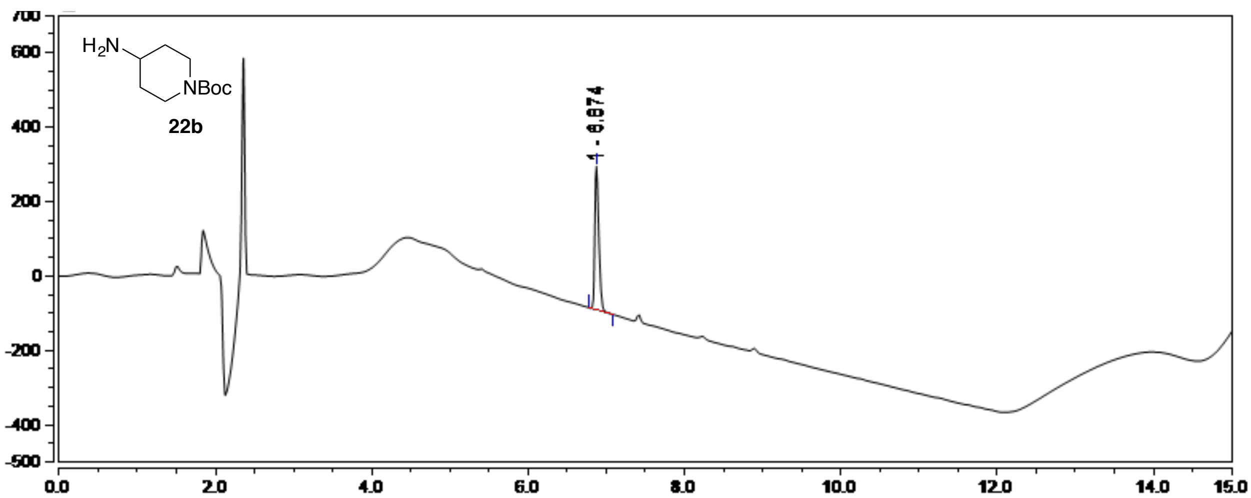

Figure S16. Chromatogram of amine **22b** using HPLC method B. Retention time 6.7 min.

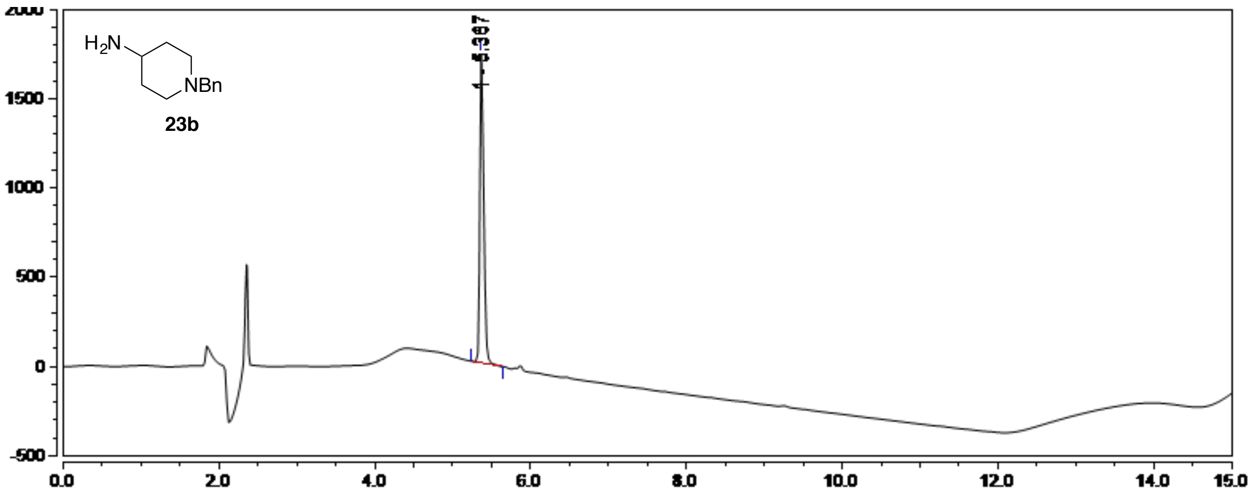

Figure S17. Chromatogram of amine **23b** using HPLC method B. Retention time 5.4 min.

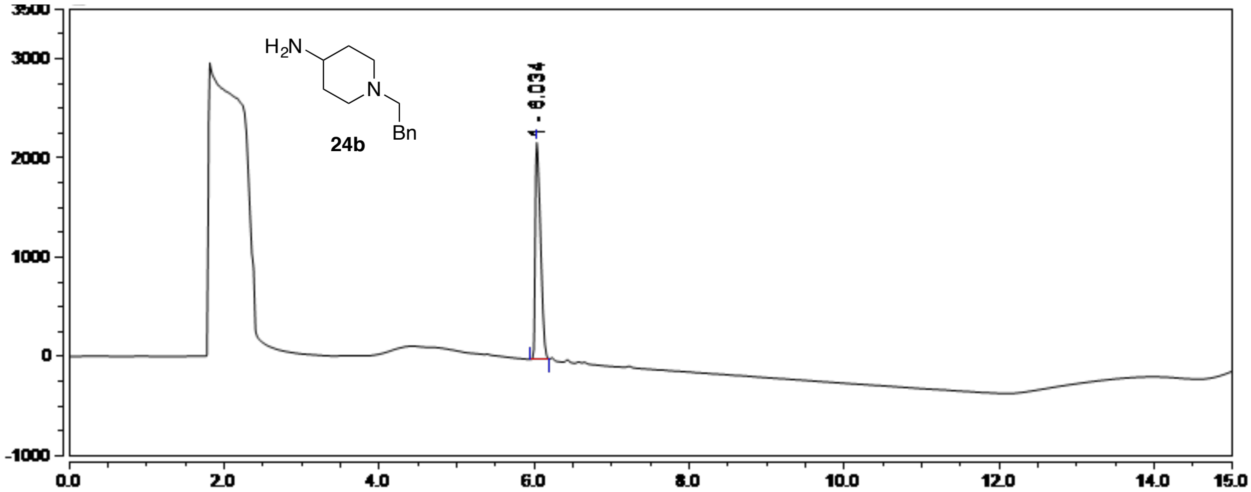

Figure S18. Chromatogram of amine **24b** using HPLC method B. Retention time 6.0 min.

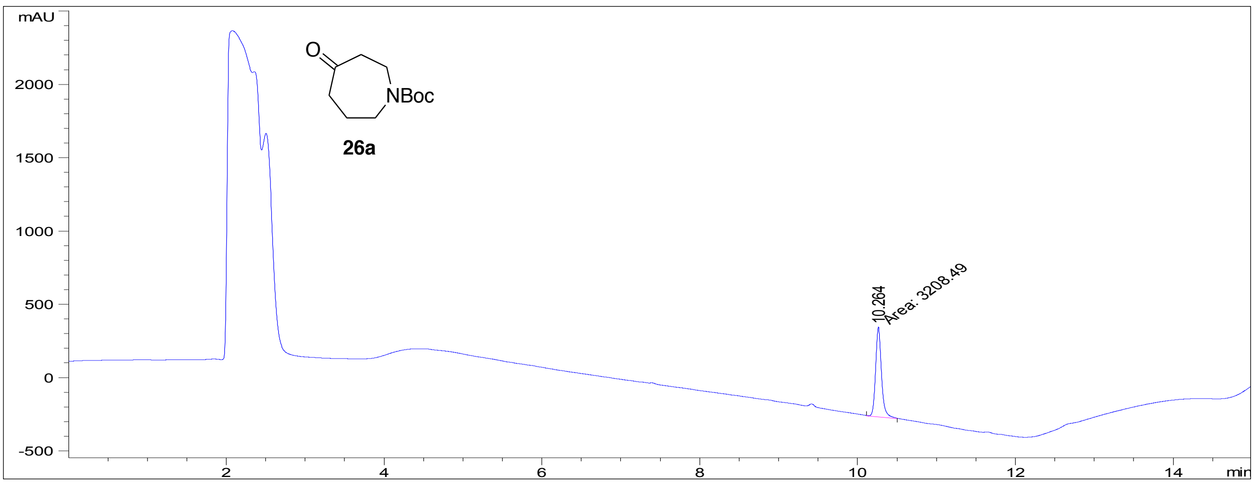

Figure S19. Chromatogram of ketone **26a** using HPLC method B. Retention time 10.3 min.

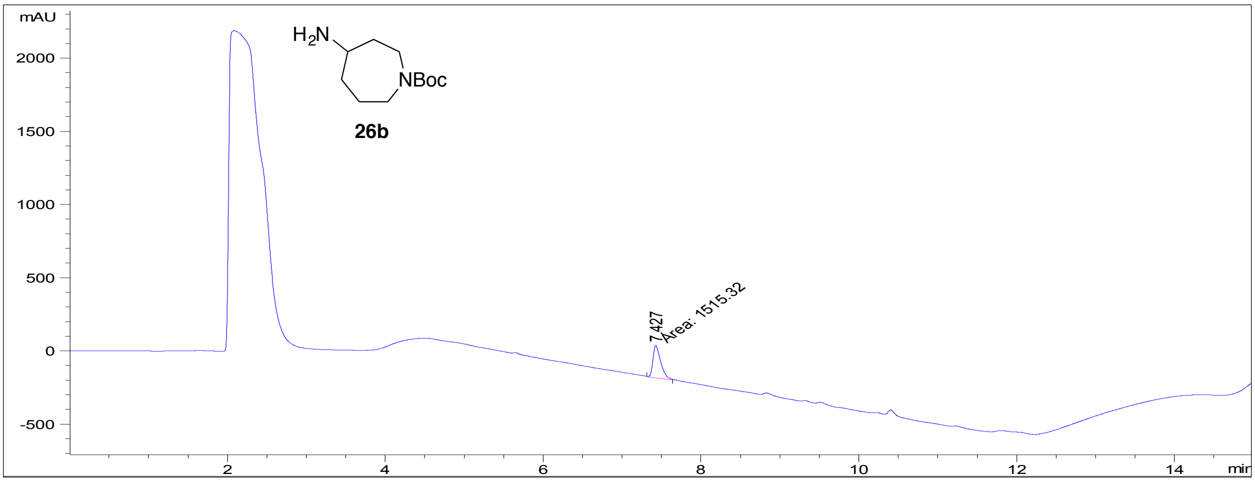

Figure 20. Chromatogram of amine **26b** using HPLC method B. Retention time 7.4 min.

8.2. GC Traces

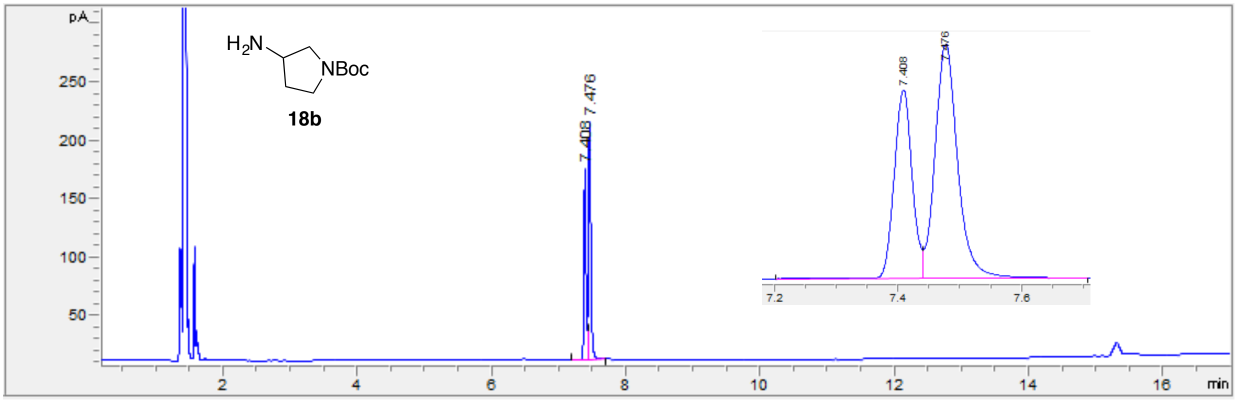

Figure S21. Chromatogram of racemic **18b** using GC method A. Retention times 7.4 &7.5 min.

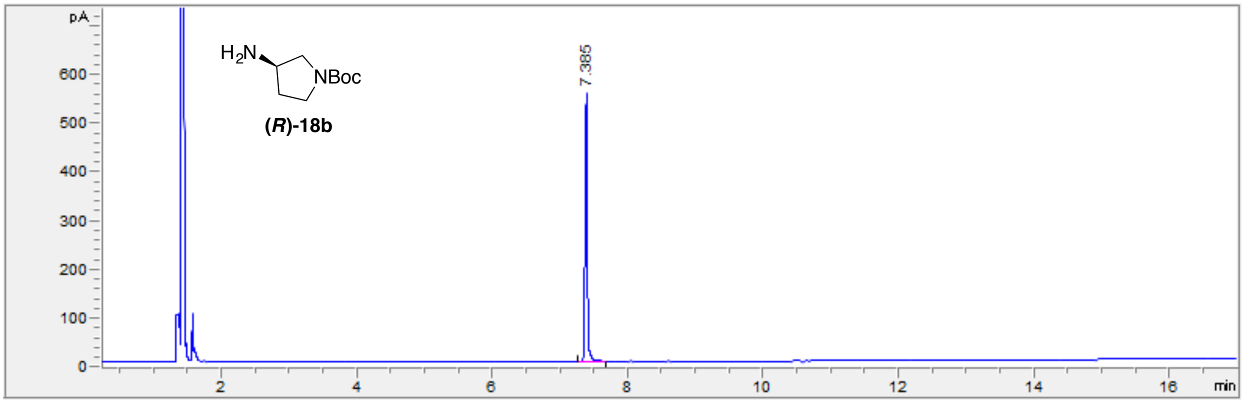

Figure S22. Chromatogram of **(*R*)-18b** using GC method A. Retention times 7.3 min.

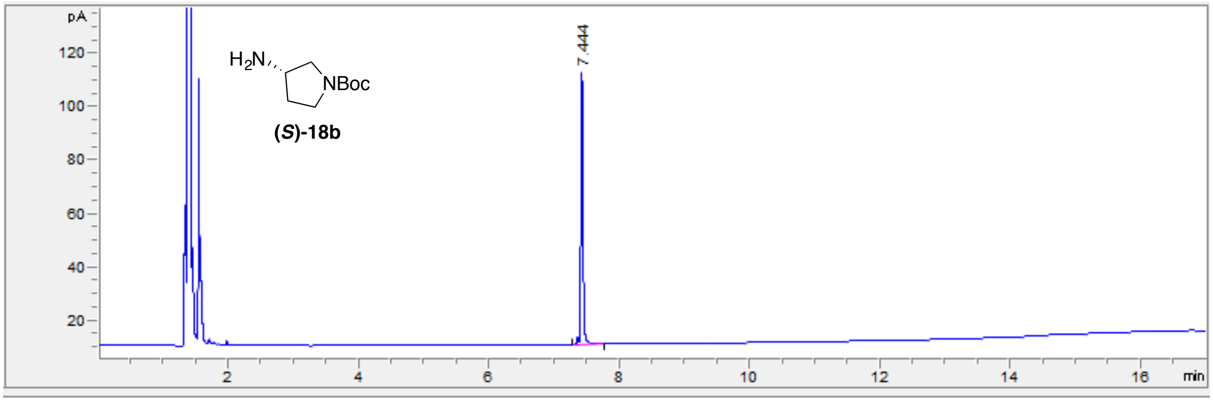

Figure S23. Chromatogram of **(*S*)-18b** using GC method A. Retention times 7.4 min.

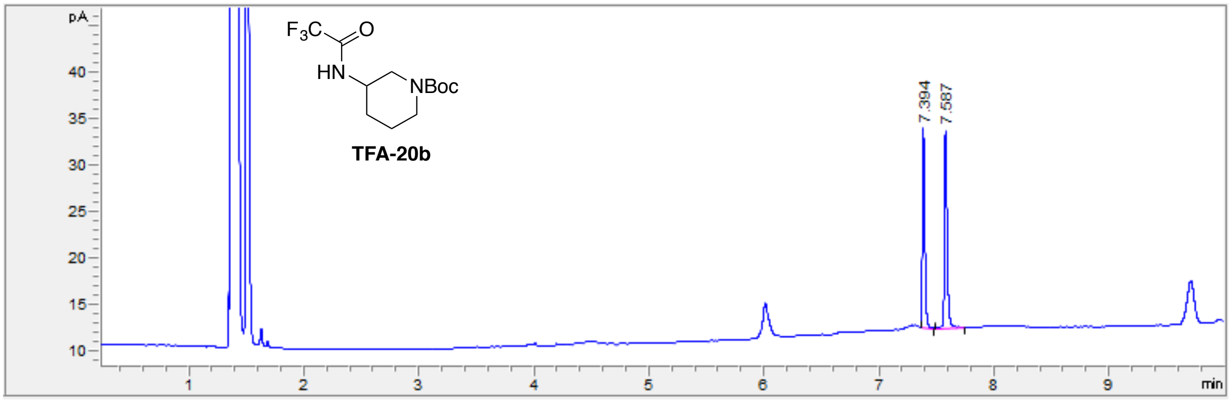

Figure S24. Chromatogram of racemic **TFA-20b** using GC method A. Retention times 7.4 and 7.6 min. Enantioselectivity inferred from **18b**.

9. Calibration Curves

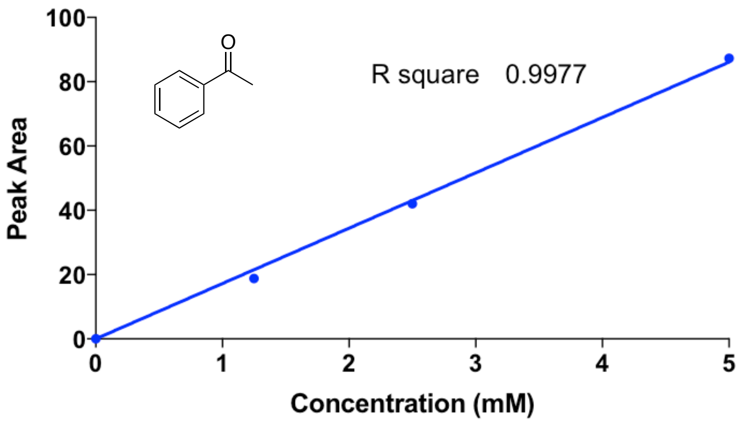

Figure S25. Calibration curve for acetophenone using HPLC method A.

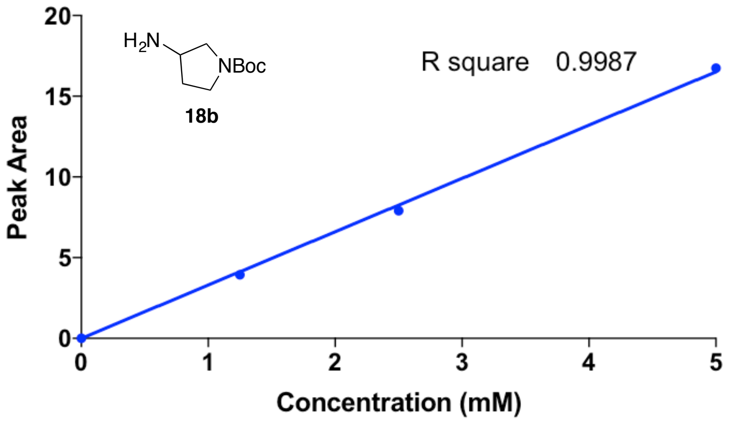

Figure S26. Calibration curve for amine **18b** using HPLC method B.

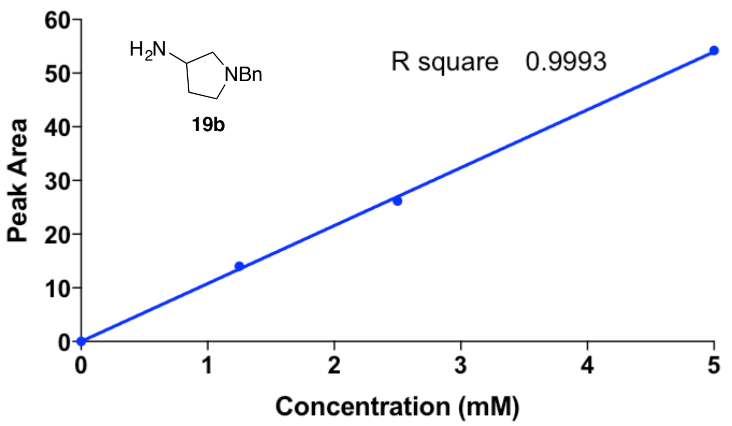

Figure S27. Calibration curve for amine **19b** using HPLC method B.

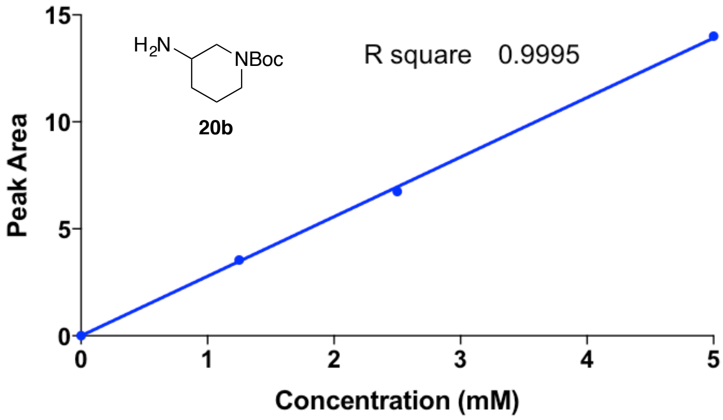

Figure S28. Calibration curve for amine **20b** using HPLC method B.

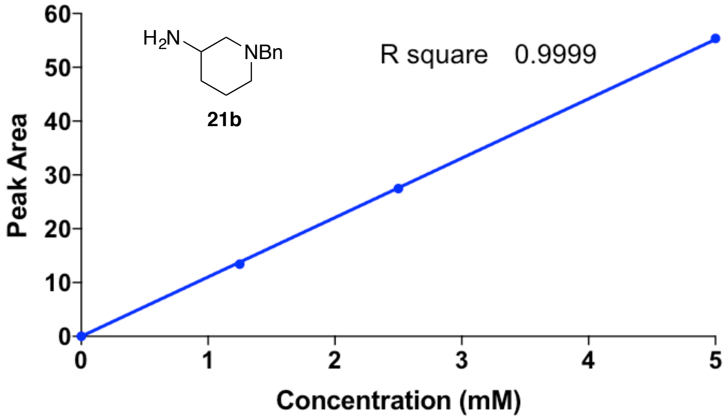

Figure S29. Calibration curve for amine **21b** using HPLC method B.

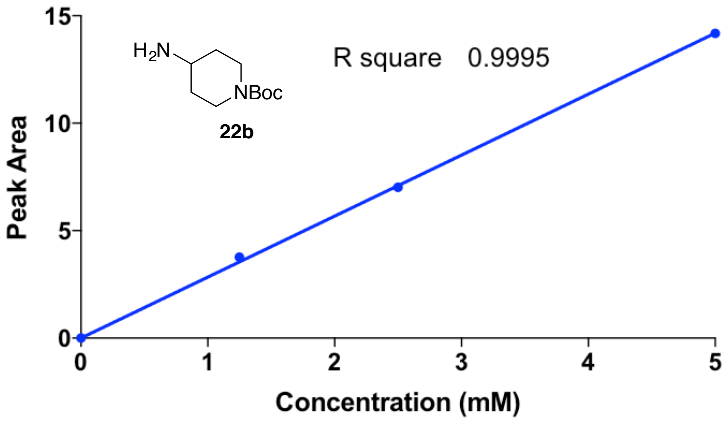

Figure S30. Calibration curve for amine **22b** using HPLC method B.

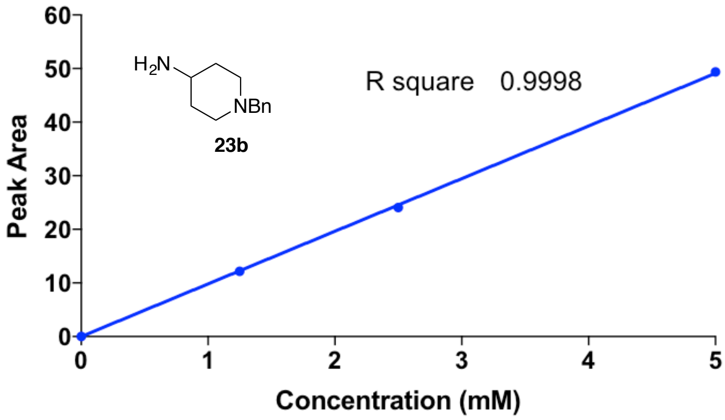

Figure S31. Calibration curve for amine **23b** using HPLC method B.

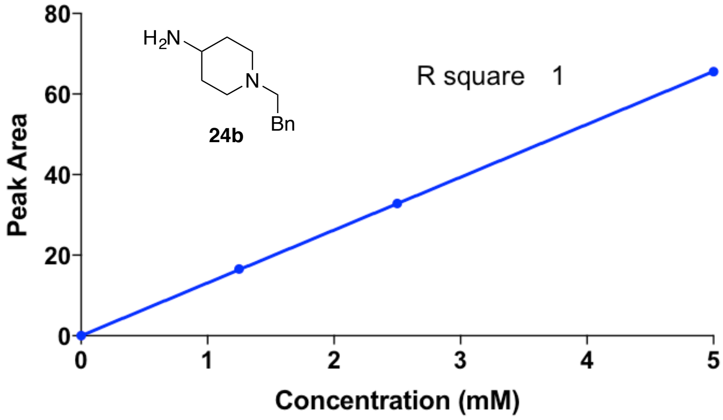

Figure S32. Calibration curve for amine **24b** using HPLC method B.

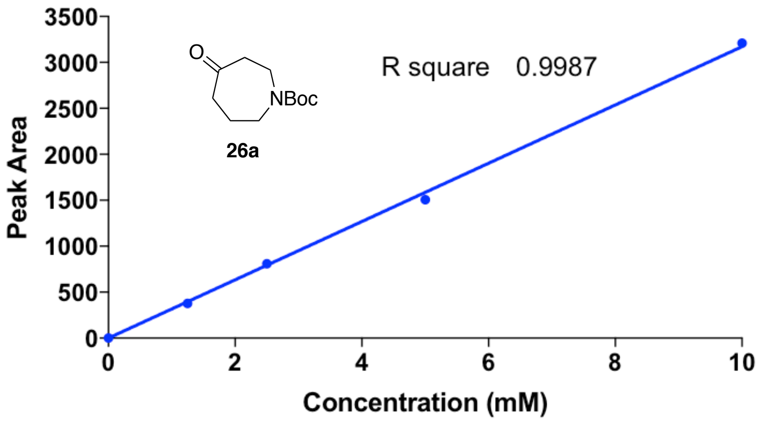

Figure S33. Calibration curve for ketone **26a** using HPLC method B.

10. Purification of pQR2189

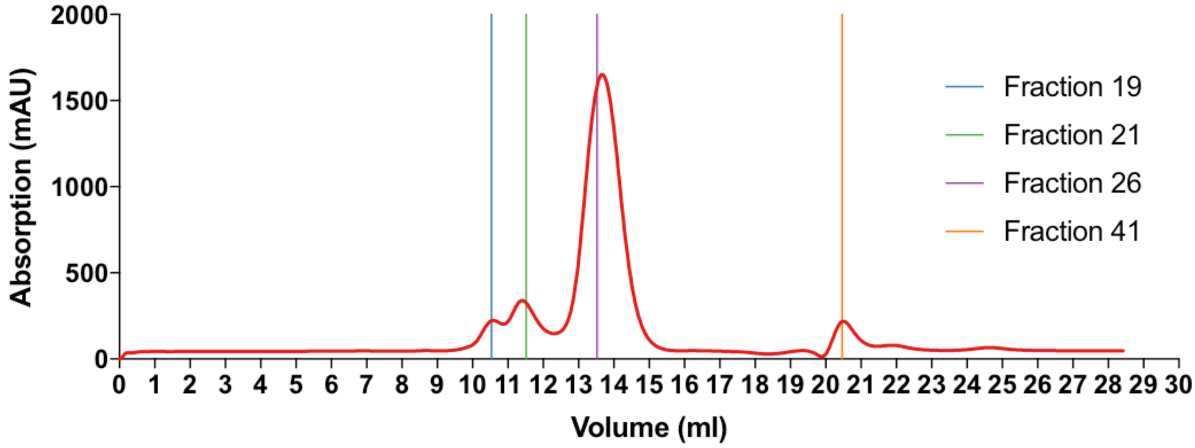

Figure S34. Trace of gel filtration of pQR2189 and His-tag purification. Vertical lines show the fractions that were run on a gel shown in Figure S35.

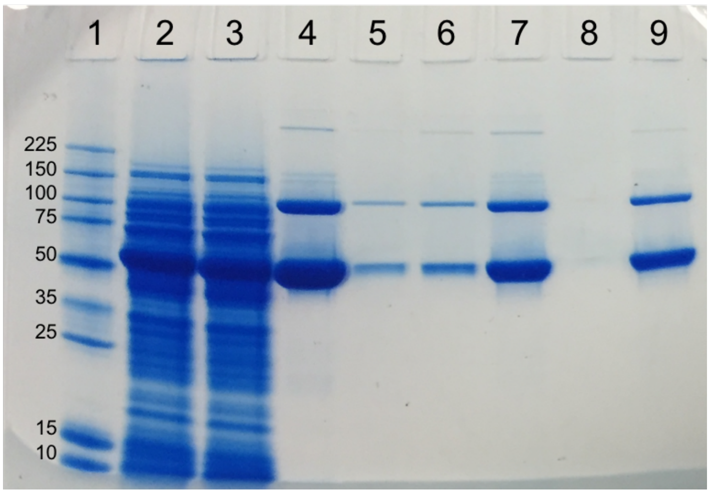

Figure S35. SDS-Page gel of pQR2189 purification. **Lane 1**: Promega broad range protein molecular marker. **Lane 2**: TP fraction. **Lane 3**: CFE fraction. **Lane 4**: pQR2189 after His-tag purification. **Lane 5**: Gel filtration fraction 19. **Lane 6**: Gel filtration fraction 21. **Lane 7**: Gel filtration fraction 26. **Lane 8**: Gel filtration fraction 41. **Lane 9**: Pure protein fractions used in kinetics studies.

11. Kinetics Graphs

11.1. Pyruvate and (*S*)-MBA

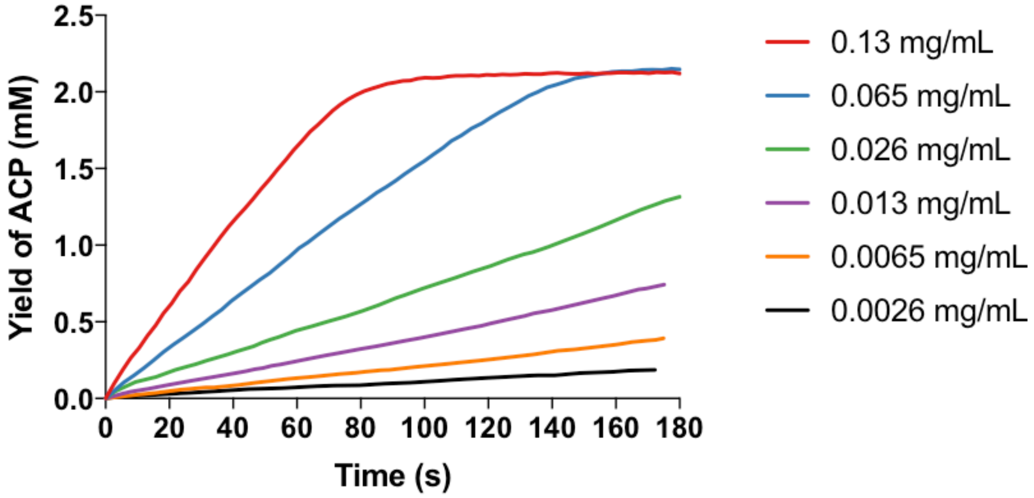

Figure S36. Effect of enzyme concentration on acetophenone formation.[1,2]

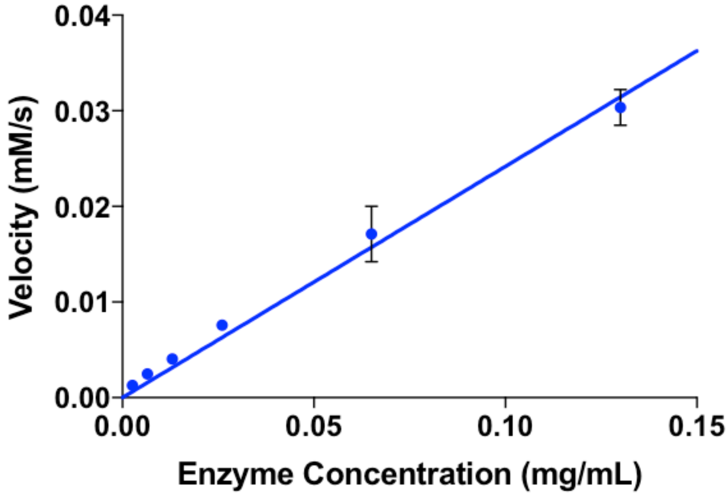

Figure S37. Enzyme concentration vs. velocity.

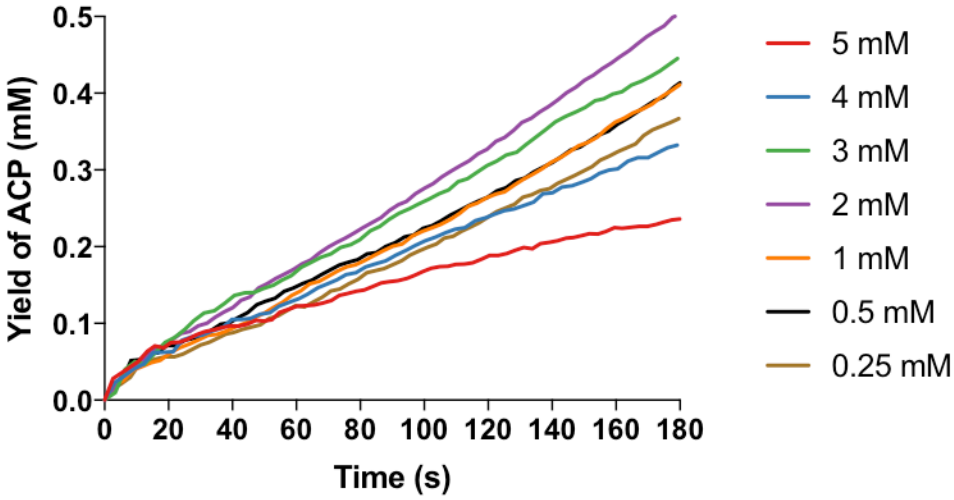

Figure S38. Effect of varying pyruvate concentration on acetophenone formation.[1,2]

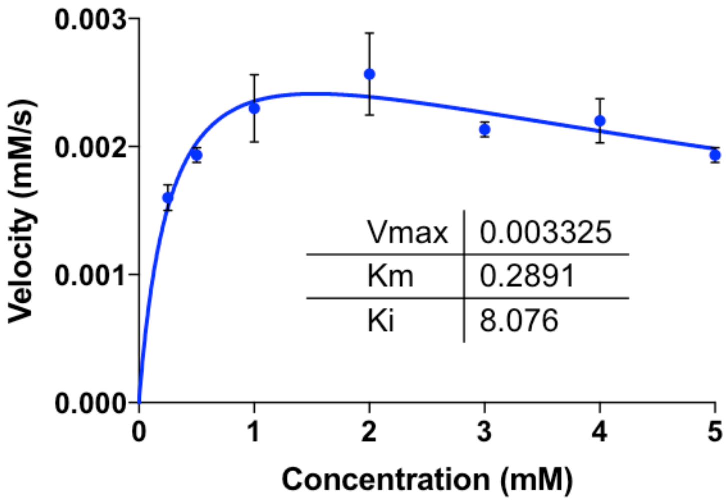

Figure S39. Michaelis-Menten plot of pyruvate concentration, K_m = 0.29 mM.

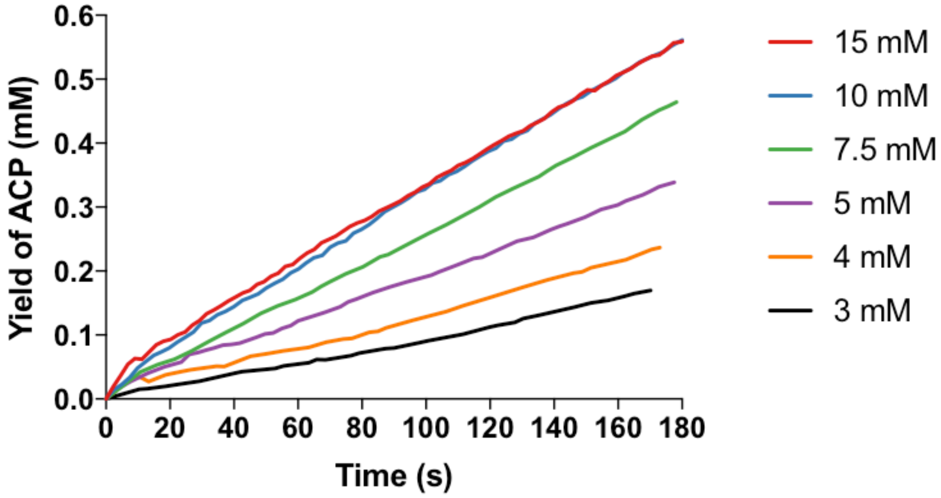

Figure S40. Effect of varying (*S*)-MBA **(*S*)-7** concentration on acetophenone formation.[1,2]

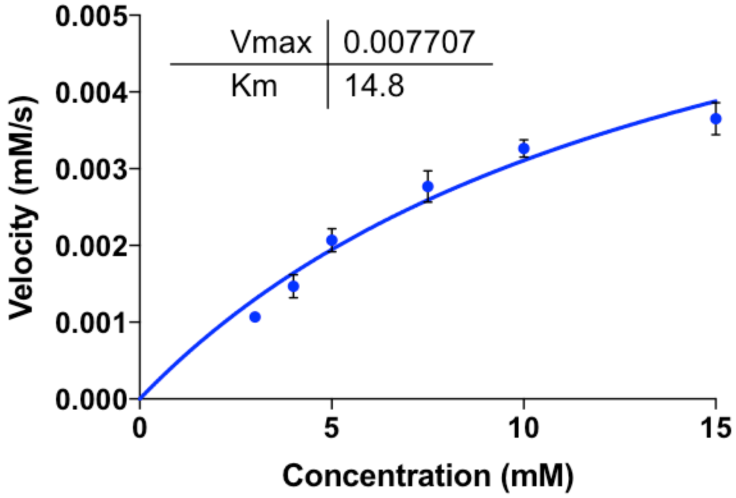

Figure S41. Michaelis-Menten plot of (*S*)-MBA concentration, K_m = 14.8 mM.

11.2. 1-Boc-3-pyrrolidinone and IPA

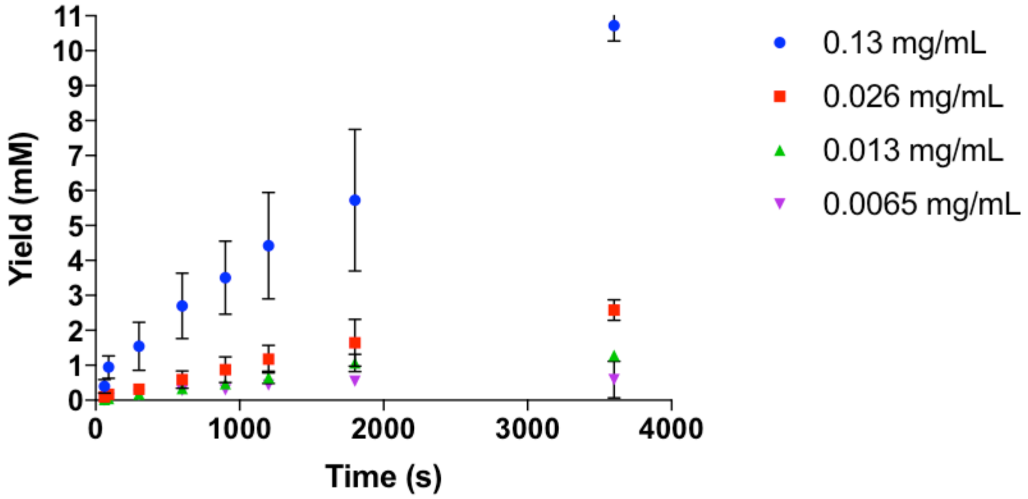

Figure S42. Effect of enzyme concentration on formation of **18b**.[2]

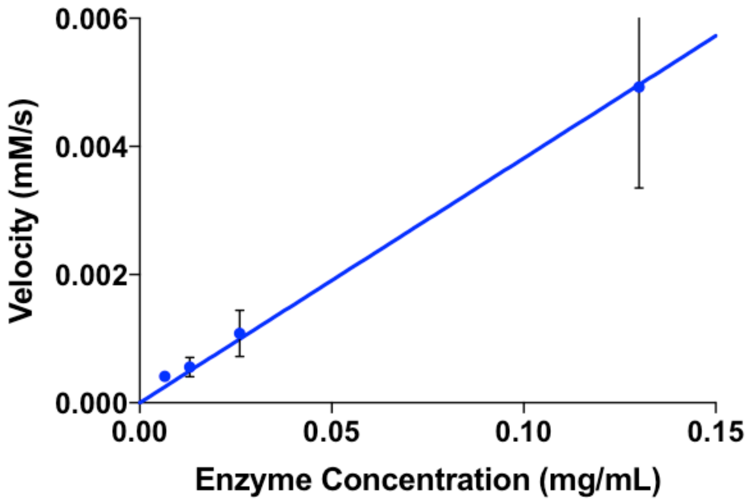

Figure S43. Enzyme concentration vs. velocity.

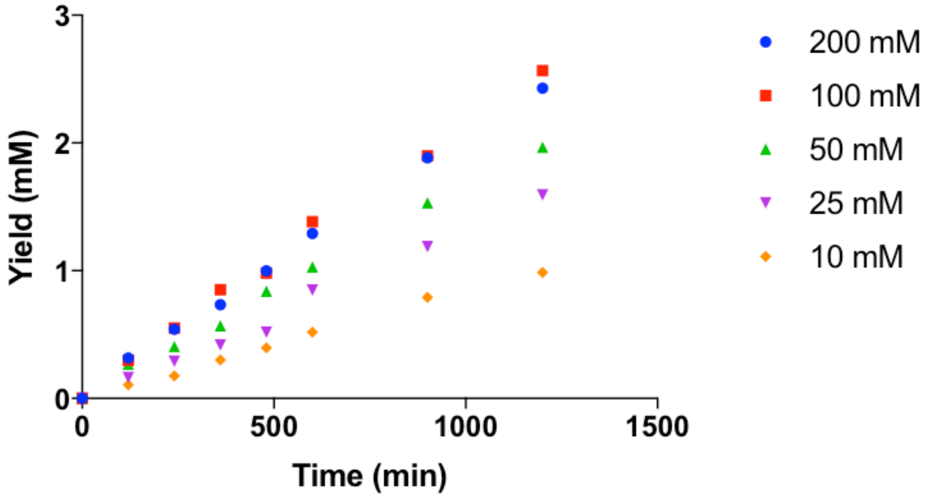

Figure S44. Effect of varying ketone **18a** concentration on formation of **18b**.[2]

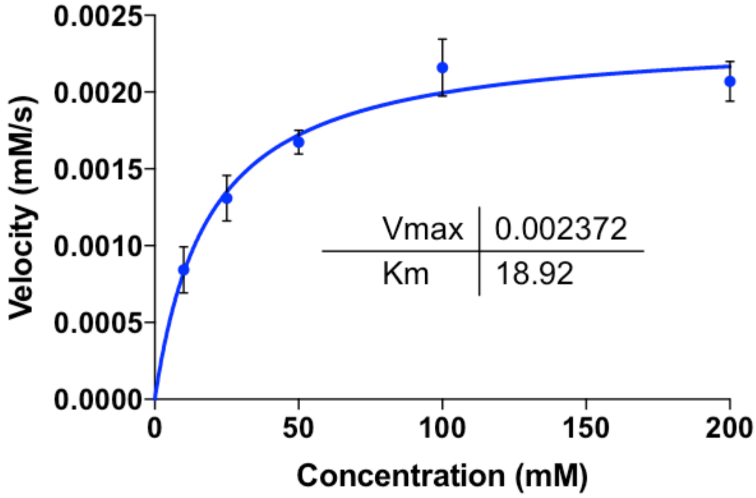

Figure S45. Michaelis-Menten plot of 1-boc-3-pyrrolidinone **18a**, K_m = 18.9 mM.

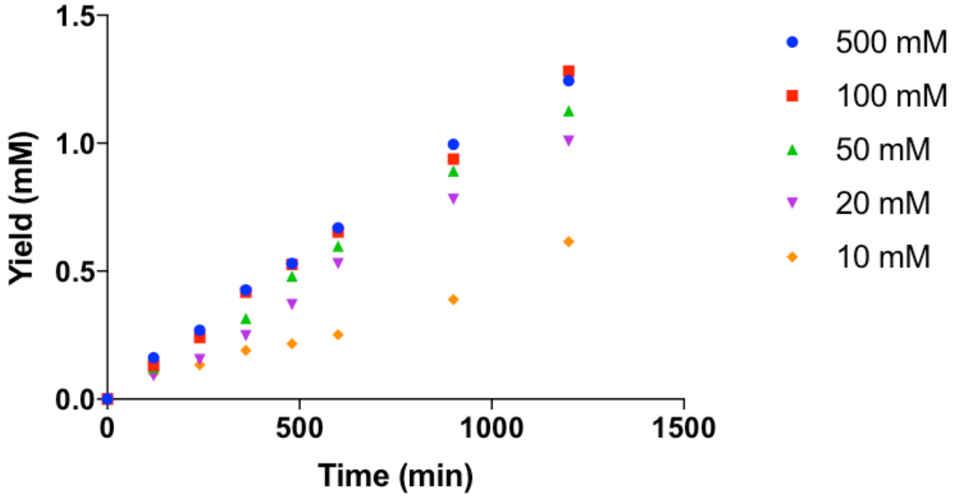

Figure S46. Effect of varying IPA **27** concentration on formation of **18b**.[2]

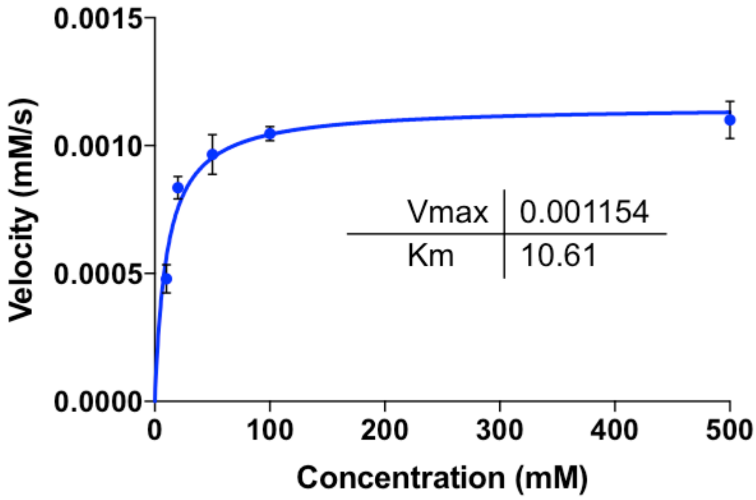

Figure S47. Michaelis-Menten plot of IPA **27**, K_m = 10.6 mM

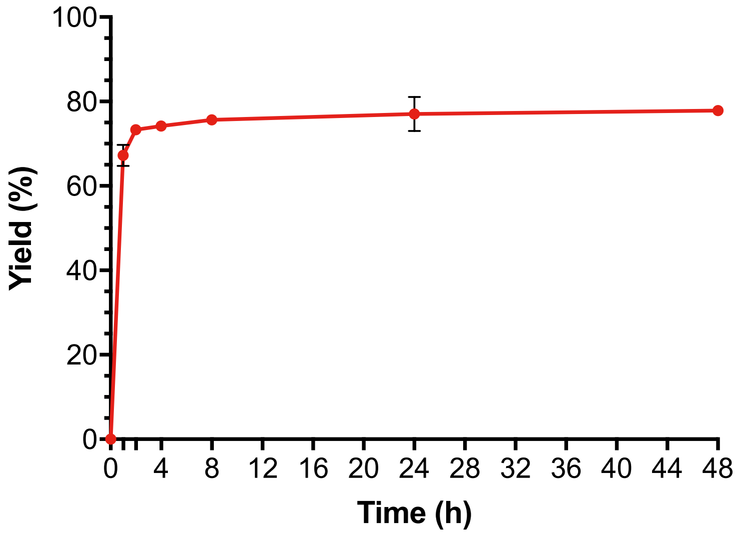

Figure S48. Percentage yield of **18b** using IPA **27** as an amine donor over time for pQR2189 using crude cell lysate.

12. NMR Spectra

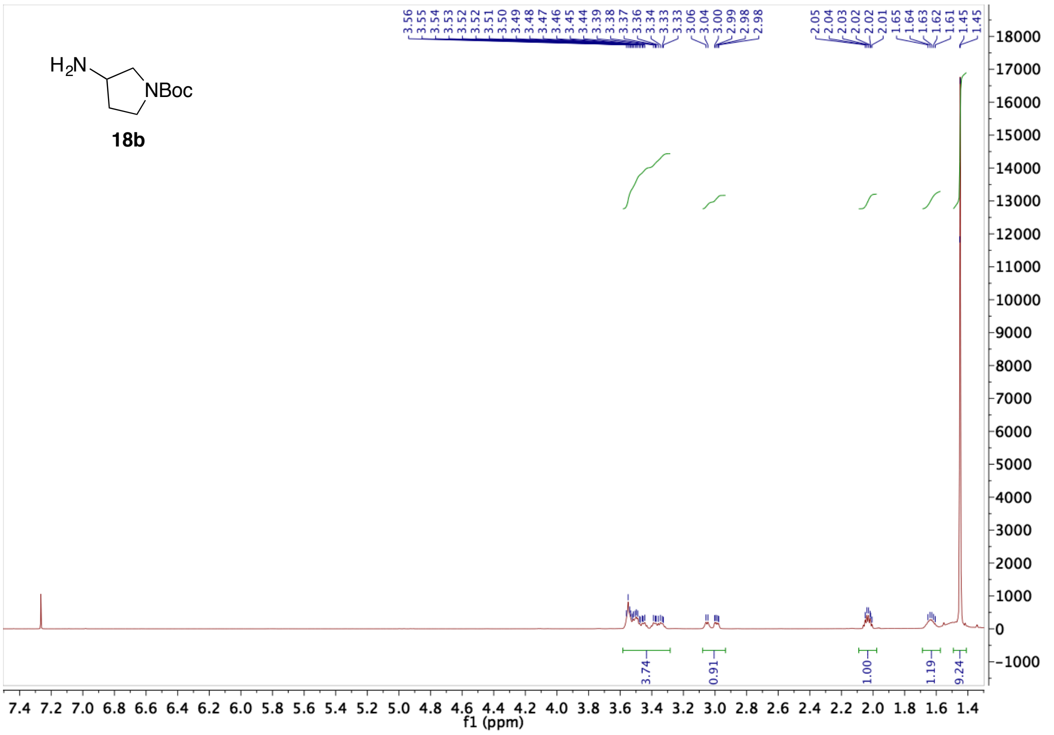

Figure S49. ^{1}H NMR spectrum of amine **18b** in $CDCl_3$.

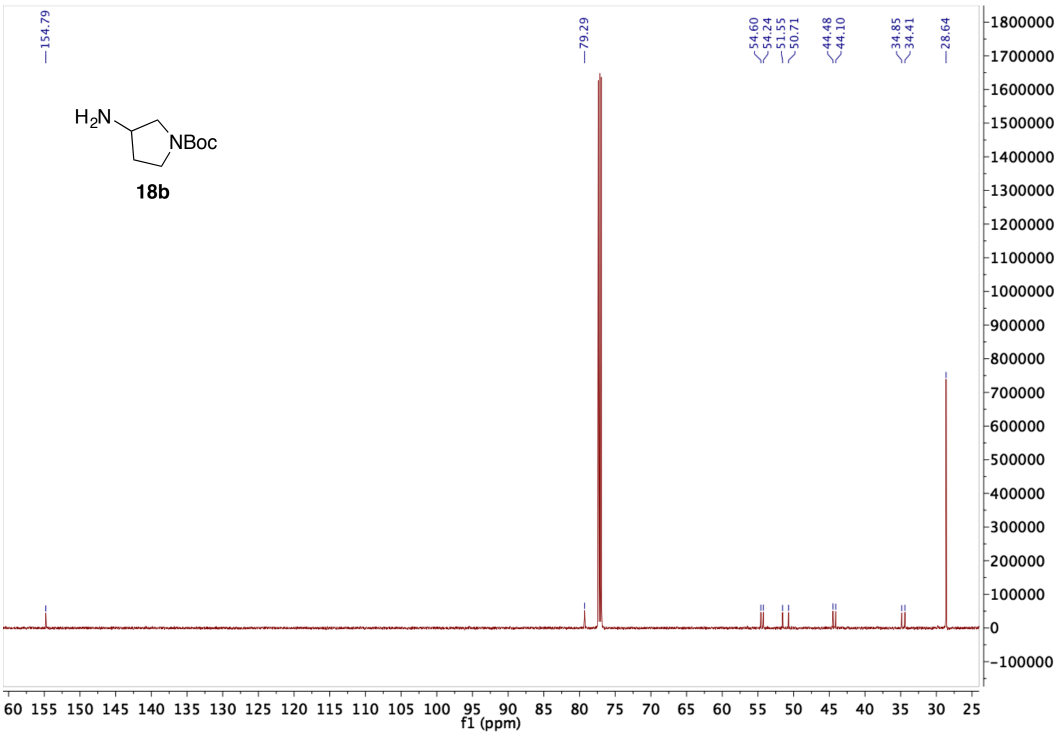

Figure S50. ^{13}C NMR spectrum of amine **18b** in $CDCl_3$.

13. References

1 S. Schätzle, M. Höhne, E. Redestad, K. Robins and U. T. Bornscheuer, *Anal. Chem.*, 2009, **81**, 8244–8248.

2 N. Al-Haque, P. A. Santacoloma, W. Neto, P. Tufvesson, R. Gani and J. M. Woodley, *Biotechnol. Prog.*, 2012, **28**, 1186–1196.