# One Pot 'Click' Reactions: Tandem Enantioselective Biocatalytic Epoxide Ring Opening and [3+2] Azide Alkyne Cycloaddition 

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## Supplementary Information

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## General

All reactions were carried out in oven dried glassware. $\mathrm{CuSO}_{4} \cdot 5 \mathrm{H}_{2} \mathrm{O}$, sodium ascorbate and sodium azide were purchased from Sigma-Aldrich and used as received. ${ }^{1} \mathrm{H}-$ and ${ }^{13} \mathrm{C}$ NMR were recorded on a Varian AMX400 ( 400 and 100.59 MHz ) using $\mathrm{CDCl}_{3}$ as solvent unless otherwise indicated. Chemical shift values are reported in ppm with the solvent resonance as the internal standard $\left(\mathrm{CHCl}_{3}: \delta 7.26\right.$ for ${ }^{1} \mathrm{H}$ and $\delta 77.0$ for $\left.{ }^{13} \mathrm{C}\right)$. Data are reported as follows: chemical shifts, multiplicity ( $\mathrm{s}=$ singlet, $\mathrm{d}=$ doublet, $\mathrm{t}=$ triplet, $\mathrm{q}=$ quartet, $\mathrm{dd}=$ doublet of doublets, $\mathrm{dt}=$ doublet of triplets, $\mathrm{td}=$ triplet of doublets, $\mathrm{m}=$ multiplet, $\mathrm{br}=\mathrm{broad}$ ), coupling constants (Hz), and integration. Flash chromatography was performed on silica gel. All thin layer chromatography was performed on Merck F254 silica gel plates. Visualization of the TLC plates was performed with $\mathrm{KMnO}_{4}$ staining reagent and UV light ( 254 nm ). Mass spectra were recorded on an AEI-MS-902 mass spectrometer by EI ( 70 eV ) measurements. Melting points are uncorrected. MonoPhos was synthesized as described in the literature (1). ${ }^{1} \mathrm{H}$ and ${ }^{13} \mathrm{C}$ NMR data are provided for all synthesized compounds. Spectra were in accordance with published experimental data and references are provided for known compounds. HRMS mass data is provided for all new compounds. Enantioselectivities were determined by HPLC analysis using a Shimadzu LC-10ADVP HPLC equipped with a Shimadzu SPDM10A $V P$ diode array detector.

## Safety

Working with azides should always be done carefully. Organic azides, particularly those of low molecular weight, or with high nitrogen content, are potentially explosive. Heat, light and pressure can cause decomposition of the azides. Furthermore, the azide ion is toxic, and sodium azide should always be handled while protected with gloves. Heavy metal azides are particularly unstable, and may explode if heated or shaken.

## Experimental Information

## General Procedure for Biocatalyzed Azidolysis of Epoxides:

To a 15.0 mL Greiner tube was added 1.0 eq of epoxide ( 0.032 mmol ) and 10.0 mL of 50 mM potassium phosphate buffer ( pH 7.5 ). To this solution was added 0.6 eq of sodium azide followed by 0.5 mL of potassium phosphate buffer containing 25 U (initial activity) of halohydrin dehalogenase. The reaction mixture was shaken overnight and the resulting solution extracted with diethyl ether ( $3 \times 5 \mathrm{~mL}$ ). The organic layers were combined and dried over $\mathrm{MgSO}_{4}$. Solvent was removed under reduced pressure to give the crude reaction products which were analyzed by HPLC.

## General Procedure for One-Pot Enzymatic Click Reactions:

## Copper Catalyzed Procedure

To a 15.0 mL Greiner tube was added $5 \mathrm{~mol} \% \mathrm{CuSO}_{4} \cdot 5 \mathrm{H}_{2} \mathrm{O}$ and $25 \mathrm{~mol} \%$ sodium ascorbate followed by 1.0 mL of 50 mM potassium phosphate buffer. To this solution was added $5.5 \mathrm{~mol} \%$ MonoPhos and the resulting mixture was shaken for 10 min . Sodium azide ( 0.6 eq ) and epoxide ( $1.0 \mathrm{eq}, 0.032 \mathrm{mmol}$ ) were added to the reaction mixture, which was diluted with a further 6 mL of buffer (volume varied depending on the substrate concentration desired). To this solution was added 2.0 eq of alkyne, followed by 0.5 mL of buffer containing 25 U (initial activity) of halohydrin dehalogenase. The Greiner tube was placed on a shaker and after 24 h , the reaction mixture was lipholized overnight. The resulting solid content was flushed over a short plug of silica with ethyl acetate, followed by methanol. Solvent was removed under reduced pressure and the crude reaction mixture was prepared for analysis by HPLC.

## Copper Free Procedure

To a 15.0 mL Greiner tube was added 1.0 eq of epoxide $(0.032 \mathrm{mmol}, 5.0 \mathrm{mg})$ and 7.0 mL of potassium phosphate buffer ( 50 mM ). To this reaction mixture was added 2.0 eq of cyclooctyne (2) followed by 0.5 mL of potassium phosphate buffer containing 25 U (initial activity) of halohydrin dehalogenase. The reaction vessel was sealed with parafilm and shaken for the indicated amount of time ( 24 or 48 h ). The reaction mixture was then lipholized overnight and the resulting solid flushed over silica with ethyl acetate followed by methanol. Solvent was removed under reduced pressure to yield the crude reaction mixture which could be analyzed by HPLC.

## Enzyme Preparation

Halohydrin dehalogenase was expressed in E. coli MC1061 using the pBAD vector $(3,4)$. Transformed cells were streaked on Luria Broth (LB) agar plates containing ampicillin and incubated overnight at $37^{\circ} \mathrm{C}$. A preculture was started by inoculating 100 mL LB containing $50 \mu \mathrm{~g} / \mathrm{ml}$ ampicillin with one colony from the plate. After overnight incubation at $37^{\circ} \mathrm{C}$, the preculture was diluted in 1 L of Terrific Broth (TB) containing $50 \mu \mathrm{~g} / \mathrm{mL}$ ampicillin, 0.5 M sorbitol and $0.02 \%$ arabinose. This main culture was incubated for two days at $37^{\circ} \mathrm{C}$.
The cells were centrifuged ( $10 \mathrm{~min}, 13000 \mathrm{~g}$ ), washed, and resuspended in $5 \mathrm{~mL} / \mathrm{g}$ pellet of TEG buffer ( 10 mM Tris- $\mathrm{SO}_{4} \mathrm{pH}=7.5,1 \mathrm{mM}$ EDTA and $10 \%$ glycerol). Cells were broken by sonication and the extract was centrifuged ( $160,000 \mathrm{~g}, 45 \mathrm{~min}, 4^{\circ} \mathrm{C}$ ). The supernatant was applied on a Q-Sepharose column ( 50 mL , GE Healthcare) (4) and elution was carried out with a gradient of 0 to $0.45 \mathrm{M}\left(\mathrm{NH}_{4}\right)_{2} \mathrm{SO}_{4}$ in TEG. The collected fractions that displayed enzymatic activity were pooled. $\left(\mathrm{NH}_{4}\right)_{2} \mathrm{SO}_{4}$ was added to a concentration of 1.5 M and the protein was applied on a Phenyl-Sepharose column ( 60 mL , GE Healthcare). Elution was carried out with a gradient of 1.5 M to $0 \mathrm{M}\left(\mathrm{NH}_{4}\right)_{2} \mathrm{SO}_{4}$ in TEG. Fractions that displayed enzymatic activity were pooled and concentrated over a 10 kDa filter (Millipore). The enzyme was $97 \%$ pure judged by SDS-PAGE. Purified enzyme was sterilized using a $0.2 \mu \mathrm{~m}$ filter and stored at $-70^{\circ} \mathrm{C}$. Enzyme assay was carried out as previously described using 1-( $p$-nitrophenyl)-2-bromoethanol (5). The concentration was determined to be $25 \mathrm{U} / \mathrm{mL}$ and the specific activity $20 \mathrm{U} / \mathrm{mg}$.

## Buffer Exchange

The enzyme was transferred from Tris-HCI buffer to potassium phosphate buffer ( $\mathrm{pH} 7.5,50 \mathrm{mM}$ ). The Tris-buffer solution was loaded onto a 10 kDa filter (Millipore) in $500 \mu \mathrm{~L}$ fractions and spun down ( $14,000 \mathrm{~g}, 30 \mathrm{~min}, 4^{\circ} \mathrm{C}$ ). After the protein was loaded, the filter was flushed twice with potassium phosphate buffer ( 500 $\mu \mathrm{L}$ each) prior to elution from the filter ( $1,000 \mathrm{~g}, 3 \mathrm{~min}, 4^{\circ} \mathrm{C}$ ).

## Characterization of substrates and reference compounds



2-azido-1-phenylethanol. 2-azido-1-phenylethanol was prepared as described in the literature (6) and NMR data was in accordance with the literature. Enantiomeric excess determined by HPLC (Chiralpak OD 99:1 Heptane: $i$ - $\mathrm{PrOH}: \mathrm{t}_{\mathrm{r}}=42.2,47.0 \mathrm{~min}$ ). ${ }^{1} \mathrm{H}$ NMR ( $400 \mathrm{MHz}, \mathrm{CDCl}_{3}$ ): $\delta 7.26-7.39(\mathrm{~m}, 5 \mathrm{H}), 4.89(\mathrm{dd}, J=4.4,3.6 \mathrm{~Hz}, 1 \mathrm{H}), 3.47(\mathrm{~m}, 2 \mathrm{H})$, 2.40 (br s, 1H).


1-phenyl-2-(4-phenyl-1H-1,2,3-triazol-1-yl)ethanol. To a 10.0 mL roundbottom flask was added $10.9 \mathrm{mg}(0.07 \mathrm{mmol})$ of 2-azido-1-phenylethanol and $15 \mu \mathrm{~L}(0.13 \mathrm{mmol})$ of phenylacetylene. $\mathrm{CuSO}_{4}{ }^{\cdot} 5 \mathrm{H}_{2} \mathrm{O}(0.85 \mathrm{mg}, 0.0034 \mathrm{mmol})$ was weighed into a small sample vial along with Na ascorbate ( $3.37 \mathrm{mg}, 0.017 \mathrm{mmol}$ ) and dissolved in 1.0 mL distilled $\mathrm{H}_{2} \mathrm{O}$. To this solution was added MonoPhos ( 1.34 mg ) and the solution was stirred at room temperature for 15 min . The solution in the sample vial was added to the roundbottom flask (with stirring) and a further 2.0 mL of water and 1.0 mL of DMSO were added to the mixture. The reaction mixture was allowed to stir at room temperature overnight, and in the morning a pale yellow precipitate had formed. 5.0 mL of ice cold distilled water was added to the reaction mixture and the precipitate was filtered off and washed with cold water. The product proved to be sufficiently pure to prevent the need for any further purification steps. Yield $=83 \% . \mathrm{mp} 156-157^{\circ} \mathrm{C}$ Enantiomeric excess determined by HPLC (Chiralcel OD-H 80:20 Heptane: $i$ - $\mathrm{PrOH}: \mathrm{t}_{\mathrm{r}}=18.0,22.2 \mathrm{~min}$ ). ${ }^{1} \mathrm{H}$ NMR ( $400 \mathrm{MHz}, \mathrm{CDCl}_{3}$ ): $\delta 7.78(\mathrm{~s}, 1 \mathrm{H}), 7.73(\mathrm{~d}, J=6.8 \mathrm{~Hz}, 1 \mathrm{H}), 7.32-7.41(\mathrm{~m}, 8 \mathrm{H})$, $5.24(\mathrm{~d}, J=6.8 \mathrm{~Hz}, 1 \mathrm{H}), 4.65(\mathrm{~d}, J=13.6 \mathrm{~Hz}, 1 \mathrm{H}), 4.46(\mathrm{q}, J=8.8 \mathrm{~Hz}, 1 \mathrm{H}), 3.61(\mathrm{~s}, 1 \mathrm{H})$; ${ }^{13} \mathrm{C}$ NMR (100.59 MHz, d6-DMSO): 146.7, 142.9, 131.7, 129.8, 129.1, 128.6, 128.4, 126.9, 125.9, 122.8, 72.3, 57.6. HRMS (EI) for $\mathrm{C}_{16} \mathrm{H}_{15} \mathrm{~N}_{3} \mathrm{O}:(\mathrm{m} / \mathrm{z})=266\left(100 \%, \mathrm{M}+\mathrm{H}^{+}\right)$. Calculated mass: 266.1293, measured mass: 266.1288 .


2-(4-nitrophenyl)oxirane. 2-chloro-1-(4-nitrophenyl)ethanol ( $1.0 \mathrm{mmol}, 201.6 \mathrm{mg}$ ) was dissolved in 3.0 mL MeOH .1 .0 mL of distilled $\mathrm{H}_{2} \mathrm{O}$ was slowly added to the solution with stirring. $\mathrm{K}_{2} \mathrm{CO}_{3}$ was then added to the solution, and the reaction mixture was
allowed to stir at room temperature for a further 3 h . The reaction was monitored by thin layer chromatography ( $4: 1$ pentane:ether). The reaction mixture was poured onto 20 mL of $\mathrm{H}_{2} \mathrm{O}$ and extracted with diethyl ether ( $3 \times 10 \mathrm{~mL}$ ). The organic fractions were collected and dried over $\mathrm{MgSO}_{4}$. Solvent was removed under reduced pressure and the resulting oil was purified by column chromatography (pentane:ether $4: 1$ ). The resulting product was a yellow solid. Yield $=89 \%$. Enantiomeric excess determined by HPLC (Chiralpak AS-H 90:10 Heptane: $i$-PrOH: $\mathrm{t}_{\mathrm{r}}=14.8,18.3 \mathrm{~min}$ ) or (Chiralcel OD-H 80:20 Heptane: $i-\mathrm{PrOH}$ : $\left.\mathrm{t}_{\mathrm{r}}=12.2,12.8 \mathrm{~min}\right) .{ }^{1} \mathrm{H}$ NMR ( $400 \mathrm{MHz}, \mathrm{CDCl}_{3}$ ): $\delta 8.17(\mathrm{~d}, J=11.2 \mathrm{~Hz}, 2 \mathrm{H}), 7.42(\mathrm{~d}, J=$ $11.2 \mathrm{~Hz}, 2 \mathrm{H}), 3.95(\mathrm{~m}, 1 \mathrm{H}), 3.22(\mathrm{~m}, 1 \mathrm{H}), 2.76(\mathrm{~m}, 1 \mathrm{H}) ;{ }^{13} \mathrm{C}$ NMR ( 100.59 MHz , $\left.\mathrm{CDCl}_{3}\right): 147.7,145.2,126.1,123.6,51.6,51.3$.


2-azido-1-(4-nitrophenyl)ethanol. 2-bromo-1-(4-nitrophenyl)ethanol ( $500.0 \mathrm{mg}, 2.25$ mmol ) was dissolved in 75.0 mL DMF in a 200 mL roundbottom flask. $\mathrm{NaN}_{3}(732.0 \mathrm{mg}$, 11.3 mmol ) was added to solution, and the flask equipped with a condenser was heated to $100^{\circ} \mathrm{C}$. The reaction mixture was left overnight with stirring. In the morning, the solution was poured onto 50 mL of brine followed by extraction with diethyl ether ( $3 \times 25 \mathrm{~mL}$ ). The organic layers were collected and dried over $\mathrm{MgSO}_{4}$ and the solvent was removed under reduced pressure. The resulting oil was purified by column chromatography ( $1: 1$ pentane:ether) to give the pure azidoalcohol as a yellow oil. Yield $=57 \%$. Enantiomeric excess determined by HPLC (Chiralcel OD-H 80:20 Heptane: $i$-PrOH: $\mathrm{t}_{\mathrm{r}}=13.0,13.6 \mathrm{~min}$, or, Chiralpak AS-H 90:10 Heptane: $i$-PrOH: $\mathrm{t}_{\mathrm{r}}: 28.6,34.8 \mathrm{~min}$ ) ${ }^{1} \mathrm{H}$ NMR ( 400 MHz , $\mathrm{CDCl}_{3}$ ): $\delta 8.20(\mathrm{~d}, J=12 \mathrm{~Hz}, 2 \mathrm{H}), 7.56(\mathrm{~d}, J=12 \mathrm{~Hz}, 2 \mathrm{H}), 4.99(\mathrm{dd}, J=4.0,3.6 \mathrm{~Hz}, 1 \mathrm{H})$, $3.49(\mathrm{~m}, 2 \mathrm{H}), 3.00(\mathrm{br} \mathrm{s}, 1 \mathrm{H}) ;{ }^{13} \mathrm{C}$ NMR ( $100.59 \mathrm{MHz}, \mathrm{CDCl}_{3}$ ): 147.7, 126.8, 123.9, 123.7, 72.3, 57.6.


1-(4-nitrophenyl)-2-(4-phenyl-1H-1,2,3-triazol-1-yl)ethanol. To a 10.0 mL roundbottom flask was added $46.9 \mathrm{mg}(0.23 \mathrm{mmol})$ of 2-azido-1-(4-nitrophenyl)ethanol and $46.0 \mathrm{mg}(0.05 \mathrm{~mL}, 0.45 \mathrm{mmol})$ of phenylacetylene. $\mathrm{CuSO}_{4}{ }^{-} 5 \mathrm{H}_{2} \mathrm{O}(0.57 \mathrm{mg}, 0.0023$ mmol ) was weighed into a small sample vial along with Na ascorbate ( $2.28 \mathrm{mg}, 0.012$ mmol ) and dissolved in 1.0 mL distilled $\mathrm{H}_{2} \mathrm{O}$. To this solution was added MonoPhos $(0.91 \mathrm{mg}, 0.0025 \mathrm{mmol})$ and the solution was stirred at room temperature for 15 min . The solution in the sample vial was added to the roundbottom flask (with stirring) and a further 2.0 mL of water was added to the mixture. The reaction mixture was allowed to
stir at room temperature for 6 h and the progress of the reaction was monitored by thin layer chromotography ( $2: 1$ pentane:ether). Upon completion of the reaction, the mixture was poured onto ice water and extracted with DCM ( $3 \times 5 \mathrm{~mL}$ ). The organic layers were combined, dried over $\mathrm{MgSO}_{4}$ and the solvent was removed under reduced pressure to yield a yellow solid. The solid was washed with chloroform and water and then dried. Yield $=74 \%$. mp 189-190 ${ }^{\circ} \mathrm{C}$. Enantiomeric excess determined by HPLC (Chiralpak ASH 80:20 Heptane: $i$ - $\mathrm{PrOH}: \mathrm{t}_{\mathrm{r}}=37.1,36.7 \mathrm{~min}$ ). ${ }^{1} \mathrm{H}$ NMR ( $400 \mathrm{MHz}, \mathrm{d} 6$-acetone): $\delta 8.34$ (s, 1H), 8.25 (d, $J=4.8 \mathrm{~Hz}, 2 \mathrm{H}$ ), 7.88 (d, $J=7.6 \mathrm{~Hz}, 2 \mathrm{H}$ ), 7.76 (d, $J=8.8 \mathrm{~Hz}, 2 \mathrm{H}), 7.43$ (t, $J=7.2 \mathrm{~Hz}, 2 \mathrm{H}), 7.34(\mathrm{t}, J=7.6 \mathrm{~Hz}, 1 \mathrm{H}), 5.43(\mathrm{~m}, 2 \mathrm{H}), 4.78(\mathrm{dd}, J=12,3.6 \mathrm{~Hz}, 1 \mathrm{H}), 4.68$ (dd, $J=16,7.2 \mathrm{~Hz}, 1 \mathrm{H}) ;{ }^{13} \mathrm{C}$ NMR ( 100.59 MHz , d6-acetone): $149.5,147.9,147.0,131.7$, 129.0, 127.9, 127.5, 125.5, 123.6, 121.9, 71.8, 56.9. HRMS (EI) for $\mathrm{C}_{16} \mathrm{H}_{14} \mathrm{~N}_{4} \mathrm{O}_{3}:(\mathrm{m} / \mathrm{z})=$ $311\left(100 \%, \mathrm{M}+\mathrm{H}^{+}\right)$. Calculated mass: 311.1144, measured mass: 311.1139 .


2-(4,5,6,7,8,9-hexahydro-1 H -cycloocta[d][1,2,3]triazol-1-yl)-1-(4-nitro phenyl) ethanol. 2-azido-1-(4-nitrophenyl)ethanol ( $10.7 \mathrm{mg}, 0.05 \mathrm{mmol}$ ) was dissolved in 1.0 mL dry THF in a Schlenk vessel under $\mathrm{N}_{2}$ atmosphere. 2.0 eq of cyclooctyne was added and the solution was stirred at room temperature overnight. Progress of the reaction was followed by GC/MS. THF and any remaining cyclooctyne were removed by rotary evaporated and the resulting viscous yellow oil was purified by column chromatography (5:1 pentane: ethyl acetate) to give the product as a yellow oil. Yield $=43 \%$.
Enantiomeric excess determined by HPLC (Chiralcel OD-H 80:20 Heptane: $i-\operatorname{PrOH}: \mathrm{t}_{\mathrm{r}}=$ $15.9,17.2 \mathrm{~min}$ ). ${ }^{1} \mathrm{H}$ NMR ( $400 \mathrm{MHz}, \mathrm{CDCl}_{3}$ ): $\delta 8.23(\mathrm{~d}, J=8.4 \mathrm{~Hz}, 2 \mathrm{H}), 7.58(\mathrm{~d}, J=8.4$ $\mathrm{Hz}, 2 \mathrm{H}), 5.45$ (dd, $J=8.0,3.2 \mathrm{~Hz}, 1 \mathrm{H}), 4.43$ (dd, $J=13.8,3.2 \mathrm{~Hz}, 1 \mathrm{H}), 4.29(J=13.8,8.0$ $\mathrm{Hz}, 1 \mathrm{H}), 2.87(\mathrm{br} \mathrm{m}, 2 \mathrm{H}), 2.66(\mathrm{br} \mathrm{m}, 2 \mathrm{H}), 1.64-1.72(\mathrm{~m}, 4 \mathrm{H}), 1.32-1.45(\mathrm{~m}, 4 \mathrm{H}) ;{ }^{13} \mathrm{C}$ NMR ( $100.59 \mathrm{MHz}, \mathrm{d} 6-\mathrm{CDCl}_{3}$ ): 148.0, 147.6, 127.1, 124.1, 118.7, 113.2, 71.8, 55.4, 27.9, 26.1, 25.9, 24.6, 23.9, 22.2. HRMS (EI) for $\mathrm{C}_{16} \mathrm{H}_{20} \mathrm{~N}_{4} \mathrm{O}_{3}:(\mathrm{m} / \mathrm{z})=317$ ( $100 \%, \mathrm{M}+$ $\mathrm{H}^{+}$). Calculated mass: 317.1608 , measured mass: 317.1608 .


1-(2-hydroxy-2-(4-nitrophenyl)ethyl)-1H-1,2,3-triazole-4-carboxylic acid. 2-azido-1-(4-nitrophenyl)ethanol ( $50.0 \mathrm{mg}, 0.24 \mathrm{mmol}$ ) was added to a 10.0 mL roundbottom flask followed by propiolic acid $(18.5 \mathrm{mg}, 0.26 \mathrm{mmol})$. In a separate vial, $\mathrm{CuSO}_{4}{ }^{5} 5 \mathrm{H}_{2} \mathrm{O}(3.0$
$\mathrm{mg}, 0.012 \mathrm{mmol}$ ) was dissolved in 1.0 mL distilled $\mathrm{H}_{2} \mathrm{O}$ along with Na ascorbate (11.9 $\mathrm{mg}, 0.06 \mathrm{mmol})$. MonoPhos ( 4.7 mg ) was then added to the sample vial along with 0.3 mL DMSO. The solution was stirred at room temperature for 15 minutes. The contents of the sample vial were then added to the roundbottom flask with stirring and a further 3.0 mL of distilled water and 0.5 mL of DMSO were added. Stirring of the reaction mixture was continued stirring at room temperature overnight. The solution was poured onto ice water and extracted with ethyl acetate ( $3 \times 10 \mathrm{~mL}$ ). The organic layers were collected and dried over $\mathrm{MgSO}_{4}$. The solvent was removed under reduced pressure and the resulting yellow oil was purified by column chromatography ( $1: 1$ pentane: ether). The product could also be purified by recrystallization from methanol to give a white solid. Yield= 75 $\%$. mp 163-164 ${ }^{\circ} \mathrm{C}$. Enantiomeric excess determined by HPLC (Chiralpak OD-H 80:20 Heptane: $i$-PrOH: $\mathrm{t}_{\mathrm{r}}=23.9,28.2 \mathrm{~min}$ ). ${ }^{1} \mathrm{H}$ NMR ( $400 \mathrm{MHz}, \mathrm{CD}_{3} \mathrm{OD}$ ): $\delta 8.48(\mathrm{~s}, 1 \mathrm{H}), 8.21$ (d, $J=8.8 \mathrm{~Hz}, 2 \mathrm{H}$ ), 7.64 (d, $J=8.8 \mathrm{~Hz}, 2 \mathrm{H}$ ), 5.26 (dd, $J=4.0,3.6 \mathrm{~Hz}, 1 \mathrm{H}$ ), 4.76 (dd, $J=$ $13.8,3.6 \mathrm{~Hz}, 1 \mathrm{H}), 4.63$ (dd, $J=13.8,8.0 \mathrm{~Hz}, 1 \mathrm{H}) ;{ }^{13} \mathrm{C}$ NMR ( $100.59 \mathrm{MHz}, \mathrm{CD}_{3} \mathrm{OD}$ ): 162.7, 148.7, 147.8, 129.2, 127.9, 127.1, 123.4, 71.1, 56.9. HRMS (EI) for $\mathrm{C}_{11} \mathrm{H}_{10} \mathrm{~N}_{4} \mathrm{O}_{5}$ : $(\mathrm{m} / \mathrm{z})=279\left(100 \%, \mathrm{M}+\mathrm{H}^{+}\right)$. Calculated mass: 279.0729, measured mass: 279.0724.


Ethyl-1-(2-hydroxy-2-(4-nitrophenyl)ethyl)-1 H-1,2,3-triazole-4-carboxylate. 2-azido-1-(4-nitrophenyl)ethanol ( $50.0 \mathrm{mg}, 0.24 \mathrm{mmol}$ ) was added to a 10.0 mL roundbottom flask. In a sample vial, $\mathrm{CuSO}_{4} 5 \mathrm{H}_{2} \mathrm{O}(3.0 \mathrm{mg}, 0.012 \mathrm{mmol})$ and Na ascorbate ( $11.9 \mathrm{mg}, 0.06 \mathrm{mmol}$ ) were dissolved in 1.0 mL of distilled water. To this solution was added 4.7 mg of MonoPhos in 0.3 mL of DMSO. The mixture was stirred at room temperature for 15 min . The solution of copper was then added to the roundbottom vial and a further 3.0 mL of water and 0.5 mL of DMSO were added. To this stirred solution was added dropwise ethyl propiolate ( $47.0 \mathrm{mg}, 48.5 \mu \mathrm{~L}$ ). The yellow solution was allowed to stir at room temperature overnight and in the morning a yellow precipitate had formed. 5.0 mL of ice cold distilled water was added to the solution and the precipitate was filtered off, washed with water and dried. Yield $=73 \% . \mathrm{mp}=152-153^{\circ} \mathrm{C}$. Enantiomeric excess determined by HPLC (Chiralcel OD-H 80:20 Heptane: $i-\mathrm{PrOH}: \mathrm{t}_{\mathrm{r}}=$ $18.9,25.2 \mathrm{~min}) .{ }^{1} \mathrm{H}$ NMR ( 400 MHz , d6-acetone): $\delta 8.52(\mathrm{~s}, 1 \mathrm{H}), 8.24(\mathrm{~d}, J=8.4 \mathrm{~Hz}, 2 \mathrm{H})$, 7.76 (d, $J=8.4 \mathrm{~Hz}, 2 \mathrm{H}), 5.48(\mathrm{~s}, 1 \mathrm{H}), 5.44(\mathrm{~m}, 1 \mathrm{H}), 4.85(\mathrm{dd}, J=14.4,3.6 \mathrm{~Hz}, 1 \mathrm{H}), 4.71$ (dd, $J=16.0,6.0 \mathrm{~Hz}, 1 \mathrm{H}), 4.34(\mathrm{q}, J=7.2 \mathrm{~Hz}, 2 \mathrm{H}), 1.34(\mathrm{t}, J=7.2 \mathrm{~Hz}, 3 \mathrm{H}) ;{ }^{13} \mathrm{C}$ NMR ( 100.59 MHz , d6-acetone): 160.2, 149.1, 129.6, 128.4, 127.5, 123.6, 116.1, 71.4, 60.6, 56.8, 13.9. HRMS (EI) for $\mathrm{C}_{13} \mathrm{H}_{14} \mathrm{~N}_{4} \mathrm{O}_{5}:(\mathrm{m} / \mathrm{z})=307\left(100 \%, \mathrm{M}+\mathrm{H}^{+}\right)$. Calculated mass: 307.1042, measured mass: 307.1037.

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