

Product Data Sheet

Product Name: Palmitic Acid Alkyne

Cat. No.: GC40292

Chemical Properties

Cas. No. 99208-90-9

Chemical Name 15-hexadecynoic acid

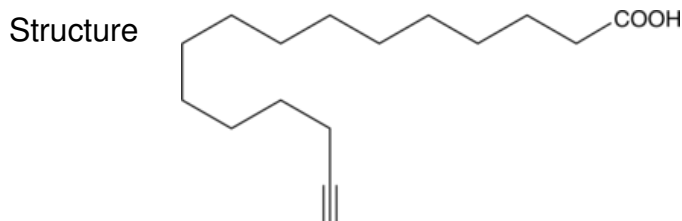
SMILES C#CCCCCCCCCCCCCCC(O)=O

Formula $C_{16}H_{28}O_2$ M.Wt 252.4

Solubility 20mg/mL in DMSO, 30mg/mL in DMF, 30mg/mL in ethanol Storage Store at -20°C

General tips For obtaining a higher solubility, please warm the tube at 37 °C and shake it in the ultrasonic bath for a while. Stock solution can be stored below -20°C for several months.

Shipping Condition Evaluation sample solution: ship with blue ice All other available size: ship with RT, or blue ice upon request.



Protocol

Click palmitoylation assay

1. Seed HEK293T cells, and transfect cells with plasmids (YKT6-WT BioID, YKT6-3E BioID and Wnt3A) in DMEM supplemented with 10% FBS.
2. Dissolve ω -Alkynyl palmitic acid (Palmitic Acid Alkyne) in ethanol to a final concentration of 50mM and stored at -80°C.
3. Dilute Palmitic acid alkyne to a final concentration of 100 μ M in DMEM supplemented with 5% FBS (fatty acid-free) sonicated for 15min at room temperature in a water bath and then allowed to

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precomplex for another 15min.

4.Add Palmitic acid alkyne-containing medium was added and replace partially after 24h.

5.At 72h post-transfection, lysis cells (PBS with 1% Triton x-100, 0.1% SDS, PIC), then centrifuge at 16,000g for 5min at 4°C.

6.Precipitate lysates with Wessel-Flugge Protein precipitation.

7.Dissolve the click labelling reaction [0.1mM biotin-azide, 1mM Tris(2-carboxyethyl)phosphine hydrochloride] in water

8.Incubate with 0.1mM Tris[(1-benzyl-1H-1,2,3-triazol-4-yl)methyl]amine dissolved in DMSO and 1mM CuSO₄ in water by shaking for 2h at 37°C under dark conditions.

9.After the click reaction, precipitate the samples were with 10× methanol overnight at –80°C, then centrifuge and wash again with ice-cold methanol

10.Resuspend the dried pellet in 4% SDS.

11.Precipitate click-biotinylated proteins with High Capacity Neutravidin Agarose Resin .

12.Wash samples with 1% SDS and eluted, then analysis further by immunoblotting.

*This protocol only provides a guideline, and should be modified according to your specific needs

References:

1.Linnemannstöns, Karen et al. “Ykt6-dependent endosomal recycling is required for Wnt secretion in the *Drosophila* wing epithelium.” *Development (Cambridge, England)* vol. 147,15 dev185421. 14 Aug. 2020, doi:10.1242/dev.185421

Background

Palmitic Acid Alkyne is a palmitic acid with an ω-terminal alkyne and a click chemistry reagent acting as an alkyl chain-based PROTAC linker with molecules containing azide groups^[1]. Palmitic acid alkyne is applied in protein detection, conjugative traffic binding, imaging assistive vectors, and PROTAC linkers [2,3,4,5].

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References:

- [1] Honcharenko, Dmytro et al. "New Alkyne and Amine Linkers for Versatile Multiple Conjugation of Oligonucleotides." *ACS omega* vol. 6,1 579-593. 18 Dec. 2020, doi:10.1021/acsomega.0c05075
- [2] Yap, Megan C et al. "Rapid and selective detection of fatty acylated proteins using omega-alkynyl-fatty acids and click chemistry." *Journal of lipid research* vol. 51,6 (2010): 1566-80.
doi:10.1194/jlr.D002790
- [3] García-Cazorla, Yolanda et al. "Conjugation inhibitors compete with palmitic acid for binding to the conjugative traffic ATPase TrwD, providing a mechanism to inhibit bacterial conjugation." *The Journal of biological chemistry* vol. 293,43 (2018): 16923-16930. doi:10.1074/jbc.RA118.004716
- [4] Paiva, Eduardo M, and Florian M Schmidt. "Ultrafast Widefield Mid-Infrared Photothermal Heterodyne Imaging." *Analytical chemistry* vol. 94,41 (2022): 14242-14250.
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- [5] Gao, Xinxin, and Rami N Hannoush. "A Decade of Click Chemistry in Protein Palmitoylation: Impact on Discovery and New Biology." *Cell chemical biology* vol. 25,3 (2018): 236-246.
doi:10.1016/j.chembiol.2017.12.002

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