Supporting Information

for

On-Resin Acetamidomethyl (Acm) Removal and Disulfide Formation in Cysteinyl Peptides Using *N*-Chlorosuccinimide (NCS) in the Presence of Other Cys-Protecting Groups

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HPLC Data:

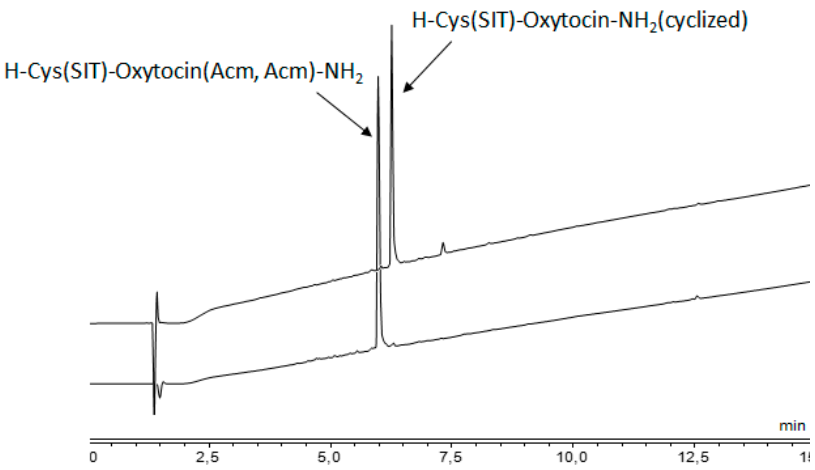


Figure S1. HPLC chromatograms of H-Cys(SIT)-oxytocin[1,6-Cys(Acm)]-NH_2 and H-Cys(SIT)-oxytocin-NH_2 obtained after NCS treatments.

LCMS Data:

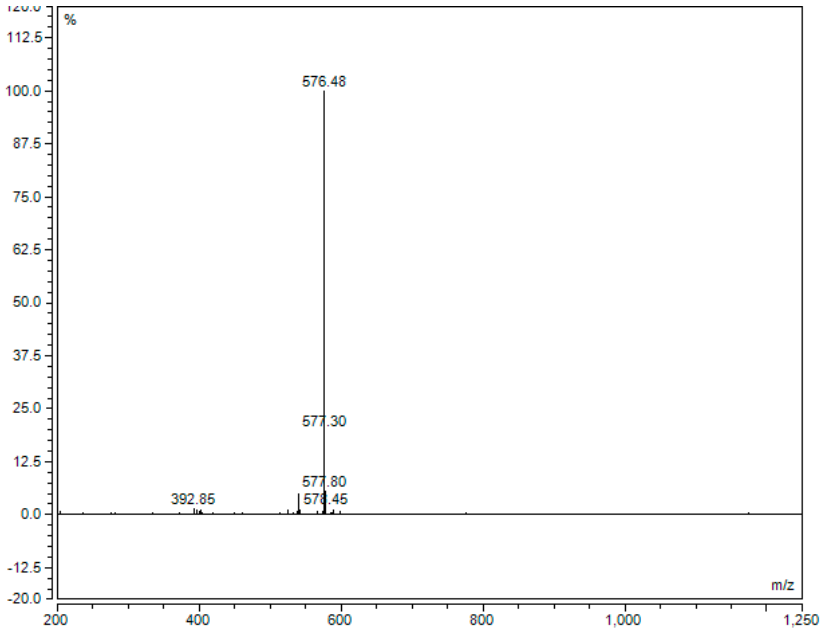


Figure S2. LCMS of H-oxytocin(Acm, Acm)-NH_2; m/z calculated as 576.27 ($M+2H^+/2$).

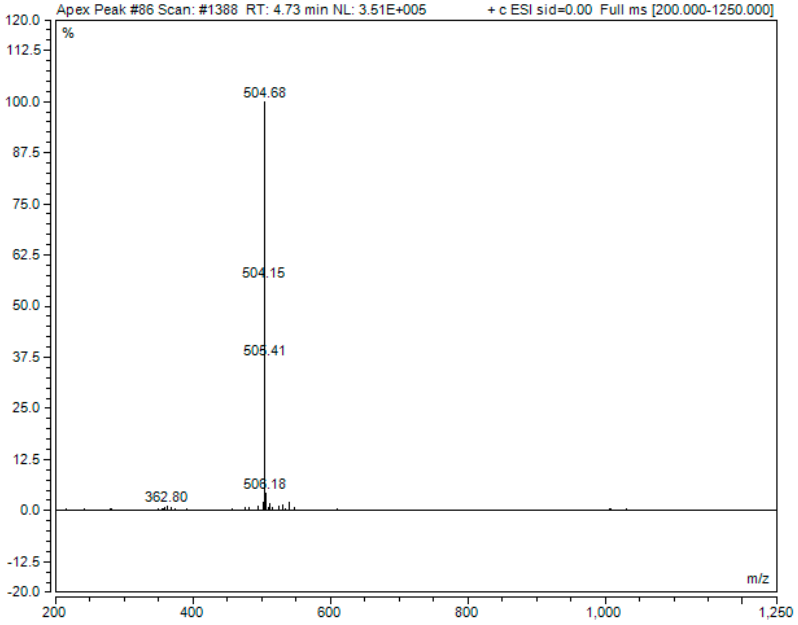


Figure S3. LCMS of cyclized H-oxytocin-NH_2; m/z calculated as 504.23 ($M+2H^+/2$).

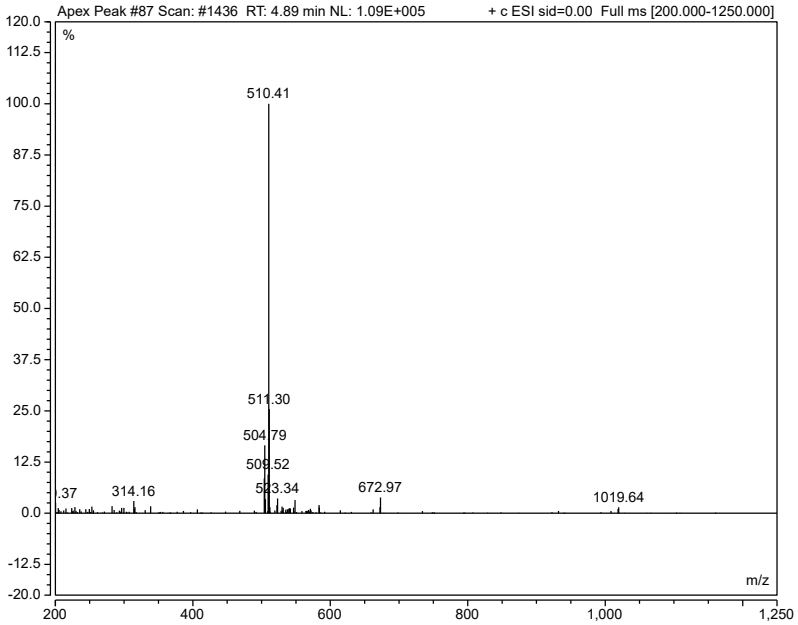


Figure S4. LCMS of the major side product during cyclization of the protected H-oxytocin-NH resin.

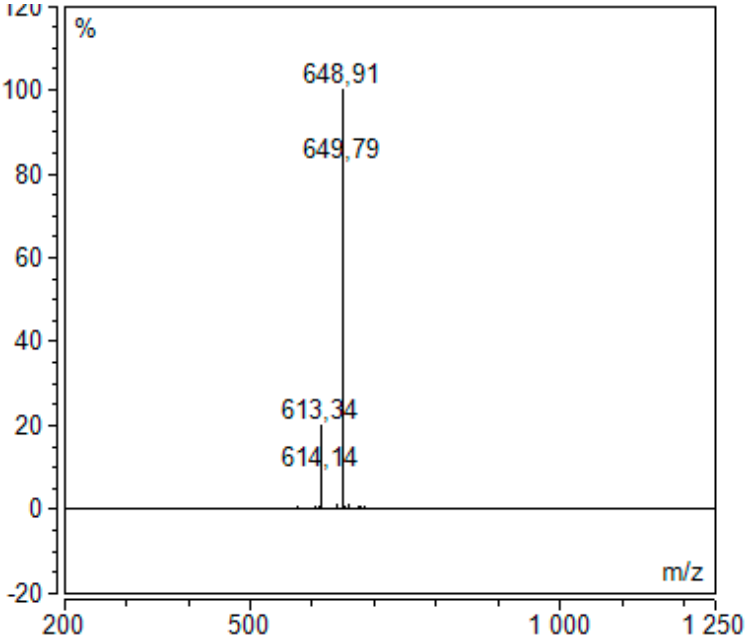


Figure S5. LCMS of Ac-Cys-oxytocin(Acm, Acm)-NH_2 obtained after cleaving Ac-Cys(Dpm)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin; m/z calculated as 648.78 ($M+2H^+/2$).

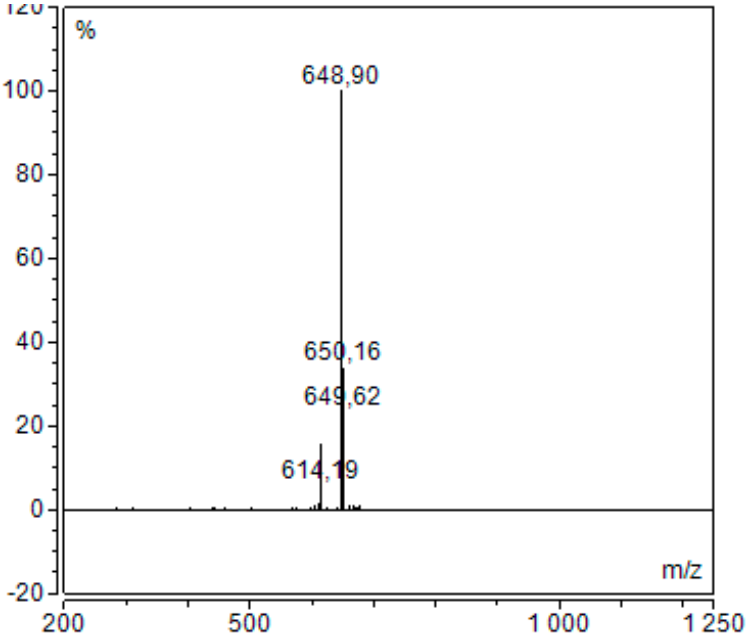


Figure S6. LCMS of Ac-Cys-oxytocin(Acm, Acm)-NH_2 obtained after cleaving Ac-Cys(Mmt)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin; m/z calculated as 648.78 ($M+2H^+/2$).

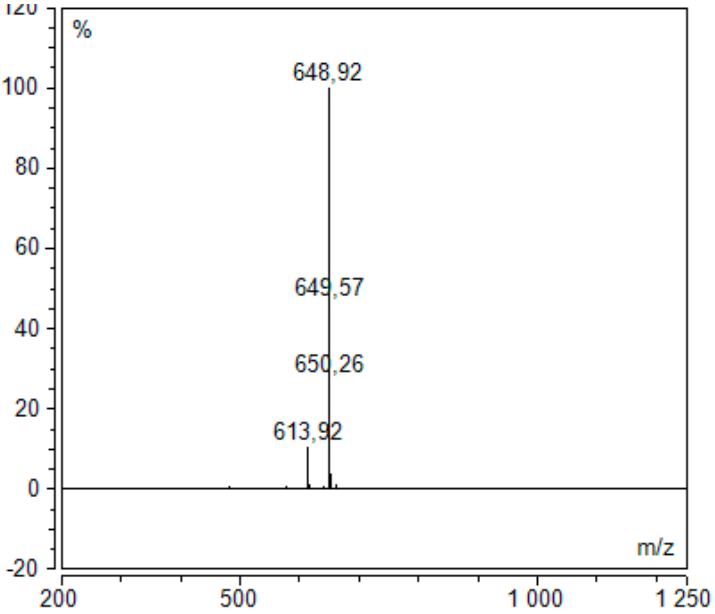


Figure S7. LCMS of Ac-Cys-oxytocin(Acm, Acm)-NH_2 obtained after cleaving Ac-Cys(Thp)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin; m/z calculated as 648.78 ($M+2H^+/2$).

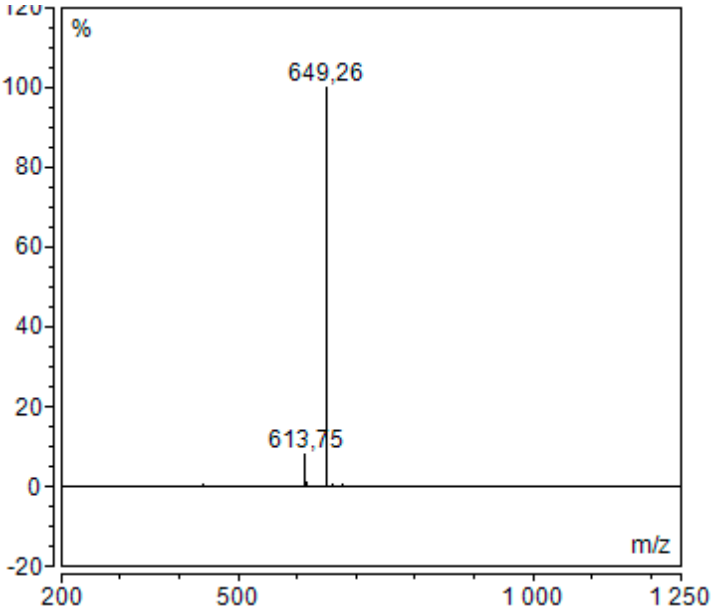


Figure S8. LCMS of Ac-Cys-oxytocin(Acm, Acm)-NH_2 obtained after cleaving Ac-Cys(Trt)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin; m/z calculated as 648.78 ($M+2H^+/2$).

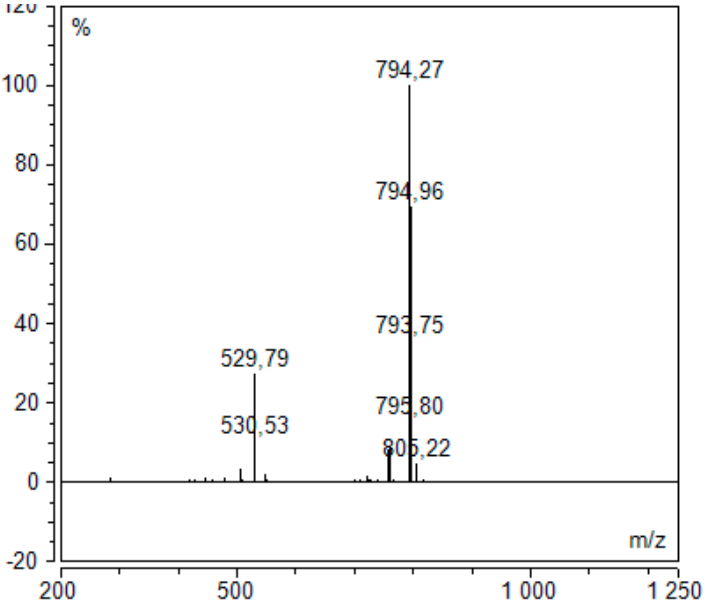


Figure S9. LCMS of Ac-Cys(Msbh)-oxytocin(Acm, Acm)-NH_2 obtained after cleaving Ac-Cys(Msbh)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin; m/z calculated as 793.8 ($M+2H^+/2$), 529.54 ($M+3H^+/3$).

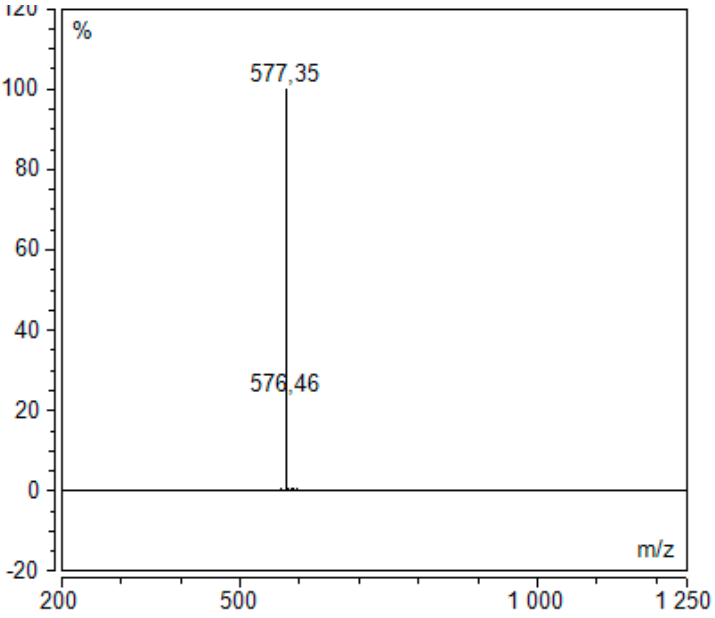


Figure S10. LCMS of cyclized Ac-Cys-oxytocin-NH_2 obtained after on-resin cyclization of Ac-Cys(Dpm)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS; m/z as calculated 576.74 ($M+2H^+/2$).

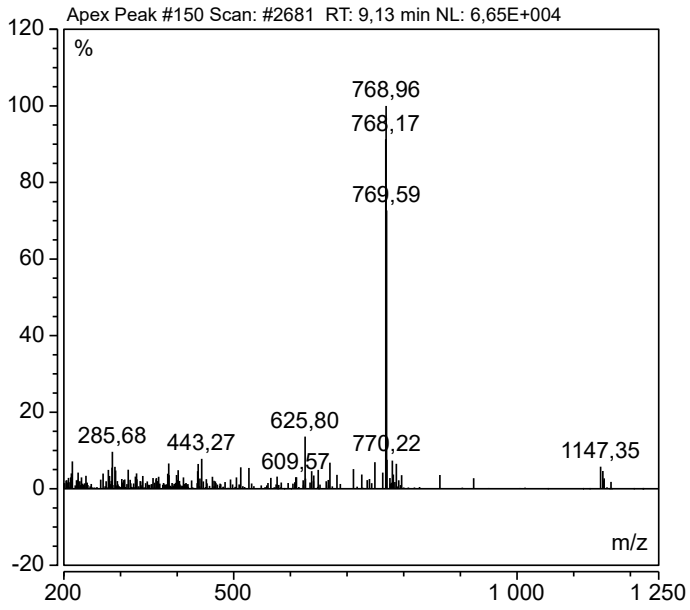


Figure S11. LCMS of major side product obtained after on-resin cyclization of Ac-Cys(Dpm)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS.

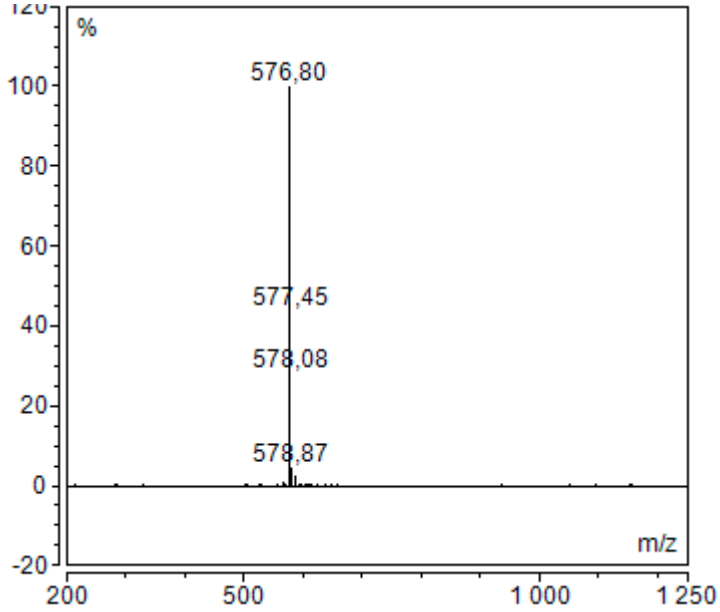


Figure S12. LCMS of cyclized Ac-Cys-oxytocin-NH_2 obtained after on-resin cyclization of Ac-Cys(Mmt)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS; m/z calculated as 576.74 ($M+2H^+/2$).

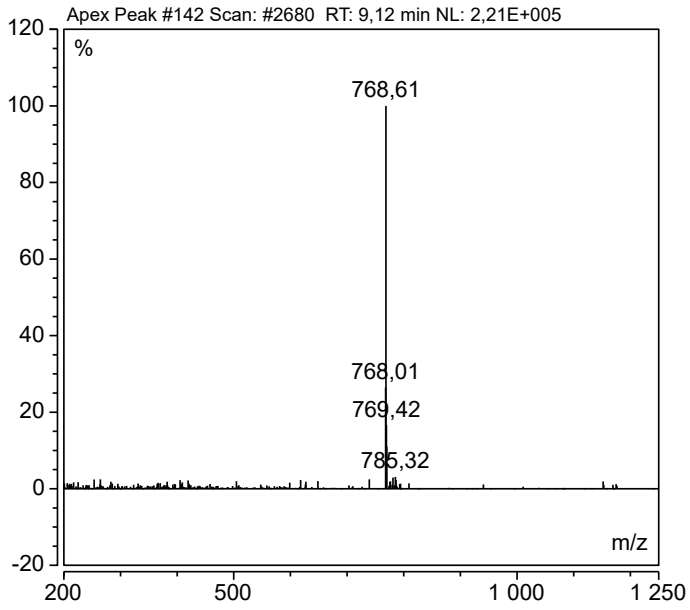


Figure S13. LCMS of major side product obtained after on-resin cyclization of Ac-Cys(Mmt)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS.

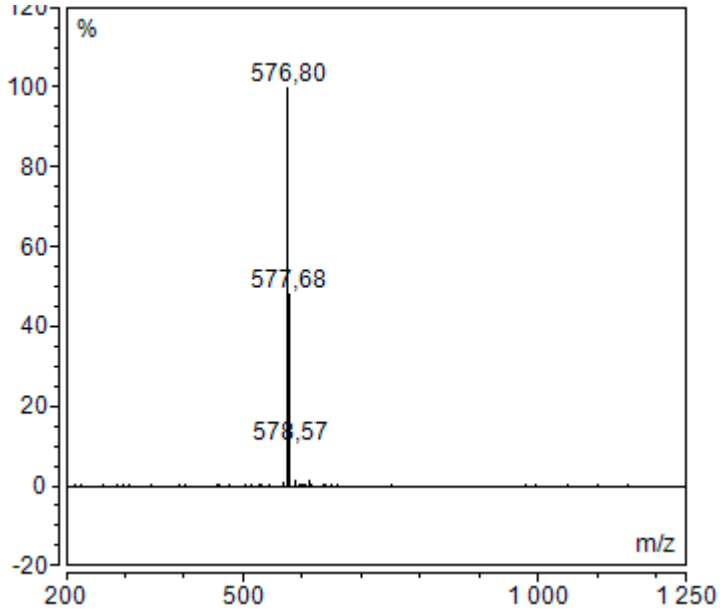


Figure S14. LCMS of cyclized Ac-Cys-oxytocin-NH_2 obtained after on-resin cyclization of Ac-Cys(Thp)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS; m/z calculated as 576.74 ($M+2H^+/2$).

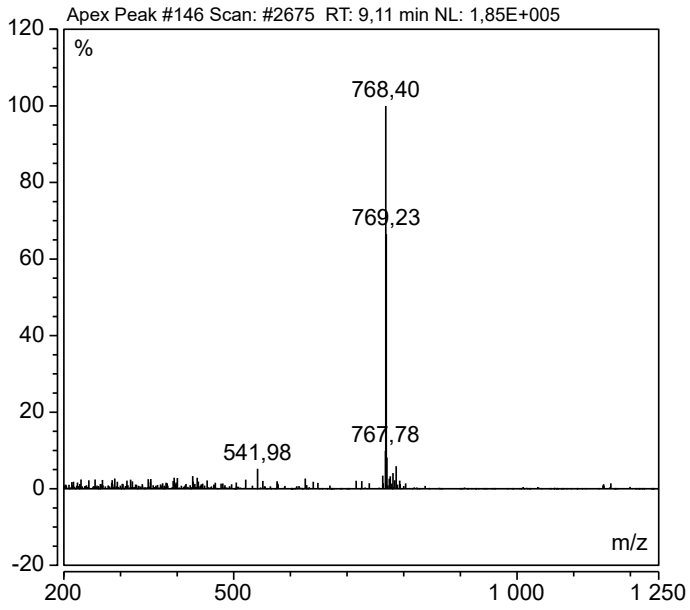


Figure S15. LCMS of major side product obtained after on-resin cyclization of Ac-Cys(Thp)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS.

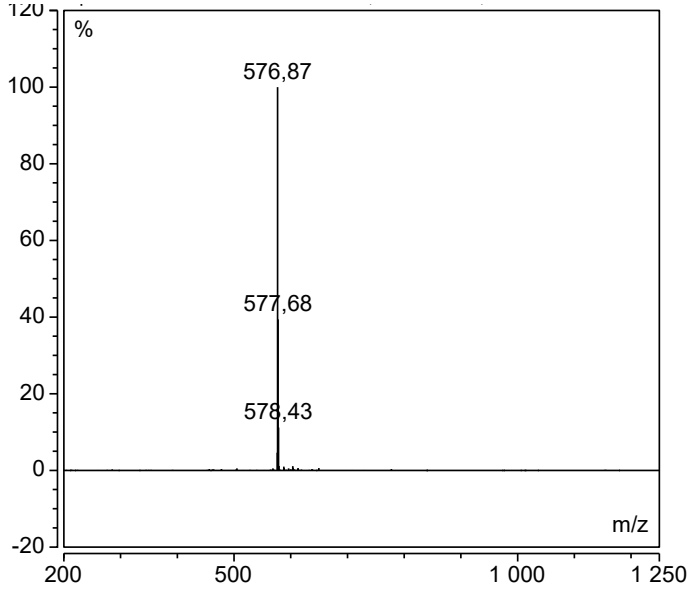


Figure S16. LCMS of cyclized Ac-Cys-oxytocin-NH_2 obtained after on-resin cyclization of Ac-Cys(Trt)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS; m/z calculated as 576.74 ($M+2H^+/2$).

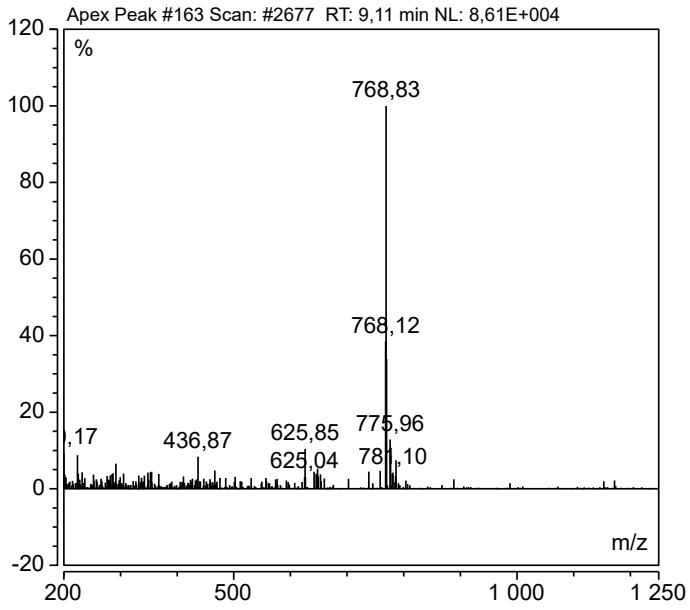


Figure S17. LCMS of major side product obtained after on-resin cyclization of Ac-Cys(Trt)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS.

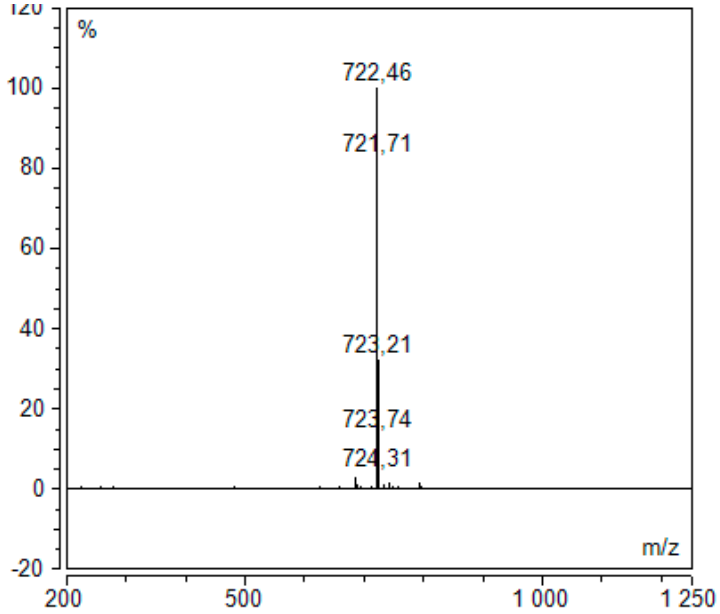


Figure S18. LCMS of cyclized Ac-Cys(Msbh)-oxytocin-NH_2 obtained after on-resin cyclization of Ac-Cys(Msbh)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS; m/z calculated as 721.76 ($M+2H^+/2$).

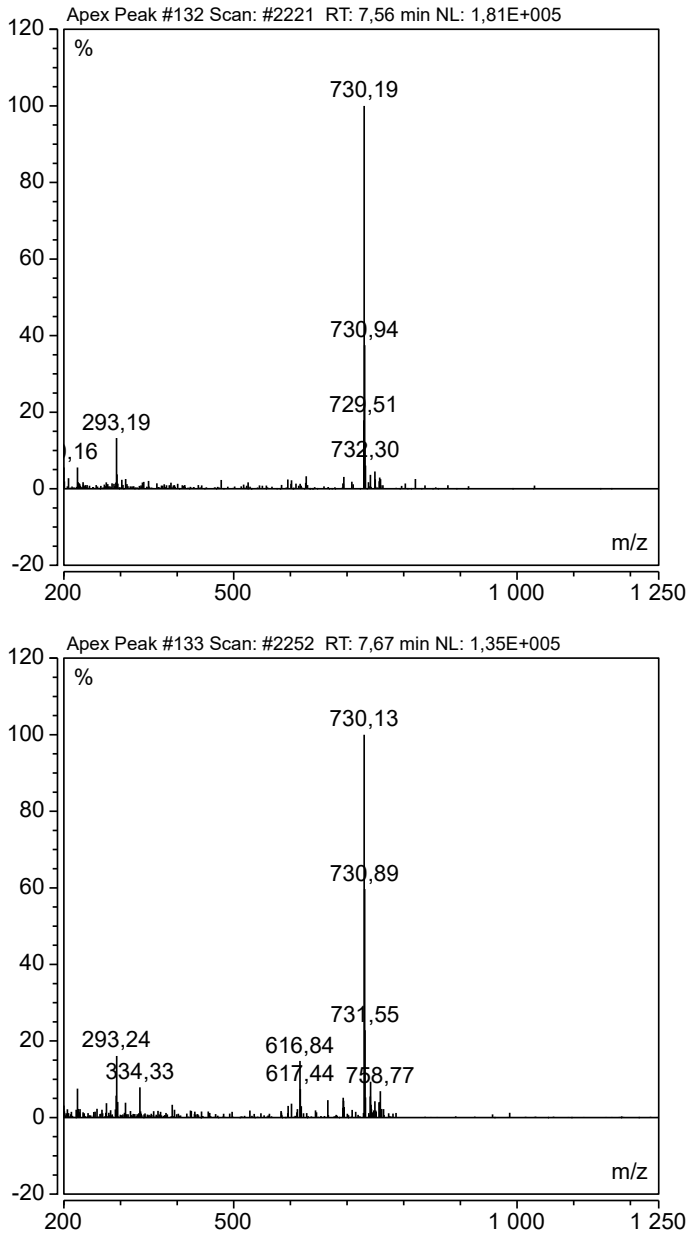


Figure S19. LCMS of major side products obtained after on-resin cyclization of Ac-Cys(Msbh)-Cys(Acm)-Tyr(tBu)-Ile-Gln(Trt)-Asn(Trt)-Cys(Acm)-Pro-Leu-Gly-NH-Rink amide AM resin using NCS.

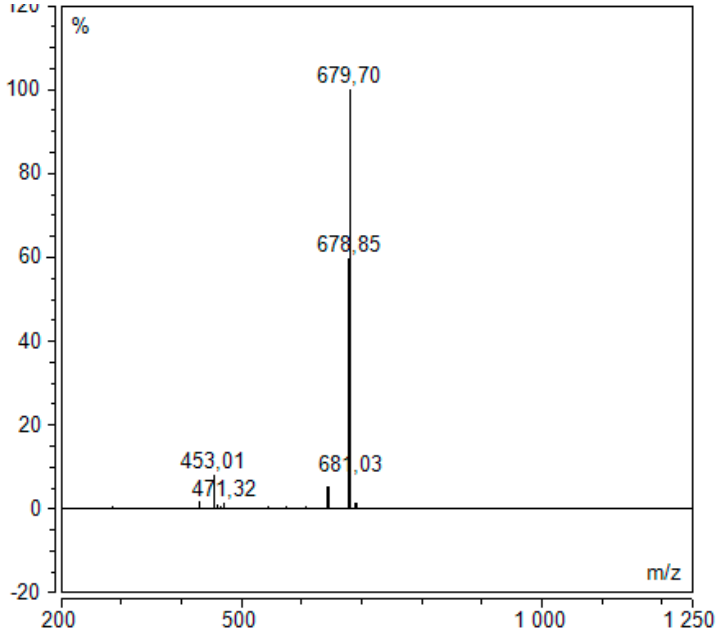


Figure S20. LCMS of H-Cys(SIT)-oxytocin[1,6 Cys(Acm)]-NH_2; m/z calculated as 678.80 ($M+2H^+/2$), 452.87 ($M+3H^+/3$).

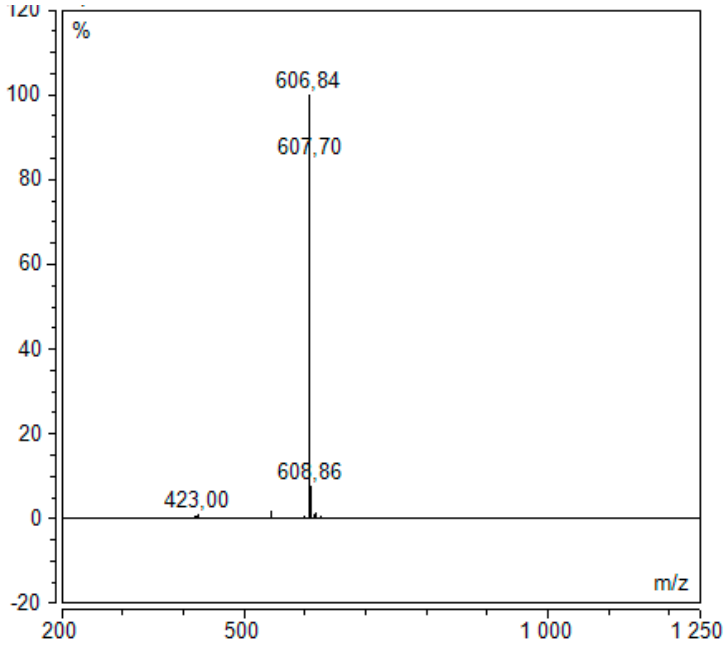


Figure S21. LCMS of cyclized H-Cys(SIT)-oxytocin-NH_2; m/z calculated as 606.76 ($M+2H^+/2$).

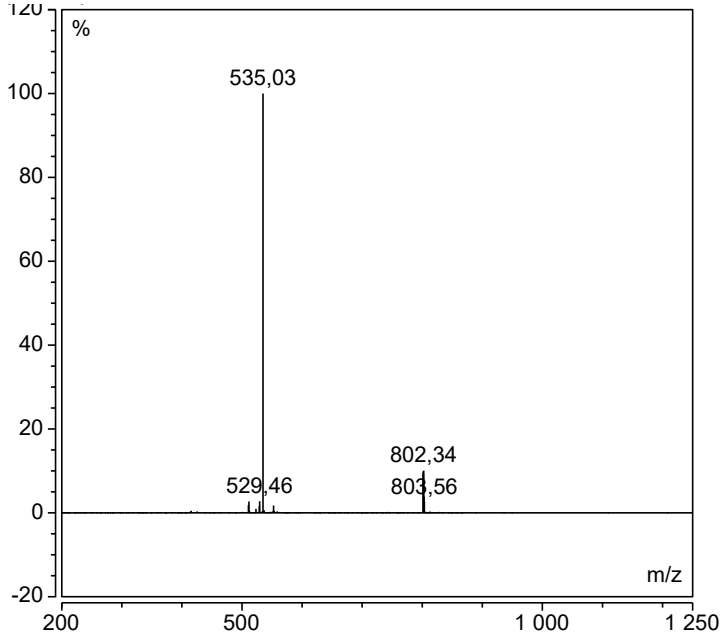


Figure S22. LCMS of H-IC(Acm)C(SIT)NPAC(Acm)GPKYSC-NH_2; m/z calculated as 802.01 ($M+2H^+/2$), 535.01 ($M+3H^+/3$).

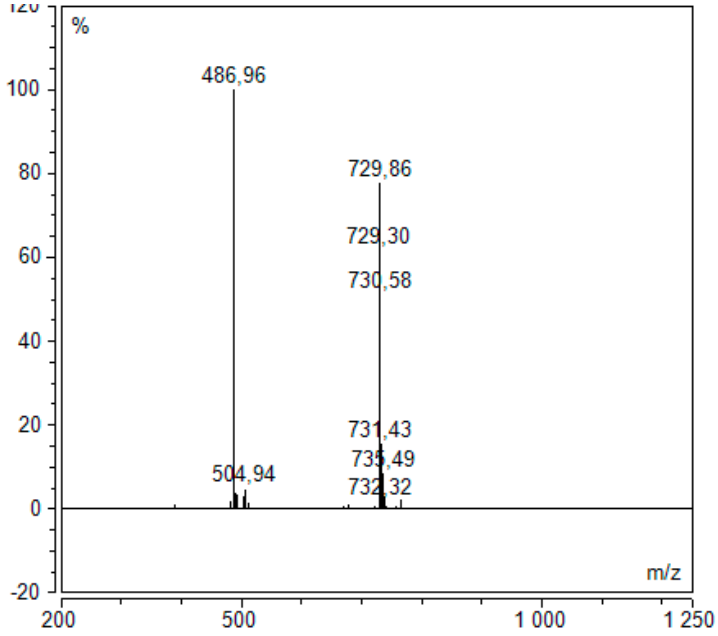


Figure S23. LCMS of cyclized H-ICC(SIT)NPACGPKYSC-NH_2 (SS, Cys2-Cys7) with one disulfide bond; m/z calculated as 729.30 ($M+2H^+/2$), 486.20 ($M+3H^+/3$).

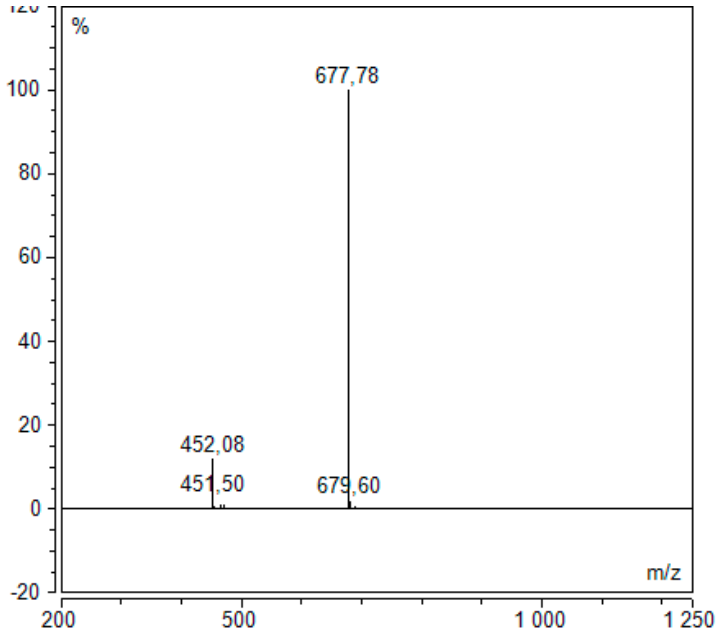


Figure S24. LCMS of fully cyclized H-ICCNPACGPKYSC-NH_2 (2SS, Cys2-Cys7 and Cys3-Cys13) with one disulfide bond; m/z calculated as 677.27 ($M+2H^+/2$), 451.85 ($M+3H^+/3$).