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DRUG DELIVERY

Long live the peptides

> “levels of testosterone were still elevated 12 hours after dosing and returned to normal after 24 hours”

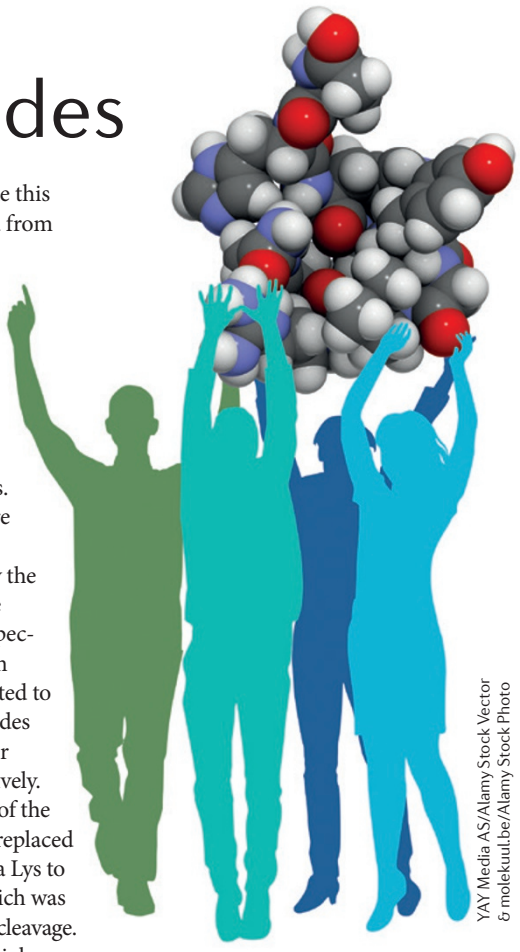
Despite their potential as specific and potent modulators of signalling, peptides are not commonly used in clinical settings owing to their short half-lives. In a recent report, Penchala and colleagues have co-opted the transthyretin system to protect peptides from serum proteases and glomerular filtration, thereby providing a much needed method for increasing peptide stability *in vivo*.

Previous efforts to increase the *in vivo* half-life of peptides include the conjugation of peptides to polyethylene glycol, unstructured proteins, antibody fragments or serum albumin. These techniques are useful for large proteins, but if peptides comprising <30 amino acids are conjugated they can be sterically hindered and thus lose efficacy. The authors therefore turned to transthyretin, a 55-kDa protein whose main function is to protect retinol-binding protein (which transports retinol or other forms of vitamin A) from protease attack and glomerular filtration during transport in the serum. Transthyretin also binds and transports about 15% of the serum thyroxine using a site that is orthogonal to the retinol-binding protein site; most thyroxine molecules are transported by thyroxine-binding globulin.

These researchers had previously developed a small, orally available molecule that binds to the thyroxine-binding pocket of transthyretin. Adapting this molecule with a short linker generated a molecule called THLE1. If conjugated to THLE1, the peptides could reside outside this pocket yet remain protected from cleavage by trypsin *in vitro*.

Two peptides, neurotensin and gonadotropin-releasing hormone (GnRH), were each conjugated to THLE1, and these molecules and derivatives thereof were investigated in serum and in animal models. Neurotensin and GnRH were substantially protected from cleavage in human serum by the THLE1: 22% and 58% of the neurotensin and GnRH, respectively, remained in the serum after 48 hours when conjugated to THLE1, whereas these peptides alone were undetectable after 4 hours and 2 hours, respectively. To further increase stability of the GnRH peptide, the authors replaced a centrally located Gly with a Lys to make THLE1-GnRH-A, which was more resistant to proteolytic cleavage. THLE1-GnRH-A retained high affinity for the GnRH receptor (dissociation constant (K_d) = 6.8 nM in the presence of excess transthyretin) relative to its affinity for transthyretin itself (K_d = 317 nM).

Administration of THLE1-GnRH-A to rats increased the serum concentration of testosterone within 1 hour of injection; the levels of testosterone were still elevated 12 hours after dosing and returned to normal after 24 hours. In rats treated with GnRH-A alone, testosterone levels returned to normal after 8 hours.



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This approach to increasing the *in vivo* half-life of molecules could potentially be applied to other short-lived molecules — including oligonucleotides, oligosaccharides, lipids and small molecules — and could provide a mechanism to move these molecules into the clinic.

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ORIGINAL RESEARCH PAPER Penchala, S. C. *et al.* A biomimetic approach for enhancing the *in vivo* half-life of peptides. *Nat. Chem. Biol.* **11**, 793–798 (2015)