contrast to conventional gene therapy, in which just one variant of the desired gene is introduced directly into cells, the expression of all the variants of the endogenous gene can be regulated using ZPF-based transcription factors, which could have important implications for the success of the approach as the authors show. The *Vegfa* gene codes for three major variants of Vegfa, which are not functionally equivalent or redundant, and the blood vessels produced using ZFP-based transcription factors seem to be more mature than those produced by expression of just the main variant, $Vegfa_{164}$. And taking things one step further, the ZFP-based approach should, in principle, be applicable to the up- or down-regulation of multiple genes, which would be considerably more challenging using conventional gene therapy.

Peter Kirkpatrick

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AMYLOID DISEASES

Assembly instructions

Whether the formation of insoluble amyloid-fibril deposits is the causative agent or merely symptomatic in amyloid diseases such as Alzheimer's disease and Creutzfeldt–Jakob disease is a point of great debate at present. But what is certain is that understanding the driving forces behind what causes proteins to misfold, aggregate and form amyloid fibrils is crucial to designing therapeutic strategies for these diseases. Now, reporting in *Proceedings of the National Academy of Sciences*, Luis Serrano and colleagues show how regions of proteins as small as six amino acids in length are all that is needed to turn a normal protein into an amyloid fibril.

The use of model proteins or fragments of proteins in *in vitro* models, which remove many of the complications that exist in the crowded environment of the cell, is revealing an emerging theme in amyloid-disease research. Despite the differences in amino-acid sequences and three-dimensional structures of the proteins involved, there seem to be common mechanistic rules for fibril formation.

To find these characteristic features, Serrano and colleagues have created an even more simplified model for fibril formation. They used a computer-designed algorithm to search for small peptide sequences of six amino acids in length that have a high propensity to form fibrils. The small size of this model system made it easy to study sequence preferences for amyloid-fibril formation as it allowed systematic substitutions of each of the six positions with any amino acid.

What they found was that fibril formation is due to a delicate balance between specific side-chain and charge–charge interactions within the sequences. Point mutations that changed the amino acid (but did not affect the overall structure) inhibited fibril formation, and peptides with charged residues only formed amyloid fibrils when the net charge was ±1 — a net charge of zero led to amorphous aggregates and a net charge of ±2 led to no aggregates being formed. This presumably occurs as a result of how fibrils form — the peptides form flat secondary protein structures, called β-sheets, which stack on top of each other to form the ordered structure of the fibril. If the net charge is zero, there are neither attractive or repulsive forces between the β-sheets and therefore non-ordered, amorphous aggregates are formed. A small net positive or negative charge favours the organization of ordered aggregates as the distances between charges of the same sign are maximized. However, too great a net charge leads to energetically unfavourable conditions for aggregation to occur, so the peptides remain in solution.

The findings show that small regions, and not the whole protein, might be involved in fibril formation. How this translates to the more complicated situation found in larger proteins, in which the number of charged residues may be high and their distribution more complex, will need to be assessed. But this does provide the fascinating possibility that fibril-forming regions of proteins can be predicted from sequence alone, from which molecules can be designed to disrupt this process.

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