

## Pressedienst Wissenschaft

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### The source of spider silk's extreme strength unveiled

#### Why don't spiders drop off of their threads?

**It has five times the tensile strength of steel and is stronger than even the best currently available synthetic fibers: Spider thread is a fascinating material. Yet, to date no one has managed to produce the material on an industrial scale. Scientists of the TU Muenchen (TUM) and the Universitaet Bayreuth (UBT) have now succeeded in unveiling a further secret of silk proteins and the mechanism that imparts spider silk with its strength. They have published the results of their work in the professional journal *Angewandte Chemie*.**

"The strength of spider dragline silk exceeds that of any material produced in laboratories, by far. All attempts to manufacture threads of similar strength have failed thus far," explains Professor Horst Kessler, Carl von Linde Professor at the Institute for Advanced Study at the TU Muenchen (TUM-IAS). In collaboration with the workgroup of Prof. Thomas Scheibel, who was a researcher at the TU Muenchen until 2007 and who now holds a chair of the Institute of Biomaterials at the Universitaet Bayreuth, Professor Kessler's team has been researching for years to unveil the secret of spider silk.

How do spiders manage to first store the silk proteins in the silk gland and to then assemble them in the spinning passage in a split second to form threads with these extraordinary characteristics? And what exactly gives the threads their tremendous tensile strength? Scientists have now come one step closer to answering these key questions for the production of artificial spider silk.

Spider threads consist of long chains of thousands of repeating sequences of protein molecules. These silk proteins are stored in the silk gland in a highly concentrated form until they are needed. The long chains with their repeating sequences of protein molecules are initially unordered and must not get too close to each other as they would immediately clump up. Only in the spinning passage, just before being used, are the threads oriented parallel to each other and form so-called micro crystallites that are, in turn, assembled to stable threads with cross links.

During the last year, the scientists in Kessler's and Scheibel's team investigated the common garden spider ("cross spider") to discover the mechanism behind the transition from individual spider silk molecules to connected threads: The individual spider silk proteins are first stored in the silk gland in small drops called micelles.

The scientists identified the regulating element that is responsible for assembling a strong thread from the individual parts. It is the so-called C terminal domain of the silk protein. It prevents the formation of threads in the silk duct with its strong salt concentrations. In the spinning passage, however, where the salt concentration is low and sheer forces are abundant, this domain becomes instable and "sticky." This causes the chains to overlap and a strong spider silk thread is formed. The discovery of the significance of this relatively small C terminal domain, when compared to the

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overall length of the protein thread, was a sensation at the time and was published in the renowned scientific journal Nature.

Now the same group of researchers has put in place a further piece in the spider silk puzzle. They showed that the other end of the long thread, the so-called N terminal domain, plays an important role in the design of strong threads with great tensile strength. This time, the scientists investigated the head ends of the spider silk proteins of the “black widow” (*Latrodectus hesperus*). The result: The N terminal head ends exist in the silk duct as single strands (monomers). Only in the spinning passage are the head to tail pairs (dimers) formed.

The process of laying together is regulated via the change in pH values and salt concentrations between the silk duct and the spinning canal. In the silk duct, a neutral pH value of 7.2 and a high salt concentration prevent the N terminal head ends from combining. In the spinning passage, however, the environment becomes acidic (pH value around 6.2) the salt is removed. Now the ends can come together. In this process, the N terminal ends connect to the respective other ends – a practically endless chain of linked up spider silk proteins is formed. “In our work we were able to show, in addition to our previous research, that both the pH value and the salt concentration influence the monomer-dimer balance,” says Franz Hagen, corresponding author of the study, in summing up the results. “Both factors influence the formation of dimers and thus the efficient cross-linking to very long silk proteins.”

Ultimately, this cross-linking is what gives the spider silk threads their enormous tensile strength. The small crystallites first formed in the parallel cross-linking of the protein chains following the controlled unfolding of the C terminal domain are connected to each other via the N terminal domains of the spider silk protein to form a very long chain. “This is the effect that eventually explains the enormous tensile strength of the spider silk thread,” says Kessler. To date, this ingenious form of cross-linking – called “multivalence” – has not been implemented in artificial polymers. “Most polymer chemists focus on the length of the thread. So far, no one has come up with the approach of cross-linking the ends of the threads and thereby opening the door to virtually unlimited lengths of polymer chains,” believes Kessler. These new findings may provide chemists with a model for manufacturing new materials with improved characteristics.

The scientists used the method of nuclear magnetic resonance (NMR) to analyze the structure of spider silk. Segments of spider silk are dissolved under conditions similar to those found in spider organs and exposed to radio wave impulses in a very strong magnetic field. The scientists can deduce the exact molecular structure from the “response” of the molecules. Using this method, environmental influences (e.g. salt concentration and pH value) can be studied accurately under simulated natural conditions. The development and application of NMR methods to biomolecules has been a longstanding focus of the Bavarian NMR Center in Garching.

### Original publication:

F. Hagn, C. Thamm, T. Scheibel, H. Kessler; pH Dependent Dimerisation and Salt Dependent Stabilisation of the N-terminal Domain of Spider Dragline Silk - Implications for Fibre Formation, *Angew.Chem. Int. Ed.* 2011, 50, 310-313.

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**Previous publications on the spinning process:**

F. Hagn, L. Eisoldt, J. Hardy, C. Vendrely, M. Coles, T. Scheibel, H. Kessler, A highly conserved spider silk domain acts as a molecular switch that controls fibre assembly, Nature 2010, 465, 239-242.

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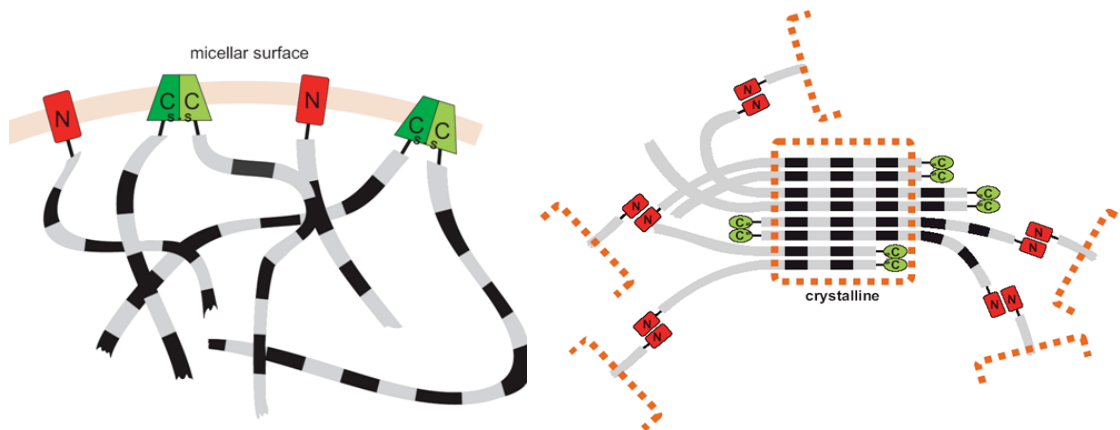


Figure: left: The spider silk protein is stored in so-called micelles, in which the folded domains (monomer N terminal domains in red, dimer C terminal domains in green) are oriented along the water on the micellar surface. The long chains with the repetitive units remain unordered. Right: In the spinning passage the threads are oriented in parallel and form so-called micro-crystallites. The free N terminal domains (red) are laid together into to dimers and from a very stable material as a result of the cross-linked micro-crystallites.