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Diversification of Insect Silk: One Name, Several Components

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Silk is a functional term used to describe protein fibers spun by a number of arthropod lineages. Silk use by humans greatly increased after the domestication of the silkworm in China in neolithic times. According to Chinese folklore, a cocoon falling into the tea cup of the Empress Leizu as she sat under a mulberry tree in the 27th century BC led to the invention of sericulture and the silk loom. While exporting large amounts of silk fabric, the process of its production was a closely guarded secret in China for centuries, and silk fabric was symbolic of wealth, elegance and power.

More recently, silks have proved to be ideal materials from which to construct parachutes, diffraction gratings, and the cross-hairs in optical device. In recent years there has been a resurgence of interest in silk in the hope of replicating its remarkable mechanical properties using modern biotechnology. Most research has centered on the cocoon silk of silkworms as well as the dragline silk of spiders. However, many insect species produce silk for a wide variety of purposes.

Silkworms (Lepidoptera) and spiders are the extensively studied silk-producing groups. The capacity to produce silk is present in many arthropod groups, including 17 out of the 30 orders of insects. Some characterization has been performed on silk materials made by web-spinners (order Embioptera); mantises (order Mantodea); the leafhopper Kahaono montana (Hemiptera: Cicadellidae) etc.

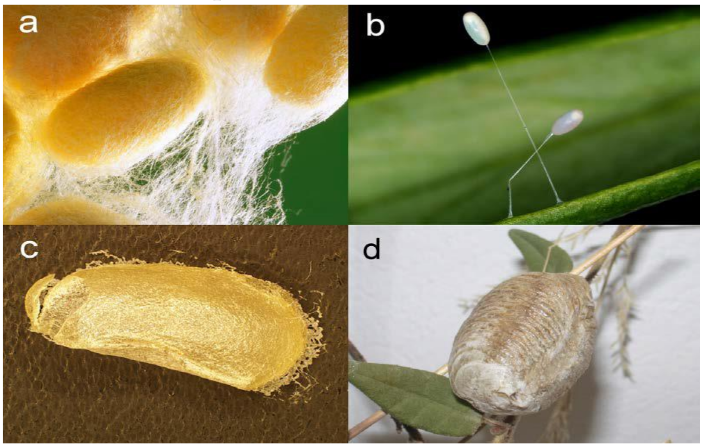

Fig 1: Examples of insect silks. (a) Silkworm cocoons (Bombyx mori). (b) Lacewing egg stalks (*Mallada signata* (c) Sawfly cocoon (*Nematus oligospilus*). (d) Praying mantis ootheca (*Tenodera australasiae*)

Insects use silk for a very wide range of purposes, including shelter, reproduction, foraging, and dispersal. For example, silk may be used for pupation cocoons, permanent communal galleries and domiciles, catching prey, transferring sperm, securing and protecting eggs, attachment during moulting, or transport by ballooning.

Identification of different silk lineages

The ability to produce silk is a complex trait requiring a suite of characteristics silk proteins, silk glands, and silk-spinning behavior. On the basis of silk gland type, silk protein molecular structure, and the phylogenetic relationship of silk-producing species, we grouped insect silks into 23 distinct categories (Sutherland *et al*.,2010). This describes the different evolutionary origins of silk glands and silk proteins with different molecular structures are not expected to be homologous. Therefore, incorporating information about silk protein structure, silk gland type, and phylogenetic relationship allows us to distinguish independently evolved silk lineages.

Diversity of Silk-Producing Glands

Glands can be classified by location and type, using as a first criterion where they are located in the body, and as a second criterion whether they are dermal glands. The categorization of silk glands into labial, Malpighian tubule, dermal glands. The last category includes the silk-secreting accessory sex glands (Sehnal and Akai,1990). By comparison, dermal gland silks are less numerous and Malpighian tubule silks are the least common.

Labial gland (Salivary gland): The labial glands of insects are commonly called salivary glands, they often perform functions besides producing saliva, such as enabling trophallaxis and synthesizing venom and silk. Labial gland (Salivary gland) are mainly in lepidopteran silks that are such a prominent feature of caterpillar and pupal biology.

Silk secreting cell of larval Sawfly: According to Kenchington, 1969 the silk- producing labial glands of sawflies possess secretory units with an end apparatus. Silk synthesized in the cytoplasm passes into a meandering intracellular duct, the two ends of which join to form a secretion canal opening in the reservoir. Labial secretory cell produce saliva in early instar and silk by last instar.

Table 1: Summary of insect silk

Common name of insect group [higher classification] (references)	Purpose of silk	Life stage/gland
1. Jumping bristletails, silverfish [Archeognatha: families Machilidae and Meinertellidae; Zygentoma]	Indirect sperm transfer, silken threads that guide the female to spermatophore, silken stalks or mats that hold droplets of sperm above the ground	Adult males/Type III secretory units
2. Mayflies: [Ephemeroptera: family Polymitarcyidae]	Lining for U-shaped tunnels in submerged wood	Larvae/apparenly in Malpighian tubules
3. Dragonflies [Odonata: family Gomphidae]	Anchoring eggs? Bundles of fibers attached to eggs that uncoil upon exposure to water	Adult female /unknown
4. Webspinners [Embiidina]	Tunnels and egg coatings	All stages/Type III secretory units in prothoracic tarsomeres
Silk proteins predominantly in beta-sheet conformation. Partial silk proteins (no N termini) contain reiterating Gly-Ser or Gly-Ala repeats. Silk fiber contains 4% Ala, 31% Ser, 44% Gly.		
5. Crickets [Orthoptera: Stenopelmatoidea in the families Gryllacrididae and Anostostomatidae]	Binding leaves together for construction of cocoon-like nests, linings for sand burrows	All stages/labial glands
6. Book lice [Psocoptera: suborders Psocomorpha and Troctomorpha]	Egg coverings and nests	Adult females/labial glands
7. Thrips [Thysanoptera: Terebrantia in the families Heterothripidae and Aeolothripidae; Tubulifera, family Phlaeothripidae in the subfamily Phlaeothripinae]	Cocoons or tent-like shelters for protection against predation, extreme temperatures, and low humidity	Larvae and possibly adults/secretions from the anal region, likely from Malpighian tubules
***8. Kahaono montana* Evans** [Hemiptera in this single species]	Protective shelters	Unknown
Related species do not make shelters. The silk proteins are predominantly beta-sheet structures. Silk fiber contains 12% Ala, 32% Ser, 13% Gly.		
9. Water beetles [Coleoptera: family Hydrophilidae]	Silken rafts to support eggs	Adultfemale /colleterial Glands
Silk proteins in a cross-beta conformation with X-ray diffraction patterns but similar to supercontracted keratins.		
10. Plant-eating beetles [Coleoptera: Cucujiformia]	Terrestrial or aquatic cocoons, silk-lined tunnels in the sand	Larvae/Malpighian tubules
Hypera species cocoon silk proteins adopt a cross-beta structure 10 amino acids in width.		
11. Lacewings [Neuroptera, found within four of the six superfamilies]	Egg stalks or egg coverings	Adult females/ colleterial glands
Species in Chrysopidae and Nymphidae have silk proteins in a cross-beta structure 8 amino acids in width. Silk fiber contains 20% Ala, 41% Ser, 24% Gly.		
12. Lacewings and ant lions [Neuroptera]	Cocoons	Larvae/Malpighian tubules
Cocoon of the green lacewing *Mallada signata* is composed of small (49 kDa) alpha-helical-structured proteins and lipids. Silk fiber contains 40% Ala, 7% Ser, 11%		
13. Sawflies and parasitic wasps [Hymenoptera]	Cocoons, nests, and webs	Larvae/labial gland

Silks contain proteins in an extended beta-sheet structure. Silk fiber contains 36–56% Ala, 8–40% Ser, 2–31% Gly.		
14. Parasitic wasps [Hymenoptera: Chalcidoidea in several Eupelmidae and a Signiphoridae species]	Egg covering and host covering	Adult female/abdomen secretion
15. Bees, ants, and wasps [Hymenoptera: Apoidea and Vespoidea]	Nests and cocoons	Larvae/labial gland
Silk fibers contain four small (30–50 kDa) homologous alpha-helical proteins that adopt a tetrameric coiled-coil structure. Silk fiber contains 23–34% Ala, 10–17% Ser, 5–6% Gly.		
16. Sawflies [Hymenoptera: Tenthredinoidea in the family Tenthredinidae]	cocoons	Adult females/labial glands
Polyglycine II structured proteins contain high levels of Gly (66%) and are present in the cocoons of some Allantinae, Heterarthrinae, and Blennocampinae species. silk fiber contains 28% Ala, 14% Ser, 11% Gly.		
17. Wasps [Hymenoptera: Apoidea in the subfamily Pemphredoninae, some species in *Microstigmus*, *Spilomena* and *Arpactophilus*]	Rope fabricated from plant fibers and silk that suspends the nest	Adult females/Type III secretory units in posterior metasoma
18. Wasps [Hymenoptera: Vespoidea in the family Vespidae the species *Dolichovespula maculate, Polistes metricus, P. annularis,* and *Ropalidia opifex*]	Nests constructed from fine silk fibers binding plant fibers	Adult females/labial glands
Silk fiber contains 14–20% Ala, 16–19% Ser, 10–27% Gly [total 49–57%].		
19. Fleas [Siphonaptera]	Cocoons	Larvae/labial glands
Proteins in the cocoon of *Xenopsylla cheopis* are inan alpha-helical conformation.		
20. Dance flies [Diptera: family Empididae in the subfamily Empidinae]	Silk-wrapped nuptial gifts	Adult males/Type III secretory units in prothoracic basal Tarsomeres
A partial protein sequence identified from mRNA extracted from male silk glands. silk protein present in the silk-producing basitarsi is repetitive with high levels of Asp (21.9%) and Gly (13.9%).		
21. Glowworms [Diptera: family Keroplatidae]	Nests/prey capture threads	Larvae/labial glands
The threads of *Arachnocampa luminosa* contain proteins with cross-beta crystalline structure		
22. Midges [Diptera: Chironomoidea in the families Chironomidae and Simulidae]	Aquatic silken tubes, adhesive lifelines, cocoons	Larvae/labial gland
The silk of Chironomidae is produced from proteins (40 kDa to very high molecular weight) containing high levels of cysteine. Silk fiber contains 6% Ala, 12% Ser, 10% Gly.		
23. Butterflies, moths, caddisflies [Lepidoptera, Trichoptera]	Cocoons (aquatic and terrestrial), tunnels, retreats, communal webs, prey capture nets	Larvae/labial gland
Silk consists of two filaments glued together by serine-rich (>15% Ser) proteins known as sericins. The filaments consist of three proteins: heavy fibroin (>200 kDa), light fibroin (25–30 kDa), and the glycoprotein P25 (~25 kDa). Around 90% of the sequence of the heavy fibroin protein of *Bombyx mori*. Silk fiber contains 5–57% Ala, 0–22% Ser, 19–48% Gly		

Malpighian silk glands: The Malpighian tubules comprise the excretory and osmoregulatory system of insects. Fibrous or silk like secretions originating in the Malpighian tubules have been reported in neuropteran larvae, coleopteran larvae, and possibly ephemeropteran larvae. The concentrated solutions of silk proteins collect in the alimentary canal, and the anus acts as a spinneret.

Dermal glands

Dermal glands are secretory units made up of a few cells that may be spaced as isolated units or clustered into compound glands, much as sense organs can be singular or clustered. Dermal silk glands can be further divided into epidermal glands and internalised glands. Typically, each glandular unit comprises a large secretory cell, a canal cell, and several supporting cells. So named the complete unit a class III dermal gland.

Internalized dermal silk glands: Most often they are sex accessory (collateral) glands, so that silk is fabricated at the opening of the reproductive canal. The glandular epithelium is composed of myriad individual class III dermal glands that secrete into a common lumen. (Kenchington 1969).

Other silk secreting dermal glands are found in a variety of locations. In sphecid wasps, glands located on the sternum of the adult female are the source of silks used for nest lining. In male hilarine flies, the basal tarsomere of the pro- thoracic legs is swollen and silk is released from ventrally located hollow bristles to make silk wrapped nuptial gifts.

Silk molecular structures

Insect silks are semi-crystalline materials, i.e., materials with regions of ordered molecular structure (crystallites) within an amorphous matrix. The structures differ dramatically from each other in molecular organization, their regularity allows the proteins to pack efficiently and form extensive networks of hydrogen bonds within or between the proteins. The hydrogen bonds con- tribute to the mechanical strength and stability of silk fibers. Each of the crystalline structures is dictated by specific sequences of amino acids in the silk proteins. Five different structures have been identified.

Common features of insect silks

Given the diversity of insect silks, it is remarkable that certain features are common among silk production systems.

- The accumulation of silk proteins at high concentrations in specialized glands
- Proteins rich in alanine, serine, and/or glycine
- The folding of silk proteins into a single dominant secondary structure with high crystallinity
- Mechanically strong and/or tough silk fibers.

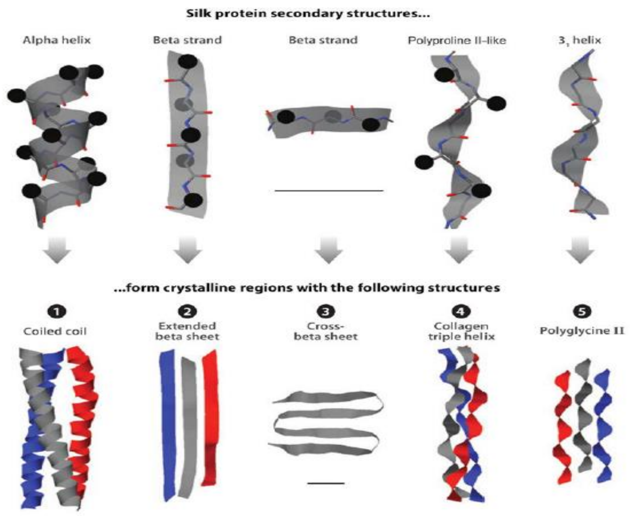

Fig. 2 Ordered protein structures of insect silks

Silk mechanical properties

The mechanical properties and solvent stability of silks are closely related to the crystallinity of the material. Highly crystalline materials have greater strength and solvent stability by virtue of the network of hydrogen bonds between and within the proteins.

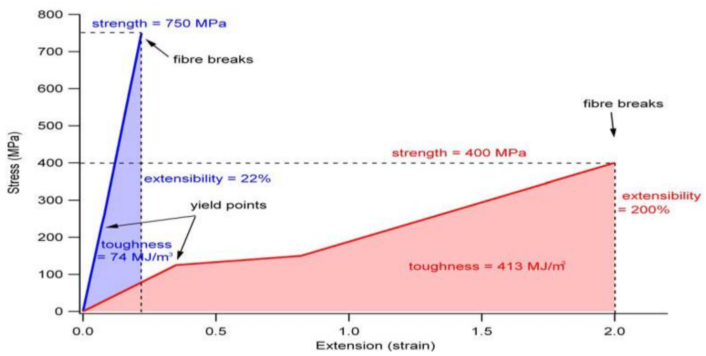

Fig. 3 Silk stress-strain plots

Stress-strain curves of silk fibres may display a `yield point' (figure 1). Up until the yield point, extension occurs by deformation of the amorphous region and crystallites. After the yield point, extension is associated with major slippages of the crystalline regions and/or structural transitions. Successive extension of silk fibres thus depends on the deformation of successively stronger structures another example of hierarchical structure in silk fibres

In general, insect silks have strengths (breaking strains) in the range 150- 750 MPa, extensibilities (engineering stresses) in the range 22-400%, and stiffnesses (Young's moduli) in the range 0.3-7 GPa. In relative terms, silkworm silk is strong, stiff, and has low extensibility. By comparison, coiled-coil silks are more extensible and weaker. The greatest extensibilities of all are found in cross-β-sheet silks, which can be extended to many times their own length. The high extensibility of α-helical and cross-β-sheet silks is associated with the ability of their crystalline regions to unravel and partially convert into extended β-sheet structures (Rudall, 1962).

Amino acid composition

Insect silks have high levels of glycine, alanine, or serine. The abundance of these amino acids is most likely due to their nonessential character and intermediate hydrophobicity. As glycine, alanine, and serine are neither strongly hydrophobic nor hydrophilic, they are able to form intermolecular interactions with water and therefore contribute to protein solubility.

Conclusion

Silks play a crucial role in the survival and reproduction of many insects. Insect silks are analyzed determine the likely evolutionary relationships among different silks. Also identified and discussed the significance of some common features shared by insect silks. Scant information is available about the composition and production of most of these silks from past 10 years have seen an explosion in molecular silk sequence discovery.

References

Kenchington,W., 1969, Silk secretion in sawflies. *J. Morphol.,* 127:355–62.

Rudall, K.M. and Kenchington ,W.,1971, Arthropod silks: the problem of fibrous proteins in animal tissues. *Annu. Rev. Entomol.,* 16:73–96.

Sehnal, F. and Akai, H., 1990, Insect silk glands: their types, development and function, and effects of environmental factors and morphogenetic hormones on them. *Int. J. Insect Morphol. Embryol.*,19:79–132.

Sutherland, T. D.,Young, J. H.,Weisman, S., Hayashi, C. and Merritt, D., 2010, Insect silk: One name, many materials. *Annu. Rev. Entomol* ., 55:171-188.

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