#### Carnivory in Byblis liniflora Revisited II: The Phenomenon of Symbiosis on Insect Trapping Plants

Hartmeyer, S. (1998). Carnivorous Plant Newsletter (ICPS), 27/4: 110-113

#### Introduction

If people consider the words carnivorous plants (CP), they mostly think those little 'killers' just catch and digest any prey they are able to overwhelm. However, meanwhile many CP-enthusiasts know that worldwide numerous arthropods as ants, true bugs, small crawfish, mosquito-larvae and spiders as well as numerous bacteria and fungi also profit from the mostly extensive prey on or inside the different traps. It makes a clear difference if e.g. some ants just steal the prey out of a sundew trap to drag it to their anthill, or if the ants are resident on the plant, defending it against herbivores and dropping their mineral-rich feces on the leaves or inside the pitchers. In the first case its just robbery (clepto-parasitism), a wide spread and certainly not spectacular strategy in nature. The second example is a typical mutualism or symbiosis where both sides benefit by the partner e.g. the plant by the accelerated decay of prey which prevents fungi development, a danger especially for plants using glue traps without enzyme production. Such a mutualism is e.g. well known on the non carnivorous (or yet) tobacco (Nicotiana tabacum) which gets its gluey protection zones cleaned by its resident true bugs (Engitatus tenuis). A comparable cleaning function is well known from the Barrier Reef in Australia, where divers can visit stationary so called 'cleaning stations', where minute specialized shrimps and fish feed parasites (their profit) from different much bigger species, that patiently line in for that health service. However, the nature hates stereotypes and so the assignment of true symbiosis is complicated by numerous 'borderline cases' e.g. the small red crawfish (Geosesarma malayanum) which visits pitchers of Nepenthes ampullaria to search for nutrition. It crushes the drowned prey with it's claws in a typically wasteful way from which numerous polyps profit, living on the snail shells inhabited by Hermit crawfish (*Paguridea*). The mechanical crushing increases the digestion speed, which can in spite of some losses by the visitor be beneficial for the plant. To suggest a true symbiosis it must first be cleared if that crawfish is just an opportunist or specialized, maybe even resident on that CP population. The understanding of such phenomena is necessary for a new discussion about the true carnivory of some plant families using glue traps (maybe even tobacco ??). The results of recent investigations about the plant-arthropod mutualism on Roridula gorgonias and about the enzyme production on Byblis liniflora are discussed below. In addition a new documentary film by Thomas Carow (Germany, 1996), which will soon be shown in the USA, is described and recommended as excellent audiovisual completion the discussed subject.

### The assignment of Roridula

The discussion whether the two species of the South African *Roridula* are carnivorous or not lasts meanwhile since Charles Darwin 1875 first suggested that it is a true CP. 1910 Marloth and 1935 Lloyd contradicted Darwin because the sticky herb, which produces a very effective India rubber containing hydrophobe glue (nonpolar) on stalked glands, has no sessile glands which produce protheolytic enzymes and absorb the dissolved nutrients on the Droseraceae. And really, it was repeatedly confirmed that *Roridula* is not able to produce own digesting enzymes. So the theory developed, that these plants with the strongest glue of all insect-trappers are just a 'useless flypaper' benefiting only from it's prey by a very ineffective soil fertilization, caused by it's decaying and down washed victims. The resident Capsid bugs (Miridae) where stated as additional loss because they steal the prey using their astonishing ability to move relatively carefree through the highly adhesive

glue of the plant. Also some species of spiders, avoiding glue contact with their threat, feed either on the plants prey or chase for the bugs. However, also Darwin's theory remained popular, as numerous *Roridulas* in several CP collections demonstrate.

Fortunately, from 1991 - 1996 I could successfully grow two plants of Roridula gorgonias (at last up to some 1.2 m) settled with a healthy and numerous Pameridea bug population in my living room in Weil am Rhein, Germany. The bugs remained on their host plant and only on rare occasions the adult insects used their wings for a reconnaissance of our house (whereby no persons were attacked in any way !). Also if placed on the balcony on sunny days, the insects showed no tendency to escape. During the winter I fed the bugs with beetle larvae, available in every pet shop, in summer the plants caught enough flies and wasps to guarantee a healthy population. If abundant prey is available, the bug population grows very quickly and the plant is endangered by suffocation beneath the leaf covering feces. But if a certain balance is reached by feeding the right quantities (in nature the bugs are reduced by spiders), the older feces on the leaves get paler and paler until they nearly disappear, even inside the living room without any washing by rain or water spraying. The plants showed their good condition by producing some 60 beautiful pink flowers one year. I filmed this phenomenon 1993 for my first video in the English language BEAUTIFUL AND HUNGRY - CARNIVOROUS PLANTS (1994) and added as comment my personal opinion, based on my for 6 years lasting observations, that I suggest a true symbiosis on this plant-arthropod mutualism. Immediately after the video was around, I experienced the intensity of the discussion about the CP state of Roridula because I maintained that the plant is a true CP because it don't needs any own enzymes to benefit from it's in abundance caught prey. The nutrition is not absorbed via soil fertilization, but much more effective through the 'predigested' feces of its Pameridea partners. I considered that the assignment as true CP (trapping of prey, decomposition of prey by protheolytic enzymes and absorbing facilities for nutrients) should include the feature of 'passive digestion' by true symbiosis if the nutrients are absorbed. In spite some CP enthusiasts shared my opinion, I could not proof my statement. But fresh light appeared 1996, when Ellis and Midgley from the University of Cape Town, published detailed the results of some highly interesting examinations concerning exact that subject.

The authors used a method published by Dixon who examined the nitrogen take in on *Drosera erythrorrhiza*, analyzing the 15N (Nitrogen) isotope. Yeast was added with 15N- Ammonium sulfate, then mixed to a feeding medium and fed for five days to a growing population of the well known laboratory-fly *Drosophila melanogaster*. Ellis and Midgeley developed some series of tests on a natural growing site and a laboratory specie of *Roridula gorgonias* populated with *Pameridea* bugs, respectively on plants on which they removed the (in normal case) resident insects for negative tests. The *Drosophila* were fixed onto the plants and sucked out by resident bugs. Some days later several leaves of the plants were removed, cleaned from all impurities and the 15N isotope content was analyzed by a mass spectrometer. The results confirmed the symbiosis theory, because those plants with resident bugs showed a clear higher 15N content, also inside new built leaves. In spite *Roridula* has no sessile glands it seems to absorb the feces via the stomata, like a lot of other non-carnivorous plants do if leaf-fertilized. So these examinations proof doubtless that *Roridula* benefits from it's caught prey much more efficient than by soil fertilization and the designation 'useless flypaper' should disappear for ever. However, the question remains if that is enough to assign it true carnivorous in spite of the lacking enzyme production.

## May Byblis and Darlingtonia not be assigned CP anymore ?

In 1953, China reports about the Capsid bug Setocornis bybliphilus observed on Byblis gigantea near Perth, and two Cyrtopeltis species (Capsid bugs as well) living on some tuberous Drosera (see table 1) in the same area. Unfortunately, we could not observe any bugs on our visit on sites of Byblis gigantea near Perth January 1991, and the tuberous Drosera got dormant at that time. My wife Irmgard and myself were the more surprised when we featured Byblis liniflora near Kununurra in North Australia May 1995 for our latest video production BEAUTIFUL AND HUNGRY PART 2 -CARNIVOROUS PLANTS (1995) and found just on our first attempt to search the Rainbow Plant numerous true bugs living on them. Apparently closely related to our well known Pameridea bugs on Roridula. The similarity was amazing. Although the insects were a little smaller they showed the same pattern on their back (typical dotting for the family Miridae), the same appetite for the plants prey and a phenomenal ability to use the glue drops as 'cooling agent' without sticking on them. Curious to find more mutualism of this kind we looked around and could film another smaller bug species (but also Miridae), obviously ideal adapted on Drosera ordensis and again later two more species near Darwin on different variations of Drosera indica. These insects living on a true CP as a sundew, need to have a protection against protheolytic enzymes, like particular mosquito larvae developing inside the pitchers of Nepenthes. Well, that is certain right in case of Drosera but some new examinations inside my greenhouse brought a surprise for Byblis liniflora, which changed my point of view.

At the beginning of this year I tried to optimize a simple enzyme testing for CP on photographic film and examined several Drosera species on which enzyme production has been proved doubtless, in addition Byblis liniflora and Roridula dentata. That resulted in a real surprise, because even after repeated tests none of two Byblis liniflora specimens showed any enzyme production whereas all sundews were clear positive, thus confirming the right function of the test. That works because the digestion of gelatin layer on photographic film corresponds with the digestion of animal protein. The other way round that means: a plant unable to digest gelatin is also unable to digest its caught prey. Inspecting the test procedure I could not find any mistakes. These results would assign the mutualism of Roridula- Pameridea and Byblis - Setocornis (and probably Cyrtopeltis) as much more similar than suggested till today, in spite Byblis liniflora develops sessile glands for nutrient absorption, which Roridula does not. But where are the enzymes that Bruce (1905) mentioned on Byblis gigantea ? If there are none, Byblis liniflora would not be a CP per definition any more. And by the way no enzymes are produced by Darlingtonia californica and some Heliamphora species too. Are they no more CP ? But there is hope for all CP enthusiasts loving their Cobra Lilly and expensive Pitcher plants, both genera show symbiosis with arthropods. Just demand a new discussion about the state of carnivory, and if you like to get more information on that subject, just have a look on a new CP footage described below.

### A new documentary by Thomas Carow features the phenomenon of symbiosis on CP

In summer 1996 the German TV (ZDF, 'Naturzeit') showed a new CP documentary by Thomas Carow (Germany). I mention that film on this place, because Thomas told me recently that it is now translated to the English language and will be shown by an American TV station soon. The film was produced with rented camera teams, very expensive special optics and without exaggeration it reaches in parts the top quality of David Attenboroughs outstanding documentary 'The Private Live of Plants'. The German title (translated) 'Deathtrap or Life donator' is well chosen, because some of

the 'deathtraps' suddenly appear as sophisticated miniaturized habitats of several different, very specialized organisms. A considerable comment on the footage, 'Some of those plants produce more life than they take'. Beside Capsid Bugs on both South African Roridula species and a Western Australian tuberous Drosera, the additional part of specialized spiders is shown in excellent pictures, not only on Roridula, on which a new Funnel spider specie was found during the camera work, but also on Nepenthes bicalcarata. Here the spider Misumenops nepenthicola fixes a threat at the peristome of the pitcher and then ropes down to dive into the digestive liquid chasing for mosquito larvae developing inside that enzyme bath. Dangerous, because without it's threat it would neither be able to get up through the liquid surface nor to leave the pitcher on the slippery walls. But that is not all about mutualism on this pitcher plant with it's 'snake fangs' beneath the lid. Even if you still know that ants are resident inside a hollow thickening (the breeding chamber) of the tendril between leafblade and pitcher, the performance of this small population of some 20 Camponotus schmitzii is really unique. They live well protected beneath the horny peristome at the inside of the pitcher and do not only walk on the slippery walls (where Misumenops survives only with her threat) without problems. They also swim and dive into the liquid to chase for the mosquito larvae, even in team work. Knowing about this miniature habitat it is remarkable that the pitchers of Nepenthes bicalcarata are more long-lived (sometimes between one and two years inside my greenhouse) than on most other pitcher plant species. But nature provides not only symbiosis, Thomas Carow also describes the parasitism of the moth *Exyra semicrocca* which is able to ruin whole *Sarracenia* sites.

If you are curious to view the film now, you will certainly not be disappointed because there are some more interesting features about CP and mutualism on it, e.g. the mentioned crawfish *Geosesarma malayanum* on *Nepenthes ampullaria*. But last not least there is another important feature on the film. Thomas Carow can be proud for his merit in protecting the natural habitats of CP by the development of methods for artificial propagation of the Venus Flytrap (*Dionaea*) and e.g. *Drosera regia*. Both species are shown on their growing sites and at his successful CP- nursery in Nüdlingen (Germany).

# Conclusion

On closer inspection the phenomenon of symbiosis on CP needs to be considered more important concerning the assignment of carnivory as well as the ability to produce protheolytic enzymes. Misjudged to be a strange exception in the past it appears clearly now that mutualism and symbiosis is not only a widespread behavior of CP, it is in some cases an integrated part of the digestion mechanisms. On all known 'CP' without enzyme production mutualism with arthropods is observed. I hope that the new examinations on *Byblis liniflora* and *Roridula gorgonias* trigger an interesting discussion on the subject.

A botanical Shakespeare could say: 'To digest or to let digest, that's the question (of carnivory)'.

### EXAMPLES OF PLANT-ANIMAL MUTUALISM ON CP (CERTAINLY NOT COMPLETE)

CP	Arthropod	Appearance
Byblis gigantea	Setocornis bybliphilus	Southwest Australia
		(Perth)
Byblis liniflora	Setocornis species	North Australia
		(Kununurra & Cairns)
Darlingtonia californica	Metriocnemus edwardsi	USA
Drosera erythrorhiza	Cyrtopeltis droserae	Southwest Australia
Drosera pallida	Cyrtopeltis russelli	(Perth)
Drosera stolonifera		
Drosera indica (different variations)	Setocornis or Cyrtopeltis species	Northern Australia
		(Kununurra & Darwin)
Drosera ordensis	The smallest Miridae species the author knows, very good adapted in color and size.	Northern Australia
Heliamphora species	Several mosquito larvae	Venezuela
Nepenthes bicalcarata	Camponotus schmitzii	Borneo
	Misumenops nepenthicola	
	Thomisus nepenthiphilus	
	Mosquito larvae (name = ?)	
Nepenthes species mostly	Several different mosquito larvae	Asia, Australia, Madagascar, Seychelles
Roridula dentata	Pameridea marlothii	South Africa
Roridula gorgonias	Pameridea roridulae	South Africa
Sarracenia flava	Sarcophaga species	USA
Sarracenia purpurea	Wyeomyia smithii	USA, Canada

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