Arthropod cuticle features and arthropod monophyly

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Summary. The following synapomorphic features of the arthropod cuticle: presence of α-chitin, similarity of codons for amino acids in cuticle protein, and occurrence of resilin, are cited as evidence for the monophyletic origin of current arthropod classes.

There has been a revived interest in the phylogeny of Arthropoda recently[1]. As a result of embryological studies[2], and functional morphological studies[3], some authors have concluded that the arthropods are a polyphyletic assemblage with 3 separate origins. Cisne[4] proposed a diphyletic origin of arthropods based on studies of the external and internal anatomy of trilobites. Boudreaux[5] concluded that the Arthropoda constitute a monophyletic group. In a table[5] he lists a series of shared apomorphic characters common to the arthropods, the first 4 dealing with cuticle biochemistry. I wish to add several more cuticular biochemical similarities in the 3 main lines of arthropod taxa.

In those cases where it has been crystallographically determined, the chitin in arthropod exoskeleton is α-chitin[6]. Certain insects are capable of forming the other known crystallographic forms of chitin (β and γ), but these are known from cocoon silk[7,8], not from exoskeleton. Thus the presence of α-chitin as the only form of chitin in the arthropod cuticle is a shared apomorphy among the arthropod taxa (and also in the Onycophora[9]).

The amino acid composition of cuticular proteins of arthropods and onycophorans have been compared[10]. There is a strong resemblance between the amino acid compositions of non-specialized soft cuticle proteins in those representatives of Chelicerata, Crustacea, Insecta, and Onycophora analyzed. This similarity is based on the 2nd letter of the codon for the individual amino acids. Annelid cuticle proteins typically have collagen[5], which although present in arthropods[11], is absent in arthropod cuticle[10]. Materials testing has also demonstrated that the mechanical properties of crustacean and insect cuticle is similar[12].

The unusual cuticular protein, resilin, is also restricted to arthropods, and is known from chelicerates[13], crustaceans[14], and insects[14].

These derived cuticular characters are unique to the arthropods, and their close relatives the Onycophora. In conjunction with the characters listed by Boudreaux[5], they imply that whatever radiation took place after the evolution of the Arthropoda, the current classes of arthropods stem from a common ancestor with a typical arthropod cuticle. The number of synapomorphies (relative to other phyla) make it unlikely that the arthropod classes arose independently from separate annelid ancestors.

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Pistia stratiotes L. in Nigerian waters

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Summary. Pistia stratiotes L., a perennial water weed found in natural water bodies in Nigeria, removes organic and inorganic nutrients as it grows and spreads in the waters. In a lake which received a variety of pollutants, the plant reduced oxidizable organic matter (4-h permanganate value) by 61%, the biochemical oxygen demand by 85.3%, ammonia nitrogen by 95.3% and water soluble phosphorus by 85%. The plant also removed other mineral matter.

Most aquatic ecosystems represent some of the most highly productive communities of the world, and require intensive investigation. In the tropics particularly, the development of aquatic weeds is favourable because there is plenty of sunlight and no growth retardation due to a winter season, as is common in other climatic zones. These factors, together with the availability of nutrients, are responsible for the excessive growth of the weeds which results in water bodies being filled up very quickly. Also, the reproductive potential of certain tropical weeds is enormous[1]. It is estimated that 10 plants of the water hyacinth *(Eichhornia crassipes)* can multiply to 600,000 in only 8 months[2]. An account of the occurrence of *Pistia stratiotes* L. in a body of water receiving organic pollutants, and the influence of this weed on the removal of nutrients from the water, is presented in this communication.

Influence of the growth of *Pistia stratiotes* L. on the removal of nutrients from the water

Characteristic	Influent from Awba stream	Sample at a distance of about 250 m after passing through *Pistia* zone
Temperature (°C)	27.4	27.4
pH value	7.3	7.5
Total solids (mg/l)	496	379
Dissolved oxygen (mg/l)	2.3	4.6
Biochemical oxygen demand (mg/l)	16.3	2.4
Ammonia N (mg/l)	3.0	0.14
Nitrate N (mg/l)	Nil	Nil
Water soluble phosphorus, PO_4 (mg/l)	0.2	0.03
Total alkalinity (mg/l)	116.3	95.1
Total hardness (mg/l)	114.8	87.8
Calcium, Ca (mg/l)	28.9	23.2
Magnesium, Mg (mg/l)	17.6	14.1
Sulphate, SO_4 (mg/l)	42.8	30.2

In Nigeria, *Pistia stratiotes* L. is a common aquatic weed which was first recorded by Cook[3] in 1965 at Kainji Lake, one of the major man-made lakes in the country. He drew attention to the fact that this plant could become a problem and a potential hazard to bodies of water. It is also found in large masses in other lakes and rivers all over the country. According to Holms et al[4], it is most troublesome in Africa, but is also a serious problem elsewhere.

In Ibadan, which is one of the largest cities in Nigeria, and West Africa as a whole, this plant had an explosive growth at various sites. One such site, which was chosen for the present study, is the Awba Lake west of the campus of the University of Ibadan. It stretches over an area of about 94,500 m^2, is 700 m long and 135 m wide, with a maximum depth of 5.5 m. The sources of water for the lake are from the Shango and Awba streams, which are supposed to be natural springs from nearby villages that existed before the development of the city. The Awba stream which has a fairly good flow, receives some organic waste from some of the residential halls on the campus, from the zoological garden and from farms along the stream.

Pistia stratiotes L. covered the whole lake in the year 1977–78. However, the staff at the lake actively fought the battle for control by manual work, and they installed a bamboo barrier across the lake which prevented the further spread of the weed on the lake surface. Now the plant is confined to almost one-third of the area (figure). This notorious tropical aquatic weed is a very pale green, free floating perennial plant consisting of a rosette of succulent leaf blades, a short stem axis and a long feathery root system.

It reproduces mainly by vegetative offshoots connected with the mother plant by stolons. Sculthorpe[5] discussed the role of *Pistia stratiotes* in the formation of dense aggregations of free floating vegetation, known as sudds, which in swampy areas have claimed many lives in the Sudan.

The growth of *Pistia stratiotes* L. over a stretch 250 m long and 135 m wide in its natural habitat in Awba Lake has certain features which prompted the present investigation. The plants at one end of the lake, where the Awba stream enters, were strikingly vigorous and large in size, with an average density of 42 plants per m^2. The plants at a distance of about 250 m from the stream inlet in the lake were small, less green and had a density of 223 plants per m^2. The whole area in between was covered by the plants and looked like a carpet. Such a vast difference in the growth characteristics at 2 different sites was investigated by examining the quality of the influent water and the water immediately at the end of the Pistia plants at a distance of 250 m.

The samples were examined at weekly intervals over a period of 4 months, for common pollution parameters: pH value, content of total solids, dissolved oxygen, oxidizable organic matter as represented by 4-h permanganate value,

Awba Lake covered with *Pistia stratiotes* L. The inset shows a single plant. The bamboo barrier prevented the plant spreading in the lake.

biochemical oxygen demand (BOD_5), ammonia nitrogen, nitrate nitrogen, water soluble phosphorus, total alkalinity, total hardness, calcium, magnesium and sulphate. The analyses were carried out according to the standard methods[6,7]. The results (table) indicate that there is a striking removal of various nutrients, which were originally present in the influent water. The permanganate value, BOD_5, ammonia nitrogen, and water soluble phosphorus contents were reduced by 61, 85.3, 95.3 and 85%, respectively. The values for total solids, total alkalinity, total hardness, calcium, magnesium, and sulphate were reduced by 24, 18, 23.5, 19.7, 19.9, and 29.4%, respectively.

The removal of nutrients by aquatic plants is not uncommon. Wolverton and McDonald[2] reported that the water hyacinth *(Eichhornia crassipes)* in a facultative lagoon treating sewage was able to reduce the BOD by 95%, total Kjeldahl nitrogen by 71.7% and total phosphorus by 56.8%. In the treatment of waste water and in water pollution control in general, a variety of microorganisms[8-10] are known to have the ability to remove pollutants. But the involvement of higher plants or weeds is of considerable importance, and *Pistia stratiotes* L. appears to be yet another in the list which may prove beneficial in the removal of nutrients from water. In the lake, at the point of entry of the pollutants, the plants grew vigorously, with an average density of 42 plants per m^2, and removed the nutrients to a great extent. As the concentration of nutrients decreased due to their removal, the plants became less vigorous and grew smaller in size with a density of 223 per m^2 at a distance of 250 m from the entry of pollutants. The plants contained about 94% water and it is commonly believed that the high water content may be uneconomical for possible use as an animal feed. While the experiments on the nutritional quality of the plant were in progress, laboratory experiments using grasshoppers as test animals showed that this plant may prove useful as a fodder or as a feed supplement in a fresh state.

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Spectroscopic study of the disulfide bond in oxidized glutathione

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Summary. Raman and circular dichroism spectra are used for obtaining structural information on the disulfide bridge in oxidized glutathione. Quantitative estimates of dihedral angles and bond angles are proposed.

Several investigations have shown that stereochemical parameters of the disulfide bond can be obtained by electronic and vibrational spectroscopy, in particular circular dichroism (CD)[1-4] and Raman scattering[5-9]. In order to extend spectra-structure correlations to disulfide bridges in proteins further work with simple peptides is necessary. On this subject we present a study of oxidized glutathione (GSSG) in solid phase and in aqueous solution by Raman and CD spectroscopy.

Raman spectra were recorded on a 25-300 Jarrel-Ash Laser Raman spectrometer. Characteristic Raman frequencies of GSSG and related disulfides are shown in the table.

CD spectra were obtained with a Cary 60 and they were in full agreement with those reported in the literature[2].

A main parameter describing the geometry of disulfide bonding is the dihedral angle χ (CS-SC), which is related both to the frequency of the sulphur-sulphur stretching vibration[6-9] and to the frequencies of electronic transitions of the disulfide chromophore[6,10]. In the Raman spectra of GSSG sulphur-sulphur stretching ν (SS) is observed at 512 cm^{-1} in the crystal and 514 cm^{-1} in aqueous solution (pH 3), suggesting that no major conformational change occurs on solution. According to an early proposal of Van Wart et al.[6], the value of ν (SS) can be related to the dihedral angle χ (CS-SC) by a linear relationship

$$\chi \text{ (CS-SC)} \simeq [2.2\ \nu \text{ (SS)} - 1060] \text{ degrees}$$

(with ν (SS) in cm^{-1}) which gives a value $\chi \simeq 70°$ for GSSG. Yet, recently Van Wart and Sheraga[9] revised their original relationship, restricting its validity from 0° to 65° and accepting the view that in the range 65-90° ν (SS) is practically invariant to χ (CS-SC). On the other hand a further evaluation of the disulfide dihedral angle can be obtained by electronic spectra, since a quasilinear correlation also exists between χ (CS-SC) and the energy of the lowest electronic transition of the disulfide chromophore–at least in simple molecules[6,10]. In GSSG the wavelength of the first electronic absorption, as seen in CD spectra (near UV absorption, spectra do not show any resolved peak), is about 260 nm, corresponding to a dihedral angle of 75°. This rather good agreement between Raman and CD results suggests that for the χ (CS-SC) angle of GSSG a value of about 70-75° can be confidently accepted; this

Characteristic Raman frequencies in GSSG and model disulfides

		(S-S) cm^{-1}	(C-S) cm^{-1}
Oxidized glutathione	(solid)	512	670
	(solution)	514	670
L-cystine	(solid)	500	680
	(solution)	507	666
L-homocystine	(solid)	510	640
	(solution)	509	640
Cystamine-2 HCl	(solid)	511	643
	(solution)	510	640