Morphological requirements in limulid and decapod gills: A case study in deducing the function of lamellipedian exopod lamellae

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According to one hypothesis, the exopods of extinct lamellipedian arthropods functioned as gills. To evaluate this hypothesis, the growth rates in *Limulus polyphemus* for total gill surface, average area per single gill lamella and number of gill lamellae are documented. The rates are compared with corresponding rates in decapod crustaceans in order to make deductions on morphological constraints in multi-foliated gills. The growth rates are given as allometric scaling exponents relative to the animal dry-body weight. The comparisons reveal that each allometric exponent is similar among examined species irrespective of differences in gill morphology or animal body plans. The numerical growth of lamellae obviously is much smaller than the growth of the total respiratory surface. To fulfill these trends in multi-foliated gills, the overall profile tends to become conical, with the result that the surface area is a couple of magnitudes larger at the base of the cone than at the tip. This geometrical shape appears to keep the numerical value of the total respiratory area (total lamellar surface) proportional to the cube of the total number of lamellae. The situation is entirely different in animals with lamellipedian exopods. In the latter, lamellae are slender structures carried in a straight row and, as exemplified by *Naraoia*, their increase in number during the growth is only half that required for the exopod lamellae to have functioned as an arthropod multi-foliated gill cone.

Key words: Arthropoda, Limulidae, Decapoda, allometrty, gills, respiration, functional morphology.

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Introduction

Lamellipedian arachnomorphs (in the sense of Hou and Bergström 1997 and Edgecombe and Ramsköld 1999) are an extinct group of arthropods (including the Trilobita), which is characterized by a wide pleural field and semipendent laterally deflected appendages (Hou and Bergström 1997: 42). The latter comprise a pair of antennae and more or less uniform trunk-type biramous limbs throughout its post-antennal body (Hou and Bergström 1997: 42). Both body and limbs tend to decrease in size towards the posterior end (Hughes 2003). The outer branch of the appendages, the exopod, carries structures that are known from the literature as either filaments, lamellar setae, or lamellae. These are flat, comparatively long, blade- or sword-like in shape, and generally arranged in a comb-like fashion in a regular row along one edge of the exopod (e.g., *Kuamaia lata*: Hou and Bergström 1997). Rarely, they occur in two rows (e.g., *Xandarella spectaculum*: Hou and Bergström 1997). Despite the increase in knowledge of the lamellipedian appendages during the last three decades, the functional interpretation of the structure is still controversial. Two major interpretations have been advocated. In one, they had a respiratory function. Accordingly each lamella would be part of a lamellar gill (e.g., Whittington 1975; Bruton and Haas 1999). In another interpretation the structures are setae, which could have ventilated the gills (Bergström 1973; Hou and Bergström 1997; Edgecombe and Ramsköld 1999), or also have had other mechanical functions, for instance digging (Seilacher 1970; Bergström 1976). The gill interpretation is based on the purported similarity in shape between lamellipedian lamellae and extant arthropod gills, particularly those of limulids, and the now outdated belief that the trilobite exopod would be homologous with the crustacean epipodite gill. Since the need of respiratory exchange is likely related to body volume and not to linear or even areal size of the animal, volumetric scaling relations should also be taken into consideration in the evaluation of this idea. However, descriptions of such relations in limulids are very limited. So far as we are aware, there are only two relevant comments. Manton (1977: 141) mentioned that there are about 150 lamellae in a branchial appendage. In the other account, Shuster (1982: 31) stated that an adult specimen with a 28 cm wide prosoma was estimated to have a total of about 1,550 gill lamellae, and that the total surface area is about 11,600 cm^2. However, we are not told about how the number of lamellae, or the total surface area, changes with body size.

In general, the scope of an important biological process (e.g., respiration) and its morphometric character (e.g., total gill surface area, number of gill lamellae) are known to be related to the body size (e.g., tissue volume, body weight; Schmidt-Nielsen 1984). This means that an organ such as the arthropod multi-foliated gill should fulfill particular morphological conditions to satisfy the above relationships, irrespective of the differences in their profiles, extent of foliations, positions within the body and so on. The first goal of this study is to clarify how the morphology of the arthropod multi-foliated gills changes in order to maintain a constant relationship between respiratory surface and body weight. This is examined by comparing allometric growths in gills of extant decapod crustaceans and limulids. The second goal is to check whether or not the lamellipedian exopods had an allometric growth that would have made a gill function possible throughout animal growth.

We first document growth-related morphological changes and allometric growth in total respiratory area, average lamellar area and number of gill lamellae of *Limulus polyphemus* Linnaeus, 1758. Reference is then made to published studies of allometric growth of these characters in marine decapod crustaceans.

Material and methods

The extant limulid used in the present study is *Limulus polyphemus*. The technical terminology follows Yamasaki et al. (1988) (Fig. 1). The respiratory organs in limulids are book gills, which are situated on the dorsal side of the opisthosomal branchial appendages (Fig. 2A). Tens of thin lamellae diverge from the base of the plate-like division of the branchial appendage, which clearly exhibits endo- and exopod divisions (Fig. 2A). The term operculate division is here used for a plate-like division of the branchial appendage, not to the gill lamellae. Each gill lamella exhibits a darkly pigmented elliptic area in the center (Fig. 2C). From an ultrastructural study, this is known as an osmoregulatory

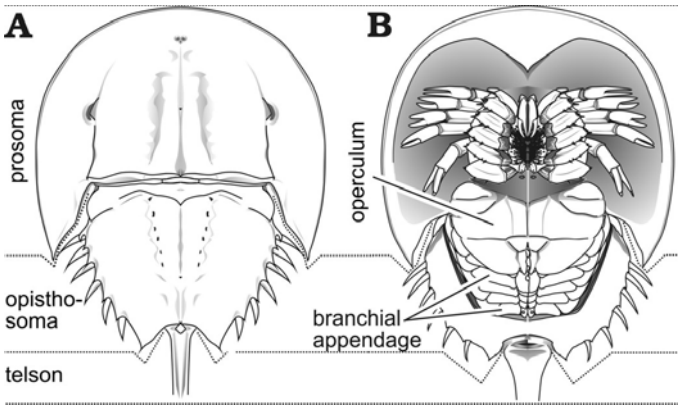


Fig. 1. Dorsal (**A**) and ventral (**B**) morphology of *Limulus polyphemus*.

area (Henry et al. 1996). Additional tissue mass is needed for osmoregulation, and the area where it occurs therefore tends to be comparatively thicker (Henry et al. 1996) and to be darker dyed than the surroundings. The surrounding area is respiratory (Mangum 1982).

Before examination, but after measuring some tergite characters, specimens were fixed and preserved in 2% formaldehyde solution. The measurements serve for the estimation of the instar stage of a sample. This estimation is based on data provided by Sekiguchi et al. (1988: tables 4-4, 4-6). Firstly all branchial appendages were cut off from the body. Of these, the limbs of the right side were used for calculation of area and for counting the number of lamellae. Every gill lamella was mechanically separated with a scalpel. The lamella was then mounted between a bi-lamellar transparent film, digitally scanned, and transformed into pixel data. Then the area for respiration and osmoregulation were calculated separately by using the free software "area calc". The gill lamellae of several earlier instar stages were too small to be individually separated. We therefore just counted the number of lamellae and made observations by Scanning Electron Microscope (SEM). To calculate a total dry-body weight, a body without branchial appendages and the left appendages

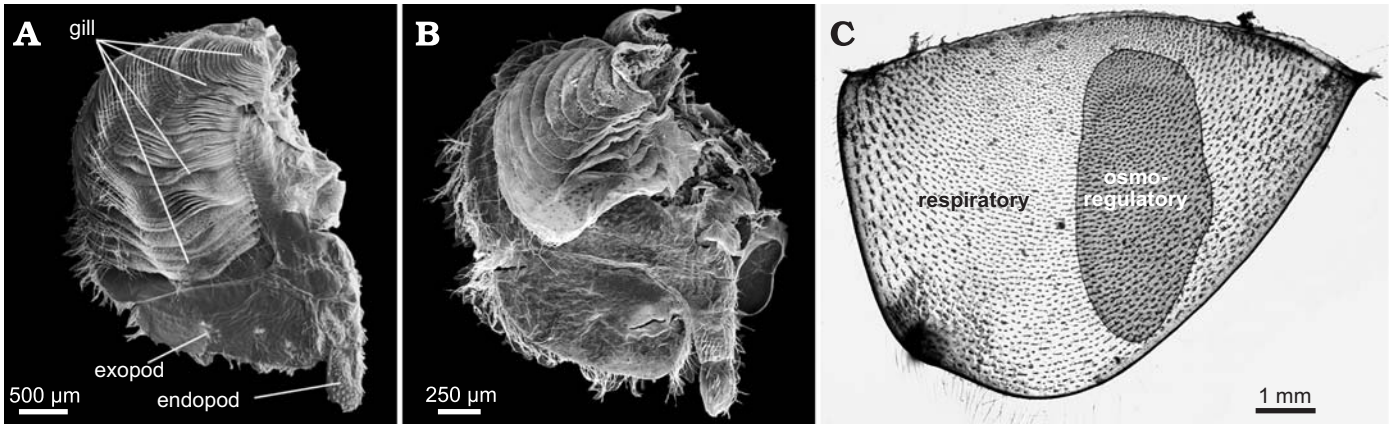


Fig. 2. Book gill of *Limulus polyphemus* Linnaeus, 1758. **A**. Dorsal view of left first branchial appendage of instar stage 14. Endopod and exopod of the operculate division of branchial appendage as well as lamellae of book gill are shown. SEM photo. **B**. Dorsal view of left first branchial appendage of instar stage 4. SEM photo. **C**. Transparent microscopic photo of a gill lamella dyed with toluidine blue, showing osmoregulatory and respiratory area.

Table 1. Number of gill lamellae and their surface area in *Limulus polyphemus*. Abbreviations: *R* = total respiratory area; *O* = total osmoregulatory area; < = less than the numerical value shown to right; br1 ~ br5 = first to fifth branchial appendage pair; — = impossible to judge or examine; × = not present; m = male; f = female.

Dry-body weight (g)	Instar stage	Sex	Prosomal width (mm)	Number of gill lamellae						Area of gill surface (cm^2)		
				br1	br2	br3	br4	br5	Total	*R*	*O*	Total
<0.01	1	–	3.21	8	12	0	x	x	20	–	–	–
<0.01	2	–	4.44	18	10	2	x	x	30	–	–	–
<0.01	2	–	–	16	18	2	x	x	36	–	–	–
<0.01	3	–	–	20	22	12	0	x	54	–	–	–
<0.01	3	–	6.65	22	24	14	0	x	60	–	–	–
<0.01	4	–	–	24	30	26	14	0	94	–	–	–
0.01	4	–	7.0	30	36	24	12	0	102	0.28	0.07	0.035
0.18	8	m	24.1	98	104	100	86	62	450	7.96	2.33	10.28
0.27	8	f	24.1	64	100	94	78	60	396	7.46	1.86	9.32
0.31	9	m	28.5	100	112	104	90	72	478	13.51	3.1	16.61
0.31	9	m	33.2	104	110	114	94	74	496	16.37	3.64	20.00
0.43	10	m	39.7	138	140	140	120	98	636	30.98	8.88	39.86
0.44	9	m	39.0	110	116	116	98	78	518	15.20	3.49	18.69
0.59	9	f	31.1	112	116	110	96	72	506	13.27	3.53	16.79
0.73	10	f	39.0	128	142	138	120	100	628	30.07	8.35	38.42
1.03	11	f	41.6	138	154	138	130	114	674	38.15	9.93	48.08
1.04	11	f	43.4	134	144	140	126	98	642	37.33	9.85	47.18
1.12	11	f	46.5	150	166	158	142	116	732	50.12	12.69	62.81
1.19	11	f	43.2	140	156	150	132	112	690	36.97	12.44	49.41
1.60	12	f	52.5	162	180	172	152	128	794	63.56	16.48	80.04
1.62	12	m	59.6	170	196	190	172	136	864	85.44	25.10	110.54
1.91	12	m	56.7	174	184	176	160	132	826	70.65	20.48	91.13
3.65	13	f	67.8	192	206	204	186	156	944	139.10	33.46	172.56
3.68	12	m	57.5	163	184	180	158	130	815	82.87	22.39	105.27
6.70	14	m	78.2	204	230	226	206	170	1036	192.59	50.57	243.17
8.93	13	f	68.8	174	194	182	166	142	858	107.88	26.39	134.27
76.08	16	m	144.0	230	254	244	220	170	1118	821.25	210.29	1031.54
87.17	17	m	150.6	276	304	292	278	230	1380	1094.55	399.32	1493.87
90.34	16	m	151.7	266	282	276	244	204	1272	1256.63	436.77	1693.40
135.98	17	m	171.0	262	282	274	254	202	1274	1409.21	453.14	1862.34
177.09	18	f	184.0	282	312	308	290	216	1408	1725.53	599.49	2325.02
261.63	18	f	198.0	272	298	296	274	226	1366	2176.03	765.69	2941.72

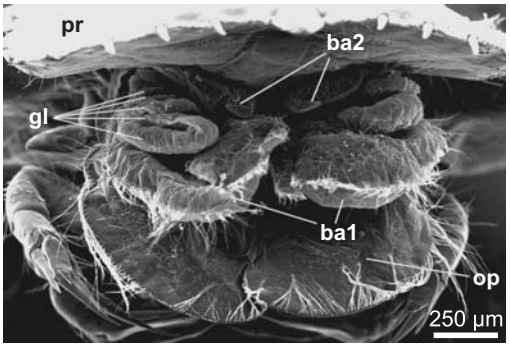


Fig. 3. Posterior view of first instar stage of *Limulus polyphemus* Linnaeus, 1758. Only five gill lamellae (gl) are visible between the operculate division of first (ba1) and second branchial appendage (ba2). Other abbreviations: op, operculum; pr, prosoma. SEM photo.

were separately kept in a desiccator at 60° centigrade until completely dry. The weight of the left appendages was doubled and added to that of the body.

Results

Growth-related changes in gill morphology of *Limulus polyphemus*.—Dry-body weight, number of gill lamellae in each branchial appendage pair, total number of lamellae, total area for respiratory, osmoregulatory and whole-lamellar surface for each examined sample with the information on the prosomal width and estimated instar stage are listed in Table 1. Each gill lamella exhibits a more or less similar sub-trapezoid shape in instar stage 14 (Fig. 2A) and instar

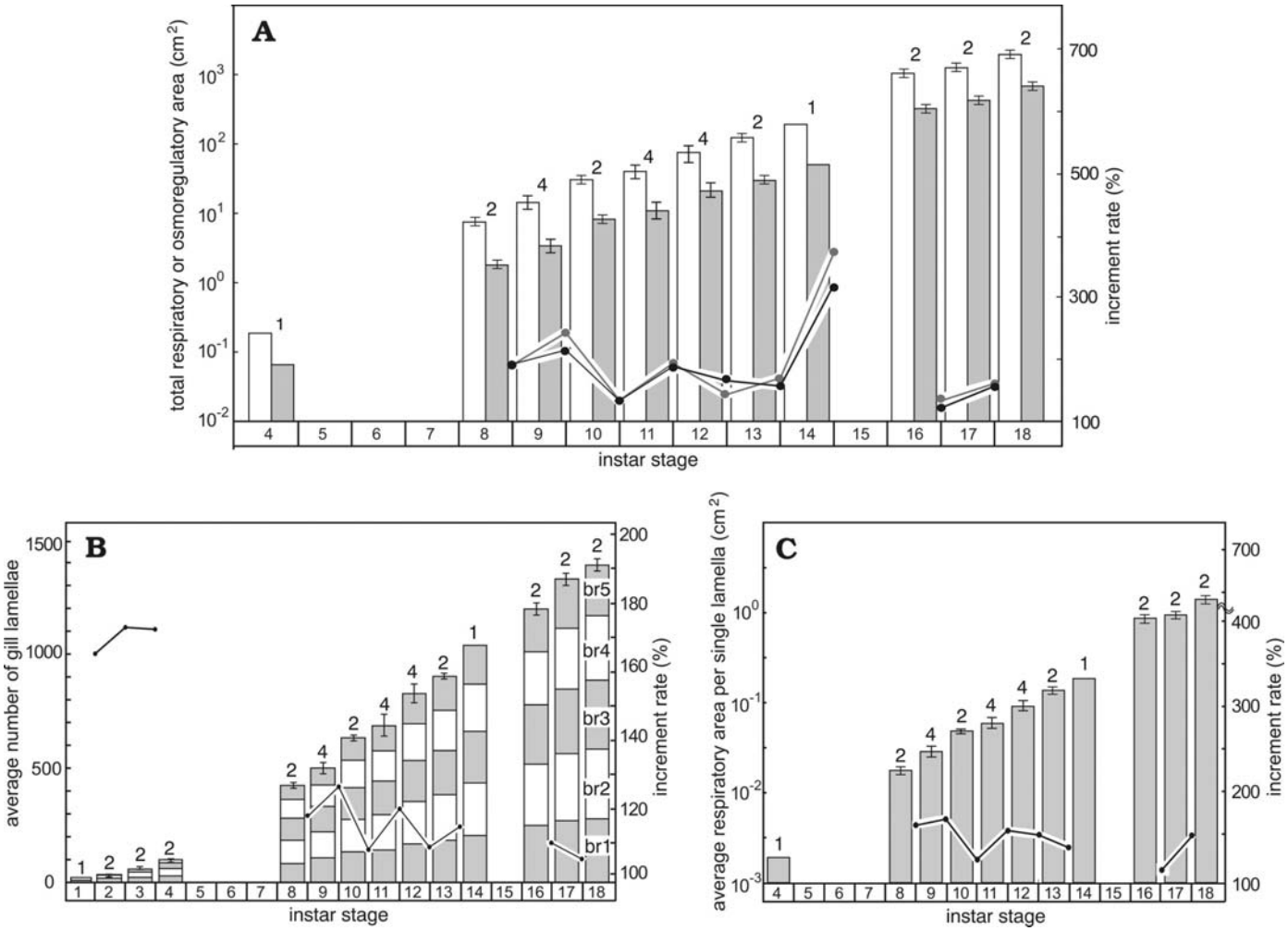


Fig. 4. Growth-related change in gill morphology of *Limulus polyphemus* shown in instar-stage series. **A**. Mean total respiratory (white bars) and osmoregulatory area for each instar stage (grey bars) and their increment rates (black and grey line graph denotes respiratory and osmoregulatory area, respectively). **B**. Average total lamellar number for each instar stage (bar graph) and its increment rates (line graph). **C**. Average area per single lamellae for each instar stage (bar graph) and its increment rates (line graph). The bar graphs should refer to left indexes shown in exponential form (**A** and **C**) or in actual numbers (**B**), and the line graphs right indexes. Error bars denote the maximum and the minimum lamellar numbers. Numbers shown above the columns represent the numbers of examined specimens.

stage 4 (Fig. 2B). The stack of gill lamellae looks like a low relief pyramid. The size difference between adjacent lamellae becomes smaller with an increasing number of lamellae (compare Fig. 2A and B). Some lamellae situated dorsally, and thus at the top of the pyramid where the lamellae are smallest, lack an osmoregulatory area. This applies to all the examined samples of branchial appendages. The number of such lamellae increases toward the growth stage 14, and gradually decreases in the later growth stages.

As briefly commented on by Yamasaki et al (1988: 93), the first instar stage possesses only three pairs of branchial appendages. The most posterior pair lacks gill lamellae, and thus consists of only operculate divisions. Branchial appendages without gill lamellae were also recognized in the fourth and the fifth pairs in instar stages 3 and 4, respectively. The number of gill lamellae in instar stages 1 to 4 is comparatively small. The first instar stage has a total of only twenty gill lamellae, which means around five gill lamellae in each limb of the two anterior pairs (Fig. 3). Among the examined specimens, the largest total number of lamellae is 1366. The total respiratory area ranges from 0.28 cm^2 (instar stage 4) to 2176 cm^2 (instar stage 18). An instar stage 1 appears to have 20 lamellae, the calculated total respiratory area of which is about 1.4×10^{-6} cm^2. There is thus a range spanning at least three orders of magnitude in the total number of gill lamellae, and a range of more than nine orders of magnitude in the total respiratory area in *Limulus polyphemus*.

Changes in total respiratory area, number of gill lamellae, and average respiratory area per single lamella with an instar series are shown on diagrams in Fig. 4. All increments (line graphs with their indexes on the right axis) tend to decrease with growth. This is especially clear in the case of the number of gill lamellae (Fig. 4B). This is because there is an exponential increase in the total respiratory area and the average single lamellar area with body growth (Fig. 4A, C: bar graphs with their indexes on the left), while the number of

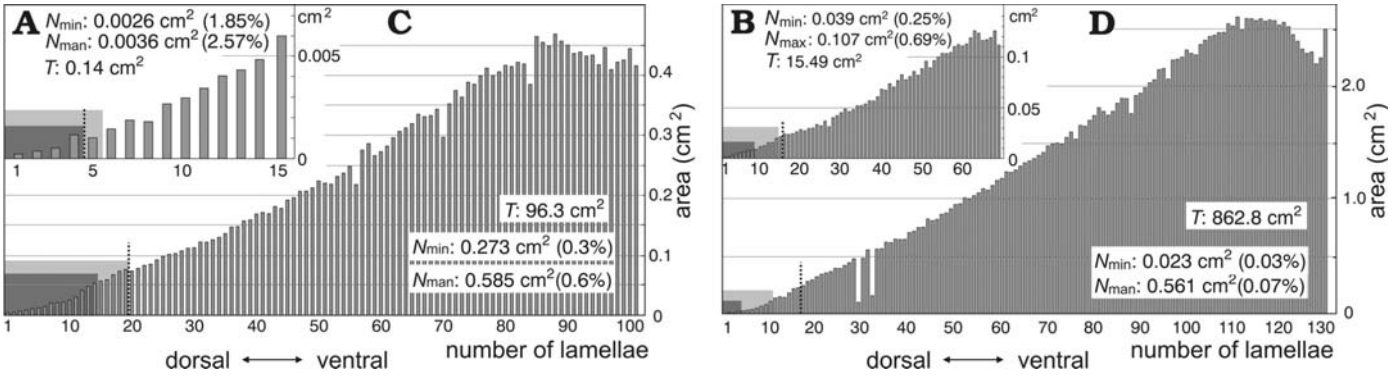


Fig. 5. The area of every gill lamella of selected first branchial appendages. **A**. Instar stage 4 of dry-body weight 0.01 g. **B**. Instar stage 10 of dry-body weight 0.43 g. **C**. Instar stage 14 of dry-body weight 6.7 g. **D**. Instar stage 18 of dry body weight 177.09 g. Grey shaded area corresponds to possible newly established lamellae in each instar stage. Darker shade ranges to minimum established number, while lighter maximum. Total respiratory area (T), respiratory area for newly established lamellae of minimum (N_{min}) and maximum value (N_{max}) are also noted. These ratios relative to the total area are shown in parentheses. The lamellae left to dashed lines lack an osmoregulatory area.

lamellae increases linearly (Fig. 4B: bar graph with its index on the left). This means that the establishment of new lamellae is much less important than the areal expansion of pre-existing lamellae. The graphs in Fig. 5 support this interpretation. They show the area of every gill lamella in the first branchial appendages of the instar stages 4, 10, 14, and 18 (Fig. 5A, B, C, and D, respectively). Newly established lamellae are shown in the shaded area in grey color. They constantly become smaller in relation to the whole as growth proceeds. Since the number of newly established lamellae is more or less constant, the ratio of these to the total area or numbers obviously decreases with growth. In addition, most of the newly established lamellae lack an osmoregulatory area. The osmoregulatory tissues apparently form some time after the establishment of new lamellae.

Allometric scaling of gill characters in *Limulus polyphemus*.—Growth-related morphological changes can often be expressed in an allometric formula with respect to changes in body weight:

Total respiratory area:

$$\log A = 0.776 \log W + 3.54 \; (A = 34.48 W^{0.776})$$

Number of lamellae:

$$\log NL = 0.165 \log W + 6.46 \; (NL = 638.4 W^{0.165})$$

Average area per lamella:

$$\log AL = 0.615 \log W - 2.92 \; (AL = 0.054 W^{0.615})$$

W is the value of the dry-body weight, A is the value of the total area for the respiratory surface, NL the total number of gill lamellae and AL the average area of single lamella of an individual sample. 0.776, 0.165, and 0.615 are the allometric scaling exponents (shown as α in Fig. 7), which appear as the slope of the log/log regression line, using the reduced major axis (RMA) regression. The exponents mean the degree of change with respect to the weight increase along the growth. To keep the respiratory area large enough during growth, the average gill area per lamella grows three times faster than the number of lamellae. This is in accord with the results shown in the previous section.

Discussion

Comparison with allometric characteristics of decapod gills: total respiratory area.—To see how the total respiratory area is related to animal weight and behavioral activity rather than to body plans or gill morphology, the allometry of the total respiratory area in *Limulus polyphemus* was compared with that of two decapods studied previously (Hughes, 1983). The decapods in question are *Callinectes sapidus* and *Libinia dubia*. The former, the so-called blue crab, is a fairly active animal (Gray 1957) with high swimming ability (Luckenbach and Orth 1992). The latter, the decorator crab, is a sluggish bottom dweller, which often places seaweeds onto the dorsal carapace as camouflage (Stachowicz and Hay 2000) probably to compensate for its inability to escape from predators by rapid movement. The former has eight pairs of gill branches, the latter nine. The type of gill found in these decapods is tall and narrow, like a spearhead, and is generally known as a phyllobranchiate gill (Fig. 6). The limulid gill, on

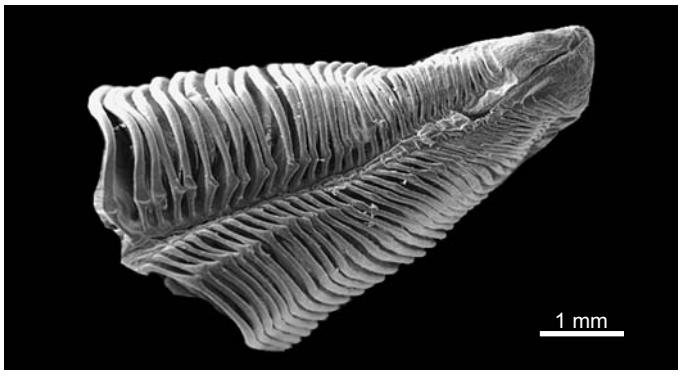


Fig. 6. Phyllobranchiate gill of a decapod crustacean *Atergatris* sp. Top one-fourth is shown. SEM photo.

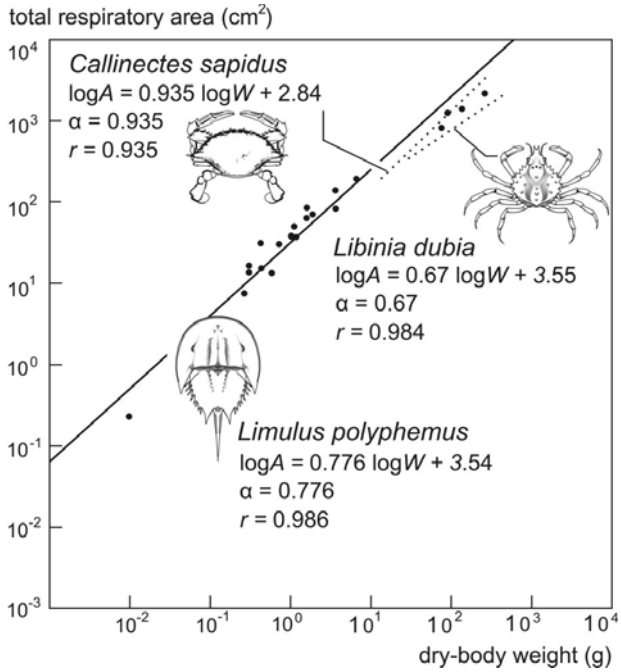
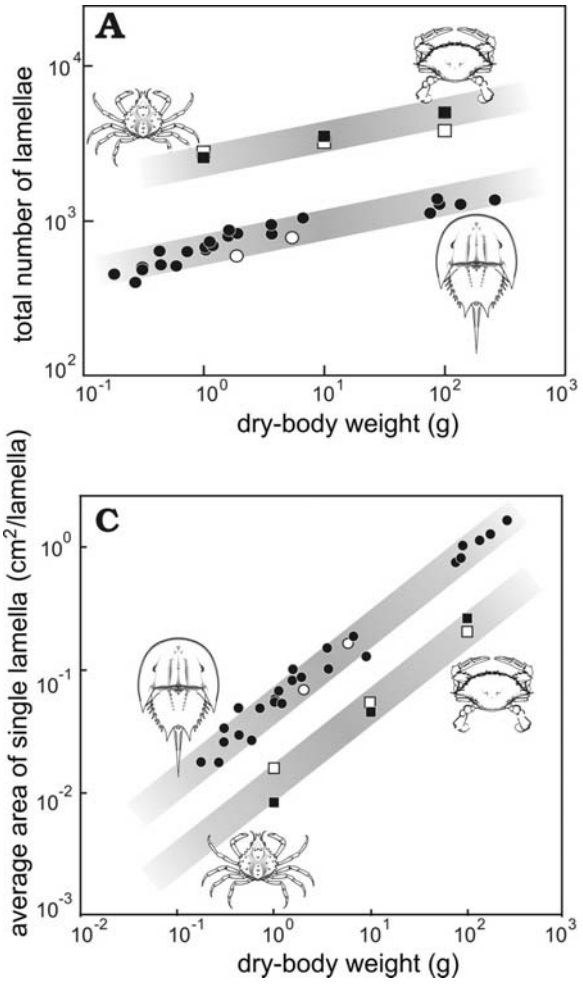


Fig. 7. Allometric relationships between respiratory surface and dry-body weight in *Limulus polyphemus* (dots and solid regression line) in bi-logarithmic coefficients. Abbreviations are: correlation coefficients (*r*); dry-body weight (*W*); total area for respiratory surface (*A*); allometric scaling exponent (α). For comparisons, the results on the gills of decapod crustaceans *Callinectes sapidus* and *Libinia dubia* are shown in dashed lines, the data are referred to Hughes (1983).

the other hand, is short and wide, with a shape reminiscent of an Egyptian pyramid (Fig. 2). Despite the great phyletic distance, the marked differences in body plans and in the profile of gills, the allometric exponent of *L. polyphemus* appears roughly similar to that of two decapods (see Fig. 7).

Comparison with allometric characteristics of decapod gills: total number of gill lamellae and average respiratory area of single lamellae.—How are then the rate of new gill lamella establishment and the rate of gill lamella expansion related in order to maintain a body-weight specific total respiratory area in decapods? Bi-logarithmic graphs (Fig. 8A, C) show the total number of gill lamellae respectively and the mean area of a single lamella with respect to the dry-body weight of *Limulus polyphemus* and the two marine decapods (same as in Fig. 7). The allometric-scaling exponent for the establishment rate of new lamellae is 0.165 for *L. polyphemus*, 0.156 in *Callinectes sapidus* and 0.083 in *L. dubia* (see α in Fig. 8B). The results on another limulid species, *Tachypleus tridentatus,* are also shown, and appear consistent with those of *L. polyphemus* (Fig. 8A and C: open circles). With these, the difference of the allometric-scaling exponents of *L. polyphemus* and *C. sapidus* appears not statistically significant at the 5% level of confidence (see *K* values in Fig. 8B). The allometric-scaling exponents of the lamellar area are 0.617, 0.778 and 0.587 in the descending order (see α in Fig. 8D). In these, *L. polyphemus* and *L. dubia*, *C. sapidus* and *L. dubia* appears not to differ significantly from each other (see *K* values in Fig. 8D). The two decapods dealt with here represent the extremes of the eight species shown in Hughes (1983). This means that decapods in general, and not only the species examined in Hughes (1983), should show allometric scaling exponents much similar to that of *L. polyphemus*. Thus, despite the great differences in the overall body plans and gill profiles between decapods and limulids, the growth rate of lamellar area and lamella numbers against body weight is much the same in the different animals. The obvious differences in the intercept values between limulids and decapods then represent the differences in the profile of the multi-foliated gills. In the process of acquiring enough respiratory surface with growth, the enlargement of lamellar surface stands for three-to-six times more of the needed modification than the establishment of new lamellae. How this relationship is accomplished in arthropod multi-foliated gills of different profiles is discussed in the following.

Morphological constraints in multi-foliated gills.—Although the height/diameter ratio differs considerably between limulid and decapod gills (less than 1 in limulids and more than 10 in decapods: compare Figs. 2A and 6), there is at least one common morphological character. This is the more or less conical shape of the multi-foliated gills. In fact, using an allometric scaling of a conical shape is conceptually in accord with the growth rates of the total number of gill lamellae and the lamellar area. As the gill lamellae are here regarded to represent horizontal sections and these keep their shape while enlarged, the shape would become a larger pyramid with truncated top (Fig. 9). New similarly shaped sections at a smaller scale (shaded pyramidal shapes in Fig. 9) need to be added onto the top. This mode of areal enlargement of pre-existing lamellae can keep the number of newly established lamellae small. As the size of the structure increases, the newly added cone top, which is constant in the number of sections (lamellae), is successively smaller relative to the total when the animal grows. From geometrical points of view, the total respiratory area of a gill cone is equivalent to its volume, because the area of a horizontal section (represented by a gill lamella) is the differential value at a certain level of the cone. Thus the number of lamellae is proportional to the height of the cone. The rate of increase in total respiratory area (cone volume) is proportional to the cube of the number (cone height), because the volume of a cube is proportional to the cube of its length (Schmidt-Nielsen 1984: 13). From the smallest to the largest stages, the range in number of gill lamellae and in total respiratory area is more than three and nine orders of magnitude, respectively. This difference is in accord with the geometric relationship between the height (number of lamellae) and the volume (total respiratory area) of a conical shape. Since the required volume of the cone (representing the total lamellar surface) is directly related to body weight, a low cone tends to have a large area for each horizontal section (representing the average lamellar area), and vice versa. Limulids with low



B

species	*N*	*r*	*α*	Log*β*	*K*	
					L. dubia	*C. sapidus*
Limulus polyphemus	25	0.95	0.165	6.46	3.66	0.40
Callinectes sapidus	39	0.60	0.156	7.8	13.18	
Libinia dubia	11	0.64	0.083	7.87		

D

species	*N*	*r*	*α*	Log*β*	*K*	
					L. dubia	*C. sapidus*
Limulus polyphemus	25	0.99	0.617	-2.89	0.27	-3.58
Callinectes sapidus	39	0.91	0.778	-2.33	1.70	
Libinia dubia	11	0.82	0.587	-1.72		

Fig. 8. Body-weight specific lamellar numbers and average single lamellar area in bi-logarithmic coefficients. **A**. Bi-logarithmic graph of total number of lamellae with respect to dry-body weight in *Limulus polyphemus* (close circle), *Tachypleus rotundicauda* (open circle), *Callinectes sapidus* (solid square) and *Libinia dubia* (open square). **B**. Results of allometric analysis shown in Fig. 5A. Sample size (*N*), correlation coefficient (*r*), reduced major axis of log$W$ = αlogNL + logβ, and $K = \alpha_1 - \alpha_2 / [(s_{\alpha 1})^2 + (s_{\alpha 2})^2]^{1/2}$ where s_α is the standard deviation of α. K is a statistic with the standard normal distribution used for discrimination of the differences of α significant or not. If K >1.96 or K <-1.96, the difference is significant. See also Fig. 7 for the abbreviations of W and NL. **C**. Bi-logarithmic graph of average area per lamella with respect to dry-body weight. Abbreviations as in Fig. 5A. **D**. Results of allometric analysis shown in Fig. 5C. Same abbreviations as in Fig. 5B. The data of two decapods are referred to Hughes (1983), which presented average, maximum and minimum specific dry-body weight and lamellar number among the examined samples as well as α and logβ of the allometric analysis with respect to their dry-body weight. Readers are referred to the results of *T. rotundicauda* as reference data, because too small numbers have been examined. This is to show the trend that the results of a species fall in a neighboring area to that of a taxonomically close species.

gill cone exhibit larger value than decapods for the average area of single lamella (Fig. 8C), while it is the other way around with respect to total number of lamellae (Fig. 8A).

Then what kind of morphological constraint is required for gill lamellae to keep a conical shape? In the case of *Limulus polyphemus* with a body weight of 0.01 g, the area of the largest lamella is fifty times that of the smallest one (see Fig. 5A), at 0.43 g eighty times (see Fig. 5B), at 6.7 g 153 times (see Fig. 5C) and at 177.09 g 2053 times (see Fig. 5D). Since the area of the smallest lamella in a gill branch is more or less constant and the others become larger as the animal grows, the minimum and the maximum area of a gill lamella in a single gill cone should span more than a couple of magnitudes in order to maintain a cone shape throughout the animal growth. Thus, if the lamellipedian exopod functioned as a gill branch, the span between the maximum and the minimum area of gill lamella should have been more than a couple of magnitudes.

Functional interpretation of lamellipedian lamellae

The camera lucida drawing (Fig. 10A) of the lamellipedian exopod of the trilobite *Olenoides*, as traced from Whittington (1980: text-fig. 6), is one of the best examples of the lamellae attached to a single exopod. The estimated area of each lamella from Fig. 10A is shown on Fig. 10B. The area of each lamella is more or less double scored, but obviously does not

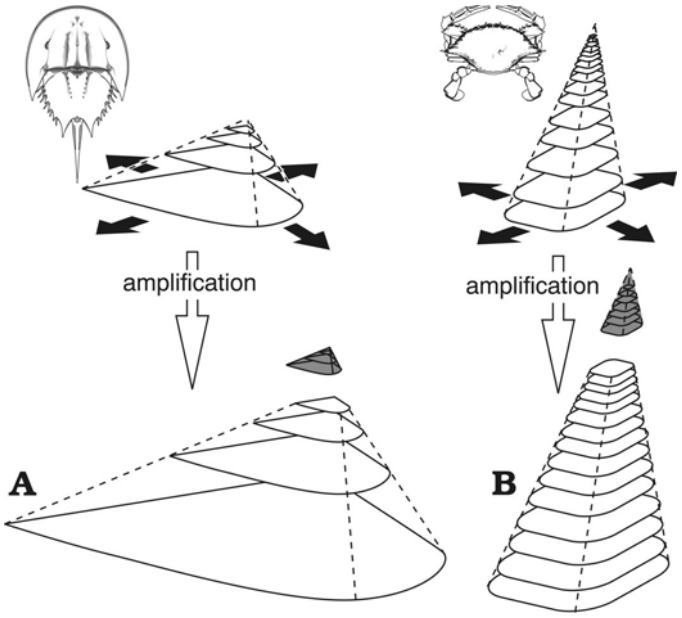


Fig. 9. Conceptual figure showing growth accompanied amplification of pyramidial shaped multi-foliated gills. **A**. Limulid type of gills with low-relief conical profile. **B**. Decapod type of gills with high relief.

span a couple of magnitudes, which is a necessity for a multi-foliate gill.

Despite the new findings of lamellipedian arthropods over the last three decades, the number of lamellae in a single exopod is often around 50 regardless of size. The minimum number is known in the case of *Ceraurus pleurexanthemus* (Trilobita) and *Kuamaia lata* (helmetiid lamellipedian), both of which is 22 (see Størmer 1939; Hou and Bergström 1997). The maximum is possibly reached in *Triarthrus eatoni* (Trilobita) and *Naraoia longicaudata* (nectaspidid lamellipedian), both of which have around 60 lamellae (see Cisne 1973; Hou et al. 2004). Data on the growth-related changes of lamellipedian exopods such as the number of lamellae, segment number of exopod shaft and length are absent from the literature. We therefore made a quick examination of the number of exopod lamellae in 11 specimens of *Naraoia longicaudata*, with a range in cephalic width from 8 to 34 mm (material sourced from the specimens illustrated by Hou and Bergström 1997, Figs. 40-43, or housed at the Department of Palaeozoology, Swedish Museum of Natural History, No. NRM Ar 60152-60156). The result, based on the counting as far as possible in one of the first few thoracic exopods, turned out to be roughly correlated, and showed an extremely low increase ratio in the numbers compared to the width. Although some extrapolations were needed because of the overlapping nature of exopods, or the differences of preserved positions and so on, the smaller ones appeared to have a number a little over 40, while the larger a little less than 70. The latter is only less than twice the number of the former. This demonstrates that the exopod growth fails by far to meet the requirements for a gill function. From theoretical points of view, the 4-fold increase in width (or body length) means an increase in body weight by 64 times. To meet the corresponding demand for respiration, the gill area would need to increase by at least 16 times ($64^{0.67}$: calculation based on the allometric exponent of the total respiratory area against dry-body weight of sluggish *Libinia dubia*). Since the cube of the number of gill lamellae in a gill cone is in proportional to the total lamellar surface as discussed above, the examined largest *N. longicaudata* should have more than 101 lamellae ($40 \times 16^{1/3}$), if its exopod lamellae function as a gill cone. The number of lamellae newly established compared with the smallest specimen, a little less than 30, is only half the minimum requirement (about 60: subtract "a little over 40" from 101), which corresponds to one eighth the requirement in the case of respiratory area (the total respiratory surface area of a gill is in proportion to the cube of lamellar number: $[1/2 \text{ lamellar number}]^3 = 1/8$ area). The extreme difference between the actual and the theoretical value constitutes strong evidence against the lamellipedian exopod-gill hypothesis. Despite the virtual constancy in the number of lamellae, the body length of adult lamellipedians spans from about 2 cm (*Marrella splendens*: Whittington 1971) to about 30 cm in the non-trilobite *Tegopelte gigas* (Whittington 1985) to over 70 cm in the trilobite *Isotelus rex* (Rudkin et al. 2003).

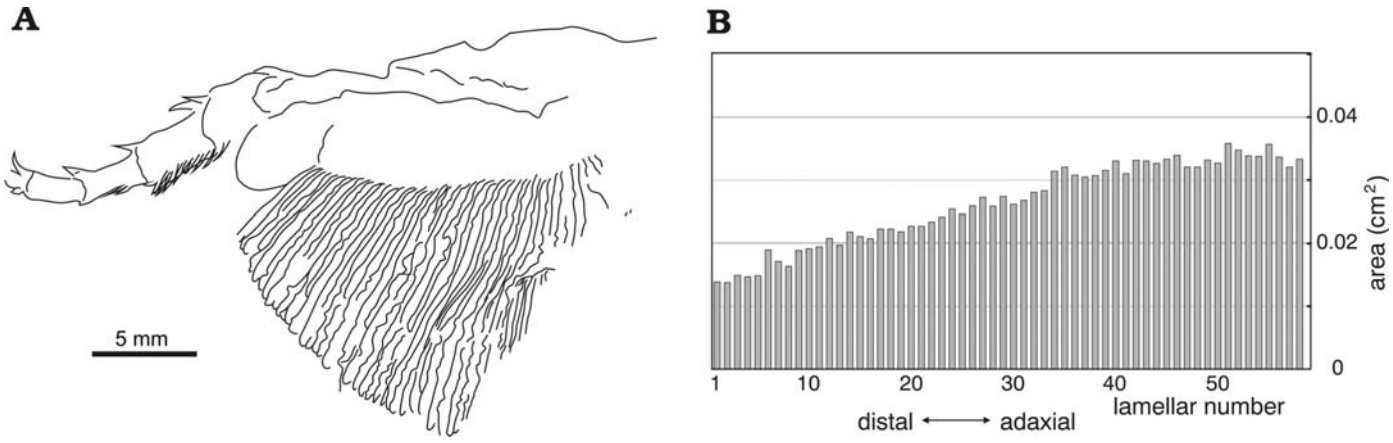


Fig. 10. Lamellipedian exopod of the trilobite *Olenoides serratus*. **A**. Camera lucida drawing; traced from Whittington (1980: text-fig. 6). **B**. Estimated area of each exite shown in A.

Unfortunately, the exopods are not known in the latter or in any close relative, although there are imprints of endopods in one isotelid trilobite. From what is said above, it is entirely clear that the expansion of lamellar surface with growth in the lamellipedians is many orders too small for a respiratory organ. We conclude that the lamellipedian exopods with their lamellae could not possibly have had (much of a) respiratory function. It leaves the possibility that they had one or more mechanical functions, for instance in ventilating the gills, in swimming, or in burrowing. Already Richter (1919: 222–223) suggested that the outer limb branch was for swimming, and that its lamellae were setae. This mechanical function also suits with an old and once rejected idea on the main place of respiratory function in trilobites (see Harrington 1959: 101), the ventro-pleural membrane. The respiratory surface of water-breathers requires oxygenated water to be pushed onto it for gaseous exchange, because of the extreme low permeation coefficient of oxygen in comparison to that of carbon dioxide or ammonia (Taylor 1998). A study of the possible respiratory area in lamellipedians as represented by trilobites is in progress.

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