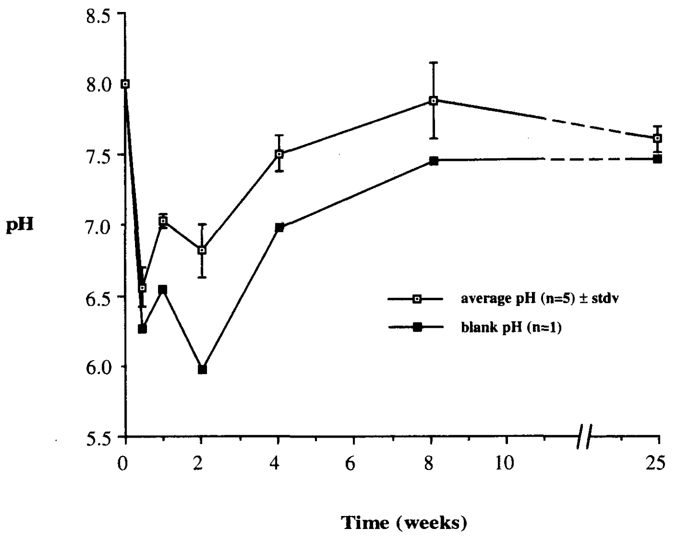

FIGURE 4—The variation in pH over time (see Appendix). The average pH recorded for vessels containing decaying carcasses (the vertical bars indicate the standard deviation) is compared with the values for the controls, which contained no carcass.

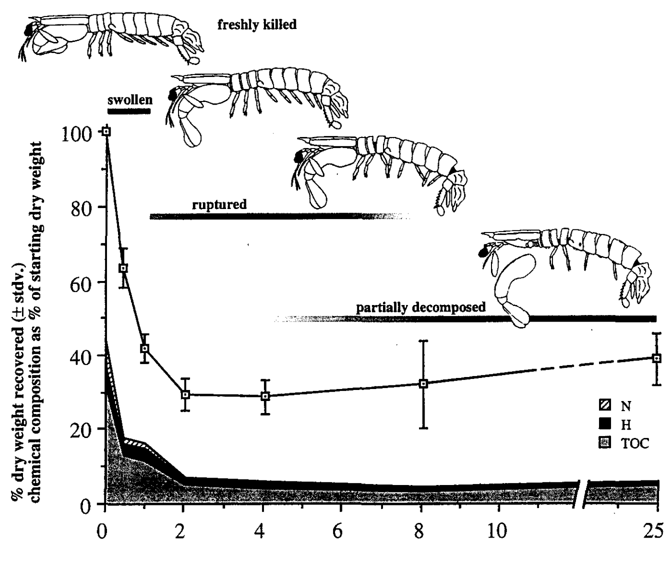

FIGURE 5 —Times through which stages of decay were observed in the decay experiments (the diffuse limits to the ranges indicate variation in the onset and persistence of stages from carcass to carcass). Percentage dry weight recovery (the vertical bars indicate the standard deviation, see Appendix), and variation in nitrogen, hydrogen, and organic carbon (expressed as a percentage of starting dry weight, see Table 1) over time.

MICROBIAL DEGRADATION OF THE CUTICLE

SEM observations revealed direct evidence of the impact of microbial activity on the cuticle. Bacteria were found on the inner surface (Fig. 2A), sometimes attached to fragments of muscle fiber (Fig. 2B). Circular pits began to appear on the cuticle within three days and were evident, varying in shape, diameter and depth (Fig. 2C) throughout the experiments. Concentrations of bacteria were often evident within and around these degradation phenomena (Fig. 2D, E). Microbial decay, particularly on the inner surface of the trunk tergites and telson, revealed the internal structure of the cuticle. Shallow subcircular depressions showed the helicoidally arranged microfibrils of the endocuticle (Fig. 3A). Sometimes a ring of cuticle was consumed by microbes, leaving a column of endocuticle (Fig. 3B). Bacteria were evident on the floor of the ring-shaped depression (Fig. 3C). Clusters of endocuticular columns (Fig. 3D) were sometimes formed by decay. These columns clearly revealed the helicoidal structure of the endocuticle (Fig. 3E), although bacteria were no longer evident on the surface (Fig. 3F). Very rarely the surrounding cuticle displayed a spongy (Fig. 3B, G, H) or labyrinthine (Fig. 3I) texture which also appeared to be bacterially induced. Bacterial decay has not been reported to reveal the helicoidal structure of the endocuticle before (A.C. Neville, pers. comm.).

Some specimens showed black spots or stains on the cuticle in life, which may have been fungal in origin. There is no evidence that these spots promoted decay, but they did form a suitable substrate for bacteria (Fig. 2F).

QUANTITATIVE CHANGES DURING DECAY

Oxygen Concentrations

Initial oxygen concentrations were ca. 50%. Throughout the experiments, the average oxygen level measured did not exceed 3% saturation. The combined effects of Merck Anaerocult® A and decay itself (Briggs and Kear, 1994) ensured that processes were O_2 limited and anaerobic almost from the outset.

pH

The pH of the ASW (Fig. 4, data in Appendix) fell from the initial value of 8.00 to a mean of 6.56 within 3 days. After 1 week, the pH had recovered to 7.03 but dropped again to 6.82 after 2 weeks. By 4 weeks the pH had risen to 7.51. It never returned to the starting value, reaching a maximum of 7.88 by 8 weeks and then falling to pH 7.61 after 25 weeks. The pH profile of the control (Fig. 4) shows a similar pattern, but with lower values. Thus the CO_2 introduced by the Anaerocult had a significant impact on pH levels. A fall in pH to below 6.5 has been recorded, however, around shrimps decaying anaerobically in aqueous experiments for 10 days where no Anaerocult was used.

Weight Changes

The rate of decay was quantified by monitoring the change in weight (Fig. 5, Appendix) and the chemical composition of the animal remains (Fig. 5, Table 1). No correlation was found between percentage weight loss (dry weight) and the initial weight of the carcass (correlation coefficient < 0.95).

Mean wet weight (see Appendix for data) increased to 115.1% of the starting weight within the first three days. This short period of osmotic uptake was followed by a steep decline to 60.7% after one week, and 34.6% after two. Thereafter, the mean wet weight remained similar, 32.6% after 4 weeks, 32.4% after 25 weeks. There was a small increase in week 8 up to 46.9%. Statistical analysis showed

TABLE 1—Composition of mantis shrimp *Neogonodactylus oerstedii* as weight % of starting dry weight (% start) and weight percent of remaining dry weight (% now). TOC is total organic carbon; C-CO_3 is carbon present as carbonate.

	TOC		C-CO_3		H		N		PO_4		Ca		S	
	% Start	% Now	% Start	% Now	% Start	% Now	% Start	% Now	% Start	% Now	% Start	% Now	% Start	% Now
Fresh	31.37		3.12		5.07		7.05		5.22		5.95		0.42	
3 days	12.96	22.47	—	—	3.05	3.04	1.64	2.84	5.61	7.99	4.06	5.78	0.44	1.07
Week 1	11.37	29.96	—	—	3.40	4.34	1.42	3.73	4.22	9.00	2.51	5.34	0.32	1.25
Week 2	5.05	14.89	0.56	1.66	1.36	2.07	0.49	1.46	4.88	15.12	2.03	6.30	0.14	1.05
Week 4	4.04	16.36	—	—	1.38	2.51	0.30	1.21	4.40	12.57	2.25	6.43	0.01	0.02
Week 8	3.14	21.74	0.20	1.40	1.10	3.29	0.21	1.47	5.37	13.42	2.94	7.35	0	0
Week 25	4.73	12.09	1.37	3.50	0.82	1.03	0.17	0.43	4.33	14.09	1.16	3.79	0	0

no significant change in wet weight after one week. The high standard deviations that characterize the wet-weight values (see Appendix) make it difficult to monitor decay in this way.

Dry weight (Fig. 5, see Appendix for data) is a much more reliable indication of the progress of decay (Briggs and Kear, 1994). Mean dry weight showed a rapid decline during the first two weeks. Just 63.5% of the initial dry weight remained after three days, 41.8% after 1 week, and 29.4% after 2 weeks. This rapid decline, which correlates with the rupturing of the carcass, was followed by a slight rise through 28.7% after 4 weeks, 32.1% after 8 weeks, to 38.9% after 25 weeks. This rise may be the result of mineral accumulation within the carcass.

CHEMICAL DEGRADATION

Changes in the composition of *Neogonodactylus* at successive sampling intervals can be expressed as a percentage of the starting dry weight (Fig. 5), or as a percentage of the material remaining (Table 1). 58.2% of the starting composition of *Neogonodactylus* consists of organic C, N, H, PO_4, C in carbonate, Ca, and S. The remainder is predominantly O with some minor elements. The starting CHN ratio of 1:1.92:0.19 changed through 1:1.61:0.11 (3 days), 1:1.72:0.11 (1 week), 1:1.66:0.08 (2 weeks), 1:1.83: 0.06 (4 weeks), 1:1.8:0.06 (8 weeks), to 1:1.01:0.03 (25 weeks). TOC, H and N all showed a rapid initial decline, which correlates with rupturing of the carcass and major weight loss (Fig. 5). TOC dropped 20% to about 10% of the starting dry weight within a week. After this decline it more or less stabilized around 4% of the starting dry weight. H dropped from 5% to about 3% in the first week, after which it slowly decreased to 0.82% after 25 weeks. N showed a rapid decline from 7% to 1.64% within 3 days, reaching 0.17% after 25 weeks. C in CO_3 showed a much less pronounced decline. PO_4 remained a more or less constant percentage of the starting dry weight throughout the experiment. Ca declined steadily from an initial 5.95% to 1.16% of the starting dry weight after 25 weeks. The percentage of S reached zero after 8 weeks.

The composition of the carcasses expressed as weight percent of the remaining dry weight shows that organic C, PO_4 and Ca are the dominant components (Table 1). TOC remained at about 16 % from week 2. PO_4 showed an increase from 5.22% to an average of 14% from week 2 to 25. Ca remained at more or less the same percentage throughout.

MINERAL PRECIPITATION

Decay of the mantis shrimp carcasses was accompanied by the precipitation of minerals. The styles of mineralization fall into four broad but distinct categories: (1) crystal bundles of $CaCO_3$ that form within the carcass, particularly within the trunk segments and telson; (2) crystal crusts of $CaCO_3$ that form on the external surface of mainly the trunk segments; (3) microspheres of $CaPO_4$, which replicate the soft tissue, particularly the hepatopancreas and muscle fibers; and (4) finely crystalline $CaCO_3$, which precipitated within the cuticle of the trunk segments, and is only clearly visible when the cuticle is sectioned.

Crystal Bundles

Crystal bundles are aggregates of hundreds of micron-sized crystals (Buczynski and Chafetz, 1991). Those that precipitated in the mantis shrimp carcasses took a variety of forms including hemispheres (Figs. 6A-C, 8A-C), spheres (Fig. 8D), rods (Fig. 8D), dumbbells (Fig. 7A-C), and almond-shaped crystals (Fig. 7D-F). They are composed of calcium carbonate (Table 2) and formed in 40%, 60%, and 40% of the carcasses sampled at 4, 8, and 25 weeks respectively.

Hemispheres were initiated as layers of elongate crystals radiating from a marginal or central nucleus on the cuticle surface. As additional radiating crystals were added and the relief increased, a crystalline hemisphere formed (Fig. 6B). Those that formed on the carapace had a porous surface produced by the action of the microbes and tended to occur in clusters (Fig. 6B). Figure 6C shows the early stage of formation of two coalescing hemispheres. The hemispheres that occurred on the inner surface of the trunk cuticle formed mainly on the lateral area of the tergites (Fig. 8A), or on the margins of the sternites (Fig. 8B). Some of them showed a small depression or a series of irregular bumps in the center of the surface (Fig. 8C). The crystal hemispheres were all bright white in color. Complete spheres were rare and only found within the clusters of rods (Fig. 8D). They were made up of triangle-shaped crystals (Fig. 8F) and were whitish in color.

Rods were sometimes found projecting from the inner surface of the trunk tergites, but they occurred mostly in massive clusters inside the abdominal somites or the telson (Fig. 8D). In some cases, rods were found within and on the mineralized hepatopancreas (Fig. 10E). These rods were bundles of overlapping wedge-shaped crystals (Fig.