

## CHITIN BIOMASS IN MARINE SEDIMENTS

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## ABSTRACT

One hundred marine sediments of various origins were screened in order to evaluate their chitin biomass. Our purpose was to assess the detritic chitin stocks in order to find some potential new source of chitin. The chitin biomass of marine sediments is very diversified, from 2 up to 2 800  $\mu\text{g g}^{-1}$  decalcified sediment (DS). Most sediments have low or very low chitin biomass (67 % under 100  $\mu\text{g g}^{-1}$  DS). No significant difference related to depth nor climatic influence was found except that all sediments richer in chitin (above 300  $\mu\text{g g}^{-1}$  DS) are on the continental shelf (above 200 m depth). Actually, the chitin content is higher in coarse, much calcified sediments of organoclastic origin; bryozoa and shelly sands and gravels are the richest. The powerful hydrolytic activity of microorganisms lower the steady state equilibrium level between chitin input and weathering, so most "unprotected" chitin is weathered very soon after settling. Marine sediments appear thus as a non competitive potential industrial chitin source.

## INTRODUCTION

Taking into account the huge surface occupied by marine sediments, the relative accessibility of this resource and the importance of potentially chitinous organoclastic remains constituting most of them, we decided to screen various marine sediments and to evaluate their chitin biomass. Our purpose was to assess the detritic chitin stocks in such biota in order to find some potential new source of chitin.

## MATERIAL AND METHODS

Hundred samples of sediments were analysed from different climatic areas (cold temperate to tropical) covering a wide diversity of ecological settings and sedimentological conditions : origin and nature of the sediments, depth ... (Table 1).

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The sediments were sampled through the usual oceanographic techniques (by diving, dredging or by grabs), washed with distilled water and stored dry until use. Chitin was estimated by the enzymatic method of JEUNIAUX [1] after thoroughful decalcification (with 0.5 N HCl) and protein extraction (with NaOH 0.5 N, 3 hours at 100 °C.). We applied the correction coefficient estimated by POULICEK and JEUNIAUX [2, 3] in order to balance the adsorption effect of chitinase on clay and sand particles

### RESULTS AND DISCUSSION

The results of chitin and  $\text{CaCO}_3$  estimations in marine sediments are exposed in Table 1. The chitin biomass of marine sediments is very diversified : from 2 up to 2 800  $\mu\text{g g}^{-1}$  DS (Decalcified Sediment). But most sediments analysed have low or very low chitin biomass : in 67 % of the samples, the chitin content is below 100  $\mu\text{g g}^{-1}$  DS and in 84 % of the samples,, it lies below 300  $\mu\text{g g}^{-1}$  DS (Table 1, Fig.1). There is no significant difference related to depth nor climatic influence except that all the sediments with a chitin biomass over 300  $\mu\text{g g}^{-1}$  DS are on the continental shelf (above 200 meters depth). Deeper samples are always poorer (63 to 157  $\mu\text{g g}^{-1}$  DS) (Table 1, Table 2, Fig. 1).

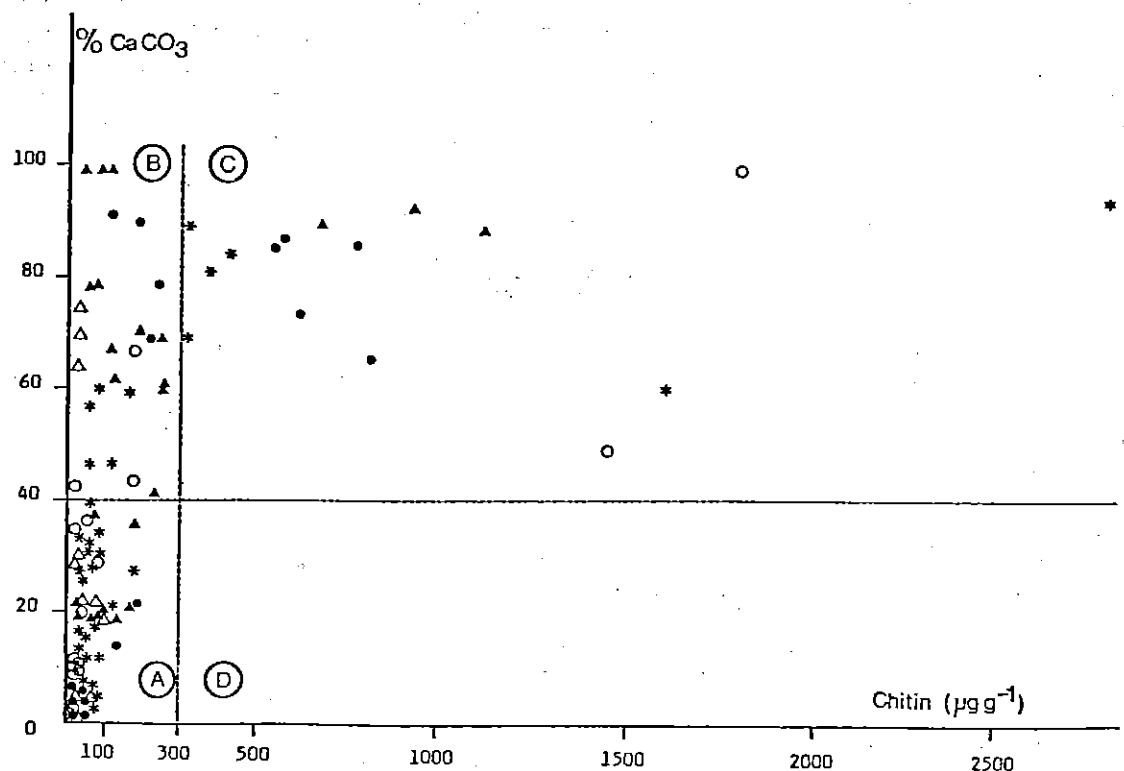


Figure 1 : relation between chitin and  $\text{CaCO}_3$  content in marine sediments (● and ○ : respectively tropical and temperate mediolittoral samples; ▲ and △ : tropical and temperate infralittoral samples; ★ : circalittoral, bathyal and abyssal samples)

As seen in figure 1, the chitin content is generally higher in the most calcified sediments, but there is no clear significant correlation between

the two variables. All samples with chitin biomass over  $300 \mu\text{g g}^{-1}$  DS contain more than 40 %  $\text{CaCO}_3$  (samples in the "C" area, [fig. 1]). No samples are to be found in the "D" area, corresponding to low calcification and high chitin content (fig. 1). This can be explained by the fact that most of the sedimentary carbonates arise from skeletal components with chitino-proteic matrix (mollusk shells, crustacean cuticles, bryozoa ectocysts,...) (Table 3). But this is not a general situation : actually, pelletoids or oolitic sediments (with more than 99.5 % non skeletal  $\text{CaCO}_3$ ) contain only around  $40\text{-}60 \mu\text{g chitin g}^{-1}$  DS, mainly in the form of adsorbed particulate organic matter and endolithic microflora (fungi,...). Similarly, coral reef sediments with high carbonate content (50 to 90 %  $\text{CaCO}_3$ ) show relatively low amounts of chitin ( $100$  to  $150 \mu\text{g g}^{-1}$  DS) since the most abundant skeletal components (mostly fragments of coralline algae and corals, *Halimeda*, foraminifera and echinoid spines with lesser amounts of mollusks and bryozoa) have no chitin or a low chitin content in their organic matrix (samples in the "B" area of figure 1) (Table 2 and Table 3).

Mollusk shells can be considered as the main contributors to chitin biomass in sediments as already showed earlier [3]. Shelly sands and gravels, either tropical or temperate, shelter the highest biomass of sedimentary chitin ( $500$  to  $1\ 800 \mu\text{g g}^{-1}$  DS). Although, there is one remarkable exception : a highly localized relatively deep (170-200 m) gravel - a thanathocenosis called "*Gryphus* beds" -, where highly calcified bryozoa, together with mollusk shells raise the chitin biomass ( $1\ 600$  to  $2\ 800 \mu\text{g g}^{-1}$  DS). Underlying that thanathocenosis, the fine sand, mainly of terrigenous origin as seen by its low  $\text{CaCO}_3$  content (30 %) is much poorer, around  $80$  to  $90 \mu\text{g g}^{-1}$  DS (Table 1).

Another important parameter governing the chitin distribution in marine sediments is the intensity of biodegradation phenomena affecting dead skeletons. We already showed that the ways and kinetics of chitin biodegradation depends on the nature of the skeleton (origin, mineralogy,...) and of the environmental depositional conditions [4-9]. The chitinous matrices screened within calcified crystallites are degraded by microboring organisms (bacteria, fungi, blue green algae) that secrete chitinolytic enzymes at the precise level of the organic sheaths [2, 5, 8, 9]. By these phenomena, 60 % of the chitin of mollusk shells can disappear within 6 months in aerobic conditions [2, 3, 9] and 40 to 90 % in anaerobic conditions [9]. Less protected chitino-proteic matrices (uncalcified or less stabilized by chemical bonding) are weathered at a much greater rate : for example, as much as 90 % of the chitin of a crab cuticle can disappear within less than 2 weeks at the aerobic water-sediment interface [8, 9], 99 % after 4 months in anaerobic conditions [9].

There is a clear correlation between the amount of chitin in marine sediments and the number of viable chitinolytic bacteria (fig. 2).

The activity of these chitinolytic microorganisms results in a quick metabolization and recycling of the chitin settling onto marine sediments although the less protected one. For example, planktonic organisms are considered as the main chitin producers in the bay of Calvi compared to benthic biocenoses [10, 11]. But most of the chitin so produced is very soon recycled (even in open water) and cannot really contribute to an increase of the sedimentary chitin biomass [12].

### CONCLUSIONS

As a conclusion, most marine sediments have very low chitin biomass, probably due to the powerful hydrolytic activities of microorganisms lowering the steady state equilibrium level between input and weathering. Even if the

chitin biomass of some selected sediments may be relatively high, the bad accessibility and/or scarcity of such environments together with the need for a complex demineralization of the material make sediment chitin not competitive as a potential industrial source.

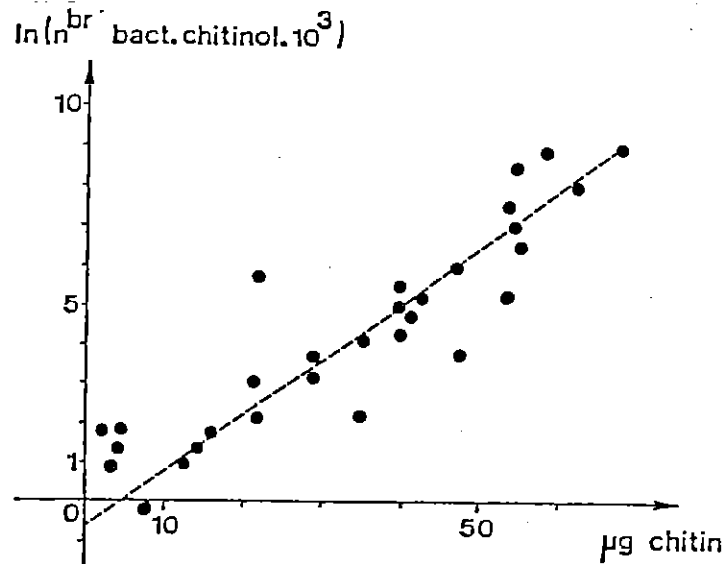


Figure 2 : relation between chitin content of marine sediments (in  $\mu\text{g g}^{-1}$  DS) and number of chitinolytic bacteria (ln number of viable strains)

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TABLE 1 : DISTRIBUTION OF CHITIN IN MARINE SEDIMENTS

SAMPLE	ORIGIN	DEPTH	% CaCO <sub>3</sub>	Chitin content ( $\mu\text{g g}^{-1}$ dec. sed.)
<u>MEDIOLITTORAL SAMPLES</u>				
Slikke (estuarine mud)	Canche estuary, Le Touquet, France	-	20.0	37.31
"	"	-	28.9	95.00
Mangrove muddy sand	Aboisso lagoon, Ivory Coast	-	14.8	86.50
"	"	-	22.5	94.36
Estuarine sand	Le Guillec estuary, Mogueeriec, France	-	2.1	1.41
"	"	-	1.6	2.17
Terrigenous sand	Crèche bay, Boulogne, France	-	11.7	7.33
"	"	-	9.4	6.40
"	"	-	43.3	8.13
"	"	-	32.6	7.66
"	Deshaies, Guadeloupe	-	6.9	11.43
"	"	-	8.3	7.00
Terrigenous muddy sand	Oysters beds, Pempoull (Roscoff), France	-	35.0	5.97
"	"	-	9.8	2.87
"	"	-	11.1	5.79
"	"	-	11.3	7.18
Terrigenous sandy mud	Perharidi (Roscoff), France	-	3.2	2.35
"	"	-	67.6	72.31
Zostera meadow sand	"Ile Verte", Roscoff, France	-	36.4	57.91
"	"	-	43.8	175.53
Coral sand	Farukolufushi, Maldives Islands	-	78.6	143.84
"	"	-	67.9	124.38
"	Takapoto lagoon, Tuamotu Islands	-	89.9	103.84
"	"	-	92.7	106.43
Volcanic sand	Basse-Terre, Guadeloupe	-	2.1	5.17
"	"	-	3.4	11.33
"	Tanah Lot, Bali	-	3.7	9.38
"	"	-	2.6	8.04
Shelly sand	La Panne, Belgium	-	49.4	1 435.39
"	"	-	99.5	1 788.51
"	Nusa Dua, Bali	-	64.6	809.41

TABLE 1 : DISTRIBUTION OF CHITIN IN MARINE SEDIMENTS (followed)

SAMPLE	ORIGIN	DEPTH	% CaCO <sub>3</sub>	Chitin content ( $\mu\text{g g}^{-1}$ dec. sed.)
Shelly gravel	Kuantan, Malaysia	-	86.7	588.24
"	"	-	83.4	776.45
"	"	-	85.9	534.50
"	"	-	73.5	604.03
<u>INFRA-LITTORAL SAMPLES</u>				
Terrigenous sand	Calvi, Corsica	40	22.4	16.38
"	"	40	23.5	19.64
"	"Lez Trepieds", Roscoff, France	30	6.7	3.51
"	"	30	6.4	4.28
Terrigenous muddy sand	Ste Croix, Virgin Islands	35	19.9	77.60
"	"	35	21.3	94.30
<u>Halophila</u> meadow sand	Nusa Dua, Bali	2	21.7	75.33
"	"	5	18.6	38.75
"	"	5	35.4	94.27
Seagrass meadow sand	Deshaises, Guadeloupe	10	36.5	61.17
"	"	10	21.3	11.03
<u>Posidonia</u> meadow sand	Calvi, Corsica	15	29.8	11.17
"	"	15	30.4	17.83
"	"	15	64.4	19.73
"	"	37	69.1	38.72
"	"	37	74.3	41.47
"	"	37	76.4	89.79
"	"	37	76.3	62.66
"	"	37	19.2	40.34
Coral Sand (reef)	Farukolufushi, Maldives Islands	17	61.4	138.11
"	Himafushi, Maldives Islands	20	59.3	133.18
"	Farukolufushi, Maldives Islands	3	62.7	118.37
"	Takapoto, Tuamotu Islands	6	42.6	140.24
Colithic sediment	Nusa Dua, Bali	10	69.8	145.81
"	"	1	99.1	41.19
"	"	3	98.3	63.84
"	"	5	99.4	64.35
Maerl	Taureau Castle, Roscoff, France	15	66.1	104.83

TABLE 1 : DISTRIBUTION OF CHITIN IN MARINE SEDIMENTS (followed)

SAMPLE	ORIGIN	DEPTH	% CaCO <sub>3</sub>	Chitin content ( $\mu\text{g g}^{-1}$ dec. sed.)
Maerl	Taureau Castle, Roscoff, France	15	71.3	198.47
Shelly gravel	Nusa Dua, Bali	3	88.9	678.38
"	"	3	87.6	1 119.62
"	"	10	92.5	932.76
<u>CIRCALITTORAL SAMPLES</u>				
Terrigenous sand	Calvi, Corsica	47	26.3	93.57
"	"	69	26.3	27.50
"	"	145	31.8	45.53
"	"	148	34.7	38.41
"	Los Angeles, California, U.S.A.	140	8.7	3.83
"	"	160	10.4	7.42
Coastal terrigenous mud	Calvi, Corsica	108	27.9	41.43
"	"	127	34.8	39.27
"FMI" muds	"	47	8.8	18.49
"	"	96	17.3	36.50
Coarse detritic sands	Calvi, Corsica	44	79.3	410.77
"	"	44	81.5	386.79
"	"	45	88.4	317.41
"	"	45	84.6	429.52
"Gryphus" beds	"	175	90.4	2 808.83
"	"	190	60.3	1 644.52
"	"	195	30.4	88.81
"	"	200	27.6	76.53
<u>BATHYAL SAMPLES</u>				
Blue bathyal mud	Calvi, Corsica	420	39.5	81.22
"	"	440	60.4	81.16
"	"	1 150	56.3	63.16
"	"	1 200	46.2	127.43
Yellow bathyal mud	"	1 170	34.7	92.75
"	"	1 600	58.2	157.03



TABLE 1 : DISTRIBUTION OF CHITIN IN MARINE SEDIMENTS (followed)

SAMPLES	ORIGIN	DEPTH	% CaCO <sub>3</sub>	Chitin content ( $\mu\text{g g}^{-1}$ dec. sed.)
<u>ABYSSAL SAMPLES</u>				
Pteropod ooze	Indian Ocean	2 200	69.8	111.16
"	"	2 200	74.3	127.32
Black abyssal mud	"	2 800	17.3	81.19
"	"	2 800	21.1	134.83
"	"	3 600	26.5	63.07
"	Pacific Ocean	3 800	14.4	66.31
"	"	4 200	13.7	73.50
Red Clay	"	4 800	7.6	65.43
"	"	5 400	3.4	81.90

TABLE 2 : RELATION BETWEEN SOME PARAMETERS OF MARINE SEDIMENTS AND THEIR CHITIN CONTENT  
(N : number of samples); (percentage)

ORIGIN	CHITIN CONTENT (in $\mu\text{g g}^{-1}$ DS)		
	< 100 (N = 67)	100 - 300 (N = 17)	> 300 (N = 16)
Temperate origin (N = 56)	43 (76.8)	5 (8.9)	8 (14.3)
Tropical origin (N = 44)	24 (54.5)	12 (27.3)	8 (18.2)
<u>MEAN GRAIN SIZE</u>			
Mud ( $\psi < 63 \mu\text{m}$ ) (N = 26)	23 (88.5)	3 (11.5)	0
Sand ( $63 < \psi < 500 \mu\text{m}$ ) (N = 55)	40 (72.7)	10 (18.2)	5 (9.1)
Gravel ( $\psi > 500 \mu\text{m}$ ) (N = 19)	4 (21.1)	4 (21.1)	11 (57.8)
<u>MAIN COMPONENTS</u>			
Terrigenous particles (N = 34)	33 (97.1)	1 (2.9)	0
Volcanic particles (N = 4)	4 (100)	0	0
Corals (N = 9)	0	9 (100)	0
Mollusk shells (N = 12)	0	2 (16.7)	10 (83.3)
Other organoclasts (N = 35)	24 (68.6)	5 (14.3)	6 (17.1)
Authigenic particles (N = 6)	6 (100)	0	0

TABLE 3 : COMPOSITION OF THE ORGANOCLASTIC FRACTION OF SOME SELECTED SEDIMENTS

Type of sediment	Coral sand Farukulufushi Maldives isld. 0 m	Coral sand Farukulufushi Maldives isld. 17 m	Coral sand Takapoto Tuamotu isld. 10 m	Shelly sand La Panne Belgium 0 m	Shelly gravel Nusa Dua Bali 0 m	Shelly gravel Kuantan Malaysia 0 m
Corals	25.8 (15-36)	30.6 (9-45)	22.6 (2-45)	0	10.3 (4-12)	14.3 (2-18)
Mollusks	10.0 (9-15)	15.5 (6-38)	9.2 (3-18)	91.4 (88-100)	54.6 (48-72)	46.3 (40-61)
Foraminifera	15.7 (10-23)	14.4 (6-31)	17.8 (2-38)	0	2.3 (tr-4)	2.2 (tr-3)
Algae	38.7 (33-54)	28.0 (19-52)	44.3 (tr-77)	3.7 (1-9)	25.6 (10-40)	27.8 (9-38)
Bryozoa	1.5 (tr-11)	4.8 (tr-10)	1.7 (0-10)	3.9 (2-12)	3.8 (1-5)	6.3 (2-9)
Miscell. Skel.*	8.3 (5-18)	6.7 (1-16)	1.4 (tr-15)	1.0 (tr-10)	1.1 (tr-6)	3.1 (tr-8)
Ooliths	0	0	0	0	2.3 (tr-8)	0

Type of sediment	Shelly gravel Nusa Dua Bali 3 m	Ooliths Nusa Dua Bali 3 m	Posidonia meadow Calvi Corsica 37 m	Gryphus beds Calvi Corsica 190 m	Bathyal mud Calvi Corsica 440 m	Pteropod ooze Indian ocean 2 200
Corals	10.5 (6-17)	tr	0	tr***	10.4 (1-28)***	0
Mollusks	44.5 (38-66)	2.4 (tr-8)	56.6 (48-74)	29.7 (20-42)	26.8 (17-42)	86.9 (82-100)
Foraminifera	2.7 (tr-5)	tr	2.8 (tr-4)	0.6 (tr-3)	2.7 (1-6)	12.2 (8-19)**
Algae	25.5 (8-39)	tr	2.8 (tr-9)	1.1 (tr-3)	tr	0
Bryozoa	4.4 (tr-8)	0	1.1 (tr-5)	38.7 (30-60)	8.6 (3-10)	0
Miscell. Skel.*	7.0 (1-11)	1.3 (tr-3)	36.7 (19-40)	29.9 (25-40)**	51.5 (33-65)	0.9 (tr-3)
Ooliths	5.4 (tr-8)	96.3 (92-100)	0	0	0	0

\* Echinoid spines and plates, crustacean cuticles, annelid tubes, ...

\*\* Up to 25 % *Gryphus vitreus* valves (Brachiopod), plus echinoid spines, ...

\*\*\* *Lophelia pertusa*, *Dendrophyllia* sp., deep ahermatypic "white corals".

\*\* *Globigerina*, planktonic foraminifera.