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# A new calanoid copepod (Spinocalanidae) swarming at a cold seep site on the Gabon continental margin (Southeast Atlantic)

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**Abstract:** Adult females, subadult males at 5<sup>th</sup> copepodid stage and adult males of *Methanocalanus gabonicus*, a new genus and species of the family Spinocalanidae Vervoort, 1951 (Copepoda, Calanoida), were found in hydrocarbon fluids, a few centimetres above vesicomyid and mytilid bivalves aggregating at the giant pockmark Regab on the Gabon continental margin (Southeast Atlantic) at a depth of 3151-3155 m. After description of the monotypic genus *Isaacsicalanus* Fleminger, 1983 from the East Pacific Rise, this is the second finding of calanoid copepods swarming in deep sea chemosynthetic environment. *Methanocalanus* gen. nov., a sister genus to the genus *Isaacsicalanus*, is characterized by the following derived characters: 3 large teeth on the masticatory blade of mandible separated from 5 others by a groove; 1 seta on the second to fourth articulating endopodal segment of maxilliped. The female exhibits, as many calanoid genera, a ventral semi-circular genital operculum covering the paired gonopores. Morphological and anatomical observations of *M. gabonicus* gen. nov., sp. nov. indicate that females of this species can feed on small particles from or near the bottom environment while the males are supposed to not feed.

**Résumé :** Un nouveau copépode calanoïde profond (Spinocalanidae) vivant en essaim sur une source de fluides froids de la marge continentale du Gabon (Atlantique sud-est). Des femelles, des mâles et des copépodites V mâles de *Methanocalanus gabonicus*, une nouvelle espèce d'un nouveau genre de la famille des Spinocalanidae Vervoort, 1951 (Copepoda, Calanoida), ont été trouvés dans des fluides chargés en hydrocarbures, à quelques centimètres au-dessus d'agrégats de bivalves Vesicomyidae et Mytilidae dans le "pockmark" géant Regab sur la pente continentale du Gabon (Atlantique sud-est) à une profondeur de 3151-3155 m. Après la description du genre monospécifique *Isaacsicalanus* Fleminger, 1983 de la Ride Est Pacifique, c'est la deuxième découverte de copépodes calanoïdes formant des essaims en environnement profond chimiosynthétique. *Methanocalanus* gen. nov., groupe frère du genre *Isaacsicalanus*, est caractérisé par les caractères dérivés suivants : trois fortes dents sur la lame mastricatrice de la mandibule séparées des cinq autres par une dépression, une seule soie sur le second et jusqu'au quatrième segment de l'endopodite du maxillipède. Les femelles, comme chez de nombreux calanoïdes, présentent ventralement un opercule génital semi-circulaire recouvrant les gonopores. Les caractères morphologiques et anatomiques suggèrent que les femelles se nourrissent de particules en suspension près ou sur le fond, alors que les mâles ne s'alimentent pas.

Keywords: Copepoda • Spinocalanidae • Deep sea • Hydrocarbon seep • Gabon continental margin • Southeast Atlantic

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# Introduction

A giant pockmark Regab, 800 m in diameter and 15 m deep, located on the Gabon continental margin (Southeast Atlantic) at 3160 m depth was explored by a remote operated vehicle (ROV) in 2001 (Ondréas et al., 2005). Massive carbonate crusts, dense aggregations of vesicomyid and mytilid bivalves as well as siboglinid tubeworms and other invertebrates were observed in sites characterized by enriched concentration of hydrocarbon fluids. The study of siboglinid tubeworms from Regab pockmark revealed 4 morphotypes of a new species belonging to the genus *Escarpia*, while the study of mytilids *Bathymodiolus* sp. discovered dual symbiosis with sulfide- and methane-oxidizing bacteria (Andersen et al., 2004; Duperron et al., 2005).

Until now, among copepods of the order Calanoida only *Isaacsicalanus paucisetus* Fleminger, 1983 (Spinocalanidae) was recorded in aggregations over a deep sea chemosynthetic environment (Fleminger, 1983). *I. paucisetus* was found at the East Pacific Rise in a clustered swarm of several thousand individuals within 1 m above a small depression framed with vesicomyids *Calyptogena magnifica* and siboglinid tubeworms *Riftia pachyptila* (Fleminger, 1983).

The present paper continues our research of copepods inhabiting deep-sea chemosynthetic environments (review and recent papers: Heptner & Ivanenko, 2002; Ivanenko, 2006; Ivanenko & Defaye, 2006a, b; Ivanenko et al., 2006; Ivanenko et al., 2007, in press). It describes a new genus and species of a spinocalanid copepod collected over bivalves from cold seep on the Gabon continental margin (Southeast Atlantic), and is the second report of calanoids swarming in deep sea chemosynthetic environment.

# **Material and Methods**

Many copepods representing only one species of a new calanoid were collected by slurp-gun a few centimetres above aggregations of the vesicomyid and mytilid bivalves (dive 146/09, samples ASPI 01 and ASPI 03; dive 147/10, samples ASPI 03). The samples have been taken during cruise Biozaire 2 (15 Nov.-4 Dec. 2001, the French *R/V L'Atalante*, chief scientist Myriam Sibuet) by a slurp-gun attached to the ROV Victor 6000 adapted to collect small and fragile organisms in separate bottles. Two samples were taken over clusters of mytilid bivalves (*Bathymodiolus* sp.) accompanied with actinians, gastropods, holothurians *Chiridota* sp., echinoids, zoarcid fishes, and shrimps *Alvinocaris muricola* Williams, 1988 (Komai & Segonzac, 2005). One sample was taken over cluster of vesicomyid bivalves (*Calyptogena* sp. and/or

*Vesicomya* cf. *chuni*) accompanied with gastropods, decapods *Munidopsis* sp. (Macpherson & Segonzac, 2005), shrimps *Alvinocaris muricola*, holothurians *Chiridota* sp., echinoids and zoarcid fishes (personal communication of J. Galeron with K. Olu, M. Sibuet and M. Segonzac).

For general morphology, the specimens have been studied with a Leica DMLB compound microscope having bright-field and differential interference optics applying the "hanging drop method" described by Ivanenko & Defaye (2004). All drawings were made via a camera lucida.

For scanning electronic microscopy (SEM), copepods were dehydrated through graded ethanol concentrations, critical point dried, mounted on aluminium stubs, coated with gold, and examined in a JEOL 840 scanning electron microscope.

For study of male digestive system and female genitalia, the specimens were dehydrated in ethanol and embedded in Epon. The semi-thin sections of female genital doublesomite and of male prosome were realized with a Microtome Ultracut E Reichert-Jung and stained with Unna Blue. To test the metabolic activity of the digestive tract, longitudinal sections of the male body were stained with hematoxylin, then with eosine. Images were recorded by using a Zeiss Axioplan 2 microscope. Digital images were taken using a Canon EOS D30 camera or a Nikon digital camera DXM 1200.

### **Systematics**

Subclass COPEPODA Milne Edwards, 1830 Order Calanoida G.O. Sars, 1903 Family Spinocalanidae Vervoort, 1951

#### Methanocalanus gen. nov.

#### Diagnosis

Female. Prosome ovoid, cephalosome distinct from first pedigerous somite; tergites of pedigerous somites 4 and 5 fused laterally. Caudal rami longer than wide, with 5 setae, distomedial seta inserted and directed asymmetrically: on the right ramus directed dorsally, on the left ramus ventrally. Rostrum an uniramous knob-like process fused to cephalosome. Antennule 24-segmented, segment 1 with 3 setae; segments 12 and 14 each with 1 seta. Antenna: coxa unarmed, basis with 1 seta; endopod shorter than exopod, first endopodal segment with 2 setae, second segment with 7 setae on each lobes; exopod 6-segmented with 3, 1, 1, 1, 1, and 3 setae. Mandibular gnathobase ending with 8 teeth and 1 seta, 3 large teeth separated from other large teeth by a groove. Maxillule: praecoxal arthrite with 13 spines; coxal epipodite and coxal endite with 9 and 4 setae, respectively; first and second basal endites each with 4 setae; endopod 3-segmented, with 2, 3 and 5 setae; exopod with 8 setae. Maxilla: first praecoxal endite with 7 setae; second praecoxal, first and second coxal endites with each 3 setae; allobasis with two lobes bearing 4 and 1 setae; endopod 3-segmented, with 1, 1 and 3 setae. Maxilliped: syncoxa with 2 setae; allobasis with 3 medial and 2 distal setae; endopod 5-segmented bearing respectively 3, 1, 1, 1, and 4 setae; outer seta on segment 4 absent. Exopods of legs 1-4 and endopods of legs 3-4, 3-segmented, endopods of legs 1-2, 1- and 2-segmented, respectively. Leg 1 without outer distal spine on second exopodal segment and with outer lobe on anterior side of endopod; legs 2-4 with row of setules on anterior side of endopodal segment 3; legs 3 and 4 with rounded outer distal corner on endopodal segment 2; leg 4 with stout inner spine on basis. Leg 5 absent.

Male. Urosome 5-segmented, genital somite with ventrolateral genital flap on left side. Labrum flat, paragnaths absent. Antennules asymmetrical: left antennule 23segmented, distal segment corresponds to two distal segments of female antennule; right antennule 22segmented, differing from left antennule in fusion of segments 19 and 20. Mandibular gnathobase tubiform pointed terminally. Maxillule: praecoxal arthrite a lob with 1-2 short spine-like outgrowths; coxal epipodite with 5 setae; basis elongate unarmed; endopod 2-segmented bearing 1 and 4 setae; exopodal lobe with 6 setae. Maxilla: praecoxa unarmed, coxa and allobasis each with 1 seta; endopod 3-segmented, with 1, 1 and 3 setae. Maxilliped: syncoxa and allobasis each with 1 distal seta; endopod 5segmented, with 3, 1, 1, 1, and 2 setae. Leg1: segment 2 of exopod elongate. Leg 2: segment 3 of exopod with middle spine directed inward. Leg 4: coxa with inner seta, instead of spine of female. Leg 5 uniramous, 5-segmented; left leg slender, about 2 times longer than urosome; segments 3 and 5 each with small seta; right leg small, reaching 1/3 of right leg proximal segment 2.

#### Type species

Methanocalanus gabonicus gen. nov., sp. nov.

# Etymology

The genus name is a combination of the word "methano" which refers to the methane seep environment and the genus name "*Calanus*" used to form names of many genera of copepods belonging to the order Calanoida; gender masculine.

#### Taxonomical notes

*Methanocalanus* gen. nov. belongs to the family Spinocalanidae Vervoort, 1951 (57 valid species in 11 genera) analyzed phylogenetically by Schulz (1989) and diagnosed recently by Markhaseva & Kosobokova (2001). The presumably carnivorous copepods of the monotypic genus *Arctokonstantinus* Markhaseva & Kosobokova, 2001 of the family Arctokonstantinidae Markhaseva & Kosobokova, 2001, a sister family of Spinocalanidae, was considered as a derived member of the Spinocalanidae by Boxshall & Halsey (2004).

The new genus *Methanocalanus* shares with the group of the 3 most derived spinocalanid genera: *Isaacsicalanus* Fleminger, 1983; *Kunihulsea* Schulz, 1992 and *Teneriforma* Grice & Hulsemann, 1967, the following synapomorphies (Schulz, 1989, 1992 & 2004):

1. The rostrum is conical and uniramous (biramous or absent in other spinocalanids);

2. The number of setae on the segments 12 and 14 of female antennule (corresponding to the ancestral segments 15 and 17) is reduced to 1 (2 setae in other spinocalanids);

3. The distomedial seta of the right caudal ramus directed dorsally, and, that of the left ramus, ventrally. The rami, are asymmetrical in all genera except the genus *Kunihulsea* (see Schulz, 2004);

In the monophyletic group of 4 genera including *Methanocalanus* gen. nov., the new genus is sister to the derived monotypic genus *Isaacsicalanus*, in considering the only females (adult and subadult males unknown in *Isaacsicalanus*). The genera share the following derived characters:

1. The rostrum is a knob-like process;

2. Tergites of the pedigerous somites 4 and 5 fused laterally (but separated dorsally and ventrally);

3. Two setae on the syncoxa of female maxilliped (in the other spinocalanids described in details, all lobes are typically armed and bear more than 2 setae);

4. The outer distal spine on the second exopodal segment of the swimming leg 1 is lost (present in other spinocalanids);

5. The outer distal corner on the second endopodal segment of the swimming legs 3 and 4 are rounded (pointed in other spinocalanids).

The presumed derived characters of *Methanocalanus* gen. nov. are as follows:

1. Three large teeth on the masticatory blade of mandible separated by a groove from 5 others (instead of 1 tooth separated by a groove from other teeth in other spinocalanids);

2. Endopodal articulating segments 3-4 of maxilliped each with one seta (instead of 2, 2, and 3 setae in *Isaacsicalanus*); the outer seta on the endopodal segment 4 is absent (present in other spinocalanids);

3. Basis of leg 4 with stout inner spine (instead of seta).

The derived characters of Isaacsicalanus are:

- 1. Tergite of the pedigerous somite 5 pointed posteriorly;
- 2. First segment of antennule armed with 1 seta;

3. Distal segment of antennule is double (corresponds to two distal segments of female antennule of *Methanocalanus* gen. nov.);

4. First and second praecoxal endites of maxilla with 5 and 2 setae, respectively;

5. Two median setae on the allobasis of the female maxilliped (instead of maximally 3).

# Methanocalanus gabonicus gen. nov., sp. nov. (Figs 1-11)

#### Type material

Dissected holotype  $\Im$  (MNHN-Cp2323), dissected allotype  $\Im$  (MNHN-Cp2324); dissected paratypes  $\Im$  on two slides (MNHN-Cp2413 and MNHN-Cp2414); golden-coated specimens paratypes:  $7 \ \Im \ \Im$ ,  $7 \ \Im \ \Im$ ,  $7 \ copepodids \ V$  (MNHN-Cp2325). The type material is deposited in the Museum national d'Histoire naturelle, Paris.

#### Type locality

The pockmark Regab, Gabon continental margin, Southeast Atlantic  $(05^{\circ}47.80^{\circ}S, 09^{\circ}42.60^{\circ}E; \text{ depth } 3151 \text{ m}; \text{ date } 28/11/01).$ 

#### Additional material

150  $\Im$   $\Im$ , 25  $\eth$   $\eth$ , 7 copepodids of stage 5  $\circlearrowright$  (same sampling data as for holotype); 30  $\Im$   $\Im$ , 6  $\circlearrowright$   $\eth$ , 4 copepodids of stage 5 (ASPI 3; 146/09; 29/11/01; 05°47.80'S, 09°42.60'E, 3154 m); 29  $\Im$   $\Im$ , 4  $\circlearrowright$   $\circlearrowright$  (ASPI 3; 147/10; 01/12/01; 05°47.80'S, 09°42.60'E, 3155 m).

# Etymology

The specific name is derived from the name of continental margin (Gabon) where the copepods were found. Gender masculine.

# Description

Adult female. Total length of holotype female (caudal setae excluded) 1.56 mm; greatest width 0.62 mm. Prosome (Fig. 1A, B): ovoid, in dorsal view 5-6-segmented; cephalosome separate from first pedigerous somite. Epimera of somites slightly developed and rounded. Tergites of fourth and fifth pedigerous fused laterally and separated dorsally and ventrally, posterior corner of fifth tergite roundish laterally (Fig. 2C). Longitudinal midventral processes between maxillipeds and leg 1 and between intercoxal sclerites of legs 1-4 sclerotized (Fig. 1A). Urosome (Fig. 3A) 4-segmented, about 1/3 as long as prosome, comprising genital double-somite and 3 abdominal somites. Genital double-somite (Fig. 3A) with midventral operculum plate (Fig. 2C, D) covering the paired gonopores. Anal somite

with anal opening on dorsal side.

Caudal rami (Fig. 3A) about 2.8 times longer than wide, slightly assymetrical, armed with 5 setae: 4 terminal setae and 1 distomedial seta inserted and directed asymmetrically: right dorsally and left rami with seta inserted and directed ventrally; seta of left ramus curved and inserted near proximal margin of caudal ramus, seta of right ramus straight.

Rostrum (Figs 1A, C & 2A): an uniramous knob-like process fused to cephalosome.

Labrum (Figs 1C & 2A, B): produced ventrally, distal part with a small row of small setules (Fig. 2B).

Paragnaths (Figs 1A, C & 2B): a pair of lobes on pedestal produced ventrally and ornamented with numerous setules.

Labrum and paragnaths forming a sort of robust oral cone protruding ventrally (Figs 1A & 2A, B).

Antennule (Fig. 3B, C): 24-segmented, extending to leg 4. Formula for armature: 3; 6 + a; 2; 2; 2 + a; 2; 2; 4 + a; 1; 2; 2 + a; 1; 2 + a; 1; 2; 1; 2; 2 + a; 1; 1; 2; 2; 2; 5 + a (a = aesthetasc). Segment 1 with 3 setae; segments 2 and 8 composed of 3 and 2 fused segments, respectively. Segments 12 and 14 bearing 1 seta. Ventral seta on segment 21 curved, with setules.

Antenna (Fig. 1D): coxa unarmed, basis with 1 seta; endopod shorter than exopod, first endopodal segment with 2 setae, second segment with 7 setae on each distal and proximal lobes; exopod indistinctly 6-segmented with 3, 1, 1, 1, and 4 setae.

Mandible (Fig. 3D, E): gnathobase elongate, masticatory blade with 8 teeth and 1 seta; 3 strong teeth separated from 5 smaller teeth by a groove ornamented with setules; basis with 3 setae; endopod 2-segmented, first endopod segment with 3 setae, second with 10 setae; exopod 5-segmented bearing 1, 1, 1, 1, and 2 setae.

Maxillule (Fig. 4A,  $A_1$ ): praecoxal arthrite with 13 spines; coxal epipodite and coxal endite with 9 and 4 setae, respectively; first and second basal endites each with 4 setae; endopod indistinctly 3-segmented bearing 2, 3 and 3 setae, distal segment with row of setules; exopod as a lobe fused to basis and bearing 8 unequal setae and row of setules.

Maxilla (Fig. 4B): first praecoxal endite with 7 setae, one very short, the others long and slender; second praecoxal, first and second coxal endites bearing each 3 stout setae; first and second endites of allobasis bearing 4 and 1 stout setae, respectively; endopod 3-segmented, bearing 1, 1 and 3 setae.

Maxilliped (Fig. 4C): syncoxa bearing 2 setae, 1 seta proximally and 1 seta on internal distal lobe ornamented with setules. Allobasis with 3 medial and 2 distal setae, belonging to endopodal segment fused with basis, inner proximal margin of basis ornamented with small spinules.



Figure 1. *Methanocalanus gabonicus* gen. nov., sp. nov., holotype  $\mathcal{Q}$ . A. Habitus, lateral. B. Habitus, dorsal. C. Anterior part of prosome, ventral. D. Antenna.

Figure 1. *Methanocalanus gabonicus* gen. nov., sp. nov., holotype  $\mathcal{Q}$ . A. Habitus, vue latérale. B. Habitus, vue dorsale. C. Partie antérieure du prosome, vue ventrale. D. Antenne.



**Figure 2.** *Methanocalanus gabonicus* gen. nov., sp. nov., paratype  $\mathcal{Q}$ , SEM photos. **A.** Anterior part of prosome, ventral. **B.** Labrum, paragnaths and mandibular gnathobases. **C.** Posterior part of prosome and genital double-somite. **D.** Genital double-somite. **E.** Endopod of left leg1, anterior. **F.** Lobe of endopod of right leg1, anterior (= Von Vaupel Klein's organ). Abbreviations: a1 – antennule, a2 – antenna, gn – gnathobase, lb – labrum, pg – paragnath, l4 – leg 4.

**Figure 2.** *Methanocalanus gabonicus* gen. nov., sp. nov., paratype  $\mathcal{Q}$ , photos en MEB. **A.** Partie antérieure du prosome, vue ventrale. **B.** Labre, paragnathes et gnathobases des mandibules. **C.** Partie postérieure du prosome et double somite génital. **D.** Double somite génital. **E.** Endopodite de P1, vue antérieure. **F.** Lobe de l'endopodite de la P1 droite, vue antérieure (= organe de Von Vaupel Klein). Abréviations : a1 – antennule, a2 – antenne, gn – gnathobase, lb – labre, pg – paragnathe, l4 – P4.



**Figure 3.** *Methanocalanus gabonicus* gen. nov., sp. nov., holotype  $\mathcal{Q}$ . **A.** Urosome, dorsal. **B.** Antennule, segments 1-17. **C.** Antennule, segments 18-24. **D.** Mandibular palp. **E.** Gnathobase of mandible.

**Figure 3.** *Methanocalanus gabonicus* gen. nov., sp. nov., holotype  $\mathcal{P}$ . **A.** Urosome, vue dorsale. **B.** Antennule, segments 1-17. **C.** Antennule, segments 18-24. **D.** Palpe mandibulaire; **E.** Gnathobase de la mandibule.



Figure 4. *Methanocalanus gabonicus* gen. nov., sp. nov., holotype  $\mathcal{Q}$ . A. Maxillule. A1. Proximal basal endite of maxillule. B. Maxilla. C. Maxilliped.

Figure 4. *Methanocalanus gabonicus* gen. nov., sp. nov., holotype  $\mathcal{Q}$ . A. Maxillule. A1. Endite basal proximal de la maxillule. B. Maxille. C. Maxillipède.



**Figure 5.** *Methanocalanus gabonicus* gen. nov., sp. nov. **A.** Leg 1 of holotype  $\mathcal{Q}$ , anterior. **B.** Leg 2 of holotype  $\mathcal{Q}$ , anterior. **C.** Distal segment of exopod of leg 2, allotype  $\mathcal{J}$ , anterior. **D.** Leg 3 of holotype  $\mathcal{Q}$ , anterior. **E.** Leg 4 of holotype  $\mathcal{Q}$ , anterior. **F.** Inner coxal seta of leg 4, allotype  $\mathcal{J}$ , anterior.

**Figure 5.** *Methanocalanus gabonicus* gen. nov., sp. nov. **A.** P1 de l'holotype  $\mathcal{D}$ , vue antérieure. **B.** P2 de l'holotype  $\mathcal{D}$ , vue antérieure. **C.** Segment distal de l'exopodite de P2, allotype  $\mathcal{J}$ , vue antérieure. **D.** P3 de l'holotype  $\mathcal{D}$ , vue antérieure. **E.** P4 de l'holotype  $\mathcal{D}$ , vue antérieure. **F.** Soie coxale interne de P4, allotype  $\mathcal{J}$ , vue antérieure.



**Figure 6.** *Methanocalanus gabonicus* gen. nov., sp. nov., allotype  $\delta$ . **A.** Habitus, lateral. **B.** Habitus, dorsal. **C.** Anterior part of prosome, ventral. Abbreviations: a1 – antennule, a2 – antenna, gn – gnathobase of mandible, pm – mandibular palp, lm – labrum.

**Figure 6.** *Methanocalanus gabonicus* gen. nov., sp. nov., allotype  $\delta$ . **A.** Habitus, vue latérale; **B.** Habitus, vue dorsale. **C.** Partie antérieure du prosome, vue ventrale. Abréviations : a1 – antennule, a2 – antenne, gn – gnathobase de la mandibule, pm – palpe mandibulaire, lm – labre.



**Figure 7.** *Methanocalanus gabonicus* gen. nov., sp. nov., paratypes  $\delta \delta$ , SEM photos. **A.** Anterior part of prosome, ventral. **B.** Mandibular gnathobase. **C.** Tip of mandibular gnathobase. **D.** Leg 5, distal part of left leg. Abbreviations: a1 – antennule, a2 – antenna, gn – gnathobase, lm – labrum, mx1 – maxillule, mx2 – maxilla, rm – rostrum.

**Figure 7.** *Methanocalanus gabonicus* gen. nov., sp. nov., paratypes  $\vec{\sigma} \vec{\sigma}$ , photos au MEB. **A.** Partie antérieure du prosome, vue ventrale. **B.** Gnathobase de la mandibule. **C.** Extrémité de la gnathobase de la mandibule. **D.** P5, partie distale de la patte gauche. Abréviations: a1 – antennule; a2 – antenne, gn – gnathobase, lm – labre, mx1 – maxillule, mx2 – maxille, rm – rostre.

Endopod consisting of 5 articulated segments armed with 3, 1, 1, 1, and 4 setae, respectively; outer seta on segment 4 absent.

Legs 1-4 (Fig. 5A, B, D & E) biramous, with 3segmented exopods on legs 1-4 and endopods on legs 3-4, 1- and 2-segmented endopods on legs 1 and 2, respectively. Formula for armature of legs 1-4 as in Table 1. Leg 1 (Figs 2E, F & 5A) without outer distal spine on first and second exopodal segment; outer lobe on anterior side of endopod with Von Vaupel Klein's organ represented by 2 rows of flat setules and one terminal pore (not shown on figure); inner **Table 1.** Formula of the armature of swimming legs 1-4, holotype  $\Im$  of *Methanocalanus gabonicus* gen. nov., sp. nov. Roman numerals indicate spines, Arabic numerals indicate setae.

**Tableau 1**. Formule des soies et des épines des pattes natatoires (P1 à P4) de l'holotype  $\mathcal{Q}$  de *Methanocalanus gabonicus* gen. nov., sp. nov. Les chiffres romains indiquent les épines, les chiffres arabes, les soies.

	Coxa	Basis	Endopod	Exopod
Leg 1	0-0	0-1	0,2,3	0-0; 0-1; I,I,4
Leg 2	0-1	0-0	0-1; 1,2,2	I-1; I-1; III,I,5
Leg 3	0-1	0-0	0-1; 0-1; 2,2,2	I-1; I-1; III,I,5
Leg 4	0-I	0-0	0-1; 0-1; 2;2,2	I-1; I-1; III,I,5



**Figure 8.** *Methanocalanus gabonicus* gen. nov., sp. nov., allotype  $\delta$ . **A.** Urosome, ventral. **B.** Left antennule, segments 1-18. **C.** Left antennule, segments 19-23. **D.** Part of right antennule, two-sided arrows indicate homologous segments of right and left antennules. **E.** Antenna. **F.** Mandibular palp.

**Figure 8.** *Methanocalanus gabonicus* gen. nov., sp. nov., allotype  $\delta$ . **A.** Urosome, vue ventrale. **B.** Antennule gauche, segments 1-18. **C.** Antennule gauche, segments 19-23. **D.** Antennule droite (part.), les flèches doubles indiquent les segments homologues des antennules droite et gauche. **E.** Antenne. **F.** Palpe mandibulaire.



**Figure 9.** *Methanocalanus gabonicus* gen. nov., sp. nov., allotype  $\delta$ . **A.** Maxillule. **B.** Setae of coxal epipodite of maxillule. **C.** Leg 1, anterior.

Figure 9. *Methanocalanus gabonicus* gen. nov., sp. nov., allotype  $\delta$ . A. Maxillule. B. Soies de l'épipodite coxal de la maxillule. C. P1, vue antérieure.

basal seta curved, with setules covering the lobe anteriorly. Legs 2-4 with a row of small setules on anterior side of distal endopodal segment. Legs 3 and 4 with rounded outer distal corner on endopodal segment 2. Basis of leg 4 (Fig. 5E) with stout unornamented inner spine.

Leg 5 absent (see Fig. 2C).

Adult male. Differs from female as follows:

Body (Fig. 6A, B): total length (caudal setae excluded) 1.57 mm; greatest width 0.61 mm. Urosome (Fig. 8A) 5-segmented, comprising genital somite and 4 abdominal somites. Genital somite with one ventro-lateral genital flap on the left side near posterior margin of somite.

Labrum (Figs 6C & 7A): flat, not produced ventrally, with pair of distolateral bifid extensions.

Paragnaths (Figs 6C & 7A) absent.

Antennules (Fig. 8B-D) asymmetrical. Left antennule 23-segmented, distal segment corresponds to two distal segments of female antennule; formula for armature: 1 + a; 6 + 4a; 2 + 2a; 2 + a; 2 + 2a; 2 + a; 2 + 2a; 4 + 2a; 1; 2 + a; 2 + a; 1; 2 + a; 1; 2 + a; 1; 1 + a; 1; 2 + a; 1; 1 + a; 1; 2 + a; 1; 1 + a; 2 + a; 2; 7 + a (a = aesthetasc). Right antennule 22-segmented, differs from left antennule in fusion of segments 19 and 20, shape of segments 13-18, and formula for armature of segments 15-19: 2; 1; 2 + a; 2.

Antenna (Fig. 8 E): first endopodal segment unarmed, second endopodal segment with 6 and 5 setae on distal and proximal lobes, respectively; exopod indistinctly 6-segmented (two proximalmost segments not clearly differentiated) with (1,1), 1, 1, 1, and 4 setae.

Mandible (Figs 6C, 7A-C & 8F): gnathobase stylet-like, thin and tubiform, pointed and separated terminally; basis with 2 setae; first endopod segment unarmed, second with 8 setae and row of minute spinules; exopod 5-segmented bearing 1, 1, 1, 1, and 2 setae as in female.

Maxillule (Fig. 9A, B): praecoxal arthrite lobed, distal part with 1-2 short spine-like outgrowths; coxal epipodite bearing 5 very long setae; coxal and basal endites absent; basis elongate; endopod indistinctly 2-segmented bearing 1 and 4 setae; exopodal lobe bearing 6 setae and row of setules.

Maxilla (Fig. 10A): praecoxa unarmed, coxa with 1 minute seta, allobasis with 1 distal seta, endopod 3-segmented bearing 1, 1 and 3 setae.

Maxilliped (Fig. 10B): syncoxa and allobasis each with 1 distal seta; endopod 5-segmented bearing 3, 1, 1, 1, and 2 setae.

Leg 1 (Fig. 9C): second segment of exopod elongate.

Leg 2 (Fig. 5C): distal segment of exopod with middle spine directed inward.

Leg 4 (Fig. 5F): coxa with slender inner seta ornamented with setules.

Leg 5 (Figs 6A, 7D & 10C): uniramous (endopod absent), 5-segmented, forming paired complex. Right leg slender, about 2 times longer than urosome; junction of

segments 2 and 3 (basis and exopodal segment 1, respectively) with lateral frill; segment 3 with small distolateral seta; segment 5 (= exopodal segment 3) with longitudinal row of spinules and terminal seta. Left leg small, reaching 1/3 of proximal segment 2 (= basis) of right leg, distal segment separated from terminal seta indistinctly.

<u>Male of copepodid stage 5</u>. Armature of appendages, except leg 5, similar with that of female. Leg 5 (Fig. 10 D) present, uniramous; right leg 5-segmented, as in adult male; segment 3 with small distolateral seta, segment 5 narrow, with small terminal spine (indistinct); left one of same structure, but 3 times shorter than right leg.

# Internal Morphology

#### Female internal morphology.

Genitalia (Fig. 11A-D). The genital area occupies a central position in the middle third of the ventral surface of the genital double-somite. In external view only the genital operculum is visible. This operculum is a semi-circular flap without anterior pad. The only ornamentation is a rounded posterior indentation. The genital area, observed in semithin sections, is similar to these observed in other operculate genera of the families Centropagidae and Pontellidae. The operculum delimits a small underlaying cavity, the genital atrium, which communicates with the exterior via the genital slit. In all observed females, the atrium was empty. The paired gonopores occupy the floor of this atrium and are visible as crescent-shaped gonoporal slits separated by a median septum and delimiting laterally the two cuticular plates or gonoporal plates. The internal part of the genital area is formed by the egg-laying ducts which are the terminal part of the oviducts; they are gutter-like cuticular ducts, invaginations of the skeleton. Their external flexible wall is continuous with the gonoporal plate and their internal rigid wall is continuous with the median septum, delimiting a narrow central space reflecting the crescent shape gonoporal slits. Two pairs of muscles are present. The opercular muscles (m1) inserted on the operculum have their origin on the ventral wall of the genital double-somite. The muscles of the egg-laying ducts (m2) originate of the lateral part of the genital doublesomite and are fixed on the flexible wall of the egg-laying ducts. No seminal receptacle has been observed.

This anatomy of the female genitalia has been observed in many calanoids, as in Diaptomoidea (Barthélémy et al., 1998; Defaye et al., 2000). Then, we can suppose that after mating, the spermatophore discharges into the atrium, under the genital operculum; after fertilization, the eggs (or the egg-sac) are extruded via this opening.

The semi-thin sections of the prosome show the presence of the ovary, but without possible interpretation of the ovarian activity.



**Figure 10.** *Methanocalanus gabonicus* gen. nov., sp. nov. **A.** Maxilla of allotype  $\delta$ . **B.** Maxilliped of allotype  $\delta$ . **C.** Leg 5 of allotype  $\delta$ , posterior. **D.** Leg 5 of paratype copepodid stage 5 ( $\delta$ ), posterior.

**Figure 10.** *Methanocalanus gabonicus* gen. nov., sp. nov. **A.** Maxille de l'allotype  $\delta$ . **B.** Maxillipède de l'allotype  $\delta$ . **C.** P5 de l'allotype  $\delta$ , vue postérieure. **D.** P5 du paratype C5 ( $\delta$ ), vue postérieure.



**Figure 11.** *Methanocalanus gabonicus* gen. nov., sp. nov. **A-D.** Photomicrographs of semithin transverse sections of genital doublesomite (section A is posteriormost, D anteriormost). Abbreviations: ga – genital atrium, go – genital operculum, m1 – opercular muscle, m2 – egg-laying ducts muscle, ed – egg-laying duct, ov – oviduct, at – atrial slit. **E-F.** Photomicrographs of semithin longitudinal sections in the male reproductive system. Note the material (asterisk) in the lumen and the rows of dense granules in the cells revealing the high metabolic activity of the testis. Scale bars: A-D: 100  $\mu$ m, E-F: 50  $\mu$ m.

**Figure 11**. *Methanocalanus gabonicus* gen. nov., sp. nov. **A-D.** Photomicrographies de coupes transversales semi-fines du double somite génital (section A la plus postérieure, D la plus antérieure). Abréviations : ga – atrium génital, go – opercule génital, m1 – muscle operculaire, m2 – muscles des canaux de ponte , ed – canal de ponte, ov – oviducte, at – fente atriale. **E-F.** Photomicrographies de coupes semi-fines longitudinales dans l'appareil reproducteur mâle. Noter le matériel (astérisque) présent dans la lumière et les rangées de granules denses dans les cellules révélant l'intense activité métabolique. Echelles : A-D : 100  $\mu$ m, E-F : 50  $\mu$ m.

Digestive system. On the semi-thin sections, in spite of the inadequate fixation and the bad state of the internal organs, the labral glands and the oesophagus could be recognized as well as some cells of the midgut even if most of this part was destroyed. These observations indicate the presence of a functional digestive system.

<u>Male internal anatomy</u> (Fig. 11E, F). In order to confirm a possible absence of digestive activity in the males, we realized semi-thin sections in the prosome. In all the males observed (4), we could not identify any structure corresponding to the digestive tract. By contrast, we could observe high cells arranged as a duct and containing two parallel rows of dense secretion grains. Moreover, in some specimens, a dense material is observed in the lumen (L). Because of the inadequate fixation, it has not been possible to realize TEM observations to precise the real activity of this apparatus. Previous observations made on many copepod species lead us to interprete these structures as the male reproductive system, in a phase of intense metabolic activity for reproduction. No semi-thin section shows the presence of digestive structures. These preliminary observations suggest that the male is lacking any digestive system, as we firstly supposed it from the morphology of the buccal appendages. However, further studies will be necessary to definitively establish the complete absence of digestive structures.

#### Remarks

The family Spinocalanidae Vervoort 1951 is "a common group of small- to medium-sized calanoid copepods found at meso- and bathypelagic depths of all oceans" (Schulz, 1996). Only two species of spinocalanids, *Isaacsicalanus paucisetus* Fleminger, 1983 from hydrothermal vent at the East Pacific Rise and *Damkaeria falcifera* Fosshagen 1983 from the Norwegian Sea, were indicated in the list of calanoids reported in the near-bottom environment (Bradford-Grieve, 2004). Among new species of deep-sea benthopelagic calanoids described recently, two more spinocalanid species (*Damkaeria bicornuta* Schulz, 2004 and *Kunihulsea antarctica* Schulz, 2004) come from epibenthic samples collected at the Antarctic continental slope of the eastern Weddell Sea (Schulz, 2004 & 2005). The finding of *Methanocalanus gabonicus* gen. nov., sp. nov. extends the list of deep-sea benthopelagic calanoids and, after *Isaacsicalanus paucisetus* Fleminger, 1983, is the second species of calanoid copepods recorded in swarms in deep sea chemosynthetic environment. The description of other calanoid copepods from deep sea chemosynthetic environments were based on the finding of one or, rarely, several specimens (Ferrari & Markhaseva, 2000; Markhaseva & Ferrari, 2005).

The feeding apparatus of the adult females, as well as the male copepodids V, of *Methanocalanus gabonicus* gen. nov., sp. nov. is characterized by the protruding oral cone formed by the well developed labrum and paragnaths (Figs. 1 A; 2 A, B), the elongate mandibular gnathobase with wide masticatory blade armed with stout teeth, the stout setae on the praecoxal arthrite of the maxillule and the endites of the maxilla, the small number of setae on the proximal segments of maxilliped. All these morphological features suggest that the females of *Methanocalanus gabonicus* gen. nov., sp. nov. probably feed on small particles such as detritus and bacterial flocks, grasping from or near the bottom environment.

On the contrary, the adult males of *Methanocalanus* gabonicus gen. nov., sp. nov. distinguish by reductions and modifications of labrum, paragraphs, mandible, maxillule, maxilla and maxilliped as well as by the probable lack of digestive apparatus or by a non-functional one. All these features, added to the lower abundance of males than females suggest that males of *M. gabonicus* gen. nov., sp. nov. do not feed and die soon after copulation.

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