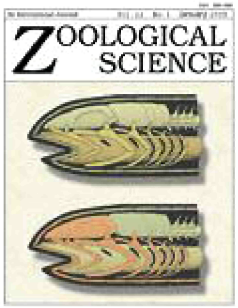

Biodiversity in the stygobiontic cirolanids (Crustacea, Isopoda) from the Mediterranean Basin: II - Systematics, ecology and historical biogeography of Typhlocirolana tiznitensis n.sp., the first representative of the genus, South of Moroccan High Atlas

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Short title: A new stygobiontic Cirolanidae from southern Morocco

Field: Taxonomy

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ABSTRACT - A number of species of the genus *Typhlocirolana* Racovitza, (1905) are already known from the Western Mediterranean countries, mainly from Spain, Sicilia and North Africa. For the first time a species, *Typhlocirolana tiznitensis* n.sp., is reported and described from a region located on the southern side of the Moroccan High Atlas. Thus the distribution area of the genus is significantly extended southwards. Ecological, biogeographic and phylogenetic relationships of the new species are presented. The species belongs to the most primitive lineage of the genus and this group of species has representatives in different other regions, which were all flooded by the sea during the Cenomano-Turonian marine transgression. The occurrence of a such a species in the Tiznit province strongly suggests that the marine paleo-gulfs of Tarfaya in the South and the High Atlas gulf in the North - apparently separated by an emerged land during the Mesozoic times - may have been in fact connected together, from the North to the South. The Cenomano-Turonian sea very probably covered the whole western margin of the present Morocco, including the Tiznit province where the mesozoic sediments have not been preserved.

Key words: Cirolanidae, Crustacea, groundwater isopods, historical biogeography, historical geology, Morocco, *Typhlocirolana* systematics.

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INTRODUCTION

Within the crustacean order Isopoda, the important family Cirolanidae – which includes more than 400 species - is still mainly marine and did not succeed in the conquest of surface freshwaters. However a good number of species are known from continental subterranean waters, in all parts of the world, either in karstic habitats or more often in interstitial milieus This number of subterranean cirolanid species was sometimes underestimated since some authors consider approximatively 50 species (Hobbs, 2004) and probably overestimated when the same author (Hobbs, 2005) give the number of 92 species. In fact the most probable number of known species, really stygobiontic, following Botosaneanu *et al.* (1986), Coineau *et al* (1994), Botosaneanu (2001) or Coineau and Boutin (2004), and considering the last discoveries (Jaume and Garcia, 1992; Boutin, 1993a; De Grave and Herrando-Pérez, 2003; Boutin *et al.,* 2002) is probably between 72 and 75 species.

The last paper dealing with the diversity of stygobiontic cirolanids from North Africa (Boutin *et al.*, 2002), included the description, the distribution pattern, the ecology and historical biogeography of *Typhlocirolana haouzensis,* a species widespread everywhere in groundwaters of the Haouz plain, North of Marrakesh (Morocco) and in neighbouring regions, all located North of the High Atlas. The main ecological characteristics and the systematic position of this species, within the genus *Typhlocirolana*, and its as well as its origin were also presented. The same paper included a brief review of the different stygobiontic cirolanids known from the Mediterranean Basin countries, belonging to the "*Typhlocirolana* group" of genera, *sensu* Wägele (1989), particularly of the species of the genus *Typhlocirolana*. The species *T. haouzensis* was only the third species formally and completely described from Morocco after the short diagnosis of *T. rifana* by Margalef (1958) and the description of a very different species, *T. leptura* by Botosaneanu *et al.* (1985), but the authors mentioned the presence of the genus in almost all provinces of Morocco where it is represented by a great number of populations, apparently belonging to a great number of different species. All these species are living in groundwaters of plains as well as in subterranean waters of High Atlas and other mountain valleys (Yacoubi-Khebiza, 1990 and 1996; Yacoubi-Khebiza *et al*, 1999). The authors also reported that previous studies of individuals of *Typhlocirolana,* occasionally collected in southern Morocco, failed to assign these Moroccan specimens to species already known from other countries or to clearly recognize new Moroccan species to be described (see especially Nourisson, 1956). In this way the systematics of *Typhlocirolana* species from North Africa seemed for a long time to be especially difficult as already pointed out by many different authors such as Monod (1930), Nourisson (1956), Benazouz (1983) and Botosaneanu *et al.* (1985).

The distinction of the different species of stygobiontic crustaceans belonging to the same genus, or to different genera - occasionally occurring together or in different stations of the same region - is nevertheless of paramount importance. Indeed it has been shown that the sensitivity of different stygobiontic crustaceans species to different pollutants which may be present in groundwaters is generally different from one species to another (Boutin 1984, 1987, Boutin et al. 1997; Fakher El Abiari et al. 1998). Consequently the regional variations in the species richness and the distribution pattern of these crustaceans may be the indication of water quality variations in the region; Yacoubi-Khebiza et al (1999). As well as the occurrence of one single species in a station where two or more related species of the same genus have previously occurred reveals a gradual change in the water quality. The use of the stygofauna as a very cheap tool for monitoring the global quality of well water used by human populations in rural zones of Maghreb countries, will be possible as soon as the diversity of this fauna will be known (Boutin, 1987; Fakher El Abiari, 1999). The first step being the identification of species.

In spite of the preceding studies, Boutin *et al.* (2002), stated that the different species of Moroccan *Typhlocirolana* are nevertheless well recognizable, and can be clearly characterized and described, if the observations and the comparisons are performed, considering a sufficient number of adult specimens, both males and females, collected at the same site, and thus forming a good representative sample of the type population. Furthermore the authors emphasized that all the morphological characteristics of the body and appendages and especially the chaetotaxic patterns of adult specimens, have to be considered in the descriptions of species, in view of possible comparisons. It seems of some interest to remind here that more than 30 years ago, Vandel (1970) in a short paper devoted to the isopodological works of Racovitza, already expressed a similar opinion and reported that the different studies of new crustaceans by E.G. Racovitza were still considered of an exceptionnal quality, fundamentally owing to the method used by the great Romanian zoologist. Indeed Racovitza has always done his utmost for presenting an extremely complete morphological description of the species he described, including very numerous figures of all parts of the body of the isopod. He emphasized that a new isopod can be known correctly and described only after a complete dissection and study - which imply the destruction of the type specimen - even if this type is too often considered by some zoologists as the main reference. It is now generally accepted that a good description of a species has necessarily to be based on the study of a group of specimens, and not only on a sole individual, the type specimen, as the characteristics of any species are at minimum that of a population (or of several populations), always exhibiting a certain variability, and not that of one "type specimen". In fact these general principles were already considered in the description of *Typhlocirolana haouzensis*. They will be also followed hereafter, since this way seems to be a necessity in order to make possible a future study of the evolutionary history of the genus, performing a phylogenetic analysis of the morphological traits of the known species. *Typhlocirolana haouzensis* is a large species which occurs in a very high number of stations, all located North of the Moroccan High Atlas (Boulanouar, 1982, 1986, 1995). *Typhlocirolana tiznitensis* n. sp. hereafter described, is a new species equally remarkable by its large body size, which occurs only South of the western High Atlas, and only very close to Tiznit, a small city of South-Western Morocco, located 90 km South of Agadir at some 15 km from the Atlantic seashore (Boulal, 1988).

DESCRIPTION OF *TYPHLOCIROLANA TIZNITENSIS N.SP.*

Study matériel and method: The description is based on the study of a dozen of individuals, including six males and six females, selected among the largest specimens. All the sudied specimens were sampled in the well P1 from Bounaamane, a small village located 20 km in the South South-East of Tiznit city (Boulal, 1988). All individuals were measured; three males and three females were completely dissected and the others observed; finally the whole group constitutes the syntype series. After clearing in lactic acid, appendages were mounted in polyvinyl-lactophenol and preserved in the author collections in Marrakesh. They are drawn using a clear chamber adapted to a CHA Olympus binocular microscope.

General aspect: Species robust, unpigmented and eyeless, clearly flattened ventrally and convex dorsally. Lateral margins of pereion parallel (Fig. 1). In dorsal view, the cephalon and the anterior part of the first pereionite as well as the pleotelson backwards, are regularly rounded. The maximum length of the body, along the medio-dorsal line, from the anterior tip of the head to the posterior tip of the pleotelson, reaches 14.0 mm in males and 14.5 mm in females. The maximum body width, dorsally measured at the level of the 6th pereionite, reaches 3.2 and 3.7 mm depending on the sex, the maximum size of females being always slightly superior to that of males.

Antennulae: Short; total length hardly oversteping the peduncle of second antennae. The A1 peduncle is formed by three segments increasingly elongated, proportionally to 1, 1.1 and 2. The flagellum, hardly as long as the peduncle, comprises 11 to 13 articles in male and 12 to 14 in female. Each flagellum article bears one or some simple setae (Fig. 2), with 2 olfactive lamina (or aesthetascs), except the first and the penultimate articles which, generally have only one single aesthetasc, and the last one which is always devoid of aestetasc or seta. The chaetotaxic formulae (giving the number of aesthetascs occurring on each article of the flagellum, from the basal one to the last one) observed in males are: 122123232210 or 122222232210 or 0222222232210, and in females: 12222232210, or 122222223210 or 1222222221110, or 13222222311210. Thus the total number of aesthetascs is 21 or 22 in males, when this same number ranges from 19 to 24 in females; so it is not significantly different in the two sexes, mainly if we consider the size of some females.

Antennae: Folded back along the lateral margin of the body, they reach the posterior edge of the 4th pereionite (Fig. 1). The lengths of the five peduncular articles (measured from the middle of the base to the apex of each article) are more or less proportional to 1, 1, 1.1, 2.4 and 3.1 (Fig. 3). The flagellum, 1.5 times longer than the peduncle, is formed by 30 to 32 articles regularly smaller in diameter and shorter from the base to the apex. All articles (except the two last segments) bear a similar equipment including one distal seta on the inner side and one tuft of 3 or 4 simple setae, also distally inserted but on the outer side.

Mandibles: As in other species, the *pars incisiva* displays three teeth, flattened in the same plan and similar in size on the right mandible and four teeth on the left mandible, including the inner tooth which is perpendicular to the plan of the three others (Fig. 4 et 5). The *lacinia mobilis* is a protuberance bearing a series of 12 to 15 strong spines, most of them denticulated (Fig. 4 to 6). Tongue-shaped *pars molaris* with 24 to 31 teeth (or strong spines). The mandibular palp includes three articles, the first one being always glabrous, the second one bearing 12 to 15 setae including one or two simple and small basal setae, near the middle of the article, about ten barbulated setae aligned along the outer edge and four more or less long and simple terminal setae. The third and shortest article bears 10 to 13 short and simple setae on the outer edge and two long barbulated setae, stronger than the others, at the tip of the palp (Fig. 4 and 5).

Maxillule: The smaller and inner lobe is rounded and bears two small simple setae and three strong and large plumose setae bearing fine hairs on their third part (Fig. 7). Outer and main lobe exhibiting 11 inner and rather similar teeth usually denticulated at their tip, and with two thinner setae, one distally denticulated on its terminal part, planted between the two more inner teeth and the other, the thinnest simple seta, planted at the base of the fifth tooth.

Maxilla: The outer lobe is armed with 5 to 7 distal pectinated setae, regularly increasing in length towards the distal part (Fig. 8). The median lobe bears 6 to 8 setae more or less similar, denticulated on their distal part, four or five are terminal and three subterminal on the inner edge. The endite or inner lobe bears a total of 13 to 15 setae including a very short and simple seta isolated at the base of the lobe, close to the outer lobe, and a dozen of setae of different length, arranged along the inner part of the endite including always a very strong plumose seta on the distal and basal angle of the lobe.

Maxilliped: More or less similar to that of other species, especially that of *T. haouzensis*; it includes a basipodite of two unequal articles, a short and a long one, an endopodite of five articles, the first one (or ischion) glabrous and the four others with a good number of setae on the two edges; a well developped endite or masticatory lobe bearing five long plumose setae and one coupling hook (Fig. 9 and 10).

Pereiopods:

First pereiopod prehensile (haptorial), shorter but stronger than the others. Basis sub-rectangular (Fig. 11) with a relatively rectilinear inner side (when it is generally curved at its terminal third in other species such as *T. houzensis*). Ischion almost triangular, bearing only some small simple setae. Inner edge of the meros (Fig. 12) bearing five teeth: three molariform spines and two denticulated strong spines with additionnal setula, and two simple setae, longer and thinner, located between the first denticulated spine and the third molariform spine. Three other short setae (two on one side and the third on the other side) of the meros are also present, like in other species. Three spines with additionnal hair are present on the inner edge of the dactyle and a "dactylian organ" (Racovitza, 1912) occurs at the end of the dactyle, formed by a terminal and transversal alignment of half a dozen of simple setae.

Pereiopods 2 to 7 are relatively slender and elongated (Fig. 13 to 18), this elongation being more and more marked from the pereiopod 2 to the pereiopod 7; the ratio of total length of P7 - including median length of the 6 articles, from basis to dactyle without the terminal nail - to that of P2 for instance, is 1.47. Ischion of pereiopods 2 and 3 exhibit, on their outer side, an angular knob well marked with a strong spine with additionnal setula on the summit. In males, the propode of pereiopods 2 and 3, is characterized, on its inner side, by the presence of the enigmatic "propodial organ" (Fig. 13 and 14) already reported in different species of the genus *Typhlocirolana* by Racovitza (1912). On the outer margin of the basis there are two to six palmate setae when the inner margin bears only four or five small simple setae. The three first articles of the endopodite (ischion, meros and carpos) bear at their end two tufts of spines with additionnal setulae, one on the inner angle and the other on outer angle, and always some small spines more on their inner margin (Fig. 13 à 18).

Pleopods:

Pleopod 1 with a short regularly rectangular protopodus, 1.7 times more wide than long, with a series of eight or nine coupling hooks (or retinacles) on the distal part of the inner margin. This sympod also bears, in pleopods 1 to 5, a small simple seta at its distal angle on the outer margin. The two ramous are single plates, the endopod slightly longer than the exopod. The exopod is a regularly oval plate, 1.5 times longer than wide, equiped with 26 to 30 plumose setae regularly arranged on the distal margin and along the 2/3 of the outer margin. The endopod slightly longer than the exopod, is a subrectangular plate (2.6 times more long than wide) with round angles; the inner margin straight and glabre is in the prolongation of the inner sympod margin; the terminal margin and the second half of the outer margin display a regular row of 23 to 26 plumose setae (Fig. 19).

Male pleopod 2 (Fig. 21): The protopod is similar to the first one but slightly shorter (two times more wide than long) and bears a series of 10, or sometimes 11 coupling setae including six to eight curved and smooth hooks and two to four long straight plumose setae (6+4, 8+3, 8+2 or 7+3, see Fig. 22). The exopod is regularly oval, 1,55 times more long than wide, bearing 47 to 52 marginal plumose setae on the distal part of the plate and a row of very small setulae on the basal outer angle. The endopod is an elongated and linguiform blade, slightly shorter than the exopod, bearing a small number of terminal plumose setae (11 to 16); near the basal and inner angle, the *appendix masculina*, a long flat stem slightly and externally curved and thicker on the inner margin, like a sabre blade is attached and jointed. In adult males, this *appendix masculina* is almost twice longer than the endopod plate and clearly longer than the whole appendage (Fig. 21) and thus well distinct on the ventral side of the isopod without dissection.

Female pleopod 2 (Fig. 23): The sympod is more or less similar to that of male, with a hooking system less important including only six or seven retinacles and one or two plumose setae, and elsewhere a row of thin setulae on the basal part of the outer margin of the sympod; these setulae are also present on the sympod of the following pleopods. The exopod, rounded and very wide (only 1,3 times longer than wide) bears 53 to 55 plumose setae on the terminal margin, and a series of small setulae on the basal and outer angle, like in male and like exopods of third to fifth pleopods. Endopod regularly rectangular with rounded angles, as long as the exopod, wider than that of pleopod 1 (2,25 times longer than wide), and with 19 to 22 marginal plumose setae planted on the terminal part.

Pleopods 3, 4 and 5 (Fig. 24, 20 and 25 are more or less similar and similar in both sexes. Sympods are very short (two times shorter than long), bearing the small simple and basal outer seta and, in pleopods 3 and 4, an inner hooking system including respectively four to six retinacles and two or three plumose setae, and in pleopod 4 three or four retinacles associated with two or three plumose setae. The endopod is modified: it is a wholly fleshly and glabrous organ, with a thin and supple tegument, apparently well adapted to a respiratory (gill) function; it is more or less regularly oval and distinctly shorter than the exopod. The exopod, also regularly oval and wide (1,5 to 1,6 time longer than wide), exhibits a transversal suture just marked in the pleopod 3, but forming a true articulation in pleopods 4 and 5 which consequently are formed of two different plates, the basal one clearly greater than the second one. The chaetotaxic equipment is more and more reduced backwards: 42 to 46 plumose setae, with only three on the basal part of exopod in pleopod 3, some 20 with only one on the basal part in pleopod 4, and a total of only four relatively short plumose setae on the pleopod 5, on the inner angle of distal article.

Uropods are relatively elongated; the proportions, shape and chaetotaxic equipment of each of the three parts are characteristic of the species.

The sympod is sub-trapezial and 1,5 times longer (along its median line) than wide (in the middle of the median line); the inner margin, longer than the outer one, is glabrous on its main part and bears 10 to 14 long plumose setae on its terminal part; the distal margin is concave and passes beyond the base of the two ramous, mainly on inner side; the outer margin bears only five or six small simple sub-marginal setae and two strong spines (with additionnal small setae) and a small simple seta at the apex.

Endopod lanceolate made of a triangular plate, broadened at the base, bearing on the inner margin seven to nine plumose setae and two or three strong spines (with additional setae); a tuft of a dozen of simple and flexible setae of different length is inserted in a small apical excavation. On the outer margin two spines occur near the apex, at the same level as that of the inner margin, with two groups of two palmate setae (often considered as acoustic organs); three other small palmate setae also occur near the outer margin, near the base of the endopod.

The exopod, clearly narrower than the endopod, seems also to be a little shorter (because of the place of its insertion point) but it is in fact slightly longer (if the lengths are measured along the median line of each ramous). There are two strong spines on the external margin and another on the inner margin planted on the fourth part of the ramous, and a fourth smaller spine, submarginal, near the middle of the outer margin, with four or five small simple setulae. The inner margin of the exopod also bears a couple of acoustic palmate setae, near the base of the ramous, these two remarkable setae being always present in all species of *Typhlocirolana*. At the exopod end, a tuft of some 15 simple and flexible setae of different length occurs in the apical cupula. The maximum width of the endopod and that of the exopod (measured perpendicularly to the median line of each ramous) are respectively 0.36 et 0.26 times as long as the median line of each ramous.

The pleotelson is sub-triangular, but slightly convex on the anterior margin and regularly rounded backwards, with a light median and apical tip (Fig.27). Its median length reaches 1/6 of the total body length. The pleotelson can be inscribed in an isoceles almost equilateral triangle (the median length of the pleotelson is about 89% of its maximum base width but the measure of this ratio is always imprecise as it is depending on the crushing more or less important of the originally convex pleotelson, between plan slides..). The apical margin of the pleotelson is gently denticulated, close to the tip, and bears a number of simple setae ranging from 10 to 14; some other small setae are sub-marginal and not very different of other setulae scattered on the whole dorsal surface of the pleotelson (Fig. 27).

Discussion and taxonomic comparisons

Typhlocirolana tiznitensis n.sp. belongs to a group of robust and large size species such as *T. buxtoni* an Algerian species described by Racovitza (1912) and *T. haouzensis* Boutin et al. 2002. A first global and morphological similarity suggests that the new species from Tiznit could be closely related to the two others (especially to the second one, due to a relatively similar shape of the pleotelson). However the methodical comparison of the morphology of the different appendages and of their chaetotaxy shows important differences with both the two other Moroccan species and especially with the Algerian species described by Gurney (1908) and Racovitza (1912) Thus we mainly list hereafter the differences between the two Moroccan species and we also mention the main differences and similarities with the Algerian ones.

Due to its adult body size (14 mm in males and 14,5 mm in females), the new species is the longest species known from Morocco (*T. haouzensis* reach only 11 and 12 mm); it is just a little smaller than *T. buxtoni* (the size of the largest males may reaches 15 mm). However if such a characteristic is of some interest in view of a diagnosis of species, surely it is not necessarily indicative of close phylogenetic relationships. Nevertheless the differences in body size between several species have to be considered for the interpretation of some chaetotaxic traits, as the number of setae or spines, regularly arranged on the rim of an article, often increases with the age and the size of individuals. Therefore some chaetotaxic differences which may be partly correlated to differences in body size are probably less informative than others which vary in the opposite direction (Boutin et al., 2002).

The maximum number of articles in the antennula flagellum in *Typhlocirolana tiznitensis* n.sp. (13 and 14 in male and female) is higher than in any other species (11 and 12 in *T. buxtoni*, the largest species, and only 8 and 10 in *T. haouzensis* which is just a little smaller than the new species). The number of aesthetascs is also significantly higher than in other species (20 to 24 in the female and 21 or 22 in males of the new species, when it is only 18 to 20 in *T. buxtoni* and 11 to 16 in females and 12 to 17 in males of *T. haouzensis*). The sensitive structures and probably the correlative functionnal role of the antennula in *T. tiznitensis* seem to be more developped than in other species.

Conversely, the second antenna seems to be more reduced than in other species: the flagellum includes 30 to 32 articles in the Tiznit species against 30 to 40 in the Algerian species and 31 to 41 in *T. haouzensis.* The relative length of the antennae is also clearly shorter than in other species as, stretched along the body side, the second antenna reaches only the posterior edge of pereionite 4 in *T. tiznitensis*, and pereionite 5 in *T. haouzensis* or *T. buxtoni*. It may be tempting to interprete the specific peculiarities of antennulae and antennae in the new species as adaptations to the life in interstitial habitats : in a subterranean habitat where food resources could be often scarse, a good antennular chemoreceptive system may be of some interest and positively selected by natural selection. But in the same interstitial habitat, the obligate exploration, usually with antennae elongated ahead, of many small holes and irregular spaces between substrate grains in which the individual has to penetrate, is easier if the antennae are not too long. Therefore the positive selection of long antennae observed in so many troglobites living in open water of caves is not acting in interstitial stygobites (Coineau, 2000) and a better fitness may be related with short antennae. Finally, a relative shortening of antennae with reduction of the number of flagellum articles as well as the increasing number of the aesthetascs could be very likely apomorphic traits of the new species.

The mandibular palp is roughly similar in the different species of the genus, except in some details of its chaetotaxy. The first of the three articles of the palp exhibits a macroseta in some species such as the Oran species *T. buxtoni* or in *T. rifana* Margalef, 1958 from Northern parts of Morocco. Conversely the basal article of the mandibular palp is perfectly glabrous in *T. tiznitensis* n.sp. like in *T. haouzensis* or *T. fontis*, a widely distributed Algerian species. The chaetotaxic pattern of the second article of the palp also leads to bring together the two Moroccan species as it includes 12 to 15 setae in the new species and 12 to 14 in *T. haouzensis* but 19 setae in *T. buxtoni*. Similarly the third and last article of the mandibular palp bears a row of 27 to 30 and more setae in *T. buxtoni* and only 13 to 16 in *T. haouzensis* and only 10 to 13 in *T. tiznitensis*. Similarly again, the mandibular *lacinia mobilis* comprises more than 16 denticulate spines in *T. buxtoni* and only 12 to 15 in *T. tiznitensis* and *T. haouzensis*. In contrast, the *pars molaris* of the mandibula comprises only 25 short spines in *T. buxtoni* and until 27 in *T. haouzensis* and until 31 in the new species. Thus the different chaetotaxic traits of the mandibula tend to bring together the two strong Moroccan species (*T. haouzensis* and *T. tiznitensis*) and allow to distinguish them from *T. buxtoni*.

The maxillula chaetotaxy and morphology provide only some distinctive traits: the internal lobe bears three strong and plumose setae and two small smooth setae in the three considered species, and the external main lobe exhibits nine strong spines and three strong setae, offering only some minute differences between the species.

The comparative chaetotaxy of the maxilliped palp is more interesting: the first article (ischion) of the endopodite is always glabrous in *T. tiznitensis*, when it exhibits one terminal seta well formed in *T. haouzensis* and in *T. buxtoni*. This autapomorphic trait characterises the new species. Conversely when *T. buxtoni* and *T. fontis* have always two coupling hooks on each maxilliped endite, *T. tiznitensis* n.sp, like *T. haouzensis,* has only one single coupling hook on each endite, another apomorphic trait which suggests a closer relation between the two Moroccan species. Another synapomorphy of the Moroccan species may be mentioned : only four or, more often, five strong plumose setae are planted on the endite in *T. tiznitensis* as well as in *T. haouzensis*, when six plumose setae are present in *T. buxtoni*.

The pereiopod 1 merus of *T. tiznitensis* n.sp. is armed with a complex row of teeth on the internal margin, including a total of seven units (three short and strong molariform teeths, two pointed and denticulate spines and two unequal simple setae), when there are only six units in *T. haouzensis*, and seven in *T. buxtoni* but of two kinds only: six short spines and one seta. Thus the merus of P1 is more apomorphic in the new species.

The propodial organ of pereiopods 2 and 3 is a remarkable secondary sexual character. It is present only in males (and always lacking in females) in *T. tiznitensis* like in *T. haouzensis,* in *T. gurnei*, *T. fontis*, when it occur both in males and females in *T. buxtoni, T moraguesi* and in another unnamed *Typhlocirolana* sp. from Morocco observed by Nourisson (1958). That is another important difference with one species from Algeria, and a similarity within the two Moroccan species and two other Algerian species of the *fontis* group.

The new species is characterized by a pleopodal chaetotaxy abundant if compared with that of related genera *Marocolana* and *Turcolana*, and also with some other species of the genus *Typhlocirolana* :

In first pleopod exopodite, the chaetotaxy (23 to 26 setae) is significantly more important than that observed in *T. haouzensis* (14 to 18 setae) but very significantly more reduced than in *T. buxtoni* (40 setae and more); similar differences characterise the chaetotaxy of the endopodites. The shape of the exopodite is more elongated than in the two other species and the part of the external margin without setae more reduced (1/3) than in the two other species (about 1/2).

In male second pleopod of adults, an important trait is the shape and the relative length of the *appendix masculina*, compared to that of the endopodite ramous. In the new species, this ratio (1.75) is higher than in *T. buxtoni* (1.6) but lower than in *T. haouzensis* (1.9). The number of plumose setae present on the endopodite (11 to 16 setae) is, on the contrary, lower than in *T. buxtoni* (30 and more) and higher than in *T. haouzensis* (only five to seven). The chaetotaxy of the exopod is very important (47 to 52 setae), compared with that observed in other species (some 30 in *T. buxtoni* and 26 to 35 in *T. haouzensis*).Similar differences exist also in female pleopod 2: the exopod bears 53 to 55 setae in *T. tiznitensis*, only 50 in *T. buxtoni* and 30 to 32 in *T. haouzensis.* The differences in the shape of exopods, more elongated in the new species than in others, already mentioned in the first pleopods, is also observable in the second pleopods.

The general shape of the uropods of the new species is more or less similar to that of *T. haouzensis* but the chaetotaxy is different: on the inner margin of the sympod there are 10 to 15 plumose setae in the new species and only 6 to 9 setae in *T. haouzensis.* Conversely, on the outer margin of the sympod there are only two terminal spines in *T. tiznitensis* when in *T. haouzensis* there is also a third spine, near the middle of the outer margin of the sympod. Similarly the outer margin of the exopodite of *T. haouzensis* exhibits more spines than the new species. The abundance of the chaetotaxy occurring on the outer margin of the uropods is not very different in *T. tiznitensis* and *T. buxtoni* but the general shape of the two ramous is different as the exopodite is broader and the endopodite more elongated and styliform in *T. buxtoni*.

The shape of the pleotelson is clearly different in the three considered species: the ratio median lenght /maximum width of the pleotelson is about 0.9 in the new species, it is 1.0 in *T. haouzensis* and 1.1 in *T. buxtoni*. More important, the pleotelson of *T. tiznitensis* is more regularly rounded than in *T. buxtoni* and moreover than in *T. haouzensis*, the narrowest one which looks like an isocele triangle. By its general shape, the pleotelson of the new species looks like the pleotelson of *T. buxtoni* rather than that of *T. haouzensis* but the chaetotaxy is similar in the two Moroccan species, including only 8 to 12 small simple setae on the posterior margin when there is an important row of some 30 large plumose setae plus 6 small spines on the posterior margin of the pleotelson of *T. buxtoni.*

Finally it appears that the species of *Typhlocirolana* from Tiznit significantly differs from the other Moroccan species described from the Haouz of Marrakech, North of the High Atlas, by a good number of characters. Some of them are very likely plesiomorphic traits more retained in *T. tiznitensis* than in *T. haouzensis* : body size and general body shape, importance of the chaetotaxic coating, especially of external plumose setae and spines of locomotor appendages, and probably the shape of the pleotelson. Conversely the two Moroccan species share some synapomorphic traits when compared with *T. buxtoni* and other species, such as the chaetotaxic pattern of the maxilliped and of the mandible, the loss of the propodial organ in pereiopods 2 and 3 of the females. The new species is also characterized by some autapomorphic traits such as the great number of aesthetascs on the antennulae, the shortening of the antennae (with conservation of a good number of aesthetascs), the differenciation of the marginal armature of the first pereiopod meros etc... The global similarities with *Typhlocirolana buxtoni,* especially when we compare the two species also with other Moroccan species, appear to be related to the conservation of plesiomorphic traits and thus they may be remarkable at first sight without being the sign of a close relationship between the new species. and *T. buxtoni*.

In other respects, *T. tiznitensis* n.sp. is very different from the two other Moroccan species not considered here, *T. rifana* Margalef, 1958 and above all from *Typhlocirolana leptura* Botosaneanu et al., 1985 which is still to date a very original species, sole representative of one of the three lineages forming the genus (Fig. 28). *T. tiznitensis* n.sp. differs also from *T. moraguesi*, Racovitza, 1905, the species from the Balearic Islands, and from the two other Algerian species, *T. fontis* (Gurney, 1908) and *T. gurneyi* Racovitza, 1912. Some features of these species are given by Boutin et al (2002) in the part including taxonomic comparisons of *T. haouzensis* with other species. In the same paper some characters of the small sized species *T. reichi* (Por, 1962) are also presented since this last species is probably the sole from the Middle East countries and especially from Israel and Palestine which may be maintained within the genus *Typhlocirolana* (Boutin, 1993a), the other species first described from these countries as *Typhlocirolana* (by Strouhal, 1961 and Herbst, 1982) having justifiably been transfered in the genus *Turcolana* Argano and Pesce, 1980 by Botosaneanu and Notenboom (1989). Finally it is clear that the Tiznit population of *Typhlocirolana*, is well distinguishable from all other known species of the genus, and has to be considered as the representative of a new species.

Derivatio nominis.

Because of the geographic localisation of its distribution area, the new species is named *tiznitensis*, as it is abundant in several wells of the Tiznit city or located in the same province, close to the city.

GEOGRAPHIC DISTRIBUTION

The type station of the new species, as above mentioned, is a well located in the Douar (= village) Bounaamane. This place is some 20.3 Km S-SW of the centre of Tiznit city and at about 15 Km from the Atlantic shore. Although several hundreds of wells have been investigated in the Tiznit Province and the surrounding region where a stygofauna including other species of *Typhlocirolana* occurs (Boulal, 1988 and 2002; Idbennacer, 1990), *Typhlocirolana tiznitensis* n.sp. is known only from eight wells: the first one is the type station of Bounaamane, another well is located 2.5 Km SW of Tiznit, and the others 2 to 4 Km North of the Tiznit city, between Tiznit and the douar Atebbane. Thus the distribution area seems to be very restricted to a small stretch of land, 25 Km long and only 3 Km wide. All other species of *Typhlocirolana* known from Morocco are also more or less widespread and can be considered as endemics of their region but *Typhlocirolana tiznitensis* n.sp., with a so restricted distribution area, appears as a good example of a steno-endemic species and probably this trait is related to the origin and historical biogeography of the species.

ECOLOGICAL DATA

The type station where the species was first discovered and where it is abundant and prolific is a well located within the schoolyard of the public school of Bounaamane village. This well is consequently carefully protected by a brickwork curb half a meter high and closed by a cement slab. The diameter of the well is 2 m but its opening and closing slab are only a 0.6 X 0.6 m square. The well is 16.3 m deep and the height of the water column ranges from 4.1 to 4.5 m throughout the year; thus the water surface is about 12 meters below the soil surface. The type biotope is a dark subterranean habitat, relatively well isolated from external influences.

All the year long the water temperature ranges between 21 and 22 °C and the electric conductivity ranges from 1400 to 1600 mS . In the region the groundwater conductivity varies mainly with the total hardness and the chlorid content of water. In the Bounaamane well the first one ranges from 250 to 450 mg.L^{-1} of $CaCO_3$ and the second one is always between 150 and 300 mg.L^{-1} of Cl^- The sulphate ions concentration stays near 44 mg.L^{-1} in Bounaamane, when the sulphate content in the water of other wells inhabited by the new species may reach 200 mg.L^{-1} and more. However this relatively high mineralisation rate of water is normal in these regions; it is not indicative of an anthropic pollution (Boulal, 1988, 2002) and well tolerated by stygionbiontic crustaceans. Other variables such as the values of nitrogenous ions, related with the organic matter content and the oxygen availability are more important ecological factors. Nitrate concentration in the well water is near 50 mg.L^{-1}, nitrites and ammonia values are respectively near 0.02 mg.L^{-1} and 0.05 mg.L^{-1}. The water content of dissolved oxygen is between 5 and 6 mg.L^{-1} with a saturation index near 80%; the mean value of pH is 7.3 and the oxydo-reduction potential higher than + 220 mV. All these values are reported here as an example of both the new species habitat characteristics and also of the well water characteristics of many wells in a poorly studied region of Morocco where a diversified stygofauna occurs (Boulal, 1988 and 2002). In spite of a relatively important mineralisation of the water from the Bounaamane well, if compared with many European wells, this well, when compared with many other wells of the Tiznit province, belongs to a group of wells containing a relatively pure and slightly mineralized water. Briefly the well provides a relatively good quality water.

Correlatively the Bounaamane well is inhabited by a good number of species including a minimum of the 10 accompanying species. Three of them are stygobionts; two are species of Gastropoda Hydrobiidae, the first one provisionally assigned to the genus *Hauffenia* by Boulal (1988) is in fact a new species of the genus *Giustia* (Ghamizi, 1998 and Boulal, 2000), the second one belongs to the genus *Heideella*; there is also a crustacean amphipod, *Metacrangonyx* sp.; other taxa include an Oligochaeta, two stygophilous Copepoda, *Eucyclops serrulatus* and *Thermocyclops dubowskii*, an Hydracarina and two larvae of Diptera insects. With a total of 11 taxa, the Bounaamane well biocenosis is moderately rich (in other wells near Tiznit the specific richness varies from 8 to 23 with a number of stygobitic species ranging from 1 to 8 according to the water quality and other ecological characteristics).

In the wells where *Typhlocirolana tiznitensis* n.sp. occurs, it is generally the sole species of the genus, except in one well, located North of Tiznit, where sometimes (in 4% of the samplings) the new species may exceptionally cohabit with a second species of *Typhlocirolana*. However near Tiznit the new species can cohabit with other stygobiontic isopods, Stenasellidae or Microcerberidae (*Magniezia* sp. or *Microcerberus* sp.). Consequently the absence of other species of *Typhlocirolana* in the stations of *T. tiznitensis* n.sp. is probably not related to competition or predation by *T. tiznitensis*. As it is no more due to abiotic ecological factors (such as water pollution or water physico-chemical characteristics), it is probably the result of some hydrogeololgical or historical factors.

Typhlocirolana tiznitensis may be considered as a carnivorous species as it feeds frequently on living preys (fragments of copepods or of metacrangonyctid amphipods were often observed in the digestive tract of this species) but it is also able to feed on dead organic matter (it is attracted in traps by a small bait of meat) and even on clay, this clay always including bacteria or organic matter. In fact the species is, like many other stygobiontic crustaceans, an opportunist polyphagous and its presence or absence is rarely due to trophic conditions.

The population dynamics of *Typhlocirolana tiznitensis* has been investigated from the analysis of the structure of monthly samples of *Typhlocirolana tiznitensis* n.sp., collected during a two year period (Boulal, 1988) with baited traps (Boutin and Boulanouar, 1983 and 1984). It is not possible to present here the complete results of this study but we can indicate the main conclusions:

- The youngest stages are present all the year long and the body size frequency histograms are bimodal. This regularity of polymodal histograms observed surely results from a continuous recruitment and a regular mortality all the year long.
- However there is apparently a higher percentage of ovigerous females in each sample during winter and spring. This increase of reproduction is probably related to the higher level of the water table during this period, due to seasonal rainfalls which also cause a "flood factor", i.e. a seasonal relatively important input of soluble or particular organic matter, sweeped down from the soil surface to groundwater, causing an increase of available food for the stygofauna.
- A more important number of individuals in each sampling is also observed in summer, resulting, from the winter and spring higher reproductive activity.
- *Typhlocirolana* species are ovo-viviparous (Boutin et al., 2002), the females laying well formed juveniles just hatched from eggs still enclosed within an internal and ventral

brood pouch of the female. Thus eggs are never exposed and the dissection of the gravid female is necessary for counting eggs or embryos. In *Typhlocirolana tiznitensis* the fecundity is relatively high; the dissection of an ovigerous female of 15 mm long, an exceptional large size, allowed to count 13 incubated eggs. The number of 11 or 12 eggs per female being common and considered as a characteristic of the new species. Therefore, *Typhlocirolana tiznitensis* exhibits some general biological traits of subterranean animals living in a relatively constant environment inducing a continuous reproduction, but in the same time, a preserved and moderate variation in the intensity of the reproduction adapted to the relative variation of the available feeding resources.

- The fecundity still relatively high, when compared with that of many subterranean species which are more typically K-strategists, indicates a plesiomorphic trait possibly related to the avaibility of food resources, not negligible in a relatively shallow groundwaters (9 to 18 m under the ground surface depending on stations and seasons); however this physiological trait, added to a relatively large body size and many other morphological traits allows to consider *Typhlocirolana tiznitensis* n.sp. - as well as a certain number of related species - as a relatively primitive species of *Typhlocirolana*. The primitive traits of these species could be related to the age of the colonization of continental groundwaters by the stygobiontic ancestral populations of the present species (Boutin, 1993b ; Boutin and Coineau, 2000 and 2005).

DISCUSSION: ORIGIN OF THE SPECIES, PHYLOGENETIC RELATIONSHIPS, HISTORICAL BIOGEOGRAPHY AND GEOLOGICAL IMPLICATIONS

The *Typhlocirolana* group of genera (Wägele, 1988; Brusca and Wilson, 1991; Boutin 1993a) is typically a group of thalassoid stygobionts (Coineau and Boutin, 1992; Botosaneanu, 2001; Boutin, 2004; Boutin and Coineau, 2005). The ancestors of each genus were first epibenthic marine crustaceans (Delamare Deboutteville, 1957). Then at certain periods of the past, different populations colonized the coastal sediments of the Tethys ocean and originated thalassostygobiontic crustaceans. Thus the first coastal stygogbiontic cirolanids resulted from the first step of the "Two step model of colonization and evolution" (Boutin and Coineau, 1990; Notenboom, 1991; Coineau and Boutin, 1992). This first step is a change in the ecology of crustaceans, sometimes called in this case the “vertical transition” (Boutin, 1997). Then the resulting coastal interstitial populations, wide spread along the marine shores, may have become adapted to changing salinity of interstitial waters and may have survived and stayed in the same place, in spite of the changing environment, first in brackish and then in fresh continental groundwaters, especially during and after marine regressions. This ecological change or second step of the "Two step model" of evolution, was also called the “horizontal transition” by Boutin and Coineau (1990) and Boutin (1997). Subsequently and progressively these populations, living more or less far from the sea, originated the different limnostygobiontic species which are now represented by the different extant species. This general scenario explains the origin of thalassoid stygobiontic crustaceans such as the cirolanids of the *Typhlocirolana* group, as well as many other limnostygobionts of marine origin, which presently occur in continental regions formerly covered by a sea.

The continental subterranean habitats are known as relatively stable and conservative milieus, especially if compared with the changing and unstable coastal habitats (Delamare Deboutteville and Botosaneanu, 1970; Boutin and Coineau, 1991; Boutin, 1997a). Thus the populations settled first in continental groundwaters may have retained ancestral traits which are not observed in species more recently settled in continental groundwaters. These latter species derived from ancestors which lived in coastal habitats for a longer period and consequently they may have evolved more significantly, showing more morphological adaptations to the life in interstitial milieus. Conversely the limnostygobiontic species resulting from more ancient colonisation generally appear more similar to their ancient marine ancestors (Boutin and Coineau, 2000).

Due to its broad and regularly rounded body shape, its relatively large body size, its long antennae, and the importance and pattern of the chaetotaxy of its appendages, *Typhlocirolana tiznitensis* n.sp. appears as phylogenetically related to species such as *T. haouzensis*, *T. buxtoni*, and *T. fontis*. All these species which have retained many plesiomorphic traits (shared with many outgroups of Cirolanidae, especially in still marine genera such as *Cirolana* or even *Bathynomus*...), may be considered as the representatives of the most "primitive" group of species within the genus *Typhlocirolana*. This group was called the « *fontis Group* » of species (Boutin and Coineau, 1991; Boutin, 1993a, 1993b, 1997). These species occur in wide regions of Morocco and Algeria, which have been covered by the seas during the important Cenomanian-Turonian marine transgression. They are considered as the Present descendants of the interstitial and coastal ancestral populations of marine cirolanids, distributed along the southern shores of the Tethys Ocean, which shifted in continental groundwaters during the Turonian marine regression, some 90 My ago.

Another lineage of *Typhlocirolana* is formed by species such as *Typhlocirolana leptura* which has also preserved a lot of plesiomorphic traits, especially in its chaetotaxy and its important body length, but which exhibits a remarkable elongation of the body and appendages, easily interpretable as an adaptation to life and locomotion in the sediment interstices, in animals having still a global large body size (Botosaneanu *et al.*, 1985). Therefore it is well known (Coineau, 2000) that the selection pressure of the small size of interstices in different interstitial habitats - both in coastal sediments and in continental porous rocks - acts positively (Bovée de *et al.*, 1995) either on the diameter of animals (leading sometimes to elongated vermiform species) or on the global body size (leading to small sized species). The *Typhlocirolana* species of the «*leptura Group*» with a normal body length are an example of the first type of adaptation. They occur in some regions

(High Atlas valleys) which became continental during the Turonian marine regression and which were subsequently uplifted by the important atlasic orogenesis. During the different steps of this orogenesis, the superficial rocks containing a moment the groundwaters were worned away by erosion and the groundwater shifted repeatedly in different deeper layers. This repeated changes in aquifer granulometry and the resulting selection pressure, probably induced a more rapid morphological change in interstitial organisms and finally the "morphological evolution" being considered as more rapid when the environment of populations is repeatidly changing or unstable (Boutin and Coineau, 1991 and 2000). Finally the origin of the «*leptura Group*» and that of the «*fontis* Group» of species in Moroccan groundwaters were in fact the same since they result from the second step of a "Two step process of colonization" of continental groundwaters by the coastal ancestors which occurred some 90 My ago, but the history of the 2 lineages in continental groundwaters of two different regions is different.

A third species group of *Typhlocirolana*, forming the third lineage, includes several known species, often still under study, such as a *Typhlocirolana sp.* from the Oued Zat valley in Morocco, and other similar species living in the alluvium of different rivers flowing down from the High Atlas range; they are characterized by a smaller body size (8 to 10 mm in length), a body shape less elongated than in the *leptura Group* but thiner, and more elongated than in the *fontis Group*, with shorter antennae and above all an appendage chaetotaxy significantly more reduced than in the two first groups of species (Yacoubi-Khebiza, 1990 and 1996). The body, smaller and smoother, exhibits a higher degree of adaptation to the life in interstitial habitats (Coineau, 2000 ; Botosaneanu, 2001). This group of species was provisionally called the «*zatensis Group»* (Boutin, 1993a, 1993b, 1997b), from the name possibly envisaged for of a Moroccan species occurring in the Zat River valley. The species of this third lineage occur in Morocco in different regions which were covered by the sea, not only during the Cenomanian-Turonian transgression, but once more (and generally for the last time) during the Senonian transgression, some 70 My ago. Compared with the species of the two first groups - which became continental during the Turonian - this third lineage have evolved significantly (Boutin and Coineau, 1991) in the changing and unstable environment of coastal habitat, favourable to a rapid adaptive morphological evolution, during some 20 My years, and gained, in this coastal interstitial environment, the morphological characteristics – including a series of apomorphies - of the future *zatensis Group* of species.

After the Senonian (as after the first wave of continental groundwater colonization of the Turonian) some marine coastal cirolanid populations survived along the Tethys shores. These thalassostygobiontic crustaceans, still coastal and interstitial, continued their morphological evolution during the late Cretaceous and the Paleocene and have originated, in western Mediterranean countries, a last wave of colonization during the marine regressions of the gulfs formed in the Eocene period. This Lutetian marine regression resulted in continental stygobiontic cirolanids smaller and so different from the different species of the genus *Typhlocirolana* that Boutin (1993a) proposed to include them in the genus *Marocolana*. Then, after the Eocene, the thalassoid coastal cirolanids seem to have disappeared of the southwestern shores of the Tethys; thus the species of *Marocolana* forms, in Morocco, the last and the most recent lineage of stygobiontic cirolanids, and in the same time, the most apomorphic forms of cirolanids (Boulanouar *et al.* 1993). However in the eastern Mediterranean countries, especially in Turkey and Palestine which emerged during later marine regressions along the Northern shores of the Tethys, different species of cirolanids form the genus *Turcolana* (Argano and Pesce, 1980 ; Botosaneanu and Notenboom, 1989) which exhibits a higher degree of apomorphic traits (Boutin, 1997a).

Finally the phylogenetic relationships among the stygobiontic cirolanids from the countries of the Mediterranean Basin can be represented on a reduced cladogramme like that schematized in Fig. 28A, where each Taxonomic Unit is not a species but a group of species sharing a series of apomorphic traits (= a lineage), which occurs in the regions mentioned on the area cladogramme Fig. 28B. This simplified and schematic phylogenetic tree of Fig. 28A, results from the analysis of morphological characters, however it shows relationships between the different lineages of *Typhlocirolana* which are perfectly supported by the first molecular classifications like that of Baratti *et al.* (1999). A more recent molecular phylogeny, from sequence alignments of two different genes (Barati *et al.*, 2003) leads either to trees in accordance with the morphological groups of species, when using the 16S rRNA sequences (but unfortunately the *tiznitensis* species is not considered) or to a tree including *T. tiznitensis* where this species seems to be more closely related to zantensis group of species than to *T. haousensis*, when the 16SrRNA sequences are compared. However, in spite of these variable results obtained from fundamentally phenetical methods applied to one gene sequences, the number of different morphological traits possible to interpret in terms of apomorphies or plesiomorphies allow to consider that *Typhlocirolana tiznitensis* n.sp. clearly belongs to the first lineage of species, also including *T. haouzensis* (the "*fontis* Group"), the origin of which being related to the Turonian marine regression.

Due to the lack of Cretaceous sediments in the Province of Tiznit, between the Atlasic Basin to the North and the Tarfaya Basin to the South, most geologists consider the two basins as evidences of two separated Cenomanian-Turonian gulfs with a question mark between them, meaning a probable emerged land during Mesozoic times (Andreu-Boussut, 1991). As a matter of fact, in this Moroccan region, between the High Atlas and the Anti Atlas, the exposed Quaternary lake limestones are directly in contact with Paleozoic metamorphic rocks (Boulal, 1988 and 2002) and not any Mesozoic rocks occur. However when considering the evolutionary history of the Cirolanids, the occurrence of species such as *Typhlocirolana tiznitensis* n.sp. - clearly belonging to the *fontis* group of species – near Tiznit, and also near Guelmim (Boutin and Idbennacer, 1989 ; Idbennacer, 1990) in the

South of the Anti Atlas, is to be considered as a strong evidence of the presence of the sea during the Cenomanian and Turonian periods, before the marine regression which allowed the settlement of some thalassoid stygobionts in continental groundwaters. In this hypothesis the absence of Mesozoic rocks in the region can be explained simply as the result of their further erosion : during the Cretaceous times the sea probably covered all the western Morocco, and receded during the late Turonian. After the emersion of the area, probably during some steps of the High Atlasic orogenesis, which occurred from the Eocene and during the Caenozoic, the raising of the southern Anti Atlasic domain very likely involved a continental erosion which removed all mesozoic rocks. Thus the Paleozoic rocks were exposed during the Caenozoic and progressively subsided, allowing the further settlement of freshwater lakes during the pluvial periods of the Quaternary.

This scenario, already presented to geologists and well accepted, during a special meeting devoted to the History of the Tethyan margins of North Africa, was briefly published (Boutin, 1997b). The use of thalassoid stygobiontic cirolanids such as *Typhlocirolana tiznitensis* n.sp. in the studied region, for infering the presence of the sea during the mid-Cretaceous, is corroborrated by the occurrence in the same region of another group of stygobiontic crustaceans, living in the same groundwaters, a metacrangonyctid amphipod of the most primitive group of genus *Metacrangonyx*. (This primitive lineage of *Metacrangonyx,* also known from different countries, settled in continental subterranean waters, after an «horizontal transition» which occurred during the Turonian regression (Boutin, 1994a and 1997a).

More generally the knowledge of the diversity of stygobiontic thalassoid species can provide useful informations on the geological history of the region where each species occurs (Boutin, 1993b). A number of other examples were studied (Por, 1975; Boutin 1993a, 1994a, 1994b, 1997a; Holsinger *et al.* 1994; Alouf 1998; Fakher El Abiari *et al.* 1999; Boutin and Coineau 2000, 2005) and this approach is efficient when a new species is discovered, as soon as it is possible to include the new species within a monophyletic lineage already known in regions geologically better known. Thus if the ecological applications of the biodiversity of stygobiont is well known, the possible use of some stygobiontic species as "living fossils", allowing to establish some geological data, is not very known and constitute another justification of the researches in favour the species diversity of the stygofauna in many parts of the world.

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FIGURE LEGENDS

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Fig. 1 to 6: *Typhlocirolana tiznitensis* n.sp.: 1 Habitus of a male 11 mm long (bar = 4 mm); 2 Antennula 1 (bar =0,5 mm); 3 Antenna (bar = 1 mm); 4 and 5 Right and left mandibles (bar = 0,5 mm); 6 *Pars molaris* and *lacinia mobilis* of left mandible (bar = 0,1 mm).

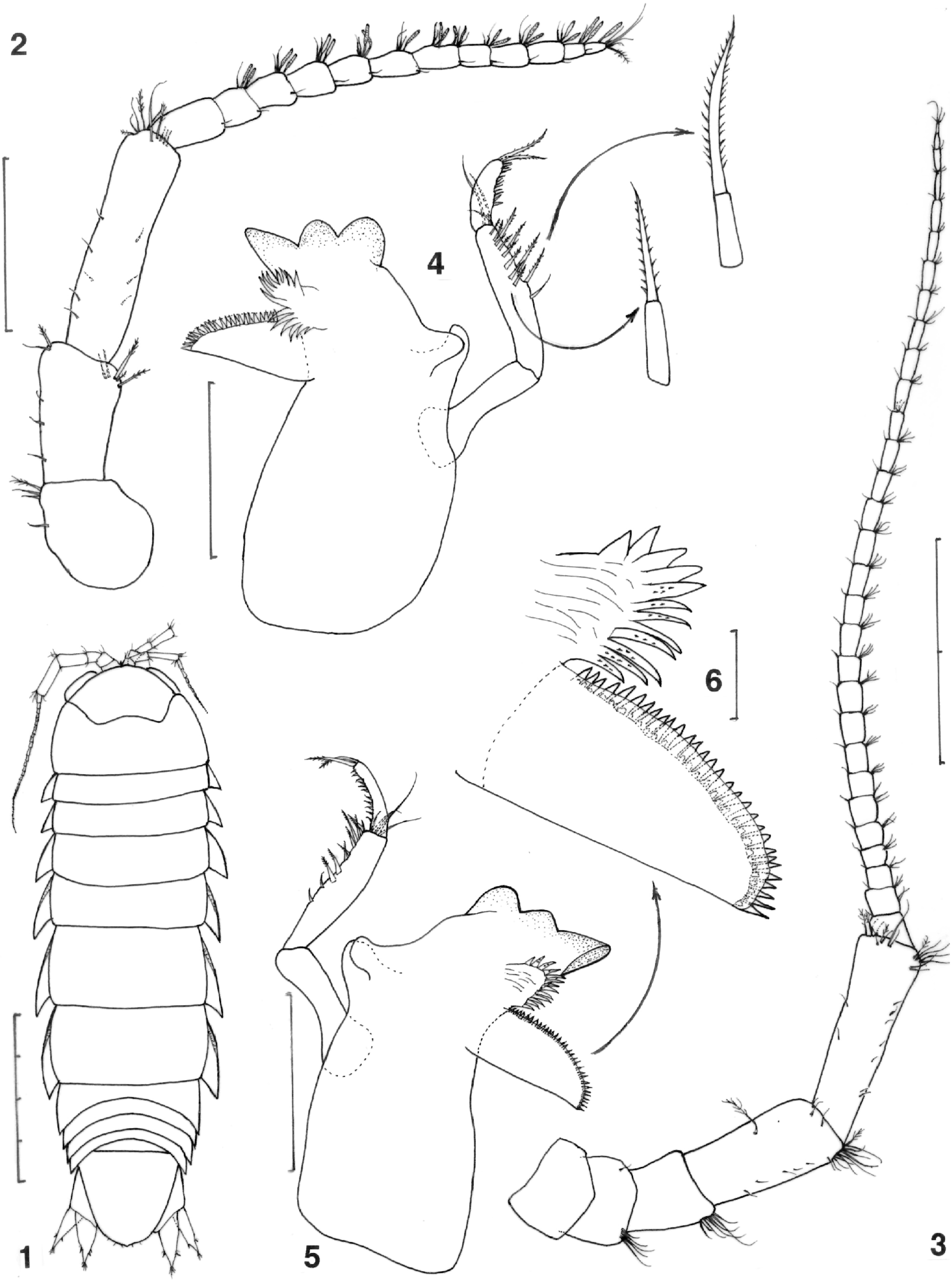
Fig. 7 to 12: *Typhlocirolana tiznitensis* n.sp.: 7 Maxillule (bar = 0,2 mm); 8 Maxilla (bar = 0,2 mm); 9 Maxilliped (bar = 0,5 mm); 10 Endite of maxilliped (bar = 0,1 mm); 11 First pereiopod (bar = 0,5 mm); 12 Inner edge of the merus of first pereiopod.

Fig. 13 to20: *Typhlocirolana tiznitensis* n.sp.: 13 to 18 Pereiopods 2 to 7 (bar = 1 mm); 19 and 20 right male pleopods 1 and 4 (bar = 0,5 mm).

Fig. 21 to 25: *Typhlocirolana tiznitensis* n.sp.: 21 left male pleopod 2 (bar = 0,5 mm); 22 detail of the coupling series of setae of male pleopod 2; 23 Pleopod 2 of a female14,5 mm long; 24 and 25 Pleopods 3 and 5 (bar =0,5 mm); 26 Ventral vew of right uropod (bar = 0,5).

Fig. 27: *Typhlocirolana tiznitensis* n.sp.: Dorsal view of pleotelson (bar = 1 mm)

Fig. 28: Origin of the *Typhlocirolana* lineages. A: Schematic cladogramme showing the phylogenetic relationships within the 3 groups of species forming the genus *Typhlocirolana* and the 2 related genera, *Marocolana* and *Turcolana*. B: Area cladogramme corresponding to the 5 taxa of cladogramme A, with indication of main events which occurred in each area and originated the 5 lineages of stygobiontic cirolanids.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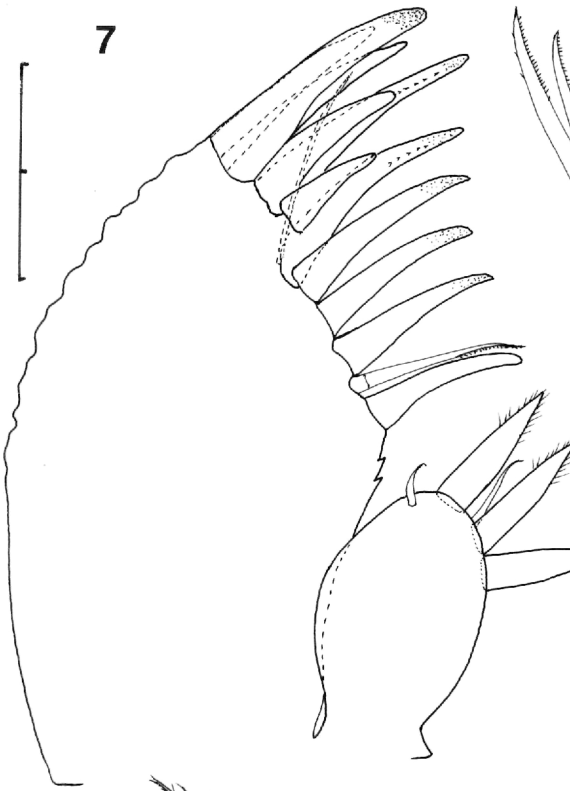

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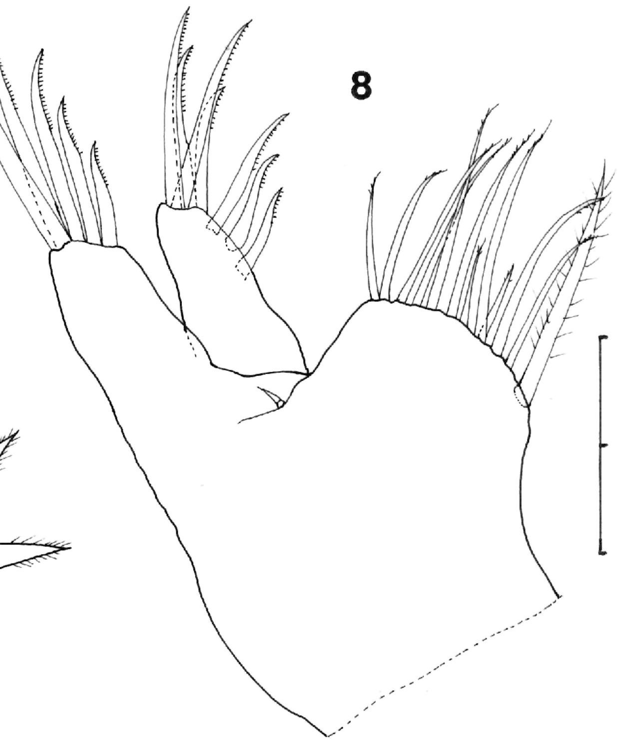

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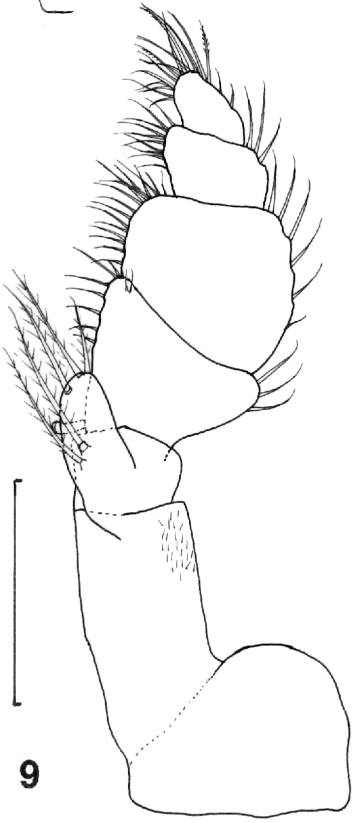

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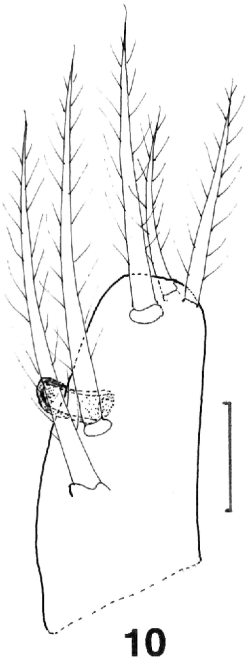

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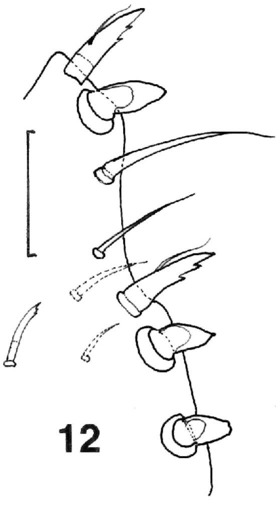

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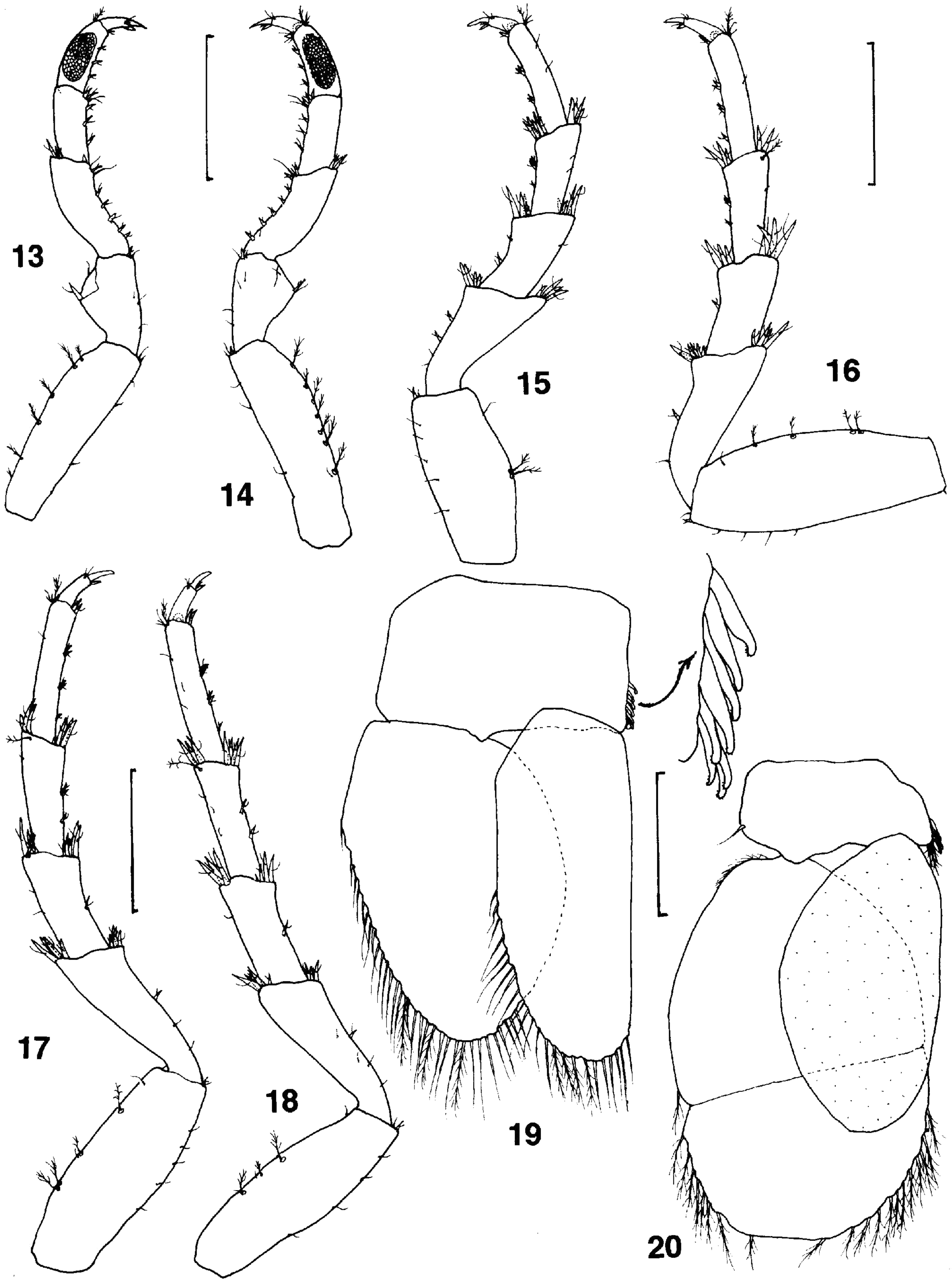
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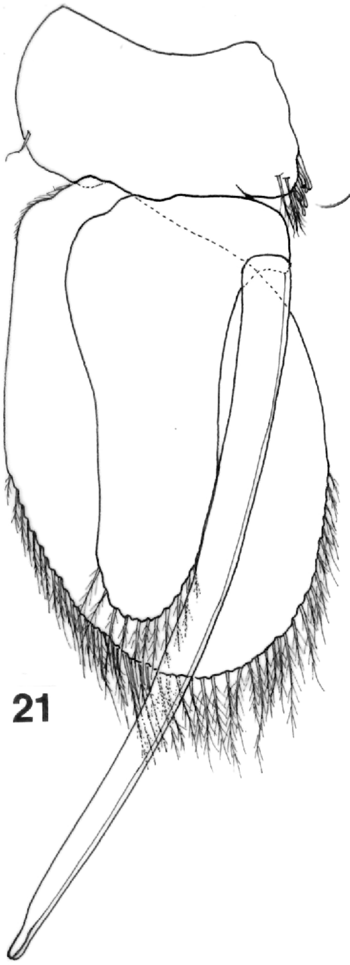

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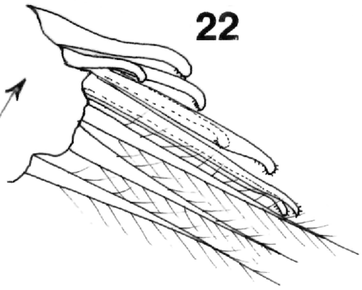

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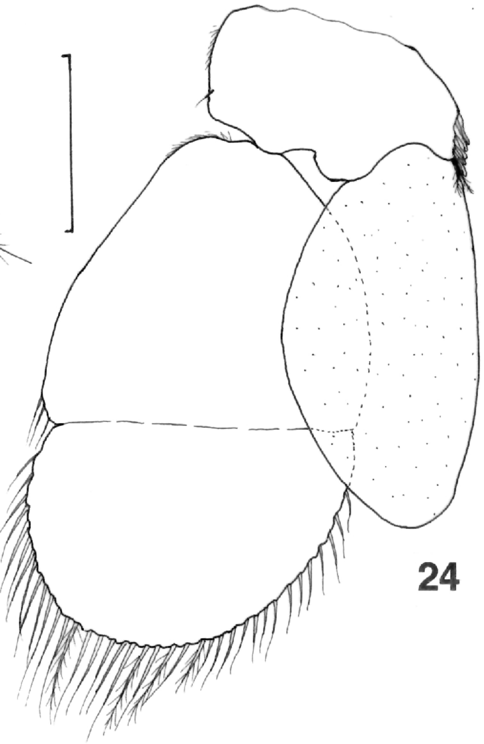

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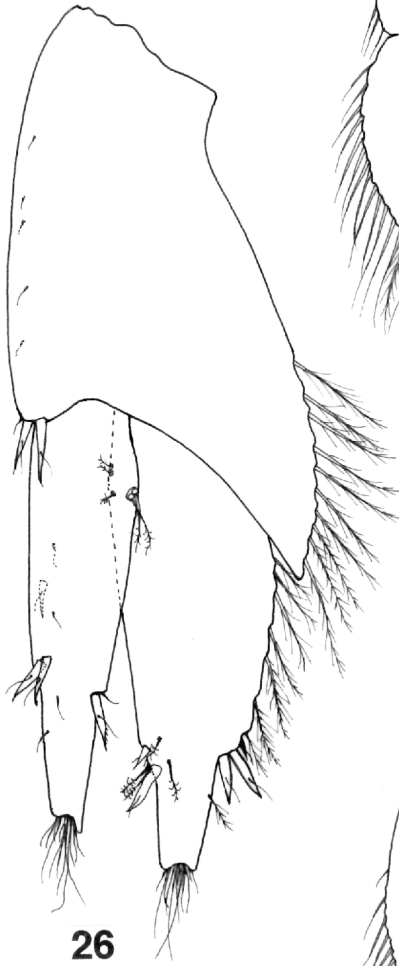

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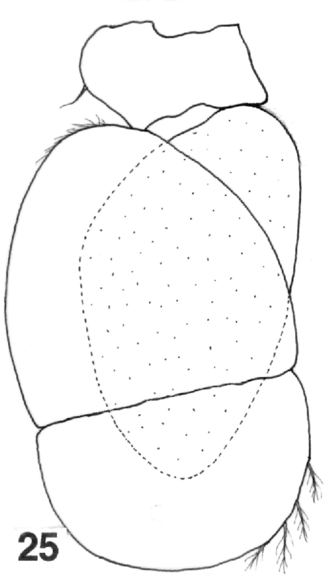

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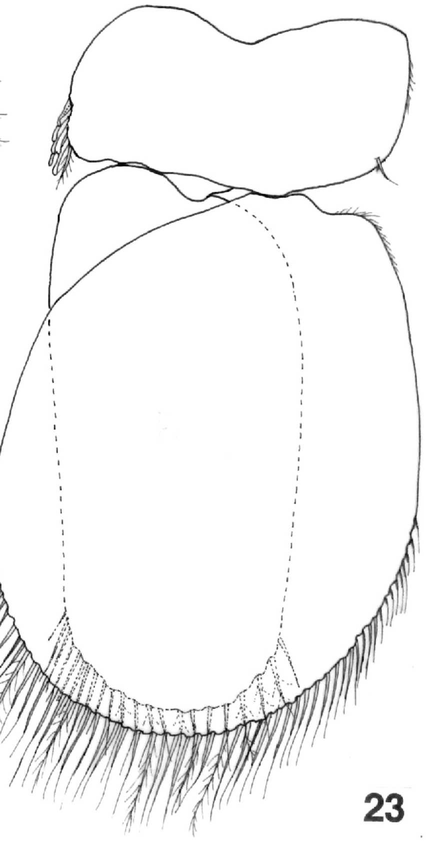

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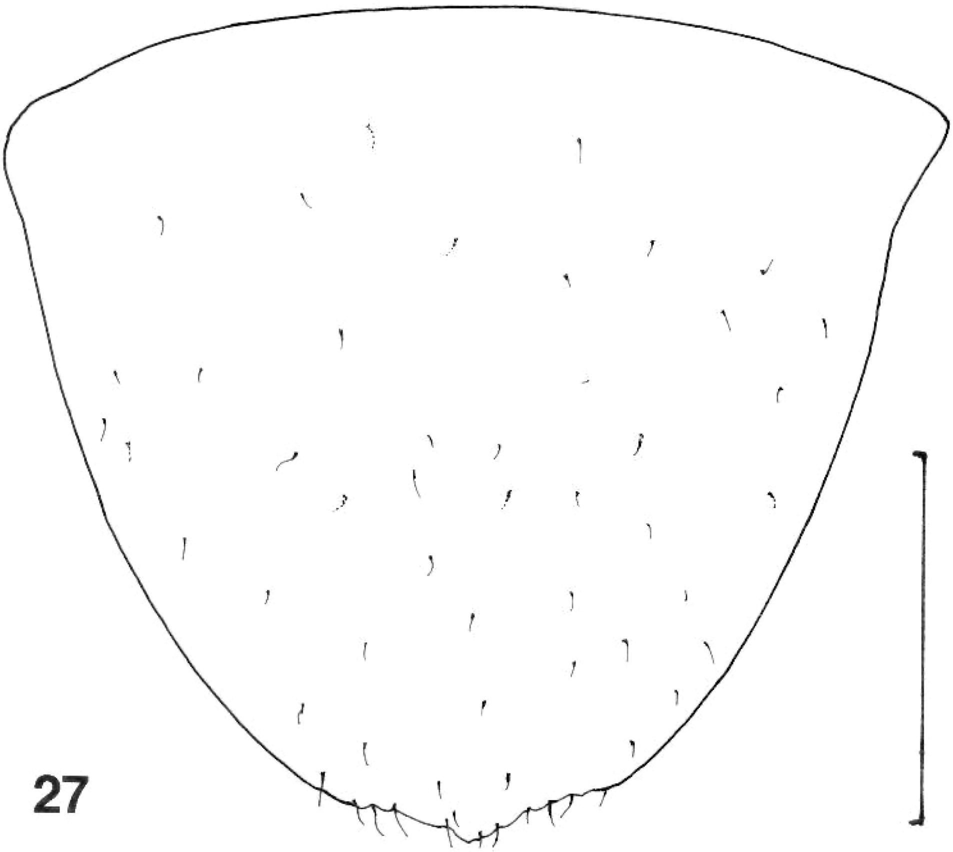

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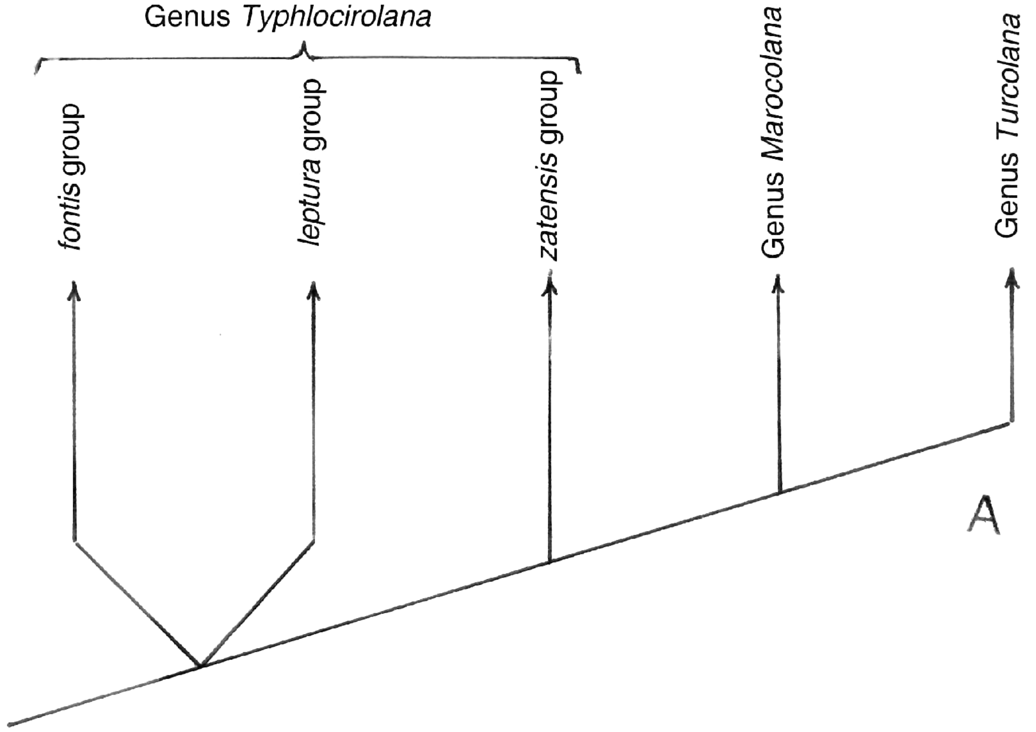
Genus *Typhlocirolana*
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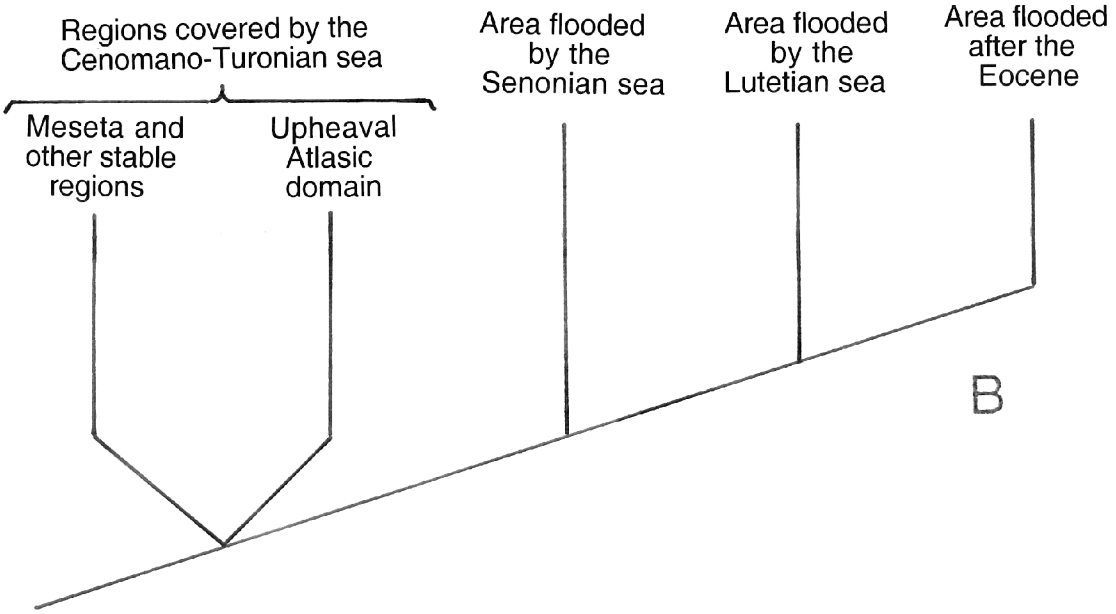
Regions covered by the Cenomano-Turonian sea
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Upheaval Atlasic domain
Area flooded by the Senonian sea
Area flooded by the Lutetian sea
Area flooded after the Eocene
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