



# The hidden agglutinated foraminifera of the mid-Cretaceous hemipelagic carbonate deposits: A method-derived bias?

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## ABSTRACT

Five different micropaleontological methods (H<sub>2</sub>O<sub>2</sub>, Glauber's salt, liquid nitrogen, acetic acid + Copper(II) sulfate, and formic acid) were applied to study the differences of obtained agglutinated foraminiferal faunas of typical hemipelagic carbonate deposits of the mid-Cretaceous of Europe, and to prove whether there is a method-derived bias of knowledge about agglutinated foraminiferal faunas in these sedimentological settings. Split samples of the same weight were treated with each method to compare overall (calcareous + agglutinated) numbers of foraminifers per gram, numbers of agglutinated foraminifers per gram, and numbers of agglutinated foraminiferal genera per sample.

The results show that the number of agglutinated foraminifers per gram strongly vary between 0.1 and 7.8 with use of standard micropaleontological methods. With application of formic acid, more agglutinated foraminifers per gram are obtained than with any other tested method. The number of agglutinated foraminifers per gram is 1.5 to 211.0 times higher in formic acid treated residues. Furthermore, with use of standard micropaleontological methods at least 2/3 of agglutinated foraminiferal genera and species are completely missing in these sedimentological settings. Consequently, standard micropaleontological methods are not applicable to study the whole agglutinated foraminiferal fauna, and a bias of knowledge and utility of agglutinated foraminifers in these sedimentological settings is obvious. A separate application of both acetic acid + Copper(II) sulfate and formic acid on samples is suggested for studies on the whole foraminiferal fauna, and a precise description of the applied method in studies is suggested.

## 1. Introduction

In shallow marine hemipelagic marl–limestone alternations, limestones, and chalks of the mid-Cretaceous, benthic and planktic foraminifera are an important tool to solve stratigraphical and paleoenvironmental questions. While agglutinated foraminifera are dominant in benthic communities in bathyal and deeper settings as well as in more clastic dominated shelf deposits such as greensands in the mid-Cretaceous of Europe, they are mostly only an accessory in shelf-related carbonates. Their relative abundance does usually not exceed 20% in these settings (e.g., Leary and Hart, 1992; Wejda, 1993; Gräfe, 1999).

Commonly used methods to receive foraminiferal faunas in calcareous rocks are the H<sub>2</sub>O<sub>2</sub>, the Glauber's salt, the liquid Nitrogen, and acetic acid with Copper(II) sulfate methods. These methods preserve specimens with both calcareous and agglutinated test, but agglutinated

ones are mostly not clean with attached sediment remaining.

Several studies showed that using dissolution methods with hydrochloric acid (e.g., Kuhnt, 1990; Coccioni et al., 1995; Kaminski et al., 2011) and formic acid (Besen et al., 2021, 2022) diverse agglutinated foraminiferal faunas can be obtained even from Cretaceous carbonates. This study focusses on the formic acid method which is suggested to have a more gentle reaction while dissolving the carbonate content and providing cleaner foraminiferal specimens (Toomey, 1974).

Through the application of different standard micropaleontological methods and the formic acid method on typical mid-Cretaceous shelf related hemipelagic carbonates, differences of the agglutinated foraminiferal faunas in all residues should be identified and quantified. The question should be addressed, if a method-derived bias of knowledge about agglutinated foraminiferal faunas in mid-Cretaceous hemipelagic carbonates exists.

In this study, we demonstrate that only a separate application of the

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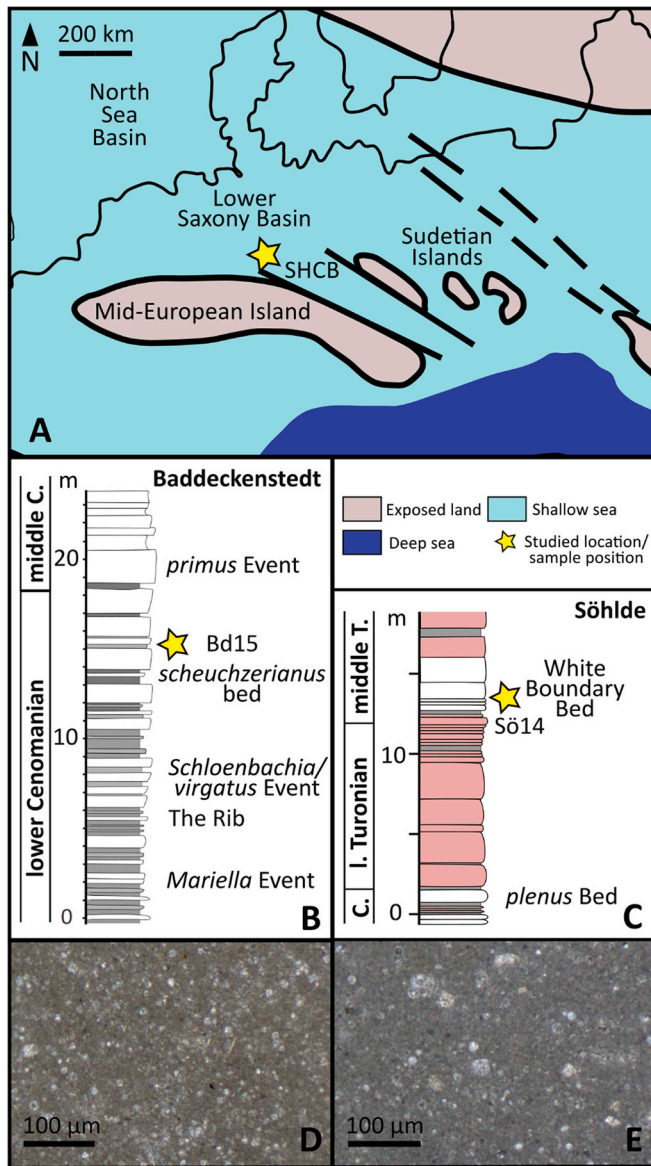
E-mail address: [rbesen@zedat.fu-berlin.de](mailto:rbesen@zedat.fu-berlin.de) (R.M. Besen).

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**Fig. 1.** A. Palaeogeographical map of middle Europe during the Cenomanian, modified after Philip and Floquet (2000), abbreviations: SHCB – Subhercynian Cretaceous Basin; asterisk – position of both locations (distance of around 10 km); B. Columnar section of the lower Cenomanian part of the Baddeckenstedt quarry section, redrawn after Wilmsen (2003); asterisk – position of sample Bd15; *scheuchzerianus* bed, *primus* Event, *Schloenbachia/virgatus* Event, and *Mariella* Event are interregional correlatable accumulations of different macro fossils, named bioevents, The Rib is an interregional marker limestone (for further explanations see Wilmsen, 2003); C. Columnar section of the Lower to Middle Turonian part of the Söhlde quarry section, abbreviations: C. – Cenomanian, redrawn after Wiese (2009); *plenus* Bed is an interregional bioevent, White Boundary Bed is an interregional marker limestone (for further explanations see Wiese, 2009); D. Microfacies: calcisphere mudstone, lower Cenomanian, Baddeckenstedt; asterisk – position of sample Sö14; E. Microfacies: coccolith mudstone, middle Turonian, Söhlde.

formic acid method is effective to obtain a reliable and well preserved agglutinated benthic foraminiferal fauna.

## 2. Material

As representative samples for the mid-Cretaceous calcareous shelf deposits, one sample from the Cenomanian of Baddeckenstedt, and one from the Turonian of Söhlde were chosen (Fig. 1A; both Subhercynian

**Table 1**

Overview and details of applied methods, and basic data and results of used samples; Ø – average of four sample batches.

Method	Hydrogen peroxide	Glauber's salt	Liquid Nitrogen	Nötzold's method	Formic acid
Chemical compounds	10% H <sub>2</sub> O <sub>2</sub>	Sodium sulfate/ Na <sub>2</sub> SO <sub>4</sub>	Liquid N <sub>2</sub>	96% Acetic acid + Cu (II) sulfate	10% CH <sub>2</sub> O <sub>2</sub>
Pre-treatment	Dried, max. 3 cm edge length (walnut size)	Dried, max. 1 cm edge length	Dried, max. 3 cm edge length (walnut size)	Dried, max. 3 cm edge length (walnut size)	Dried, max. 3 cm edge length (walnut size)
Cycles	1	5	20	1	1
Duration(per cycle)	24 h	12 h	14 h	20 h	24 h
Sample	Bd15 Sö14	Bd15 Sö14	Bd15 Sö14	Bd15 Sö14	Bd15 Sö14
CaCO <sub>3</sub> %	93.9 82.2	93.9 82.2	93.9 82.2	93.9 82.2	93.9 82.2
Calcareous/g	2.8 0.8	12.4 4.3	7.2 13.6	135.7 252.1	- -
Agglutinated/g	0.1 0.2	2 0.8	0.4 0.1	7.8 6.3	Ø21.1 09.9

Cretaceous Basin, Germany).

The sample from the abandoned quarry Baddeckenstedt was taken from about one metre above the *scheuchzerianus* bed in the lower Cenomanian (Fig. 1B; compare Wilmsen and Niebuhr, 2002, Wilmsen, 2003, Besen et al., 2021), a bioevent expressed by the mass occurrence of heterotroph ammonites, especially *Turrilites scheuchzerianus* Bosc. The lithology is classified as a compact marly limestone with a carbonate content of 93.9%, and the lithofacies as a typical calcisphere mudstone (Fig. 1D) based on Dunham classification (Dunham, 1962). For the lower Cenomanian strata of Baddeckenstedt, water depths of around 30 m are estimated by Wilmsen (2003).

In the Söhlde-Loges quarry one sample was taken from the lower part of the White Boundary Bed ('Weiße Grenzbank'), an important interregional marker limestone in Westphalia, Lower Saxony, and Sachsen-Anhalt in middle Turonian strata (Fig. 1C; compare Wiese, 2009, Besen et al., 2021). The material is classified as porous and pure limestone/chalk with a carbonate content of 82.2%, a typical Turonian mudstone (Fig. 1E) based on Dunham classification (Dunham, 1962). For this middle Turonian interval, Wiese (2009) suggests a water depth slightly below storm water base. Coccolithophores, which are the main component of the sediments deposited at Söhlde, are known in large abundances and diversity from shallow marine settings in Cretaceous times (Püttmann and Mutterlose, 2021).

## 3. Methods

### 3.1. Pre-treatment

Each of the two samples were subdivided in eight parts of around 40 g (one for each standard method, four for the formic acid procedure). All samples were gently crushed into pieces of maximal 3 cm edge length (walnut size; smaller pieces of maximal 1 cm edge length for the Glauber's salt method) and dried overnight at 50 °C.

### 3.2. H<sub>2</sub>O<sub>2</sub> (hydrogen peroxide)

This method from Wick (1947) is based on a combination of chemical dissolution of organic compounds and mechanical dissection through CO<sub>2</sub> pressure in pores of the rock material by reaction of 10% H<sub>2</sub>O<sub>2</sub> (Table 1).

Samples were covered with a 10% H<sub>2</sub>O<sub>2</sub> solution for 24 h following

Wick (1947). This method was frequently modified from many authors regarding H<sub>2</sub>O<sub>2</sub> concentration and/or temperature conditions (see Boltovskoy and Wright, 1976; Green, 2001; Jones, 2014).

### 3.3. Glauber's salt (sodium sulfate/Na<sub>2</sub>SO<sub>4</sub>)

Rock samples are covered with an oversaturated sodium sulfate solution. The water portion evaporates over time while sodium sulfate crystals form. This crystal growth within the rock pores breaks the sample along weak zones. Afterwards, the solution is heated while maintaining temperatures below boiling point. The procedure of alternating freezing and heating is repeated 5 times for each sample (Table 1; Franke, 1922, Wicher, 1942, Green, 2001) This method falls within the freeze-thaw techniques.

### 3.4. Liquid Nitrogen (LN2)

This freeze-thaw method is applied by using liquid Nitrogen and boiling water by Remin et al. (2012). The force of ice crystals growing in pores of the rock sample and thermal expansion of different components and rock matrix effectuated by a thermal amplitude of almost 300 °C cause the rock to break. The procedure of freezing and subsequently heating is repeated up to 20 times (Table 1).

### 3.5. Nötzold's method

The method using 96% acetic acid and Copper(II) sulfate after Nötzold (1965) works with a low degree of dissociation of the highly concentrated acetic acid, dehydration through Copper(II) sulfate and formed calcium acetate which loses the mineral compound (see also Nielsen and Jakobsen, 2004). For this method, 2/3 of the sample material is covered by highly concentrated 96% acetic acid and anhydrous Copper(II) sulfate for up to 20 h (Table 1). Afterwards, the solution is removed and replaced by several liters of cold water.

### 3.6. Formic acid (CH<sub>2</sub>O<sub>2</sub>)

This method initially invented to receive conodonts in Paleozoic rocks (Rixon, 1949) was first applied to receive agglutinated foraminifera by Toomey (1974). In this procedure, the rock samples are treated with 10% formic acid. With strong reaction in application on carbonates, all calcareous content is dissolved usually within <24 h depending on the porosity and proportion of clay fraction of the rock sample (Table 1). This procedure was applied in four separate batches for each sample to get precise data and avoid straying results.

This acid extraction technique is limited to agglutinated foraminifera with an acid resistant test cementing media, non-calcareous detrital grains, or diagenetically replaced, acid resistant test material (Green, 2001). No calcareous foraminifera are preserved applying this method.

### 3.7. Post-treatment

All samples were washed, sieved using a 63 µm mesh, and dried. A micropaleontological splitter was used for some samples to obtain representative sample splits (see also table supplements). From selected residues or splits, all agglutinated foraminiferal specimens were counted and determined on species level, while calcareous foraminifera were only counted and classified as either benthic or planktic forms. The taxonomy of agglutinated foraminifera is based on Frieg (1980), Loebllich and Tappan (1987), Frieg and Kemper (1989), Kaminski and Gradstein (2005), Kaminski (2014), Kaminski et al. (2011), Setoyama et al. (2017), and Besen et al. (2021). Fragmented specimens (< half of the test) were not further considered. From absolute foraminiferal counts, specimens per gram of dissolved or disaggregated rock material were calculated. Photographs were taken with a Keyence Digital Microscope and Zeiss Stereo Microscope.

**Table 2**

Overall (calcareous + agglutinated) foraminiferal specimens/g, relative abundances of planktic foraminifera, agglutinated foraminiferal specimens/g, and relative abundances of agglutinated foraminifera from the samples at Baddeckenstedt and Söhlde-Loges by applied methods, all values from residues treated with formic acid are averages from four separate analyses.

Method	Hydrogen peroxide	Glauber's salt	Liquid Nitrogen	Nötzold's method	Formic acid
marly limestone (Baddeckenstedt, lower Cenomanian)					
specimens/g	2.8	14.4	7.2	143.3	21.1
plankt. %	69.9	27.8	60.2	35.7	0
aggl./g	0.1	2	0.4	7.8	21.1
aggl. %	2.7	13.9	5.5	5.4	100
genera (aggl.)	1	9	7	7	36
species (aggl.)	1	9	7	8	53.5
limestone (Söhlde-Loges, middle Turonian)					
specimens/g	1	5.1	13.7	258.4	9.9
plankt. %	70.7	49	62.4	64.4	0
aggl./g	0.2	0.8	0.1	6.3	9.9
aggl. %	20	15.7	0.7	2.4	100
genera (aggl.)	4	7	1	5	32.3
species (aggl.)	4	7	1	6	43.3

### 3.8. Carbonate content

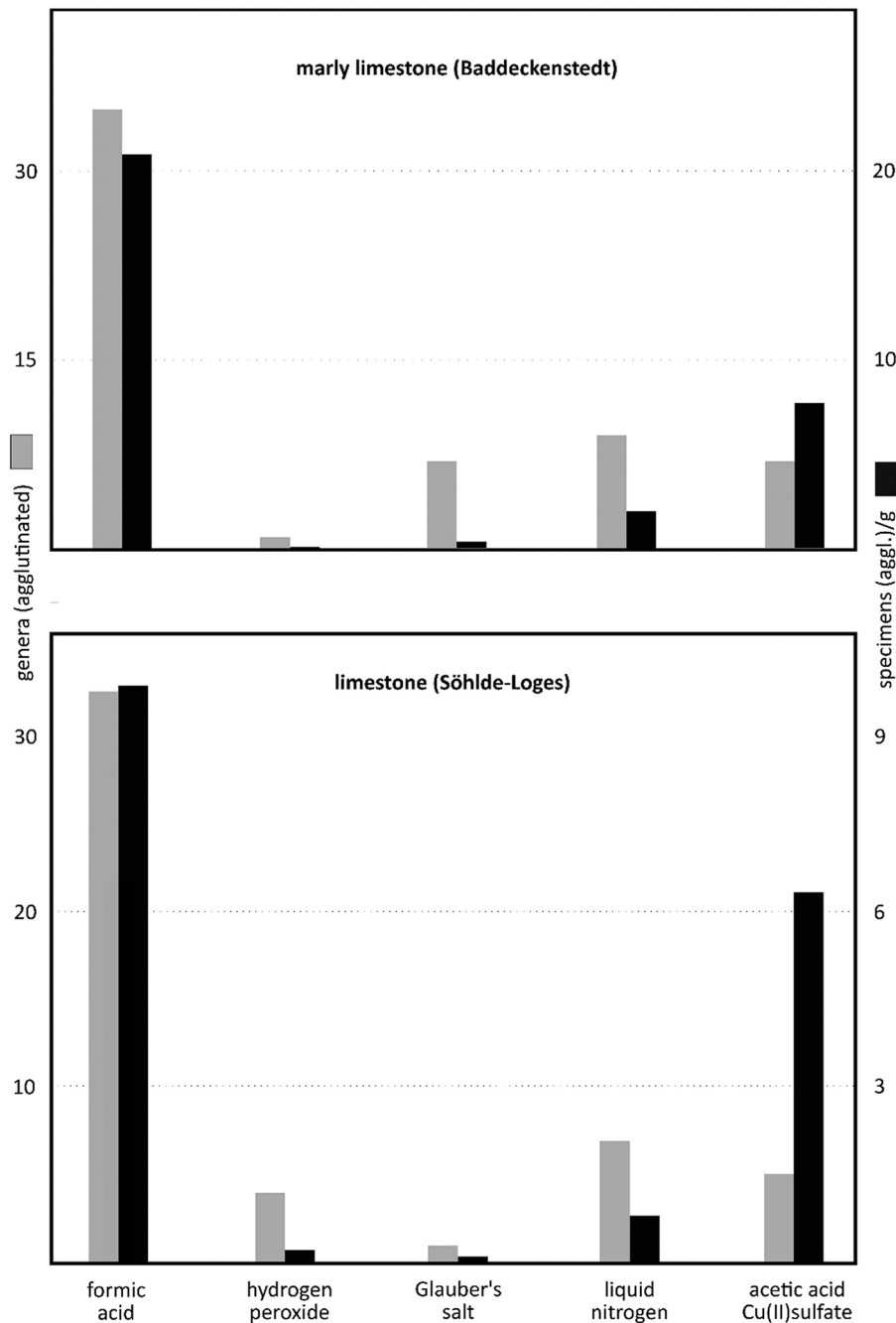
For the analysis of carbonate contents, approximately 2 g of sample powder was weighted into pre-weighted centrifuge vials and treated with 2 M hydrochloric acid until no further reaction was observed. Afterward we rinsed the remaining sample repeatedly with distilled water and dried the samples overnight at 50 °C in a drying oven. The loss of weight was then calculated as the carbonate content in weight % (Table 1). 3.9. Corrected relative abundances.

To get reliable relative abundances of agglutinated foraminifera for each sample, calcareous/g obtained from residues from the acetic acid + Copper(II) sulfate method and agglutinated/g from formic acid treated residues were summed up, and thereof percentages were calculated. These percentages should reflect the true relative abundance of agglutinated foraminifera.

## 4. Results: comparison of methods

In general, the washed residues that contain the greatest number of foraminifera (calcareous and agglutinated) per gram (143.3 specimens/g for the marly limestone, 93.9% carbonate content; 258.4 specimens/g for the pure limestone, 82.2% carbonate content) were obtained from the acetic acid and Copper(II) sulfate method. Other adopted methods such as H<sub>2</sub>O<sub>2</sub>, Glauber's salt, and liquid Nitrogen provided less foraminiferal specimens (up to 14.5 specimens/g for both samples; Table 1 and 2; see also supplementary table). Among all methods, the overall less effective method is the application of H<sub>2</sub>O<sub>2</sub> which provides very few specimens/g and agglutinated foraminiferal specimens/g from these lithologies. For limestone samples from Söhlde, the lowest amount of agglutinated foraminiferal specimens/g were recovered in application of the liquid Nitrogen method. Among the standard procedures, the Glauber's salt method provides the highest relative abundances of agglutinated foraminifera (aggl. %: Table 1 and 2; see also supplementary table).

Relative abundances of planktic foraminifera vary within the different applied methods. They lie between 35.7 and 69.9% for the lower Cenomanian limestone from Baddeckenstedt and between 49 and 70.7% for the middle Turonian limestone from Söhlde (Table 2).



**Fig. 2.** Number of genera and number of agglutinated foraminifera per gram in both samples in application of different processing methods; values from formic acid treated residues are averages derived from four separate analyses.

Relative abundances of planktic foraminifera obtained from the acetic acid procedure (Baddeckenstedt: 35.7%; Söhlde: 64.4%) are most reliable basing on the largest number of foraminiferal counts and detected specimens per gram.

Agglutinated foraminifera appear in different relative abundances within the different standard micropaleontological methods. Their relative abundances lie between 2.7% and 13.9% for the lower Cenomanian marly limestone and between 0.7% and 22.0% for the middle Turonian pure limestone (Table 1 and 2). These strong differences of relative abundances of agglutinated foraminifera from a single sample are interpreted to be related to a different degree of mechanical destruction during the laboratory procedures, especially affecting coarsely agglutinated foraminifera, and/or leading to ineffective cleaning of often rough test surfaces of agglutinated foraminifera.

Considering the standard procedures, the acetic acid + Copper(II) sulfate method provided the highest number of agglutinated foraminifera (up to 7.8 specimens/g) whereas the other methods proved to be less applicable (< 2.0 specimens/g; Fig. 2). In residues of samples treated with formic acid, the numbers of agglutinated foraminifera are highest with up to 26.22 specimens/g for the marly limestone at Baddeckenstedt, on average 21.12 specimens/g (Fig. 2). From the Turonian limestone at Söhlde, on average 9.93 agglutinated specimens/g were obtained by use of formic acid (Fig. 2; see also supplementary table).

The differences of the obtained agglutinated foraminiferal fauna from different methods are even more severe regarding the number of genera. While for the marly limestone, on average 36 different genera are recorded from formic acid residue only between 1 and 9 genera are recorded by application of standard micropaleontological methods

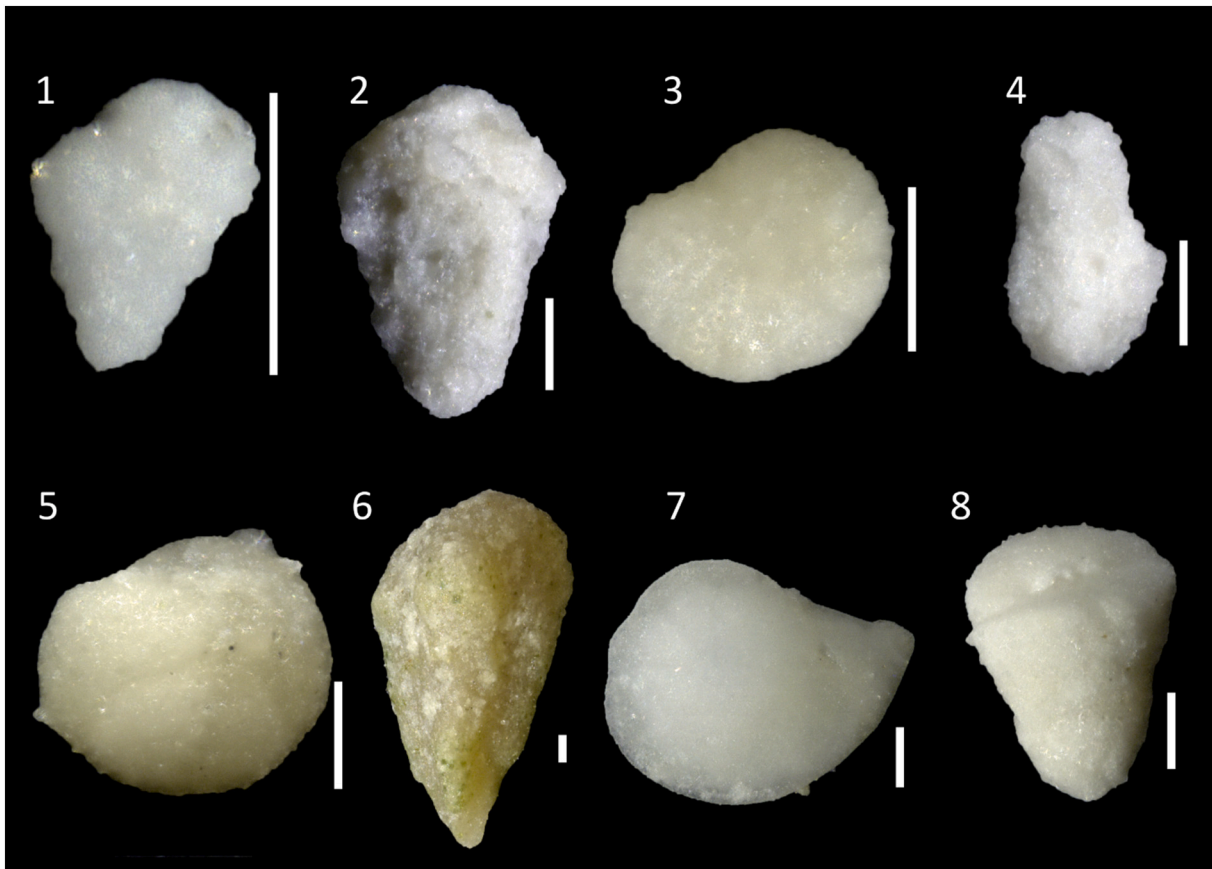


Fig. 3. Calcareous and agglutinated foraminifera obtained by the application of standard micropaleontological procedures; 1. *Heterohelix* sp. (Söhlde, H<sub>2</sub>O<sub>2</sub>), 2. *Verneulinoides* sp. (Söhlde, H<sub>2</sub>O<sub>2</sub>), 3. *Gavelinella* sp. (Söhlde, Glauber's salt), 4. *Bulbobaculites* sp. (Söhlde, Glauber's salt), 5. *Gyroidinoides* sp. (Baddeckenstedt, liquid Nitrogen), 6. *Tritaxia* sp. (Baddeckenstedt, liquid Nitrogen), 7. *Lenticulina* sp. (Söhlde, acetic acid + Copper(II) sulfate), 8. *Marssonella* sp. (Söhlde, acetic acid + Copper(II) sulfate); scale bars – 100  $\mu$ m.

(Fig. 2, Table 2). Similarly, on average 32 genera are recorded in the formic acid residue from the pure limestone of Söhlde, and between 1 and 7 genera with standard micropaleontological methods (Fig. 2, Table 2). Important taxa for paleoecological reasons such as taxa with a tubular shape (e.g., *Nothia*, *Bathysiphon*, *Psammosiphonella*), and genera such as *Ammobaculites*, *Ammolagena*, *Ataxophragmium*, *Parvigenerina*, *Pseudonodosinella*, *Pseudotextulariella*, and *Saccamina* are completely missing in samples treated with standard micropaleontological methods. With standard micropaleontological procedures, only 1 to 9 different agglutinated foraminiferal species were obtained, while in average 43.3 (Söhlde) and 53.5 (Baddeckenstedt) from formic acid residues could be determined (see Table 2). In summary, 35 agglutinated foraminiferal genera and 58 species are not found in residues obtained with standard micropaleontological methods differently from the formic acid procedures (see also Appendix A).

Our study therefore highlights that by applying the standard micropaleontological procedures agglutinated foraminifera occur only in low abundance with the loss of genera and species (Fig. 2). In the same rocks the application of the formic acid method, agglutinated foraminiferal faunas occur in numbers with a statistical utility, with many genera, and high diversities. In summary, standard micropaleontological methods used to obtain foraminifera are not applicable to study agglutinated foraminifera in hemipelagic carbonates because at least 2/3 of agglutinated foraminiferal genera are missing. To study the whole fauna of foraminifera in the analyzed hemipelagic marlstones to limestones, including both calcareous and agglutinated foraminifera, two separate processes, the formic acid treatment and the acetic acid + Copper(II) sulfate method are recommended. Otherwise, information on one important part of the fauna is missing.

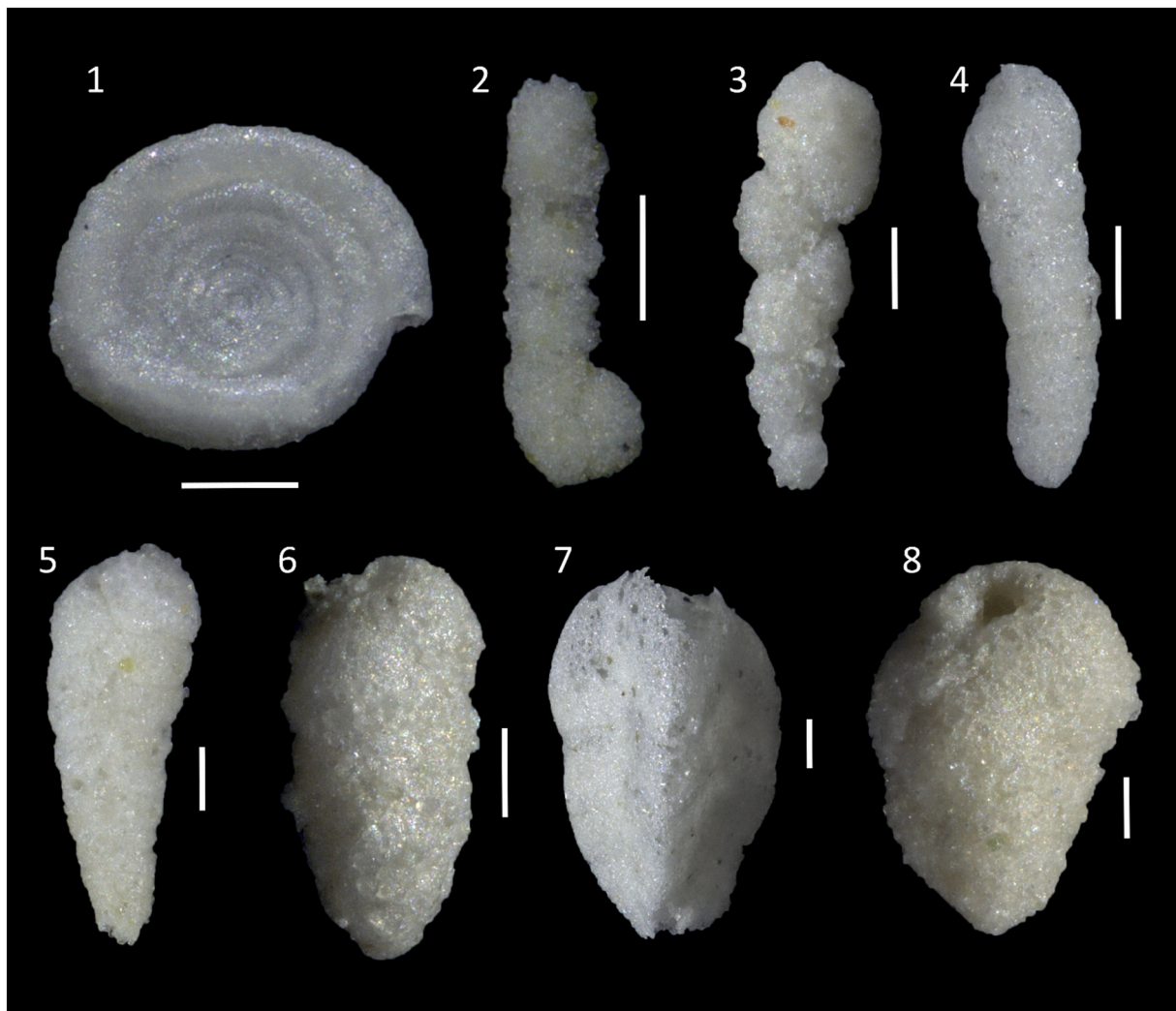
Conclusively, our study demonstrates that bias concerning the loss of agglutinated foraminifera can be introduced by standard laboratory methods adopted to disaggregate samples from the analysis of mid-Cretaceous hemipelagic rocks. Such bias can be expected also in previous studies focusing similar lithologies also from different time intervals and regions for which only the standard laboratory procedures are applied.

## 5. Preservation (standard procedures)

Different types of damages on foraminiferal tests can result of different processing techniques in purpose of rock disintegration. While corrosion and etching of calcareous tests is reported for the hydrogen peroxide and acetic acid + Copper(II) sulfate methods, no verified damages are known for freeze-thaw techniques (Hodgkinson, 1991; Remin et al., 2012) such as the Glauber's salt and the liquid Nitrogen procedures. No evidence of corrosion or etching, nor signs of enhanced fragmentation of foraminiferal tests obtained by standard procedures could be found in this study (Fig. 3). However, especially in residues obtained by the hydrogen peroxide and both freeze-thaw methods, foraminiferal tests are often not clean with sediment matrix attached to them (Fig. 3). Remin et al. (2012) reported similar observations for the Glauber's salt method, while Wissing and Herrig (1999) described the necessity of additional cleaning of foraminiferal tests after application of the Nötzold's method.

## 6. Preservation and effectiveness (formic acid)

The agglutinated foraminiferal fauna from the mid-Cretaceous marly



**Fig. 4.** Clean agglutinated foraminifera obtained by formic acid method; 1. *Ammodiscus cretaceus* (Baddekenstedt), 2. *Ammobaculites agglutinans* (Söhlde), 3. *Parvigenerina* sp. 3 (Söhlde), 4. *Gerochammina stanislawi* (Söhlde), 5. *Spiroplectammmina navarroana* (Söhlde), 6. *Plectina cenomana* (Baddekenstedt), 7. *Tritaxia tricarinata* (Söhlde), 8. *Flourensina intermedia* (Baddekenstedt); scale bars – 100  $\mu\text{m}$ .

limestone and limestone processed with formic acid are generally well preserved. Specimens are mostly not damaged despite of tubular specimens which are preserved in fragments. The fragmentation of tubular taxa is the usual preservation of these taxa (Bubík, 2019). Test surfaces are clean from attached rock matrix (Fig. 4). Even genera, such as *Dorothia*, *Eggerellina*, *Falsogaudryinella*, and *Marssonella*, are preserved which use calcareous particles or cement for their test. Nevertheless, a loss of calcareous agglutinated foraminifera due to dissolution during the acid attack is much likely to occur according to observations from previous studies (Murray and Alve, 1994, 1999a, 2011), but cannot be proven in this study.

The effectiveness of the formic acid method related to the carbonate content of the treated sample is given in the minimum range from 82.2 to 93.4% carbonate content.

### 7. Impact on paleoecological interpretation

To correctly estimate the relative abundance of agglutinated foraminifera in carbonate samples, abundance per gram data from acetic acid and formic acid treatments were compared. Corrected relative abundances of agglutinated foraminifera are 14.7% for the lower Cenomanian marly limestone (acetic acid: 5.4%), and 3.8% for the middle Turonian limestone (acetic acid: 2.4%). Therefore, relative abundances

of agglutinated foraminifera are likely underestimated in all calcareous deposits.

In addition, the loss of agglutinated foraminifera deriving from standard procedures also affects the planktic/benthic ratio as it artificially increases the planktic foraminiferal relative abundance. A corrected planktic benthic ratio combining reliable agglutinated foraminiferal specimens per gram obtained by the formic acid procedure with results from the acetic acid method ((Planktic foraminifera count (acetic acid) x 100) / (planktic foraminifera count (acetic acid) + calcareous benthic foraminifera count (acetic acid) + agglutinated foraminiferal specimens per gram (formic acid) x weight of sample (acetic acid)) decreases (Baddekenstedt, acetic acid: 35.7%, corrected: 32.9%; Söhlde, acetic acid: 64.4%, corrected 63.5%).

The paleoecological utility of exclusively agglutinated foraminiferal assemblages was shown for in another way reduced foraminiferal assemblages due to dissolution effects in sedimentary deposits (e.g., Kuhnt, 1990; Murray and Alve, 1999b; Murray et al., 2003; Kaminski et al., 2011) or artificial by laboratory experiments by Murray and Alve (1994, 1999a, 2011). By application of the formic acid procedure, a partial or complete loss of calcareous agglutinated foraminifera is possible to occur. Similar results were observed in obtained foraminiferal assemblages by application of acetic acid or hydrochloric acid (e.g., Kuhnt, 1990; Murray and Alve, 1994, 1999a, 2011; Kaminski et al.,

2011).

It is highly recommended to accurately explain the methodologies adopted to disaggregate rocks. This allows to correctly identify the reliability of data acquired and their interpretation in micropaleontological studies, especially concerning the agglutinated foraminiferal content.

## 8. Conclusions

Five different processing methods to obtain foraminifera were applied on samples of lower Cenomanian marly limestone and middle Turonian pure limestone to evaluate the real abundance and the proportion of agglutinated foraminifera in such rock types. Our results show that among the five methods ( $H_2O_2$ , Glauber's salt, liquid Nitrogen, and acetic acid + Copper(II) sulfate, formic acid) that can be applied to obtain the foraminiferal fauna only a separate application of the formic acid procedure and acetic acid + Copper(II) sulfate method is effective to obtain the whole fauna and to isolate the agglutinated foraminifera.

Specifically, we demonstrate that the formic acid method is the best procedure to provide the highest agglutinated foraminiferal specimens per gram. The agglutinated specimens/g are always lower with standard micropaleontological methods, especially with  $H_2O_2$ , liquid Nitrogen and Glauber's salt not exceeding 2.0 specimens/g, while agglutinated specimens/g lies between 6.7 and 18.1 with formic acid. Furthermore, at least 2/3 of agglutinated foraminiferal genera and species are completely missing in washed residues prepared with the standard methods. Therefore, the standard procedures introduce a significant bias about the knowledge of agglutinated foraminifera.

Our analysis reveals also that the formic acid method provides the best agglutinated foraminifera preservation because of low mechanical strain and the complete removal of calcareous rock matrix during acetolyses.

Concluding, we suggest the application of both the acetic acid + Copper(II) sulfate and formic acid method on mid-Cretaceous hemipelagic carbonate deposits to obtain the complete foraminiferal fauna. In consideration of the different functionality of different standard micropaleontological methods (soaking vs. mechanical dissection), a precise description of the applied method is highly recommended for forthcoming studies on foraminiferal faunas.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## Appendix A. Taxonomical reference list

Original descriptions and taxonomical reference of mentioned foraminiferal genera and species were given by [Loeblich and Tappan \(1987\)](#) and [WoRMS Editorial Board \(2022; www.marinespecies.org\)](#). They are

not given in the reference list.

*Ammobaculites* Cushman 1910.  
*Ammobaculites agglutinans* (d'Orbigny) 1846).  
*Ammodiscus cretaceus* (Reuss 1845).  
*Ammolagena* Eimer and Fickert 1899.  
*Ataxophragmium* Reuss 1860.  
*Bulbobaculites* Maync 1952.  
*Dorothia* Plummer 1931.  
*Eggerellina* Marie 1941.  
*Falsogaudryinella* Bartenstein 1977.  
*Flourensina intermedia* Ten Dam 1950.  
*Gavelinella* Brotzen 1942.  
*Gerochammina stanislawi* Neagu 1990.  
*Gyroidinoides* Brotzen 1942.  
*Heterohelix* Ehrenberg 1843.  
*Lenticulina* Lamarck 1804.  
*Marssonella* Cushman 1933.  
*Parvigenerina* Vella 1957.  
*Parvigenerina* sp. 3 ([Kuhnt, 1990](#)).  
*Plectina cenomana* Carter and Hart 1977.  
*Pseudonodosinella* Saidova 1970.  
*Pseudotextulariella* Barnard 1953.  
*Saccamina* Carpenter 1869.  
*Spiroplectammina navarroana* Cushman 1932.  
*Tritaxia* Reuss 1860.  
*Tritaxia tricarinata* (Reuss 1844).  
*Verneulinoides* Loeblich and Tappan 1949.

## Appendix B. Recovered genera and species for each method

### B.1. $H_2O_2$

*Psammosphaera irregularis*  
*Spiroplectammina* sp.  
*Tipeammina* sp.  
*Trochammina* sp.  
*Verneulinoides* sp.

### B.2. Glauber's salt

*Arenobulimina* sp.  
*Bulbobaculites problematicus*  
*Eggerellina brevis*  
*Hagenowella elevata*  
*Haplophragmoides* sp.  
*Plectina cenomana*  
*Repmanina charoides*  
*Tritaxia tricarinata*  
*Trochammina* sp.  
*Trochaminoides* sp.  
*Uvigerinammina praejankoi*  
*Verneulinoides* sp.  
*Vialovella frankei*

### B.3. Liquid Nitrogen ( $LN_2$ )

*Ammodiscus* sp.  
*Arenobulimina* sp.  
*Plectina cenomana*  
*Psammosphaera irregularis*  
*Tritaxia tricarinata*  
*Trochammina* sp.  
*Voloshinoides advenus*

## B.4. Nötzold's method/ acetic acid + Copper(II) sulfate

*Ammodiscus glabratus*  
*Ammodiscus peruvianus*  
*Arenobulimina truncata*  
*Arenobulimina* sp.  
*Dorothia gradata*  
*Hagenowella elevata*  
*Haplophragmoides* sp.  
*Marssonella trochus*  
*Spiroplectinella cretosa*  
*Trochammina* sp.  
*Trochamminoides* sp.  
*Vialovella frankei*

## B.5. Formic acid

*Ammobaculites agglutinatus*  
*Ammobaculites* sp.  
*Ammodiscus cretaceus*  
*Ammodiscus glabratus*  
*Ammodiscus peruvianus*  
*Ammodiscus* sp.  
*Ammodiscus tenuissimus*  
*Ammolagena clavata*  
*Ammolagena contorta*  
*Arenobulimina barnardi*  
*Arenobulimina bochumensis*  
*Arenobulimina preslii*  
*Arenobulimina* sp.  
*Arenobulimina truncata*  
*Ataxophragmium* sp.  
*Bathysiphon* sp.  
*Bicazammina lagenaria*  
*Bulbobaculites problematicus*  
*Caudammina ovula*  
*Caudammina ovuloides*  
*Caudammina* sp.  
*Clavulinoides* sp.  
*Dolgenia pennyi*  
*Dorothia gradata*  
*Eggerellina brevis*  
*Eggerellina mariae*  
*Eobigenerina kuhnti*  
*Eobigenerina variabilis*  
*Falsogaudryinella* sp.  
*Flourensina intermedia*  
*Gaudryinella irregularis*  
*Gerochammina stanislawi*  
*Glomospira diffundens*  
*Glomospira gordialis*  
*Hagenowella elevata*  
*Hagenowella obesa*  
*Haplophragmoides eggeri*  
*Haplophragmoides* sp.  
*Haplophragmoides stomatus*  
*Haplophragmoides walteri*  
*Hemisphaerammina batalleri*  
*Hemisphaerammina glandiformis*  
*Hormosinella fusiformis*  
*Hyperammina* sp.  
*Kalamopsis grzybowskii*  
*Lagenammina difflugiformis*  
*Lituotuba lituotuba*  
*Marssonella trochus*  
*Nothia* sp.

*Parvigenerina* sp. 3  
*Placentammina placenta*  
*Placopsilina* sp.  
*Plectina cenomana*  
*Plectina mariae*  
*Praecystammina* sp.  
*Psammosiphonella* sp.  
*Psammosphaera fusca*  
*Psammosphaera irregularis*  
*Pseudonodosinella nodulosa*  
*Pseudonodosinella parvula*  
*Pseudonodosinella troyeri*  
*Pseudotextulariella cretosa*  
*Rectogerochammina eugubina*  
*Recurvoides* sp.  
*Reophax* sp.  
*Repmanina charoides*  
*Rhabdammina* sp.  
*Rzehakina minima*  
*Saccammina grzybowskii*  
*Spiroplectammina navarroana*  
*Spiroplectammina* sp.  
*Spiroplectinella cretosa*  
*Subreophax scalaris*  
*Textulariopsis rioensis*  
*Tipeammina elliptica*  
*Tipeammina* sp. 1  
*Tritaxia tricarinata*  
*Trochammina* sp.  
*Trochamminoides* sp.  
*Uvigerinammina jankoi*  
*Uvigerinammina praejankoi*  
*Verneuilioides* sp.  
*Vialovella frankei*  
*Voloshinoides advenus*  
*Voloshinoides anglicus*

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