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# Organic biogeochemical techniques to evaluate the zones of methanogenic activity in sediments of Western Offshore Basin

## Abstract

Biogenic gas is becoming increasingly important as an exploration target in the petroleum industry because it occurs in large quantities at shallow depths as free gas or gas hydrates. About 20% of conventional natural gas resources in the world are biogenic and more economic reserves of biogenic gas are expected to be discovered in the future. Effective exploration of biogenic gas requires a better understanding of processes that lead to its generation, migration and entrapment. In this context, certain biogeochemical parameters and conditions viz., sediment temperature, TOC content, lipid content, gross isotopic composition of lipids, sulphate-reducing bacteria (SRB) count, trace elements (Fe, Ni, Co, Mn, Zn and Cu), pyrites and carbonates have been identified as indicators of methanogenic activity. Based on these, probable generative intervals of biogenic gas have been identified in three of the four studied wells in Shelf margin area of Western Offshore Basin viz., AM-1 (555-705m, 805-905m & 955-1005m), CO-1 (858-903m, 1002-1056m, 1158-1203m, 1257-1302m & 1551-1605m) and DD-1 (2190-2240m, 2390-2440m & 2490-2580m). Methanogenic activity could not be detected in the fourth well BS-1. However, the organic richness (TOC) is low in all the four wells suggesting that the volumes of gas generated may not be significant.

## Introduction

Biogenic gas is produced during the decomposition of organic matter by microorganisms called methanogens. These microorganisms generate methane using a variety of organic substrates such as $H_2$ plus $CO_2$, acetate and methylated compounds (methanol and methylamines) (Vandecasteele, 2008). However, methanogens, being distinct from Bacteria and Eukarya, are confined to certain environments that are necessary for their growth. Unlike thermogenic gas, biogenic gas is often generated in self-sourced gas reservoirs rich in organic carbon, such as shallow coal-seams and organic rich shale units. The organic matter consumed by microorganisms may be of two origins: i) remains of organisms accumulated within the sediments (“primary” biogenic methane quickly generated after the sedimentation close to the surface), ii) or mature thermogenic oils (“secondary” biogenic methane produced by the biodegradation of deeper oil pools) (Schneider *et al.*, 2016). This study deals with the primary biogenic gas.

## Favourable conditions for generation of biogenic gas

One of the primary requirements for the growth of methanogens is anoxic environment as methanogens are strictly anaerobes and cannot tolerate even traces of oxygen. Methanogenesis occurs at depths that are equivalent to temperatures less than 75°C which is assumed to be the pasteurization temperature (Buswell and Mueller, 1952). Higher temperatures lead to denaturation of bacterial tissues inhibiting bacterial growth and transformation of organic matter to a form (humin/ kerogen) which is less susceptible to attack by microorganisms.

A minimum of metabolizable organic matter equivalent to about 0.5% organic carbon is required to support methane production in marine sediments (Claypool and Kaplan, 1974). However, low abundance organic matter in deposits was once regarded as not being an obstacle to biogenic gas formation and even 0.18% was proposed as the minimum TOC for an effective source rock. (Zhang *et al.*, 2014). In order to produce methane, the organic matter, once buried, should be preserved partially through the oxidation and sulphate reduction zone. Thus, a minimum hydrolysable fraction is required, at a given sedimentation rate, in order to enter the fermentation zone and then to produce methane (Schneider *et al.*, 2016).

Methanogens can flourish only in those intervals where sulphate reducing bacteria are either absent or present in low numbers. This is primarily because sulphate reducing bacteria out-compete methanogens for substrates, particularly, acetate and $H_2$, thereby inhibiting methanogenesis (Jørgensen, 2005). Also, microbial metabolism is inhibited in high salinity waters and methanogenesis occurs only where chloride concentrations are <1 to 2 M (Shuai *et al.*, 2013).

Trace elements are essential components of enzymes or co-factors of metabolic pathways associated with methanogenesis. The production of methane in the subsurface could be affected by the presence or absence of metals. Of the different elements, iron, nickel, cobalt, copper, zinc and manganese are considered to be crucial in methanogenesis (Ünal *et al.*, 2012). The mobilization of manganese (conversion of ions present in the insoluble state to the soluble state by redox, acidic and complexation reactions) appropriately coincides with increase in total cell numbers, increased methane levels and maximum abundance of methanogens (Sujith *et al.*, 2014). Nickel (Ni), cobalt (Co) and Zinc (Zn) have a fundamental role in structural or catalytic functions in cellular enzymes involved in methanogenesis. High concentrations of Ni, Co and Zn viz., 470 ppm, 1770 ppm and 10 ppm respectively, are reported to be toxic to methanogens (Paulo *et al.*, 2017; Bhattacharya *et al.*, 1996). Copper as divalent ion ($Cu^{2+}$) is toxic to methanogens at concentrations greater than 800 ppm and decreases methane emission along with the diversity and abundance of methanogens and methanotrophs (Mao *et al.*, 2015). Iron as divalent ion ($Fe^{2+}$) stimulates methanogenesis and increases methane production (Ahmed *et al.*, 2001). However, methanogenesis is inhibited under $Fe^{3+}$ reducing conditions. This is usually explained by competition of methanogens and $Fe^{3+}$ reducing bacteria for the common substrates acetate and hydrogen. In addition, $Fe^{3+}$ reduction by methanogens may also contribute to $Fe^{3+}$ inhibition of methanogenesis (Bodegom *et al.*, 2004).

However, toxicity of heavy metals is alleviated in the presence of sulphides. Sulphide reacts quickly with several metal ions forming insoluble metal sulphides (Paulo *et al.*, 2017). Iron sulphide partially controls the soluble concentration of heavy metals and their toxicity in aquatic environments. Heavy metals displace the Fe from iron sulphide forming poorly soluble metal sulphides in the iron sulphide matrix. Therefore, iron sulphide may be expected to attenuate the heavy metal toxicity. But, not all the sulphide in iron sulphide is readily available to react with the soluble ions (such as, Cu and Zn) which may explain the need for a large stoichiometric excess of iron sulphide to highly attenuate toxicity of heavy metals (Gonzalez-Estrella *et al.*, 2016). Moreover, microbial methane production is less favourable in the presence of sulphate due to the energetic advantage gained by sulphate reducing bacteria. The presence of framboidal pyrite in sediments suggests sequestration of sulphur in the system as pyrite, thereby reducing the competition for substrate and allowing methanogenic archaea to flourish for a time. (Clark *et al.*, 2015).

## Favourable conditions for accumulation of biogenic gas

In nonmarine and (or) brackish-water environments that are generally low in sulphate, methane production begins close to the surface and most of the initially formed gas is lost by aerobic bacterial oxidation or escape to the atmosphere because of lack/paucity of adequate seal. In marine sediments, methane production begins at a depth below the sediment-water interface because sulphate reduction must precede it. In carbonate-evaporite sequences, biogenic methane production may be inhibited by sulphate. Because of higher hydrostatic pressures due to the overlying water column, a larger amount of biogenic methane in marine sediments can be retained in solution in the interstitial (pore) waters. This can serve as a holding mechanism until the sediments are compacted and, traps and seals are formed. If conditions are such that methane generation results in saturation of waters above the 82°C minimum solubility isotherm, the waters will become super-saturated with further burial and will produce a free gas state. At very shallow depths of burial, free gas will probably bubble to the surface and eventually will enter the atmosphere. This is a situation similar to nonmarine and (or) brackish water environments.

A large amount of biogenic methane is probably trapped stratigraphically, at least initially, in rocks of low porosity and permeability. In the marine-shelf environment, the deposition of discontinuous and relatively impermeable silt and sand enveloped by organic-rich mud and clay provides extremely favourable conditions for both in-situ generation and entrapment of biogenic methane. Another key trapping mechanism of biogenic methane may be the formation of early diagenetic carbonate cements as either layers or concretions; obviously the layers are more effective traps. Early diagenetic carbonates may be formed in two ways. First, when $CO_2$ is removed from the dissolved bicarbonate reservoirs of interstitial waters by reduction and formation of methane, the pH increases which can result in the precipitation of authigenic carbonates in anoxic sediments. A second major mechanism for precipitation of isotopically anomalous carbonate layers is from $CO_2$ generated from oxidation of methane. The hydrates can serve both as a trap for the methane enclosed in the clathrate structure, and also as a seal for hydrocarbons, including biogenic methane generated below the hydrate, or liberated at the base of a gas hydrate zone subsiding into a region of temperature instability (Rice and Claypool, 1981).

## Elucidation of zones with methanogenic activity

Indirect methods to ascertain the zones of methanogenic activity in the subsurface sediments include separation and characterization of compounds that are peculiar to methanogens. Such compounds include polar lipids present in archaeal cell membrane (Koga *et al.*, 1993) and specific acyclic isoprenoids as biological markers (Brasell *et al.*, 1981). This study is based on the extraction of lipids from sediments to ascertain zones of methanogenic activity.

Considering the above discussed conditions and parameters as indicators of biogenic gas generation and accumulation, zones of biogenic gas generation have been evaluated in four wells in Shelf margin area of Western Offshore Basin viz., AM-1, BS-1, CO-1 and DD-1.

Well AM-1 produced gas from the intervals 932-938 m, 581-583 m and 585-589 m ($\delta^{13}C_1$: -55.0 ‰ and -60.0 ‰ respectively). Lipids have been found in certain intervals between 505 and 1005 m. The intervals with low SRB count are more likely to witness methanogenic activity as sulphate reducing bacteria are known to out-compete methanogens for substrates. Thus, zones with high lipid content and low SRB count are most likely generative intervals of biogenic gas. Since methanogens, like all other bacteria, preferentially consume $^{12}C$, lipids present in their cell membranes are isotopically lighter. Thus, intervals where decrease in $\delta^{13}C$ of lipids is observed can be the probable zones of methanogenic activity. Moreover, generative intervals of biogenic gas are marked by concentrations of trace elements below their toxicity limits, such as, Ni <470 ppm, Co <1770 ppm and Cu <800 ppm. However, Zn may be present as insoluble sulphides or carbonates and hence, is not toxic to methanogens which is corroborated by presence of pyrite against 545 m depth in the Master log of the well. Concentration of Mn which is known to stimulate methanogenesis, is also high, being comparatively higher in the shallower identified zones (555-705 m) than the deeper ones. In addition, limestone sections have been encountered in this well in several depths below 795 m. Early diagenetic carbonates may be formed when $CO_2$ is removed from the dissolved bicarbonate reservoirs of interstitial waters by reduction and formation of methane, resulting in increase in pH and subsequent precipitation of authigenic carbonates in anoxic sediments. Based on these observations, three zones have been identified as probable generative intervals of biogenic gas viz., 555-705 m, 805-905 m and 955-1005 m (Figure 1). Absence of lipids below 1005 m rules out the microbial activity in the deeper sediments**.**

Figure 1: Variation of biogeochemical parameters with depth in well AM-1. Three zones (555-705 m, 805-905 m and 955-1005 m) identified as probable generative intervals of biogenic gas.

In well BS-1, lipids have been encountered in only the shallowest studied interval, i.e. 745-790 m. Though lipid content is high in this interval, this zone is not being considered as generative interval of biogenic gas because of very high SRB count in this interval (1000-10000 MPN). Well is devoid of lipids in sediments below 790 m, hence methanogenic activity can be ruled out in sediments deeper than 745 m depth in the well (Figure 2).

Figure 2: Variation of biogeochemical parameters with depth in well BS-1. No zone could be identified as probable generative interval of biogenic gas.

In well CO-1, lipids have been found almost throughout the studied column, except five intervals. Intervals with high lipid content, low SRB count, decrease in $\delta^{13}C$ of lipids and concentrations of trace elements below their toxicity limits are most likely generative intervals of biogenic gas. Zn may be present as insoluble sulphide due to which its toxicity is alleviated. This is corroborated by the presence of pyrite in each of the identified zones as seen in Composite Log. Concentration of Mn, which is known to stimulate methanogenesis, is strikingly higher in the interval 858-1100 m. Interestingly, this interval corresponds to strikingly low concentration of Zn. This trend of a sharp increase in the concentration of Mn (a methanogenesis stimulating element) and a sharp decrease in the concentration of Zn (a methanogenesis inhibiting element) is very peculiar. Composite Log shows a marked increase in the total gas (methane) concentration in the interval 850-905 m. So a high concentration of Mn and low concentration of Zn do suggest favourable conditions for methanogenic activity. In addition, dolomite sections have been encountered in this well in several depths below 725 m. The depth intervals where dolomite sections have been encountered correlate well with the identified zones of probable generative intervals of biogenic gas.  Based on these observations, five zones have been identified as probable generative intervals of biogenic gas viz., 858-903 m, 1002-1056 m, 1158-1203 m, 1257-1302 m and 1551-1605 m (Figure 3). Since TOC is very low in these identified intervals (0.1-0.32 %), so the volumes of biogenic gas generated would be very less.

Figure 3: Variation of biogeochemical parameters with depth in well CO-1. Five zones (858-903 m, 1002-1056 m, 1158-1203 m, 1257-1302 m and 1551-1605 m) identified as probable generative intervals of biogenic gas.

In well DD-1, the ROV monitored and photographed hydrate formation below the BOP stack indicating the possibility of shallow gas or gas hydrates being present at the location. The well has water depth of 1077m, and no cuttings are available till 2034m as the well was drilled in riserless phase. The hydrate formation confirms the methanogenic activity in the well and this would be in all probability in the sediments at shallower depths. Lipids have been found almost throughout the studied column, except one interval (2640-2700 m). Sulphate reducing bacteria are completely absent throughout the sedimentary column, barring few intervals where SRB count is very low (10-100 MPN). Lipids in this well are much lighter isotopically than in the other studied wells. $\delta^{13}C$ of lipids is as low as -37.7 ‰. Also, concentrations of trace elements (Ni, Co and Cu) are below their toxicity limits. Zn may be present as insoluble sulphides as suggested by the presence of pyrites in the Composite Log. Mn (which is known to stimulate methanogenesis) is also present throughout the sedimentary column. Based on these observations, three zones have been identified as probable generative intervals of biogenic gas viz., 2190-2240 m, 2390-2440 m and 2490-2580 m (Figure 4).

Figure 4: Variation of biogeochemical parameters with depth in well DD-1. Three zones (2190-2240 m, 2390-2440 m and 2490-2580 m) identified as probable generative intervals of biogenic gas.

## Conclusions

Zones of methanogenic activity have been identified in three wells viz., AM-1, CO-1 and DD-1 of Shelf Margin area of Western Offshore Basin based on high lipid content, relative decrease in gross isotopic composition of lipids, low sulphate-reducing bacteria count, presence of trace elements (Ni, Co, Mn, Zn, Cu) below their toxicity limits, pyrites and carbonates. Well BS-1 is devoid of lipids and hence, studied section is not favourable for biogenic gas generation.

Though methanogenic activity has been observed, TOC is quite low and so the volumes of biogenic gas that would have been generated would be less.

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