

## Characterization of light gaseous hydrocarbons of the surface soils of Krishna-Godavari basin, India

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### Abstract

Several techniques are used for the exploration of hydrocarbons, of which; the geochemical techniques involving the microbiological technique use the principle of detecting the light hydrocarbon seepage activities for indication of sub-surface petroleum accumulations. A survey was carried out to characterize the light gaseous hydrocarbons seeping in oil and gas fields of Krishna-Godavari basin of Andhra Pradesh. A set of 50 sub-soil samples were collected at depths of about 3 m for geochemical analyses and 1 m for microbiological analysis. The microbial prospecting studies showed the presence of high bacterial population for methane  $2.5 \times 10^2$  to  $6.0 \times 10^6$  cfu g<sup>-1</sup>, propane  $1 \times 10^2$  to  $8.0 \times 10^6$  cfu g<sup>-1</sup> in soil samples. The adsorbed soil gas analysis showed the presence of moderate to low concentrations of methane (26 to 139 ppb), ethane (0 to 17 ppb), propane (0 to 8 ppb), butane (0 to 5 ppb) and pentane (0 to 2 ppb) in the soil samples of the study area. Carbon isotope analysis for methane (<sup>13</sup>C<sub>1</sub>) ranging from -36.6 to -22.7‰ Pee Dee Belemnite (PDB) suggests these gases are of thermogenic origin. Geo-microbial prospecting method coupled with adsorbed soil gas and carbon isotope ratio analysis have thus shown good correlation with existing oil/ gas fields of Krishna-Godavari basin.

### Publication Data

Paper received:  
25 September 2010

Revised received:  
13 January 2011

Accepted:  
05 February 2011

### Key words

Light gaseous hydrocarbons, Adsorbed soil gas, Carbon isotope analysis, Hydrocarbon oxidizing bacteria, Thermogenic

### Introduction

Surface geochemical methods detect the minute quantities of migrating hydrocarbons at the sub-surface levels. The migrated hydrocarbons reside in the near surface as free and bound gases; however, only free gases migrate from depth. Free gases occur as either vapor in pore spaces or as gas dissolved in aqueous solution. If the gas is attached to the sediment matrix or contained within the interstices of rocks or certain minerals such as calcite or oxide

coatings, it is considered to be bound (Jones *et al.*, 2000). Bound gases include adsorbed and chemi-adsorbed gases. Gases that have reached the soil horizon may also contain biogenic, thermogenic, and/or abiogenic gases that migrated to the surface from deep sources (Saunders *et al.*, 1999). Near-surface free gases are dominated by gases from deep sources but may also contain gases formed during diagenesis, such as biogenic methane (Jenden *et al.*, 1993). To minimize the influence of biogenic or

another source of C1, some other hydrocarbon constituent (ethane, propane and butanes) should be measured (Jones *et al.*, 2000).

Most of the studies on organic geochemical exploration concern the analysis of light gaseous hydrocarbons in recent sediments because these compounds migrate easier than the heavier, liquid hydrocarbons. Hydrocarbons reaching to the surface can be measured directly - both in the sediment itself and in the overlying air or water or through geochemical changes they induce. These hydrocarbons are extracted from adsorbed gas by processing of fine sediment fractions as hydrocarbons are sorbed within clay lattices. Trace amounts of light hydrocarbons collected near the earth's surface provide clues to present day subsurface fluid composition and migration (Gevirtz, 2002). Recent advances in analytical technology as well as fundamental and applied research have contributed towards lowering detection limits. In particular, the measurement of carbon isotopic ratios of gas quantities in the micro-liter range has increased the value of the analyses. In sediments, two gas fractions are generally distinguished: 1) interstitial gas which is dissolved in the pore water and can be released by mechanical disintegration of the sediment (Kvenvolden *et al.*, 1981) and 2) adsorbed gas which is bound on mineral surfaces and can be released by acid treatment (Horvitz, 1981).

Microbial prospecting method for hydrocarbon research and exploration is based on the premise that the light gaseous hydrocarbons, namely methane (C1), ethane (C2), propane (C3) migrate upward from subsurface petroleum accumulations by diffusion and effusion, and are utilized by a variety of microorganisms present in the sub-soil ecosystem. The methane, propane oxidizing bacteria exclusively use these gases as carbon source for their metabolic activities and growth. These bacteria are mostly found enriched in the shallow soils /sediments above hydrocarbon bearing structures and can differentiate between hydrocarbon prospective and non prospective areas (Tucker and Hitzman, 1994). The methane oxidizing bacteria are usually predominant over gas fields as the gas reservoirs are commonly dominated by methane (Jones *et al.*, 2000). The isolation and enumeration of light gaseous oxidizing bacteria are used as indirect indicators in petroleum prospecting. Microbial Prospecting for Oil and Gas (MPOG) method success rate has been reported to be 90% (Wagner *et al.*, 2002). This method can be integrated with geological, geochemical, geophysical methods to evaluate the hydrocarbon prospect of an area and to prioritize the drilling locations thereby reducing drilling risks and achieving higher success in petroleum exploration (Wagner *et al.*, 2002).

This paper reports the amount and composition of adsorbed light gaseous hydrocarbon, carbon isotopic analysis and microbiological analysis of samples collected in part of Krishna-Godavari basin in order to define the geochemical signature and characterization of light hydrocarbons over oil and gas proven areas. The originality is in terms of one of the first published works in the area of microbiological prospecting over a known hydrocarbon occurrence in Krishna-Godavari basin to establish feasibility of its application in the basin.

## Materials and Methods

**Geology of the study area:** The Krishna-Godavari basin is a pericratonic rift margin system with an archean basement on the east coast of the Indian peninsula. It covers an area of 28,000 km<sup>2</sup> on land and 24,000 km<sup>2</sup> off land up to 200 m bathymetry. The basin lies between 15°30' to 17°N latitudes and 80° to 82° 30' E longitudes. The basin is divided into Krishna, East Godavari and West Godavari depressions separated by basement highs at Bapatla and Tanuku horsts, respectively. The average temperature ranges between a maximum of 32° to 36°C and a minimum of 23° to 24°C; and the average rainfall is recorded between 800-1100 mm. The soil types vary from deltaic alluvium, red soils with clay, red loams, coastal sands and saline soils. The crops grown are paddy, coconut, mango, flowers and sweet orange. Despite the severity of tropical climate and vagaries of soil breathing, geochemical techniques can credibly identify the signatures of gaseous thermogenic hydrocarbons emanating from a subsurface petroleum accumulation and the microbiological methods are still able to provide results which are very much complimentary to the results obtained from geochemical methods. Thus microbiological methods can be employed with confidence as cheaper and faster methods to carry out a rapid reconnaissance and semi-detailed surveys to provide credible focus for likely hydrocarbon occurrence demanding a priority acquisition of costlier seismic surveys. The depositional environment of the basin varies from continental to lagoonal, marine, littoral, infraneric and deltaic conditions. The sediment yield rich faunal assemblages. Geological location map of Krishna-Godavari basin is shown in Fig. 1.

**Sampling:** A total of 50 soil samples were collected from a depth of 1 - 2.5 m using a hollow metal pipe by manual hammering to the required depth. About 100 g soil samples were collected in pre-sterilized whirl-pack bags under aseptic conditions from a depth of about 1 m (Wagner *et al.*, 2002) and stored at 2-4°C for microbial analysis. For adsorbed soil gas analysis, the cores collected were wrapped in aluminum foil and sealed in poly-metal packs. The samples were collected along a reconnaissance pattern with a spacing of 3 km (Fig. 2).

**Isolation of hydrocarbon oxidizing bacteria:** Isolation and enumeration of methane and propane oxidizing bacteria for each sample was carried out by standard plate count (SPC) method. One gram of soil sample was suspended in 9 ml of pre-sterilized water for the preparation of decimal dilutions (10<sup>-1</sup> to 10<sup>-5</sup>). A 0.1 ml aliquot of each dilution was plated on to mineral salts medium (MSM) (Ronald and Lawrence, 1996). These plates were placed in a glass desiccator, filled with the desired hydrocarbon gas (methane/propane with 99.99% purity) and zero air (purified atmospheric gas devoid of hydrocarbons) in a ratio of 1:1. For isolation of methane oxidizing bacteria, the desiccator was filled with methane gas and zero air. Similarly, for isolation of propane oxidizing bacteria, the desiccators were filled with propane gas and zero air respectively. These desiccators were kept in bacteriological incubators at 35 ± 2°C for 10 days. After incubation, the developed bacterial colonies

of methane and propane oxidizing bacteria were manually counted using colony counter and reported in colony forming units (cfu g<sup>-1</sup> of soil sample) (Sreenivas *et al.*, 2005; Rasheed *et al.*, 2008).

**Analysis of hydrocarbons:** The light gaseous hydrocarbons were extracted from the soil samples using a gas extraction system (Horvitz, 1981). One gram of 63  $\mu$  wet sieved soil sample was used to extract light gaseous hydrocarbons after acid treatment in glass degasification apparatus and its subsequent analyses on gas chromatograph (GC) for all samples and gas chromatograph-combustion-isotope ratio mass spectrometer (GC-C-IRMS) analysis for samples that had higher concentrations of hydrocarbons. During acid treatment, the dominant gas released was CO<sub>2</sub> and was trapped in KOH solution. The light gaseous hydrocarbons were collected by water displacement in a graduated tube fitted with rubber septa (Kumar *et al.*, 2002). The volume of desorbed gas was recorded and 500  $\mu$ l of desorbed gas sample was injected into Varian CP 3380 gas chromatograph fitted with Porapak Q column, equipped with flame ionization detector. The gas chromatograph was calibrated using external standards with known concentrations of methane, ethane, propane, n-butane and n-pentane. The quantitative estimation of light gaseous hydrocarbon constituents in each sample was made using peak area measurements and the correction for moisture content on wet basis was also applied. The hydrocarbon concentration values of individual hydrocarbons from methane through pentane are expressed in parts per billion (ppb). The GC accuracy of measurement of methane (C<sub>1</sub>), ethane (C<sub>2</sub>), propane (C<sub>3</sub>), butane (C<sub>4</sub>) and pentane (C<sub>5</sub>) components is ~ 1ppb.

## Results and Discussion

**Microbiological study:** The soil samples collected from Krishna-Godavari oil and gas fields were analyzed for the presence of propane oxidizing bacteria using standard plate count (SPC) method. The bacteria, which are able to utilize propane gas as a sole carbon source, are merely developed as bacterial colonies on the MSM plates. Positive control of known hydrocarbon oxidizing bacterial strain namely, *Rhodococcus rhodochrous* MTCC 291 were obtained from the Microbial Type Culture Collection and Gene Bank (MTCC), IMTECH, Chandigarh, which were inoculated onto MSM plates and incubated along with the test soil samples. The growth was observed in the positive control and in the test samples after incubation. Results of methane and propane oxidizing bacteria of Krishna-Godavari basin are given in Table 1.

The bacterial count of methane oxidizing bacteria ranged from 2.5 x 10<sup>2</sup> to 6.0 x 10<sup>6</sup> cfu gm<sup>-1</sup> of soil with a mean of 1.12x10<sup>6</sup> cfu gm<sup>-1</sup>. The standard deviation value was 1.71 x 10<sup>6</sup> cfu gm<sup>-1</sup> of soil. The bacterial count of propane oxidizing bacteria ranged from 1x10<sup>2</sup> to 8.0 x 10<sup>6</sup> cfu gm<sup>-1</sup> of soil with a mean of 1.1x10<sup>5</sup> cfu gm<sup>-1</sup>. The standard deviation value was 1.74 x 10<sup>5</sup> cfu gm<sup>-1</sup> of soil (Table 2). A statistical approach was followed and standard deviation value was taken as a background value for the demarcation of anomalous zones. The minimum, maximum, average and standard deviation values of methane (C<sub>1</sub>), ethane (C<sub>2</sub>), propane (C<sub>3</sub>), butane (nC<sub>4</sub>)

and pentane (nC<sub>5</sub>) are given in Table 2. The results of hydrocarbon oxidizing bacterial population *i.e.*, methane and propane oxidizing bacteria were plotted on the surveyed map. The samples showing bacterial population less than the background values indicate negative prospects, while the value above the standard deviation value gives the anomalies concentration of these gaseous hydrocarbon oxidizers. In the present study, the anomalous zones for methane and propane oxidizing bacteria were observed in the study area (Fig. 3,4).

**Integration with geochemical studies:** To minimize the influence of biogenic or other sources of C<sub>1</sub> methane, it is important to measure C<sub>2+</sub> hydrocarbons such as propane. The geo-chemical signature of the near surface sediment, determined mostly by the molecular composition of light hydrocarbons and the carbon isotope ratio of methane, ethane, is indicative of its seepage/migration and genetic origin (biogenic versus thermal). The magnitudes of each of the five organic constituents of the alkane series methane (C<sub>1</sub>), ethane (C<sub>2</sub>), propane (C<sub>3</sub>), butane (nC<sub>4</sub>) and pentane (nC<sub>5</sub>) were measured and expressed in ppb of gas per unit volume of the soil gas mixture. Results of adsorbed soil gas data of Krishna-Godavari basin are given in Table 3.

The adsorbed soil gas analysis showed the presence of moderate concentrations of methane C<sub>1</sub> (26 to 139 ppb), ethane C<sub>2</sub> (0 to 17 ppb), propane C<sub>3</sub> (0 to 8 ppb), butane nC<sub>4</sub> (0 to 5 ppb) and pentane nC<sub>5</sub> (0 to 2 ppb). Adsorbed methane, ethane, propane, butane and pentane concentration in the soil samples are given in Fig. 5, 6, 7, 8 and 9. Pixler's plot shown in Fig. 10 (Madhavi *et al.*, 2009) is indicative of the zone of occurrence of the accumulated hydrocarbons; here, the samples fall in oil and oil-gas zone.

The higher hydrocarbon concentration in the samples is the characteristic of the gases of petroliferous origin and follow the trend C<sub>1</sub>>C<sub>2</sub>>C<sub>3</sub>>C<sub>4</sub>>nC<sub>4</sub>>iC<sub>4</sub>>nC<sub>5</sub>, which further confirms the petroliferous nature of these hydrocarbon gases. The Pearson correlation coefficient for adsorbed soil gas between C<sub>1</sub>-C<sub>2</sub>, C<sub>1</sub>-C<sub>3</sub>, C<sub>1</sub>-C<sub>4</sub>, C<sub>1</sub>-C<sub>5</sub>, C<sub>2</sub>-C<sub>3</sub> (r=0.88), C<sub>2</sub>-C<sub>4</sub>, C<sub>2</sub>-C<sub>5</sub>, C<sub>3</sub>-C<sub>4</sub>, C<sub>3</sub>-C<sub>5</sub>, C<sub>4</sub>-C<sub>5</sub>, C<sub>2</sub>- $\delta^{13}C_{2+}$  (r=0.93), C<sub>3</sub>- $\delta^{13}C_{2+}$  (r=0.91) (Table 4) show moderate correlation respectively and indicate that these hydrocarbons are genetically related and they are not affected by secondary alteration during their migration from the sub-surface to subsequent adsorption onto the surface soil and they might have been generated from a thermogenic source (Madhavi *et al.*, 2009). Adsorbed soil  $\Sigma C_{2+}$  concentration map in the study area is shown in Fig. 11.

This suggests that all the hydrocarbon constituents in the microseep are genetically related and not influenced by secondary alterations during migration of microseeps from the sub surface to the surface and were subsequently adsorbed on to the soil. Geo-microbial counts were integrated with light hydrocarbon (adsorbed soil gas, C<sub>1</sub> – nC<sub>5</sub>) concentrations. Composite maps for light hydrocarbons and hydrocarbon oxidizing bacteria *i.e.* C<sub>1</sub> vs MOB and C<sub>3</sub> vs POB shown in Fig. 12, 13 indicate that anomalies are adjacent and follow the natural model depicting 'halo' pattern. This

**Table - 1:** Results of methane and propane oxidizing bacteria of Krishna-Godavari basin

Sample Id	Latitude	Longitude	MOB (cfu gm <sup>-1</sup> )*	POB (cfu gm <sup>-1</sup> )*
KG/03	81.875	16.505	4800000	2900
KG/05	81.875	16.500	1100000	28400
KG/07	81.885	16.481	2200000	70000
KG/08	81.885	16.477	580000	57200
KG/09	81.885	16.472	5000	612000
KG/10	81.884	16.467	330000	280000
KG/11	81.885	16.496	4700000	14000
KG/12	81.885	16.500	7000	260000
KG/13	81.885	16.505	6000000	2400
KG/18	81.894	16.519	18100	1600
KG/19	81.894	16.514	4800000	170000
KG/25	81.912	16.514	2000000	459200
KG/26	81.913	16.519	3500000	284800
KG/27	81.903	16.519	3100000	10000
KG/29	81.903	16.514	2800000	86400
KG/47	81.941	16.486	10000	171000
KG/48	81.941	16.490	600	700
KG/58	81.951	16.519	22700	4000
KG/59	81.941	16.519	3000	47200
KG/61	81.951	16.514	2500000	45200
KG/64	81.932	16.519	4800000	9000
KG/66	81.922	16.514	4000000	11000
KG/67	81.924	16.524	2800000	600
KG/68	81.927	16.524	22000	800000
KG/73	81.941	16.531	250	400
KG/76	81.951	16.524	500000	236000
KG/77	81.951	16.528	32000	0
KG/92	81.903	16.496	2000	124000
KG/99	81.922	16.467	2000	520000
KG/110	81.889	16.501	10000	22000
KG/201	81.913	16.477	20000	312000
KG/205	81.951	16.519	2400000	100000
KG/208	81.951	16.505	18500	50000
KG/209	81.960	16.505	30000	260000
KG/211	81.960	16.496	3000	11000
KG/212	81.957	16.486	20000	4000
KG/213	81.958	16.491	1200000	0
KG/214	81.951	16.496	4000	29000
KG/215	81.951	16.500	100000	32000
KG/217	81.951	16.486	20000	41000
KG/218	81.951	16.491	1200000	3900
KG/222	81.941	16.500	26000	3000
KG/223	81.943	16.496	11000	144000
KG/238	81.922	16.519	44000	140000
KG/249	81.885	16.519	9000	68000
KG/267	81.960	16.533	7200	100
KG/282	81.913	16.533	2200	2500
KG/301	81.866	16.491	200000	7300
KG/378	81.903	16.467	6300	0
KG/379	81.903	16.477	189000	0

\* cfu gm<sup>-1</sup> = Colony forming units per gram, \* MOB = Methane oxidizing bacteria, \*POB = Propane oxidizing bacteria

**Table - 2:** Statistical analysis of the adsorbed soil gas samples and microbial samples from parts of Krishna-Godavari basin

Parameter	Methane (C <sub>1</sub> )	Ethane (C <sub>2</sub> )	Propane (C <sub>3</sub> )	Normal butane (nC <sub>4</sub> )	Normal pentane (nC <sub>5</sub> )	Methane oxidizing bacteria (MOB)	Propane oxidizing bacteria (POB)
Minimum	26	0	0	0	0	2.5x10 <sup>2</sup>	0
Maximum	139	17	8	5	2	6.0x10 <sup>6</sup>	8.0x10 <sup>6</sup>
Mean	46	6	3	0	0	1.12x10 <sup>6</sup>	1.1x10 <sup>5</sup>
S.D.	20	3.8	2	1	0.25	1.71x10 <sup>6</sup>	1.74x10 <sup>5</sup>

**Table - 3:** Results of adsorbed soil gas data of Krishna-Godavari basin

Sample Id	Latitude	Longitude	Methane (C <sub>1</sub> )	Ethane (C <sub>2</sub> )	Propane (C <sub>3</sub> )	nButane (C <sub>4</sub> )	nPentane (C <sub>5</sub> )
KG/03	81.875	16.505	139*	17	8	0	0
KG/05	81.875	16.500	82	4	0	0	0
KG/07	81.885	16.481	39	0	0	0	2
KG/08	81.885	16.477	44	0	0	0	0
KG/09	81.885	16.472	32	0	0	0	0
KG/10	81.884	16.467	26	0	0	0	0
KG/11	81.885	16.496	67	0	0	0	0
KG/12	81.885	16.500	31	0	0	0	0
KG/13	81.885	16.505	49	4	2	0	0
KG/18	81.894	16.519	29	4	0	0	0
KG/19	81.894	16.514	76	13	6	0	0
KG/25	81.912	16.514	66	6	2	0	0
KG/26	81.913	16.519	73	13	7	0	0
KG/27	81.903	16.519	71	10	3	0	0
KG/29	81.903	16.514	61	10	5	0	0
KG/47	81.941	16.486	43	7	4	0	0
KG/48	81.941	16.490	30	5	4	0	0
KG/58	81.951	16.519	34	4	2	0	0
KG/59	81.941	16.519	38	7	3	0	0
KG/61	81.951	16.514	61	10	5	5	0
KG/64	81.932	16.519	54	9	5	0	0
KG/66	81.922	16.514	38	7	3	0	0
KG/67	81.924	16.524	57	12	5	4	0
KG/68	81.927	16.524	42	8	6	0	0
KG/73	81.941	16.531	42	6	2	0	0
KG/76	81.951	16.524	29	4	2	0	0
KG/77	81.951	16.528	39	8	3	0	0
KG/92	81.903	16.496	31	4	0	0	0
KG/99	81.922	16.467	34	6	2	0	0
KG/110	81.889	16.501	30	6	4	0	0
KG/201	81.913	16.477	26	4	2	0	0
KG/205	81.951	16.519	63	9	4	0	0
KG/208	81.951	16.505	28	5	2	0	0
KG/209	81.960	16.505	28	5	1	0	0
KG/211	81.960	16.496	54	9	5	0	0
KG/212	81.957	16.486	33	7	3	0	0
KG/213	81.958	16.491	55	9	4	0	0
KG/214	81.951	16.496	29	5	2	0	0
KG/215	81.951	16.500	47	5	0	0	0
KG/217	81.951	16.486	41	8	4	0	0
KG/218	81.951	16.491	61	11	4	0	0
KG/222	81.941	16.500	50	6	4	0	0
KG/223	81.943	16.496	26	3	0	0	0
KG/238	81.922	16.519	38	4	4	0	0
KG/249	81.885	16.519	40	7	2	0	0
KG/267	81.960	16.533	32	6	3	0	0
KG/282	81.913	16.533	45	9	5	0	0
KG/301	81.866	16.491	61	8	4	0	0
KG/378	81.903	16.467	38	0	0	0	0
KG/379	81.903	16.477	31	0	0	0	0

\*ppb = Parts per billion



Fig. 1: India map showing location of Krishna-Godavari basin

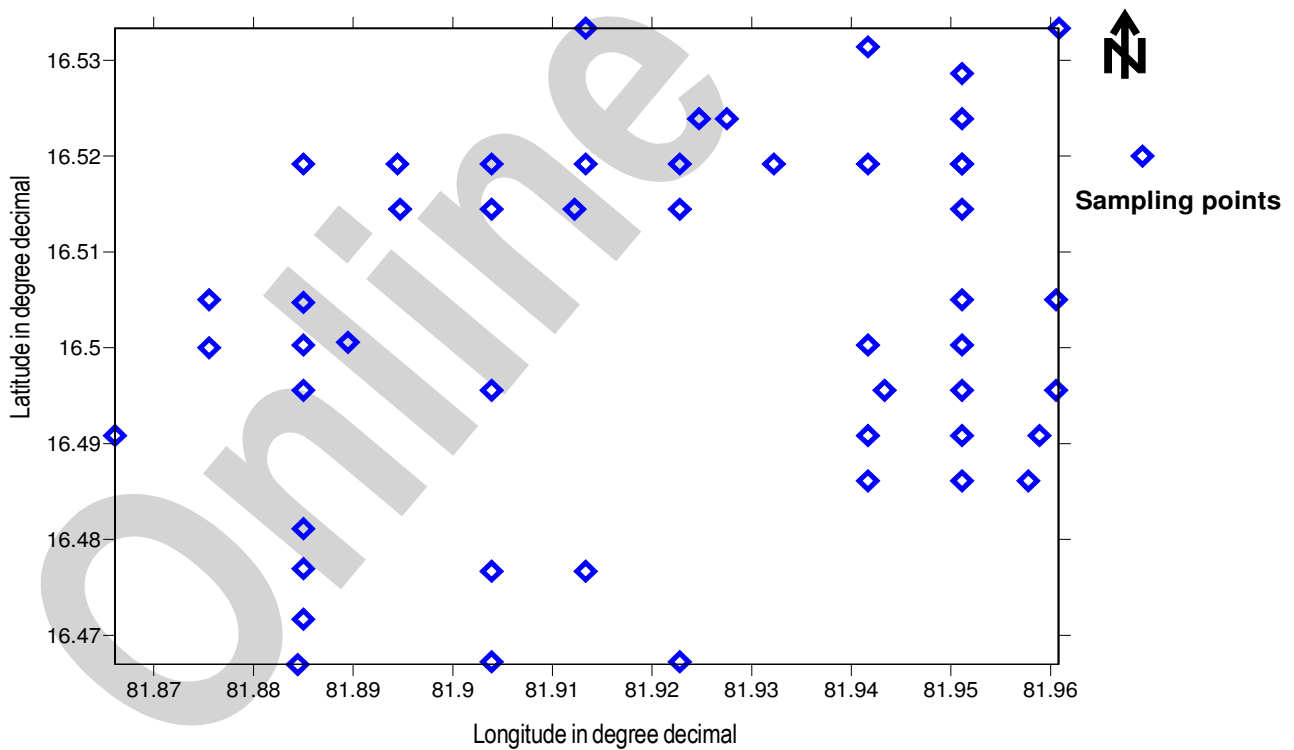


Fig. 2: Sampling points (◇) in the study area of Krishna-Godavari basin

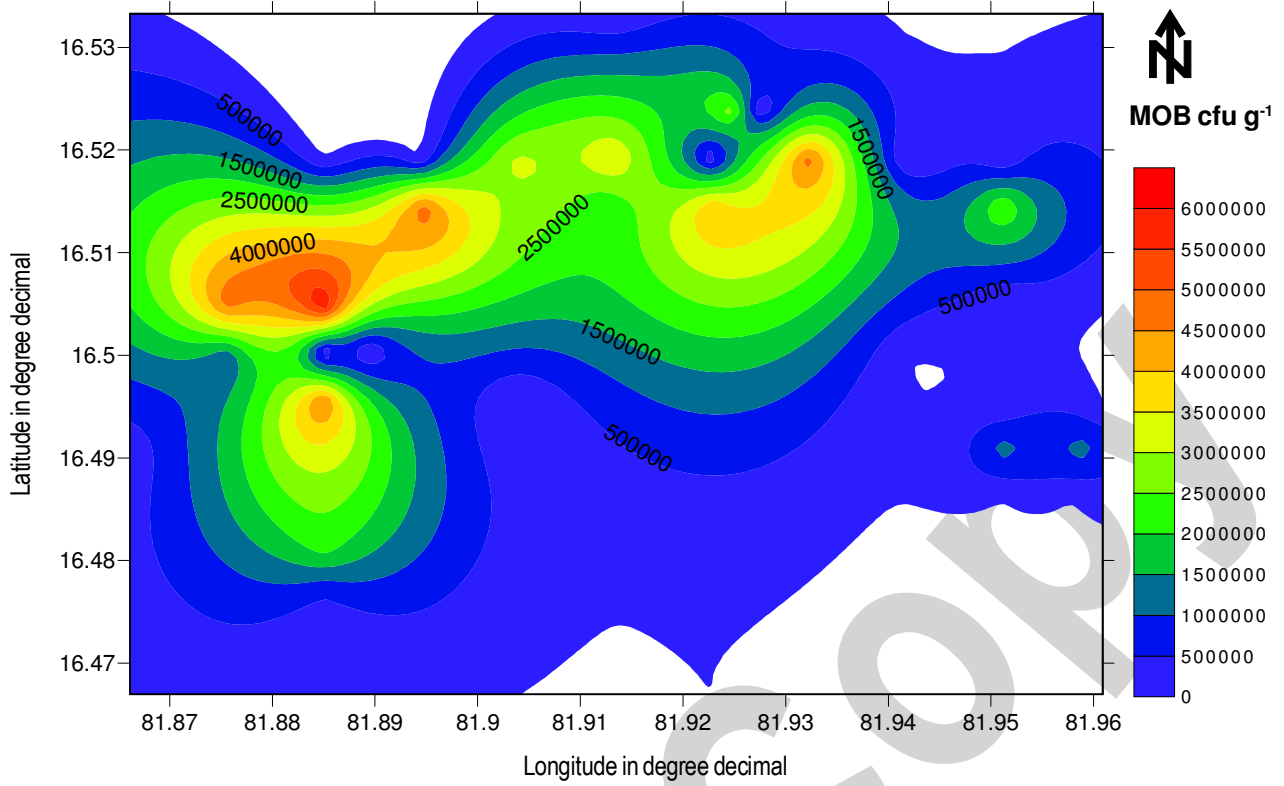


Fig. 3: Methane oxidizing bacterial concentration map in the study area

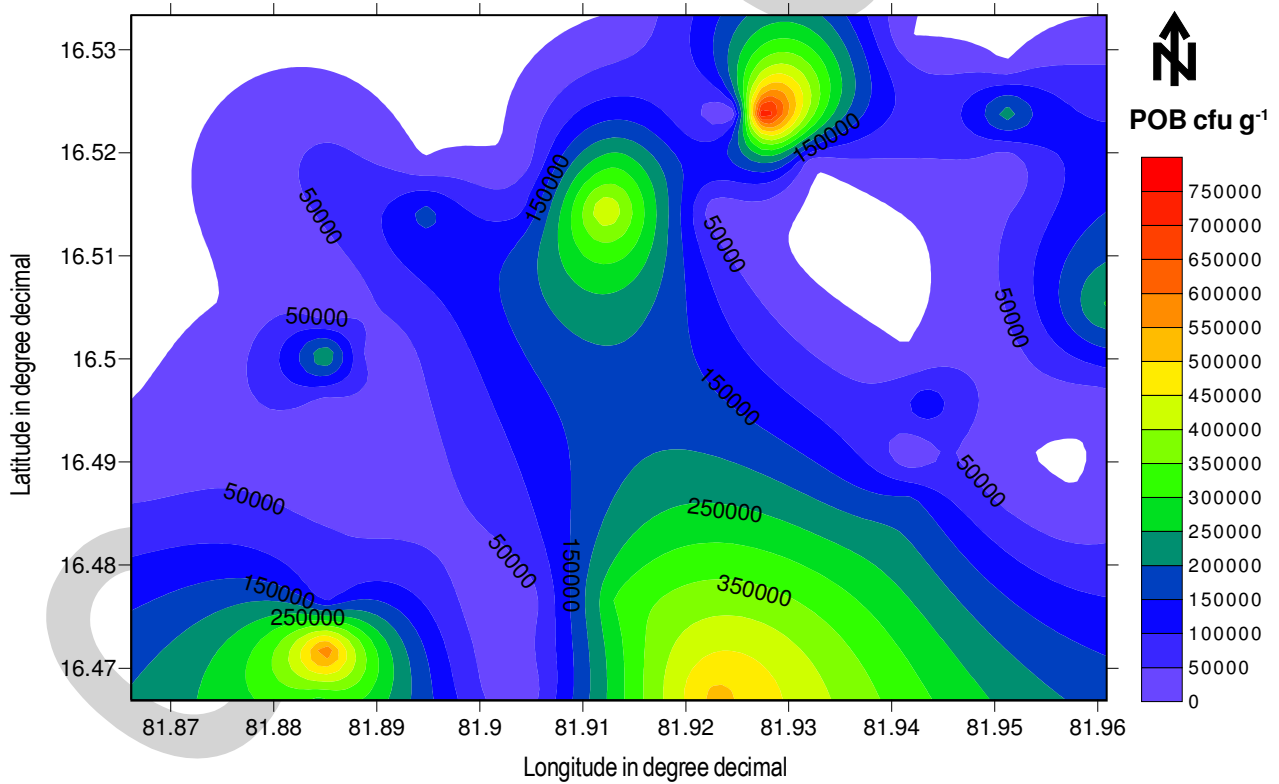


Fig. 4: Propane oxidizing bacterial concentration map in the study area

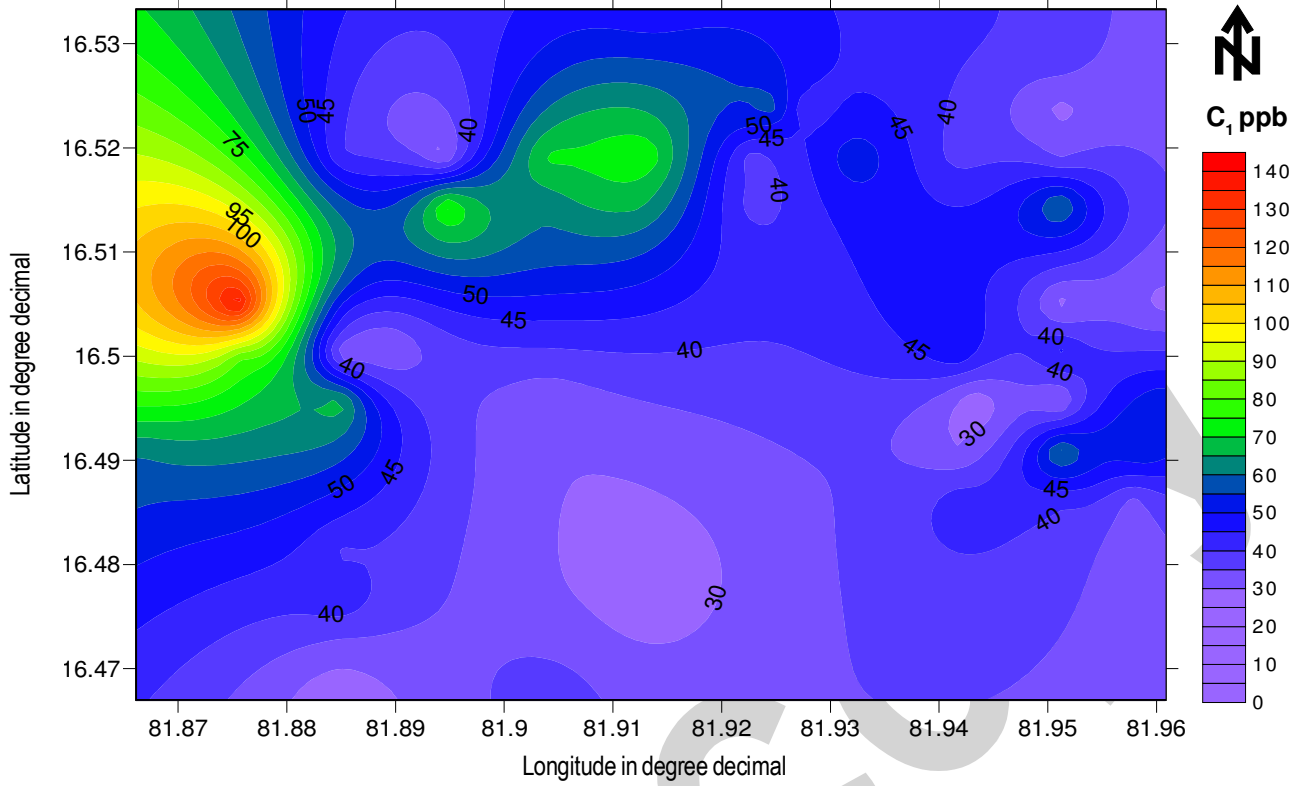


Fig. 5: Adsorbed soil methane concentration map in the study area

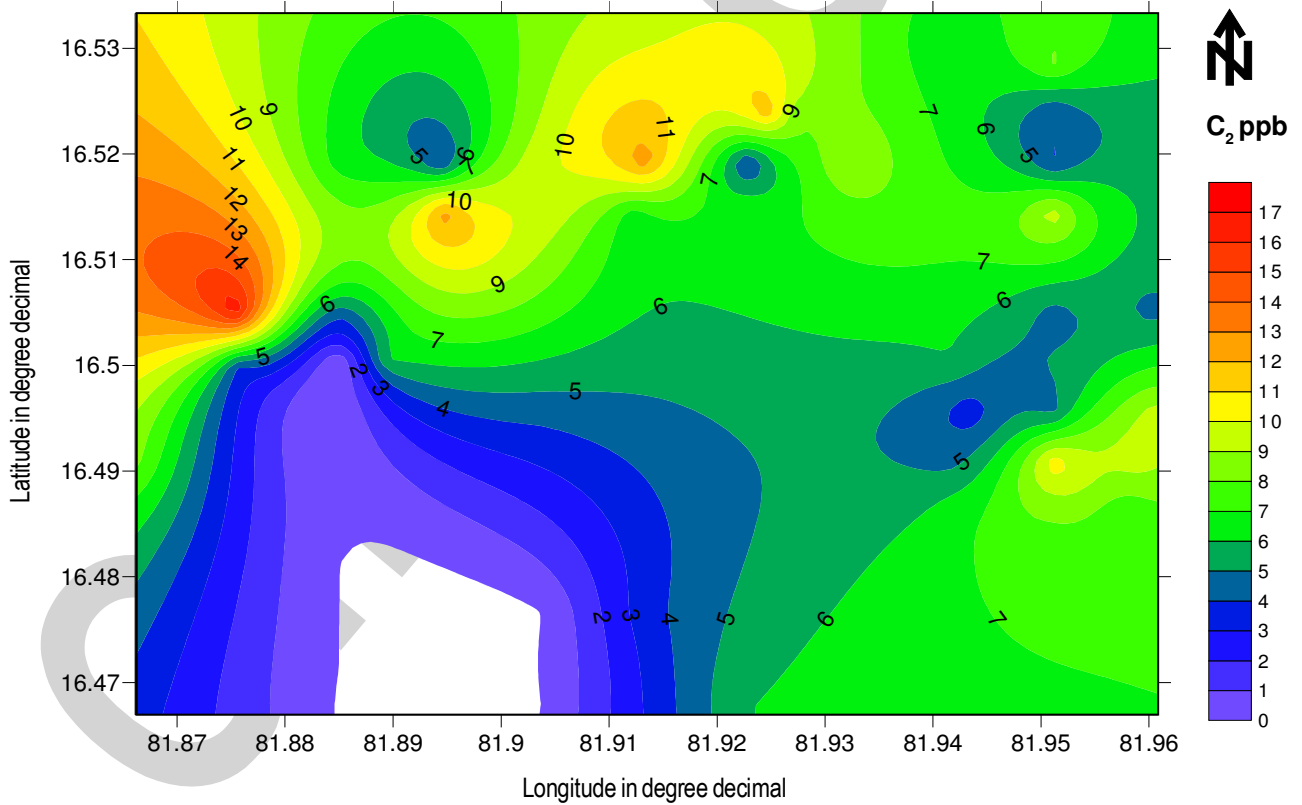


Fig. 6: Adsorbed soil ethane concentration map in the study area



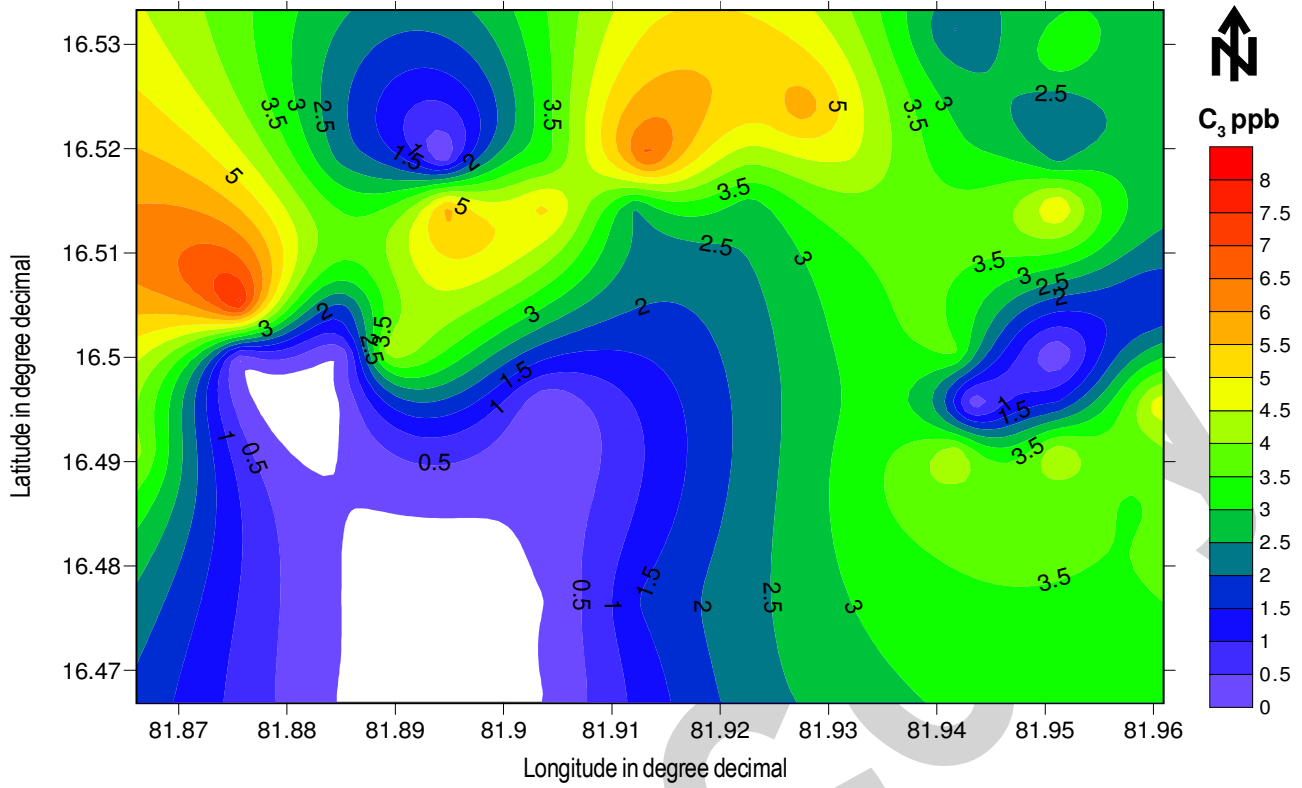


Fig. 7: Adsorbed soil propane concentration map in the study area

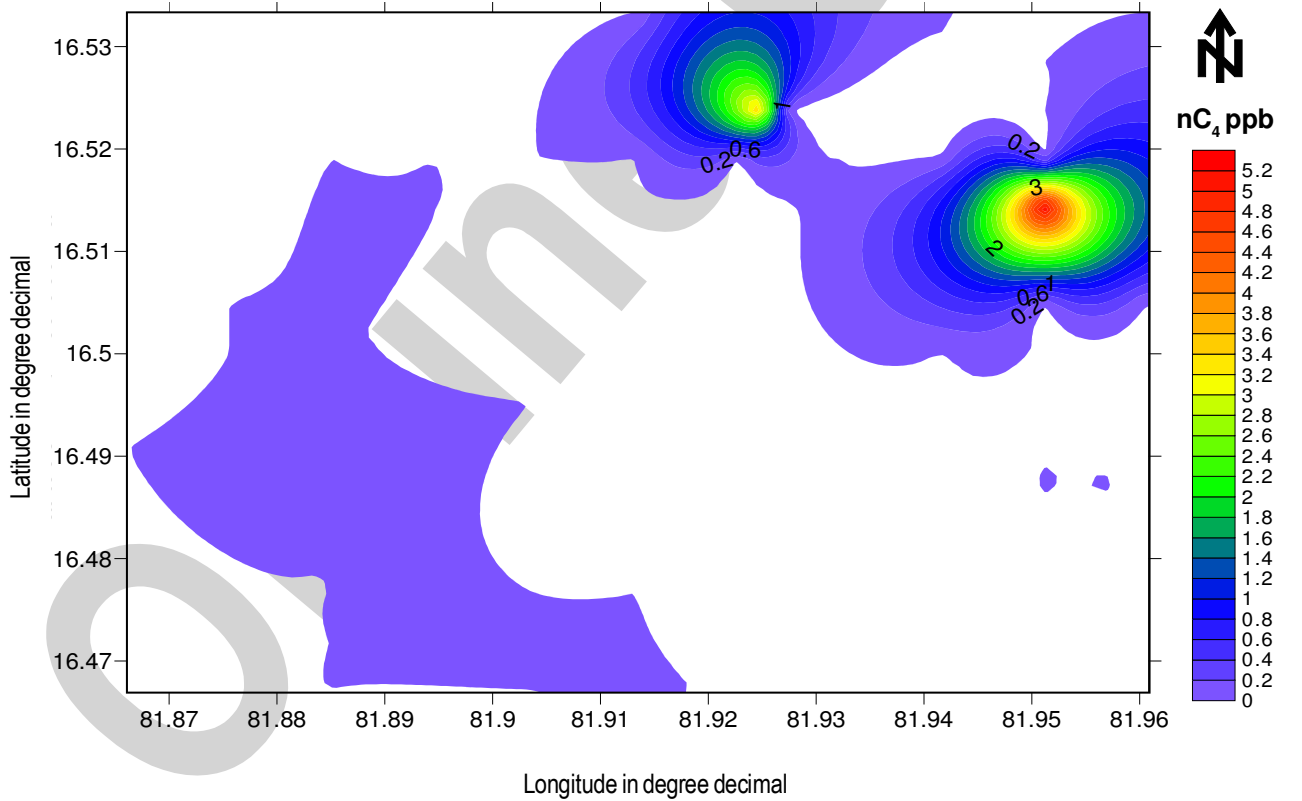


Fig. 8: Adsorbed soil butane concentration map in the study area

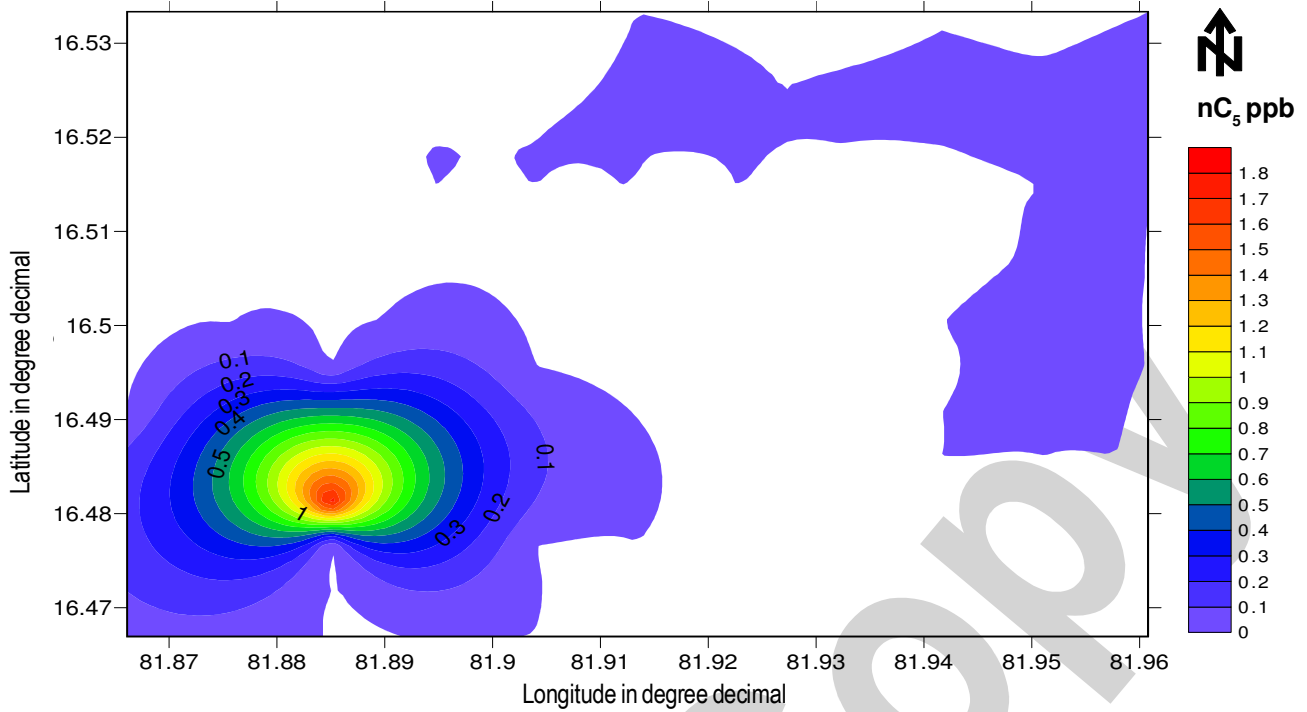


Fig. 9: Adsorbed soil pentane concentration map in the study area

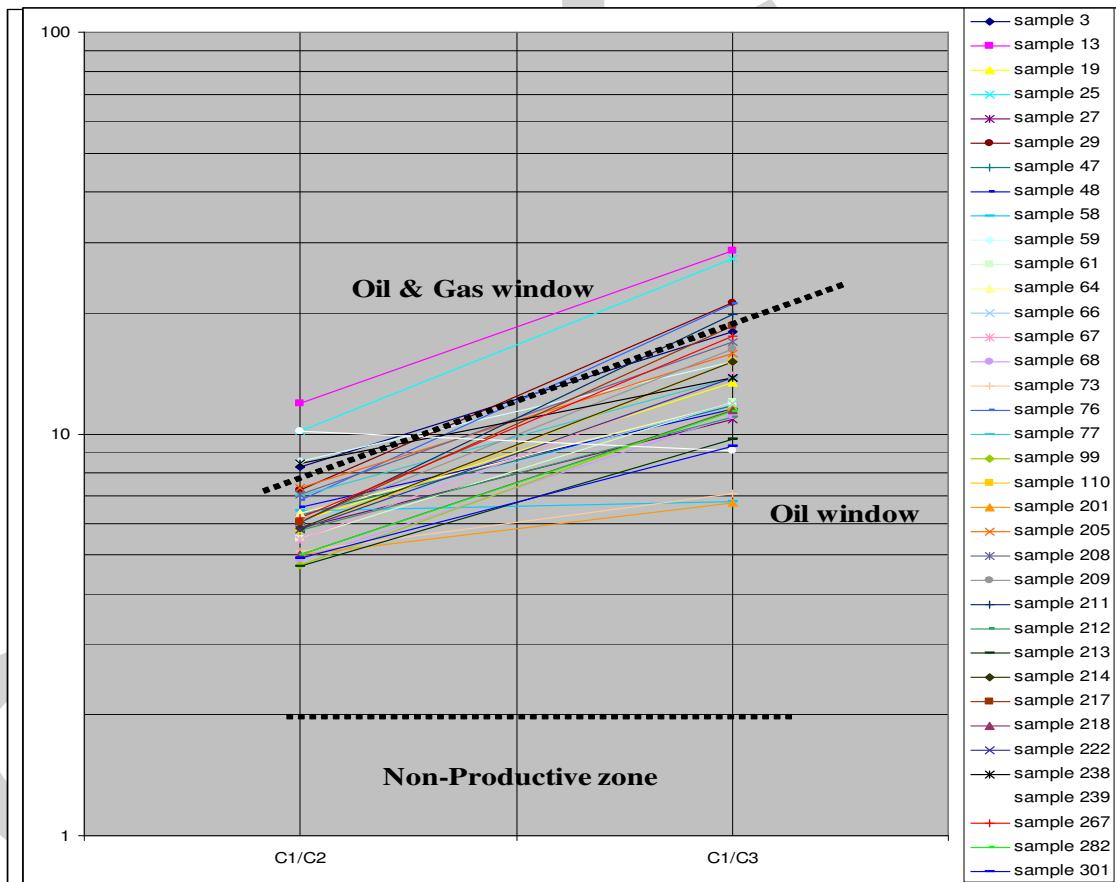


Fig. 10: Pixler's plot for discriminating oil, oil and gas and gas windows using  $C_1/C_2$  and  $C_1/C_3$  ratios

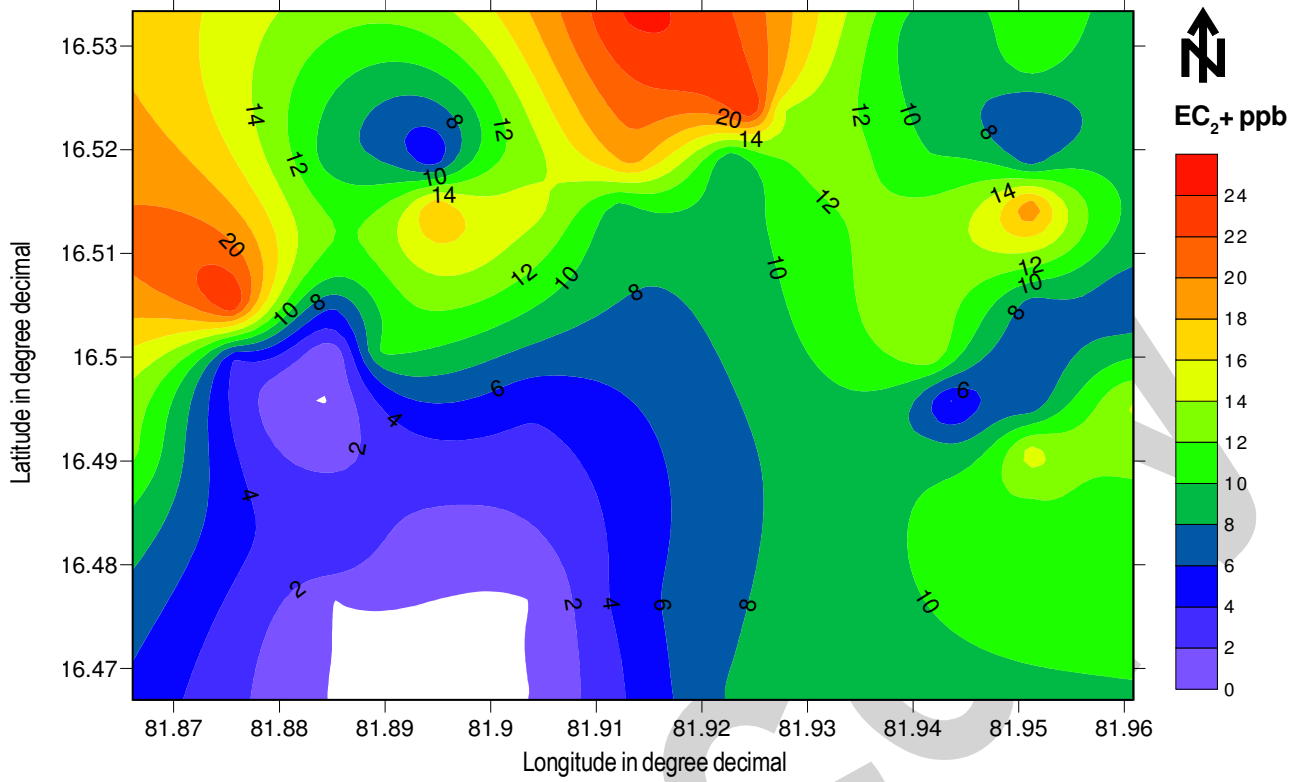


Fig. 11: Adsorbed soil  $\Sigma C_2+$  concentration map in the study area

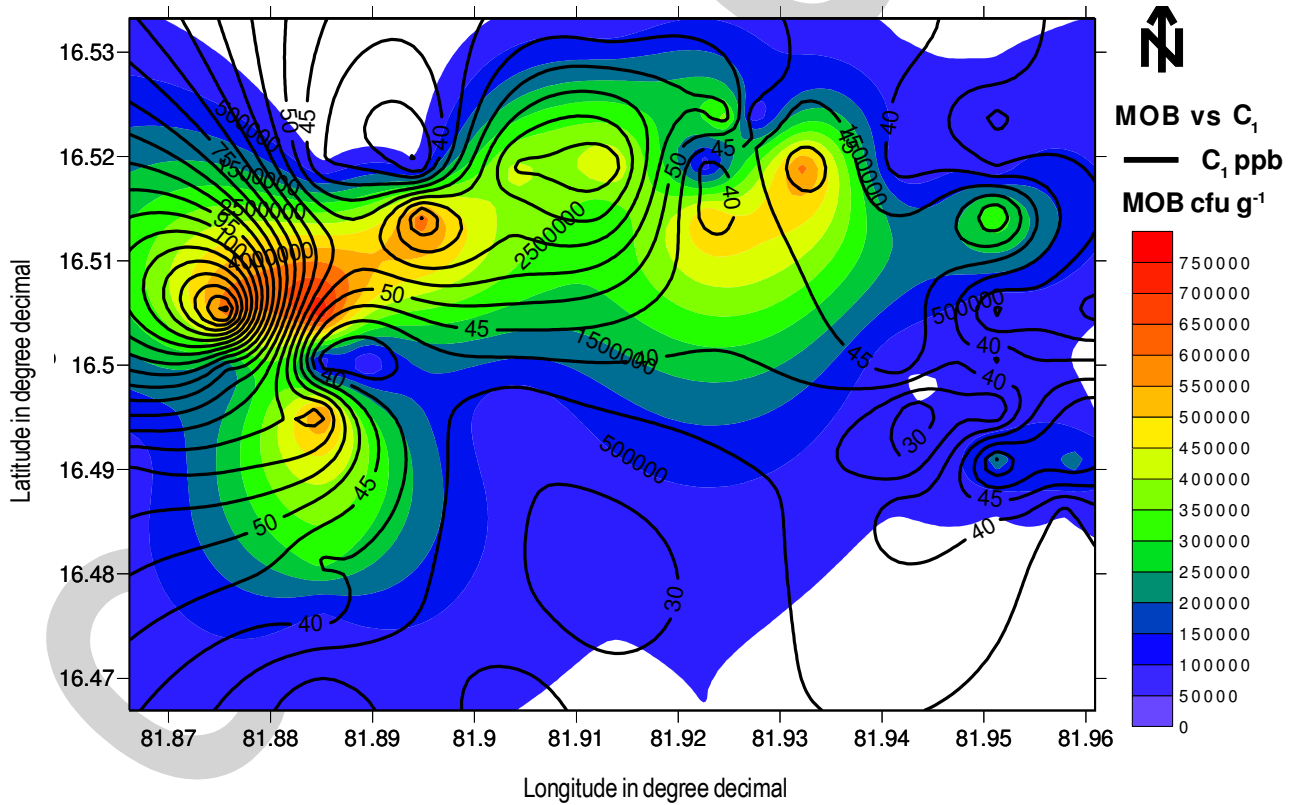


Fig. 12: Composite anomaly map of adsorbed soil gas methane and methane oxidizing bacteria in the study area

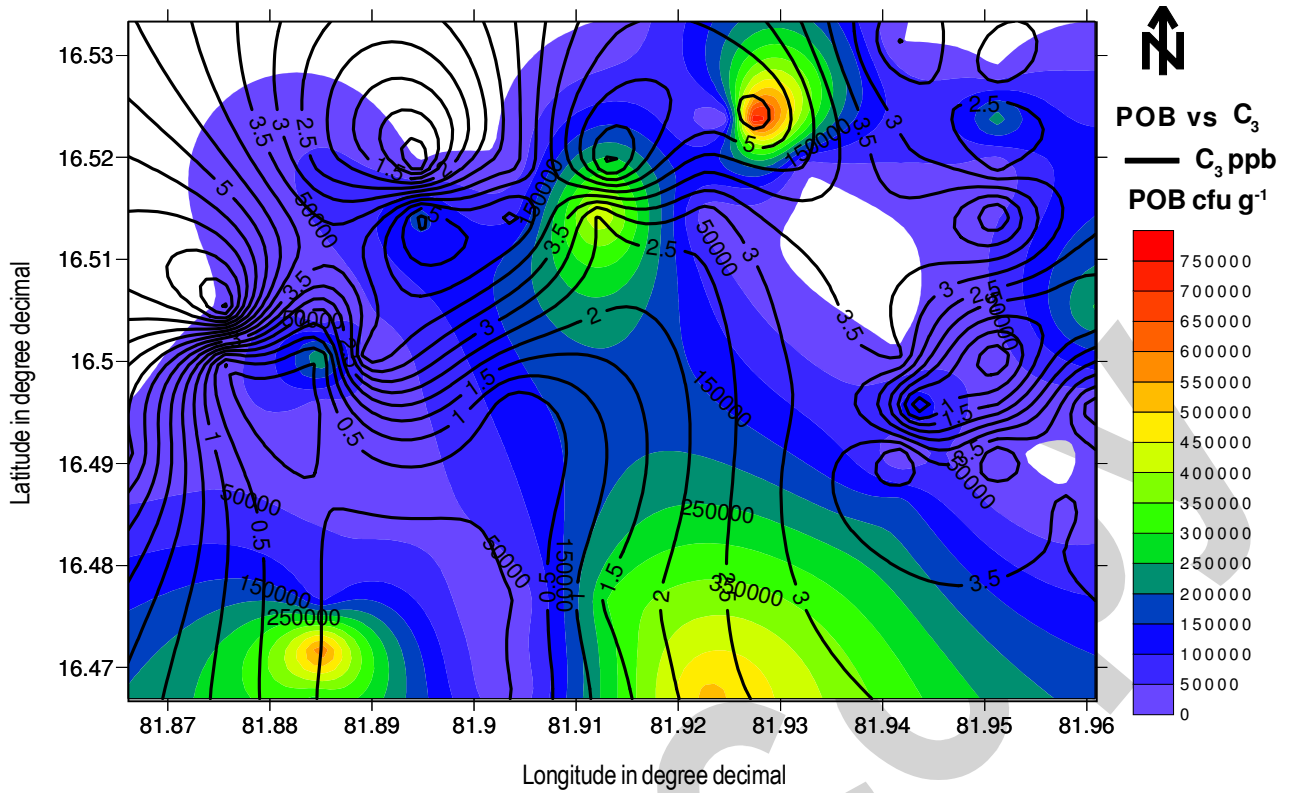


Fig. 13: Composite anomaly map of adsorbed soil gas propane and propane oxidizing bacteria in the study area

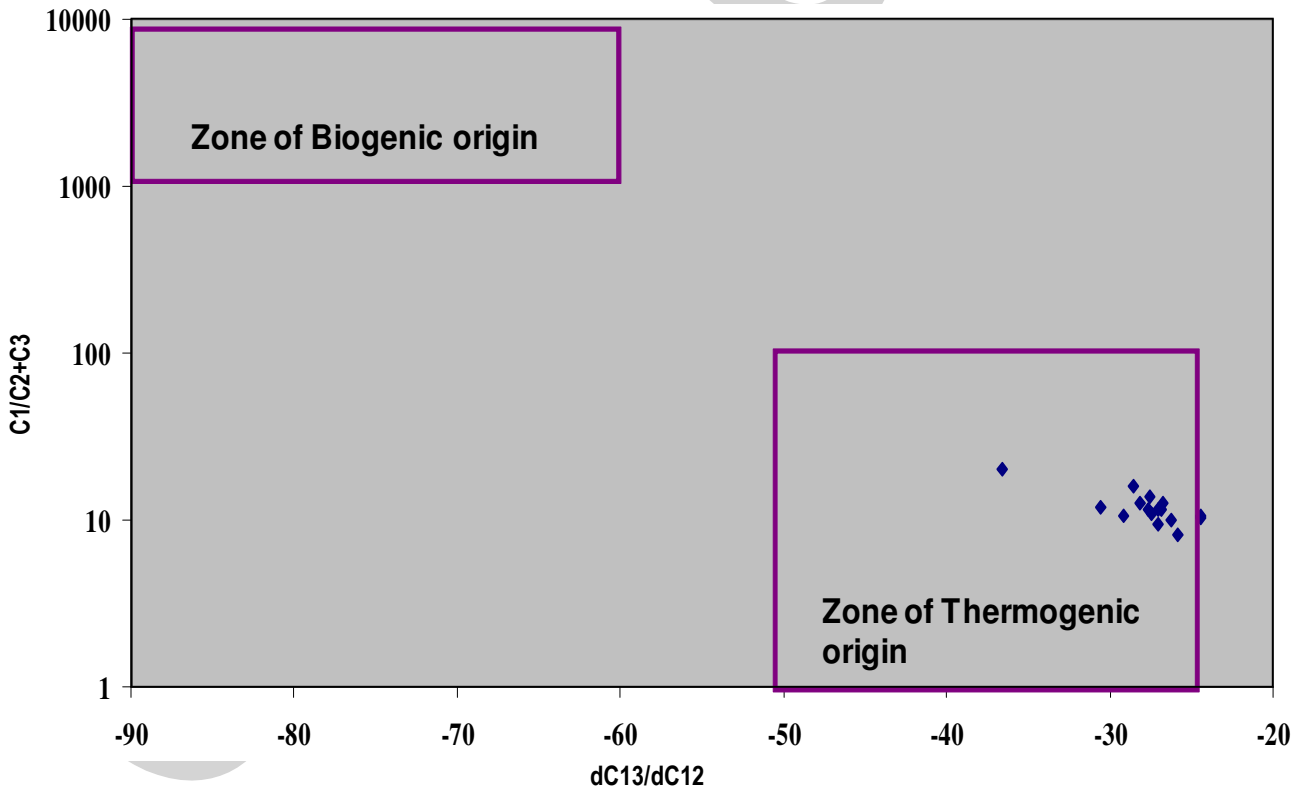


Fig. 14: Bernard plot indicating the possible source for the light gaseous hydrocarbons in the study area

**Table - 4:** Pearson correlation co-efficient matrix for adsorbed soil gas from parts of Krishna-Godavari basin

	C <sub>1</sub>	C <sub>2</sub>	C <sub>3</sub>	C <sub>4</sub>	C <sub>5</sub>	ΣC <sub>2+</sub>
C <sub>1</sub>	1.0					
C <sub>2</sub>	0.64	1.0				
C <sub>3</sub>	0.52	0.88	1.0			
C <sub>4</sub>	0.13	0.23	0.23	1.0		
C <sub>5</sub>	0.05	0.23	0.19	0.09	1.0	
ΣC <sub>2+</sub>	0.57	0.93	0.91	0.39	0.12	1.0

implies significant microbial anomaly above the hydrocarbon reservoir with lower values for adsorbed soil gases. Relatively higher concentrations of adsorbed gases were observed surrounding the microbial anomaly demonstrating the 'halo' effect (Rasheed *et al.*, 2008).

**Carbon isotopic study:** A genetic classification diagram can be drawn by correlating gas wetness *i.e.*, C<sub>1</sub>/(C<sub>2</sub>+C<sub>3</sub>) ratios with the δ<sup>13</sup>C of methane to classify natural gas types as biogenic or thermogenic (Madhavi *et al.*, 2009). Molecular ratios of C<sub>1</sub>/(C<sub>2</sub>+C<sub>3</sub>) less than -50‰ are typical for thermogenic hydrocarbon gases with δ<sup>13</sup>C<sub>1</sub> values between -25 and -50‰ (PDB) and ratios of C<sub>1</sub>/(C<sub>2</sub>+C<sub>3</sub>) above 1000 with δ<sup>13</sup>C<sub>1</sub> values between -60 and -85‰ (PDB) are indicative of biogenic origin of hydrocarbon gases. Biogenic gases thus have exclusively methane with very low concentrations of ethane and propane. Methane usually has a quite isotopically depleted C<sub>13</sub> ratio of less than -60‰. Thermogenic gases have greater gas wetness *i.e.*, more of ethane and propane with lesser methane. The carbon isotopic composition of δ<sup>13</sup>C<sub>1</sub> of the samples ranged between -36.6 to -22.7‰ (PDB). Hence, none of the isotopic values of the samples are characteristic of biogenic range. In fact, all the samples from the study area fall in the thermogenic range (Fig. 14) which presents convincing evidence that the gases collected from these sediments are of catagenetic origin. The microbiological results correlate with existing well data of the study area. The microbial anomalies were found to be in good agreement with geological information available for the area and correlated with the major oil and gas fields of Krishna-Godavari basin. Therefore, the microbiological study confirmed the seepage of lighter hydrocarbon accumulations from the subsurface.

The published experimental work or research employing methodologies, which have potential to reduce risk in petroleum exploration, especially now when globally petroleum reserves replacement is becoming a challenge is of high significance. The present work has an excellent significance as it is yet another example that shows that

geomicrobiological and adsorbed gas based geochemical methods give very complementary results.

### Acknowledgments

The authors are thankful to the Director, National Geophysical Research Institute, (CSIR) for granting permission to publish this work. The authors Ms. M. Lakshmi (SRF-CSIR), Dr. M.A. Rasheed (RA-CSIR) and T. Madhavi (SRF-CSIR) acknowledge CSIR for providing fellowship.

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