Original Res	earch Paper Volume - 14 Issue - 03 March - 2024 PRINT ISSN No. 2249 - 555X DOI : 10.36106/ijar
anal OS Applica Por contract of the second	Environmental Science COMPREHENDING THE ENVIRONMENTAL PRODUCTION AND ATTRIBUTES OF NITROUS OXIDES (N2O) IN THE NORTH-EASTERN MAHARASHTRIAN CLIMATIC CONTEXT
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The microbial ecological conditions that boost the emission of fluxes of green house gases are typically anoxic processes like denitrification, eutrophication, putrification and obviously aerobic processes like enhanced organic matter decomposition and nitrification. The temporal, causal, spatial and economic reasons for the existence of such conditions have been traced, studied, estimated and experimented to generate data, correlate it with the existing and paradigmatic scenario and find the solutions to it. The extensive interdisciplinary work related to classical meteorology, fundamental microbiology, grass root biochemistry, natural biodiversity, copitrophic ecosystem, healthy environment and durable sustainability related to climate change has been undertaken and analyzed. The initiation, generation, parameters affecting, the microbes responsible, the occurrence of the favourable conditions, the management practices to have solutions for minimization of the effect for the terrestrial microbial contribution of the atmospheric trace green house gases has been monitored. Each of the parameter was experimented by sophisticated and calibrated instrumentation, standardized techniques, preservation methods, data storage, statistics, supported by peer reviewed literature. Overall, the execution of this research would enhance the understanding of the mechanisms lying below the selected topic and find the regional solutions in more innovative ways for mitigation and adaptation to the ever-changing climate in more sustainable ways in favour of the society, region, nation and planet.

1. Scope of the proposed research work

North-eastern region of the Maharashtra state and central parts of India i.e. *Vidarbha* and *Khandesh* majorly covers the areas of *Wardha*, *Kanhan*, *Wainganga* and *Tapi-Purna* river basins, respectively and forest covers with the soils of alluvial to black cotton type that have poor to appropriate fertilization capacity (Phirke, 2014). The *Vidarbha* further constitutes of the area of *Varhad* and *Zadipatti*. The *Varhad* region also has the Sant Gadge Baba Amravati University's (SGBAU) jurisdiction prevailing over the part of this region constituting five districts including *Amravati* (a revenue divisional government head quarter), *Akola, Buldhana, Washim* and *Yavatmal*.

This region is traversed at the middle by the National Highway No. 6 (NH-6) starting from Dhule up to Gondia and offers the shelter, food, protection and geography to the human inhabitants that belongs to various ethnicities, races, communities, faiths and tribes and use and exploit the available natural resources for their livelihood. Among the non-human inhabitants, it includes several plant species including naturally grown vegetation, agriculture and forests; animal species including domestic livestock, wild and native genera and microbial communities that make these eco-systems sustainable through their contribution in biogeo-chemical cycling of all required vital elements. This has resulted in the multifaceted interactions of all these inhabitants with the biotic and abiotic factors of the regional environment. This region has participated successfully in national population increase as well as green and white revolutions and also, enjoyed its socio-economic benefits. Increase in human population per unit area per unit time has reduced the equal and justified use of natural resources specifically air, water and land for generating food and space for activities. This has forced several people to seek professional

alternatives. Many have tried to increase the availability of land by deforestation and removing the cover of vegetation for cultivation, construction and industry.

The greed towards the continuous power generation and consumption for increase in the overall standard of living and transportation; lead to the pollution through fuel consumption. Therefore, human activity was among the major biotic factor that makes the impact on the everchanging environment of the region that is contributed to the national and global levels. Among the notable feature of pollution is the steady rise in the gaseous inorganic carbon emission (CO₂) with the least ways for its recycling back into organic biomass.

On this background, this region is witnessing the change in its climate especially intolerable temperature rise since last two decades during different seasons like monsoon, winter and summer. This change is further affecting the environmental conditions like relative humidity, water availability, drought and heavy rains etc. causing alterations in the hydrological cycles, making the regions agro-based economy unequal, bias and unsustainable for all levels.

Cumulatively, this led to depletion of overall pleasant climate for maintaining the sustainable standards of living for the people of the region. Therefore, the study and execution of this research would be limited to the area under north-eastern Maharashtra.

2. Background and supporting review of literature to begin the proposed research work:

The following is the brief background of the proposed project and a review of literature from the exhaustive review taken by the investigator for the study and the actual execution of this project. Only selected information that was easily grasped and comprehended by the investigator and considered needful to emphasise the significance of this topic has been included herewith for the design of this project. Thus, it is merely representative and not the complete. Thus, simple and easily comprehensible issues are discussed for the perusal of the readers at Phirke, 2014.

b) What was pre-industrial era in north-east Maharashtra?

The careful observation of the meteorological parameters could be concluded with the following facts (Challa et al., 1995). Prior to the decade of 1960, climatic conditions in North-eastern region of the Maharashtra were moderately extreme, mesophillic winter being quite cold; temperatures dropping to 6+1°C and summers were pleasantly hot, temperatures rising to $40\pm2^{\circ}$ C. Rains were confined to monsoon with short to appropriate annual precipitation; varying place-wise appropriately from 4 to 781 mm. Most of the districts are under the assured rainfall area. Humidity was highest in the months of July and August and day temperature varied from 33 to 38°C. Dust storms were occurring occasionally during April to June, with severity in May (sometimes hell's stones). Soils were alluvial on the banks of the rivers, but black cotton soil was found in other parts of plains. Although these are the general human observations that match with appropriate analysis of the past instrumentally recorded data from each district's meteorological observatories.

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The green house effect and global warming:

According to Schifer and Unninayar, 1991, the 'greenhouse effect' is the sum of the interactions between the heat that is attempting to escape from earth to space and the molecules of various gases that trap this heat, reradiating it within the atmosphere, and impeding its loss to space. Without the greenhouse effect, the earth would be about 33°C cooler than it is for all practical purposes, uninhabitable. The problem our planet now faces is 'global warming', the potential augmentation of the greenhouse effect due to the build up of the gases that trap heat in the atmosphere. These gases are largely anthropogenic in origin, and are now at greater concentrations than at any time in the past 160,000 years (Milich, 1999). The major anthropogenic greenhouse gases are depicted in the following figure 1 (Boyle, 1990). Concentrations are estimated high and low ranges. According Milich, 1999 to the sources of global is a release of nitrous oxide (N2O), mainly via the application of fertilizer to arable lands through nitrification and denitrification; and the production of tropospheric ozone, the highly irritating constituent of smog. There are another 30 or so greenhouse gases that have been identified so far, but they do not add much atleast at the current situation. In this project our major interests are N₂O gases as majority of these yields from microbiological processes.

c) Implications of climate change at different spatial levels:

At global level: The Inter-Governmental Panel on Climate Change (IPCC, 2007a) in its recent report has affirmed that the global atmospheric concentrations of nitrous oxide have increased markedly due to human activities. Between 1000 and 1750 AD, nitrous oxide concentrations were 270 ppb. In 2005, this value has increased to 319 ppb. The global increases in concentration of nitrous oxides are primarily due to injudicious agricultural production practices, landfills with organic wastes and improper waste management. These increases in GHGs have resulted in warming of the climate system by 0.74°C over a century between 1906 and 2005. Eleven of the last twelve years (1995-2006) ranked amongst the 12 warmest years in the instrumental record of global surface temperature, since 1850. The rate of warming has been much higher in the recent decades and the night-time minimum temperature has been increasing at twice the rate of day-time maximum temperature. The quantity of rainfall and its distribution has also become more uncertain. In some places, climatic extremes such as droughts, floods, timing of rainfall and snowmelt have also increased. The sea level has risen by 10-20 cm with regional variations. Similarly, snow cover is also believed to be gradually decreasing.

At national level: A study of overall national meteorological records (Aggarwal, 2008) showed the incremental trends in the average temperature across our nation by 0.4°C during 1901-2000, whilst sea levels rose by 10-25cm, with a continuation and acceleration pattern during the current 21st century if precautions are not appropriately taken. Temperature has a direct impact on crop yields and the survival of life forms and biodiversity. Even 1°C rise in temperature reduces the wheat growing season by a week. Past studies suggested that a metre rise in sea level would displace over 7 million people. Three of the world's major Indian cities Mumbai, Kolkata and Chennai must contemplate this risk. Surat, a sprawling and rich business city is also on the western coast of *Tapi* estuary. Over 60 million people living in low-lying areas along the country's vast coastline may experience difficulties as aquifers become degraded by saline intrusion.

The indirect impact of rising temperatures on rainfall is more difficult to predict (Houghton *et al.*, 1996). High resolution climate models for India remain embryonic, a shortcoming of special relevance for a country whose climate ranges from the desert to the humid sub-tropical states of South India. The broad expectation is for fewer wet days, but more intense rainfall, implying a shorter monsoon with heightened risk of floods and drought. The future may have been glimpsed in the exceptional Bihar floods of 2008 and in the failure of the 2009 monsoon. The incidence of cyclones is also the subject of uncertainty, but the risk of increased intensity is regarded high. The bulge of population in coastal regions is such that 370 million people in India experience one or more cyclones each year.

At regional (north eastern Maharashtra) level:

Since 1970s, climatic conditions in North-eastern region of the Maharashtra have very abruptly changed. Weather is severely extreme with rise in temperatures almost by 5°C, experiencing hot winters and unbearable summers. The onset of Monsoon showers has postponed by one to two months turning the districts towards irregular rainfall area. Humid and hot rainy seasons and extremely dry hottest summers are blocking the regular traditional work routines absolutely intolerable. Summer provides the day temperatures upto 48°C. Cyclonic dust storms and hell's stones are now regular visitors. The grass, forest and wetlands have already disappeared or fighting hard to maintain their existence. Streams, fountains and rivers are flowing only in rainy season and for the rest of 8 to 10 months appear completely dry. These are the general human observations compounded with that instrumentally recorded by each district's meteorological observatories.

Much attention focuses on the observed retreat of springs originating from *Satpuda* and *Vindhya* Mountain ranges, the source region for India's few major rivers *Narmada, Tapi, Purna, Wardha, Kanhan* and *Wainganga* etc that use to be flowing throughout the year. As the science of spring drought develops, the working assumption is that the rivers will experience 2-3 decades of greater dry droughts risk, with diminishing summer flows in the longer term. Few million people live in the catchments of these rivers.

Terrestrial soils and soil microorganisms participating in global warming: According to Conrad, 1996, when characterizing green house gas metabolism in soil, the soil in the various atmospheric budgets of these trace gases usually assumes as a terrestrial site which is covered by some vegetation. These sites comprise (i) farmland, grassland, forest soils that are dominated by minerals and (ii) wetland, riparian soils which are often dominated by living and dead plant biomass e.g. Sphagnum bogs. These various productive soils usually also contain animals, which may produce trace gases on their own. From the viewpoint of a microbiologist, it is appropriate to assess the effects of plants and animals (mainly worms and arthropods) separately from those of microorganisms. Often the term 'microorganisms' is used as a proxy for members of the Bacteria, Archaea, fungi, and protozoa. However, from a practical point of view, the soil microbiota is best defined as including all organisms that are too small to be separated from the soil matrix by mechanic tools (e.g., sucking pipettes, forceps and sieves). Thus, soil microorganisms in a practical sense would comprise bacteria (Bacteria and Archaea), fungi, algae, and protozoa but also the micro- and mesofauna smaller than about 1 mm, e.g., rotifers, nematodes, acari, and collembols.

Timing of N-applications to periods when plant uptake is highest should limit the accumulation of nitrate and ammonium in the soil, and thus also limit the substrate availability for the production of nitrous oxide. By so limiting the nitrogen turnover rates, the agro-ecosystem's capacity to consume methane will be maintained. Timing and frequency of irrigation can also minimize the production of nitrous oxide and maximize the consumption of methane. Large, less-frequent irrigations will tend to accomplish this goal.

d) Comprehending the production and attributes of nitrous oxides (N₂O):

Although nitrous oxide's atmospheric concentration is very low 310 ppby, but is increasing at a rate of about 0.25% per annum. In spite of its low concentration and less rapid rise, N₂O is important due to its longer lifetime of 150 years and greater global warming potential almost 300 times more than CO₂ (IPCC 2007a, GOI, 2010). Both fertilized and unfertilized soils contribute to the release of this laughing and anaesthetic gas. Estimates of total nitrous oxides released from Indian

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agriculture as a whole are low due to generally low native soil fertility of our soils and relatively lower amounts of fertilizer used compared to western countries. But, still there exist the zones and watersheds where cash crops and fruits are produced. These areas are routinely being chemically fertilized agriculture orchards and farmlands exist in north east Maharashtra that are turning hot spots even those are regularly irrigated, not chilling the surrounding as much as required. Studies must be conducted in such regions in India to precisely quantify the magnitude of N_2O emission apart from other different agroecosystems.

Soil N₂O production in upland and riparian soils with natural vegetation cover is influenced by various microbiological, chemical, and physical properties and processes in the soil. N₂O emissions from soils are produced predominantly by the microbial processes of nitrification and denitrification. Nitrification is the main source of N₂O under aerobic conditions, while denitrification dominates under anoxic conditions. Both processes can occur simultaneously in soils, and the production of N₂O depends on the balance between these two microbial processes (Butterbach-Bahl *et al.*, 2004; Conrad, 1996).

In addition to these microbiological processes, chemical processes in neutral and acidic soil pH contribute a small fraction of soil N₂O (Bremner, 1997). Also, the physical composition of the soil influence emissions, as increasing soil clay content increases emissions of N₂O (Chatskikh *et al.*, 2005). The majority of soils of north Maharashtra region are neutral and have high clay contents heavily enriched with nitrogen either through artificial chemical fertilization or natural leguminous biological nitrogen fixation, making suitable for N₂O augmentation to atmosphere.

Therefore, composition of microbial community, physical and chemical characteristics of soils all influences the rates of N₂O production and emission. Worldwide, upland soils exhibit a mix of these various attributes. Beside agricultural soils, tropical rainforest soils have been found to be the main source of atmospheric N₂O (Werner *et al.*, 2007). Nitrogen availability is high in these clay-rich, weathered tropical soils, and the high temperature, high moisture, and aggregation of clays create conditions where N₂O can be produced both by nitrification and denitrification (Neill *et al.*, 2005). The region under study is rich in tropical rain forests of wildlife sanctuaries of *Pench, Tadoba, Navegaon Bandh, Melghat, Gugamal, Yawal, Toranmal* and so. These are suspected to contribute N₂O emission and study must be undertaken to understand their contribution and control of GHGs to this region.

One method of identifying factors that influence N₂O emissions has been the development of process-based models, which simulate trace gas fluxes from soils. In addition, summaries of emission measurement data from field studies, using statistical techniques, have led to the development of emission factors such as those used by the IPCC. Soil organic C content, vegetation type, soil pH, bulk density and drainage were the major factors influencing N2O emissions. As soil organic content increases, emissions of N₂O increase as well due to increased availability of C for denitrifying bacteria (Kanerva et al., 2007; Stehfest and Bouwman, 2006). Vegetation type also influences emissions, as N2O emissions decrease with increasing plant species diversity and increase in the presence of legumes (Niklaus et al., 2006). Chemical and physical characteristics of soil also influence emissions of N2O. As soil pH increases, N2O emissions decrease. As soil bulk density decreases, so do N₂O emissions. Soil moisture also plays a role in N2O emissions, as Khalil and Baggs (2005) reported; N2O emissions were the highest from the wettest soils (at 75 percent water-filled pore spaces). In the same study, it was found that 90 percent of the N₂O was produced through denitrification proof that these water-filled soil micro sites were primarily anaerobic.

The various microbiological, chemical, and physical properties of soil that influence N_2O emissions are distributed throughout upland and riparian soils worldwide. However, some general trends for particular biomes (ecological communities in particular climates) appear. Specifically, emissions of N_2O from rainforests are significantly higher than from grasslands, savannah, and tropical dry forest and emissions from grasslands are significantly lower than those from deciduous forests and rainforests (Stehfest and Bouwman, 2006). High nitrogen availability, coupled with high moisture content, makes tropical soils especially likely to emit N_2O .



Production of NO and N,O: The fluxes of NO and N,O between the soil and the atmosphere result from complex and little-understood interactions of different processes in soil that occur simultaneously. Since processes involved in production (e.g., nitrification) often differ from those involved in consumption (e.g., denitrification), production and consumption are regulated differently (Aleem, 1977, Aleem and Alexander, 1958). The following figure 3 shows the simple nitrogen cycle and oxidation states of different nitrogen compounds. (X) and (Y) indicate intermediates in the two processes denitrification and nitrification [Source: Codispoti et al., 2001] Furthermore, NO production and N₂O production are a composite of different processes, such as nitrification and denitrification that vary in relative significance depending on soil conditions. Even one particular process, e.g., denitrification, can be a composite of different bacterial species that may be phylogenetically distant from each other and express different types of enzymes. For example, the NO-producing nitrite reductase can be either a copper-type or a cytochrome cd-type enzyme. Both NO production and N2O production in denitrifiers are a function of the relative activity of the producing and the consuming enzymes, i.e., nitrite reductase and NO reductase for NO production and NO reductase and N2O reductase for N2O production.

The actual turnover of nitrogen by these enzymes depends on substrate concentrations and the kinetic properties of the enzymes. The specific activities of the enzymes depend on biosynthesis (transcription and translation) and inactivation. The available information on the regulation of enzyme synthesis suggests that O₂ partial pressure and concentrations of nitrogen substrates are major regulators. More importantly, however, the patterns of regulation seem to be different in different species of denitrifiers.

The expression of denitrifying enzymes shows different O_2 sensitivities for the different bacterial strains. There are also bacteria that denitrify under aerobic conditions. The different effects of O_2 on the expression of the various bacterial nitrite reductases, NO reductases, and N₂O reductases will affect whether NO or N₂O is the dominant product when the O_2 availability in soil changes. Therefore, knowledge of the composition of the community of denitrifiers in soil is important for understanding of the production of NO and N₂O.

Unfortunately, there are only a few studies of expression of denitrifying enzymes in soil after the onset of anaerobiosis. The results indicate that synthesis of nitrate reductase and nitrite reductase starts within hours whereas synthesis of nitrous oxide reductase is delayed for more than 1 day, so that N_2O is produced. None of these studies involved the investigation of the composition the microbial community or looked for the metabolism of NO. Therefore, it remains unclear whether the pattern of NO and N_2O production are affected by the composition of the denitrifier community in natural soil. Community effects are not unlikely, since soil which is amended with different denitrifiers exhibits completely different patterns of production of NO and N_2O .

There are reports that the soil microbial communities which reduce nitrate differ as a function of temperature and pH. There are also numerous reports that the production and release of NO and N_3O from soil into the atmosphere are affected by soil temperature and pH. However, the available information does not allow a synthesis of the observed patterns of release or production of NO and N_2O with patterns of microbial community composition. These examples provide a

narrow focus on denitrification and on three of the most conspicuous soil variables, i.e., aeration (soil water), temperature, and pH.

It is much more complex and involves a hierarchy of environmental controls that affect denitrification rates on the one hand and the fraction of NO and N_2O produced on the other hand. Furthermore, the real situation in soil probably involves many more types of microbial metabolism, such as autotrophic and heterotrophic nitrification, nitrate respiration, and dissimilatory nitrate reduction to ammonium (DNRA), which produces and consumes NO and N_2O . This complexity will be understood only when more will be known about the microorganisms involved and their metabolism through this project.

g) Grasping the microbial control over the ecosystem:

Trace gases are produced and consumed by defined reactions in individual microorganisms. Any control of the production or consumption process is exerted at this level initially. However, it is also relevant to ask whether the microbial diversity and community structure in soil have any consequences at the ecosystem level.

Although a microbiologist may tend to answer this question in the affirmative way, the true answer is by no means clear. In fact, much of the spatial and temporal variability of trace gas fluxes between the soil and the atmosphere can often be explained by only a limited number of factors, including soil and parent material, climate, vegetation, and topography. Thus, empirical models that are based on correlation analysis involving easily measurable soil variables (temperature, moisture, texture, and organic carbon) often predict trace gas fluxes quite well. Statistical techniques, such as multiple regression or multivariate data analysis, help to assess the optimum set of variables to be measured. For example, water table position, temperature, and degree of peat humification are expected to explain 91% of the variance in the logs of CH4 flux from a peatland. Weaker but still significant correlations of CH₄ flux to a few predictors have also been reported for other sites. Emission rates of N₂O from some soil environments (grassland ecosystems) have also been predicted amazingly well from a few soil variables such as temperature, moisture, and inorganic nitrogen. The strong correlations of CH4 emission with water table height and of N2O emission to inorganic nitrogen compounds are easily understood, since microbial methanogenesis requires anoxia and microbial N2O production requires either ammonium or nitrate as a substrate. Although these empirical models based on various physical and chemical parameters are successful without considering the structure of the microbial community, knowing the exact microbial community causing production and consumption of GHGs is very much significant.

Even largely mechanistic models of NO and N₂O production that differentiate between nitrification and denitrification neglect microbial community structure. In some studies, it was found that trace gas fluxes were controlled largely by a single ecosystem function that created a bottleneck in substrate flow. For example, rates of N₂O emission from tropical soils were strongly correlated with rates of net nitrogen mineralization which limited nitrification and denitrification and thus, also N₂O production.

On the other hand, although NO release should be limited by nitrogen mineralization in the same way as N₂O release is limited, the release rates of these gases have often been found to be anticorrelated because of the different reactions of the gases to changing soil moisture. Annual fluxes of N₂O were correlated with the annual aboveground net primary production. Similarly, rates of CH₄ emissions from wetlands correlated well with the net ecosystem productivity, i.e., the amount of the net primary production that is left in the ecosystem and is not immediately liberated by microbial decomposition.

Relevant trace gas flux measurements on the ecosystem or landscape level are still scarce. In addition, the classification of ecosystems is not satisfactory, and the possible mosaic structure of an ecosystem is also subject of debate. The microbial community structure in the soil of the various ecosystems and ecosystem mosaics is largely unknown. Therefore, it cannot be assessed safely at present whether the soil community structure is important for the trace gas flux.

In addition, we do not know the scale in space and time on which it is possibly important. Therefore, it remains unclear whether trace gas flux at the ecosystem level is finally controlled by a few processes only or whether changes in the diversity and composition of microbial species are also important. However, by analogy to ecological studies of plant and animal communities, microbial species diversity is an equally important factor in ecosystem function.

3. Objectives of the proposed project:

On this background of worries of the local people, weather and environment, exhaustive review and design of the strategic work plan and with the aim to study and understand the terrestrial microbial share in atmospheric heating through emission of GHGs, the following are the major objectives derived for this proposed project to be sanctioned.

(I) Location of the natural and anthropogenic conditions for GHG emissions:

Identify, locate and study the naturally or artificially occurring aerobic and anoxic conditions in the subtropical, terrestrial, domestic, agricultural and forest, dry and wet land, rainfed and irrigated, flooded, industry and social situations and sources, upland and riparian soil ecosystems of the north-eastern Maharashtra for the share of various soil microbial populations towards the contribution to the greenhouse gases especially nitrous oxide (N,O).

(ii) Duplication of the conditions:

Reproduce nitrous oxide generating conditions *in vivo*, *in vitro* and *in situ* at laboratory or pilot scale for its detailed study and possibly understanding mechanisms lying underneath.

(iii) Study of N_2O production:

Isolate, characterise and enumerate the N_2O producing microbes and their biomass from representative soils from the selected high activity locations and determine their ability of N_2O production efficiency to calculate the annual N_2O production per hectare and overall, its tentative share in the atmosphere and the environment.

(iv) Quantification of emission of significant GHGs:

Estimate the NO and N₂O fluxes from different sources from the above-mentioned ecosystems due to microbial reasons and generate the data for statistical analysis along with the collection of classical meteorological temporal and spatial reading.

(v) Co-relation and regression:

Co-relate the emission of these green house gases with that of CO_2 and establish the cumulative effect with the change in climate, especially rise and drop in temperature during various seasons and irregularly occurring hydrological cycle.

(vi) Comprehension and path forward:

Look for the necessary methods for sustainable budgeting of the gases under study and suggest the mechanisms, ways and methods for its control. Also, look for the possible and available ways of the adaptation to the existing scenario.

Such are the major objectives that are imperative for the execution this project. These major elects would result into several minor objectives that are not listed herewith, but would be important to increase our scientific understanding and the exact nature of the interactions between microbes, eco-system, GHGs, global warming, vulnerability of life, their adaptation, mitigation and survival.

4. Tentatively visualised materials and methodology:

The materials will be used of fine quality, AR/LR grade reagents, distilled water, demineralised (DM) water, sophisticated and microprocessor-based instruments to reduce manual error, standard laboratory glassware, plastic ware and consumables for accurate experimentation, calibrated instruments for maintaining recommended aseptic techniques and preservation methods for microbial isolates, biochemicals for biological characterisation etc. The following methodology (Burlage *et al.*, 1998) will be undertaken to successfully complete this research project and achieve the objectives to meet the aimed goal.

 Sample collection and its systematic analysis: (i) Gas sample: The standard operating procedure (SOP) outlined by U.S.E.P.A. will be used. (ii) Soil sampling will be done as per Tandon, 1992, Sonar et al., 1984, Somwanshi et al., 1997. (iii) Water sampling will be done as per Somwanshi et al., 1997. All the samples collected will be preserved well, analysed and results will be recorded. Apart from this, the ecosystems selected for the study will be monitored on site for different ecological parameters related to the topic and

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its varying profiles will be maintained (Burlage et al., 1998).

- Isolation, characterization and analysis of microorganisms involved in nitrous oxide production will be undertaken by recommended standard microbiological techniques as per Bergey's Manual of Determinative Bacteriology (Holt *et al.*, 1994, Jayraman, 1996, Dubey, 2005, Paerl, 1998). The cultures will be purified, preserved and deposited at culture collections for subsequent use in technology and process development.
- 3. Determination of microbial efficiencies for their GHG contribution will be done according to the suggested procedures given in Burlage *et al.*, 1998. The data will be recorded and analysed.
- 4. In situ analysis of GHG's emission for N₂O. The emission of CO₂, CO, H₂ and NO will also be undertaken to generate subsidiary data for finding the correlation and cumulative effect upon atmospheric heating. This will be done on sites and the data will be recorded.
- Statistical analysis will be done for each parameter, experiment and findings using computing techniques and statistical software etc.
- 6. Interpretation and conclusions will be done finally to make the objectives of this project successful and within reach.

5. Accountable deliverables of this project

The following are the accountable deliverables of this project:

- 1. Atmospheric monitoring for the changes in GHG concentration in air
- 2. Microbial biodiversity that is contributing to GHG emission
- 3. Mechanisms for the interactions between microbes and weather
- 4. Path forward for the no-regret adaptation strategies
- 5. Development of simple, judicious and sustainable technologies
- 6. Modern and unexplored avenues for employment

6. Social relevance of the project:

This project has identified the need for sustainable development of agriculture, trade and rural industrialisation without over and injudicious exploitation of natural resources in the region.

These adoptions can be at the level of individual farmer or entrepreneur, farm or entrepreneurship, society or organisation, village or town or watershed or even at a regional, state and national level (Phirke *et al.*, 2009). Overall, regional interests, livelihoods and standard of living of all sections of the society must be improved and life's vulnerability due to climatic variations has to be nullified.

7. National relevance of the current project:

The study undertaken in this project will be helpful, nationally (IPCC, 2007b, Phirke *et al.*, 2003 and 2008) in the following aspects.

- 1. Augmentation of agriculture with sustainable production practices
- 2. Development of the watershed management programme.
- 3. Promoting conservation agriculture (Phirke *et al.*, 2003, 2005 and 2010).
- 4. Developing agricultural and rural enterprises
- 5. Improved risk management through an early warning system
- 6. Recycling waste water and solid wastes in agriculture
- 7. Harvest and post-harvest management for minimising the losses due to extreme climatic events or mean climate change conditions.

Adaptation strategies can assist in providing some relief in future provided these strategies could be operationalised in field.

8. Global relevance of this project:

The study undertaken in this project will be helpful, rationally (IPCC, 2007b, Phirke *et al.*, 2003 and 2008) in the following aspects.

9. Strength of the project:

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The narrated project began with careful observations, it's linking to the education and knowledge, and ideas put forth, supported by review of literature, evidenced by vigorous experimentation in available laboratory infrastructure, perpetuated by keen supervision, resulted into aided demonstration trials and extension activities. This has a potential to cumulatively improve the agronomy, environment and living standards of *Khandesh*, *Varhad* and *Zadipatti* region farmers, traders and entrepreneurs with the above stated advantages. This was a real strength of this project, initiated by socially relevant research.

10. Facilities provided/ to be made available at the host institute: The following are the facilities provided by the Department for this project.

Sr.	Equipment	Qty	Status	Remark
No.				
1.	UV-Visible	1	Working	Available for use
	Spectrophotometer			
2.	Olympus biological	1	Working	Available for use
	microscope			
3.	Laminar Air Flow	3	Working	Available for use
	Bench			
4.	Vertical Autoclaves	4	Working	Available for use
	Small			
5.	Semi-automatic baby	1	Working	Available for use
	fermentor (2L)			
6.	Thermal Cycler	1	Working	Available for use
7.	Refrigerators	6	Working	Unavailable due to
			_	rush
8.	Rotary shaker	2	Working	Available for use
9.	Deep freeze	1	Working	Available for use
10.	Incubators	3	Working	Available for use
11.	B.O.D. Incubators	2	Working	Unavailable due to
				rush
12.	Flame photometer	1	Working	Available for use

Other Infrastructural facilities: Laboratory space, furniture, water, power and gas supplies, botanical garden, university and personal library, FAB lab, university campus location surrounded by forests and agricultural fields.

The facilities to be made available at the Department through this project are as following for the use of interested students, faculty and experts.

- Sophisticated and in depth microscopic, micro-metric and biochemical examination of methanogenic, nitrifying and denitrifying bacteria, fungi and actinomycetes for developing taxonomic expertise among investigators, faculty members and students.
- Facility to ferment and further process the various sources of organic matter to methane and nitrous oxide.
- Characterization of the fermentative microbes and its culture collection for its subsequent use in experimentation or technology and product development.
- Gaseous analytical facilities for the estimation of GHGs under study.
- Facility to monitor the classical meteorological parameters and correlate it with the surrounding changes in the environment.
- The academic and industrial expertise and temperament in the area of biomethanation, climate change among students, faculty and society.

11. Present work engaged in:

The professional work culture adopted at the laboratory by PI is of six working days-a-week, having Saturday as a half day and Sunday as a weekly holiday. As a discipline, PI maintains the minimum of eight hour working a day that regularly extended generally to ten hours. The general distribution of the work hours for imparting education, conducting research and offering extension/ other works is in the ratio of 40:40:20. The PI engages his regular teaching schedule prescribed by the administration as per the norms of UGC, New Delhi at postgraduate level in Microbiology in the sustainable areas of soil, food and environment. He is currently engaged in the following type of research work with the mutual participation of the P.G. students, research scholars, faculty members and few of the national level subject experts.

- 1. Soil nitrification studies in the region for the minimization of nitrate losses from commercial fertilization, nitrate pollution, eutrophication and emission of N_2O to the atmosphere. This project is likely to be funded by UGC, New Delhi.
- 2. Searching the ways to improve and preserve the food quality of preferred millets like *sorghum, kodo, kuttaki* through the use of microbes and microbial processes like fermentation for feeding the poverty, famine, nutrient and culture deficiency stricken our aboriginal tribes of *Melghat* of *Satpuda* mountain ranges like *Bhilla, Korku, Thakar, Paradhi, Gond, Madia* and *Pawara,* falling under the jurisdiction of this university and the region described herewith, during the ever-changing climatic and environmental conditions. This project is recommended for funding by SERB, DST, New Delhi.
- 3. Understanding the mechanisms underlying below the millet grain fermentation, nutrient especially nitrogen and phosphorus cycling

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and atmospheric change due to microbial reasons to troubleshoot the problems to be faced in near or distant future. In house minor projects without external funding are in operation through the projects of post-graduate students.

12. Future plans:

The following future plans become imperative after the project funding and successful execution.

- Creation of awareness among the students, faculty, society and 1. policy makers.
- Creation of literature on the said project like books, educational 2 aids, research articles.
- Development of cost-effective, sustainable and eco-friendly 3. technologies to mitigate the climate change in the field of agriculture, staple food and environment.
- 4 Open a public campaign for reducing the emission of green house gases, pollution and its hazards for creating and maintaining a sustainable environment and ecosystem.
- 5. Develop a cluster for promoting cumulative rural industrialization in the region.

13. Prospects after the fellowship:

The current research project and topic have the following major and genuine prospects after the funding is awarded.

- Change in the understanding of the people about the climate change, its importance, severity and the irreparable damages caused by it.
- 2. Educate and create adequate human resource through incorporation of the climate related issues in the syllabi of the educational courses of regional state universities, colleges and schools
- Evolve the region and society specific climate change mitigation 3. and adaptation strategies.
- 4 Make an impact on the socio-political and economic scenario by improving livelihood through achieving overall sustainability.
- Provide modern inputs for inclusion or improvement in our 5 National Action Plan on Climate Change (NAPCC), Government of India.

14. How are these related to the strengthening of basic research in the country?

In spite of the enormous efforts made by the various ministries including that of Science and Technology of the Government of India through design of appropriate policies and provision of research funds, the fundamental research in our country related to the climate change has remained limited to the national institutes mainly those of ICAR, CSIR and IITs, TIFR, IISc and NGOs like TERI etc. as it is evident from the literature these organisations has published and the state universities found no role in such type of extensively important and down to earth type of research. Therefore, the data, literature, knowledge, technologies generated through this project would be really conducted at the regional level and in the area, where the implications of weather changes have been witnessed, experienced and mitigated by the common people, citizens of India for their survival in this region over a period of extremely harsh conditions like water scarcity and drought, hot and dry summers with scorching radiations, famine, heavy rains, floods and hell's stones, cyclones etc. This shall result into sound, in-depth, grass root and people's participatory data related to the topic that would help the investigators to find region-specific sustainable solutions for the adaptation to the ever-changing climate in this region.

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18. Minimal references that supported this research work plan of the proposed project:

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