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SIGNIFICANCE OF SOIL PROPERTIES AND MICROBIAL ACTIVITY ON SOIL CO_2 EMISSION IN COASTAL SAND DUNES OF ODISHA, INDIA

T. PANDA

Department of Botany, Chandbali College, Chandbali, Bhadrak- 756133, Odisha, India

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Seasonal changes in soil respiration, soil temperature, soil moisture, soil organic carbon and microbial activity are compared between a barren sand dune without vegetation and sand dune with monoculture plantation of *Casuarina equisetifolia* (L.) of a coastal ecosystem. The result shows consistent patterns of change in CO_2 efflux rate and soil biotic and abiotic properties between barren sand dune and sand dune with monoculture plantation of *Casuarina equisetifolia* (L.). The abundance of fungi (*Absidia glauca, Aspergillus awamori, Cladosporium cladosporioides, Curvularia eragrostidis, Drechslera australiensis, Fusarium oxysporum, Paecilomyces varioti, Penicillim citrinum, Rhizopus nigricans, Trichoderma viride*) and bacteria increased significantly with plantation than the barren sand dune, and these differences in microbial community composition are strongly correlated with greater soil moisture and soil C content. The variation of soil CO_2 efflux rates is observed with soil properties and microbial activity and it is consistently higher in sand dune with vegetation than in barren sand dunes. Linear regression analysis revealed that soil respiration responses to changing biotic and abiotic factors are best predicted by soil moisture, soil organic carbon and microbial community composition in coastal ecosystem of Odisha. The result shows a significant and negative correlation between soil temperature and soil respiration. Additionally, no correlation is observed between soil moisture and soil temperature in both the sites. In contrast to the case for many other ecosystems, the relation of soil moisture and soil temperature failed to explain variations in respiration among the studied sites. The present study suggests that vegetation cover may alter the soil C balance in this ecosystem.

Keywords: *Casuarina equisetifolia*, soil moisture, soil respiration, soil temperature, fungi

Coastal sand dunes are common in different parts of the world, covering 6×10^6 km^2 of its land surface (Tropek *et al.* 2013). In contrast to the relatively stable nature of soil, sand dune ecosystems are dynamic and are mostly in a state of successional change (Jones *et al*. 2008). Additionally, coastal dunes are characterized by the impact of the marine ecosystem, including tidal rhythms, climatic conditions and sedimentary deposition (McLachlan and Brown 2006). Thus typically a distinct gradient across a coastal dune-field is formed from the shore to the inland, with the typical sandy character as a common property. These natural structures protect the coastal environment by absorbing energy from wind, tide and wave action. The hostile sand dune environment limits the plant growth and distribution (Rajaniemi and Allison 2009). Due to high plasticity, microorganisms are very successful inhabitants of coastal soil, and adopt various forms in response to adverse or unfavorable conditions. The microbes are known to contribute significantly to the stabilization of sand dune and development of plant community structure(Koske *et al.* 1996) through the process of organic matter decomposition and play a key role in regulating the balance of carbon and nutrients (Zifcakova *et al.* 2016).

Soil contains the largest pool of terrestrial organic carbon in the biosphere, storing more carbon than plants and atmosphere combined (Xu *et al.* 2014), with 73% of soil C contained in soil organic matter (Eswaran et al. 1993). Carbon dioxide (CO_2) is released from soils in the process referred to as soil respiration, soil-CO_2 evolution or soil CO_2 efflux. It is a major way in which C fixed by terrestrial ecosystems is released to the atmosphere (Barba *et al.* 2018). The amount of carbon released by soil respiration accounts 50-95% of the total ecosystem respiration (Xu *et al.* 2001; Wang *et al.* 2004), of which a substantial contribution comes from tropical ecosystem (Raich *et al.* 2002). It mainly comprises two carbon flux components: an autotrophic component, originating from the respiratory activity of roots and the associated rhizosphere, and a heterotrophic component,

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arising from microbe-associated soil organic matter decomposition (Chen *et al.* 2017). Soil CO_2 efflux differs among ecosystems, even among two neighboring forest types (Saviozzi *et al.* 2001). Therefore, diverse ecosystems should be taken into account in measuring the soil CO_2 flux.

Furthermore, several reports have shown that multiple soil abiotic and biotic factors such as soil temperature, soil moisture, soil oxygen, soil nutrient supply quantity, soil texture, soil pH, vegetation type, substrate quantity, availability and supply, soil microbial activity, concentration and quality of litter in decomposition, differentially affect the respiration process, making the interpretation of soil respiration complex (Bauhaus et al. 1998, Raich *et al.* 2002, Chen *et al.* 2017, Tang *et al.*2018). Evidence suggests that the effects of these limiting factors on soil respiration will differ among different ecosystems (Ohashi et al. 2008). For example, soil temperature is the dominant factor in determining soil respiration rates in a temperate forest stand (Luan *et al.* 2012). However, soil temperature and water content are the key factors responsible for variation in soil respiration in tropical regions (Ohashi *et al.* 2008, Adachi *et al.* 2009; Hanpattanakit *et al.* 2009). Zhou *et al.* (2013a) stated that in a mountain forest climatic factors did not affect soil respiration, where the climate is hot and humid, while Zhu *et al.* (2015b) stressed on the availability of quality substrate for microbial degradation; Epron *et al.* (2006) and Mo *et al.* (2008) suggested that soil respiration is strongly influenced by soil nutrient input, physico-chemical properties of soil, and activity of decomposing organisms. High accessibility of sufficiently moistened litter input stimulated soil microbial growth and activity (Singh and Gupta 1977, Song *et al.* 2010, Tlaskal *et al.* 2016). Finally, litter mass loss or decay encompasses ample breakdown of organic matter into CO_2 and nutrients. It returns carbon, as CO_2, into the atmosphere through the respiration of soil microorganisms. Consequently, measurement of CO_2 efflux from the soil surface is probably the most widely used system of estimating the rate of soil respiration and is one of the best approaches to evaluate soil biological activities in relation to carbon and energy flow in terrestrial ecosystems.

In sand dune ecosystems microbial respiration is the major source of respiration and needs to be investigated in order to assess the role of edaphic factors and microbial decomposers. Various authors have documented the role of microorganisms in decomposition of organic substances as well as their substantial contribution to soil respiration (Anderson 1982, Auffret *et al.* 2016, Batubara *et al.* 2019). Similarly, many published literature have emphasized the role of edaphic factors in controlling the soil respiration rate (Ohashi *et al.* 2008, Adachi *et al.* 2009; Hanpattanakit et al. 2009, Arora and Chaudhry 2017, Zoua *et al.* 2018; Prasad and Baishya 2019; Meena *et al.* 2020)). However, such type of study is missing in coastal sand dunes of Odisha. In the current study, I sought to address how differences in soil moisture, temperature and soil organic carbon as well as microbial community composition, determine the response of soil respiration in coastal sand dunes of Odisha.

MATERIALS AND METHODS

The present study was conducted in Ganjam district of Odisha which falls at 84º50'E longitude and 19º15'N latitude at an altitude of 6-8m from mean sea level. The study area has a tropical monsoonal climate with coastal characteristics. The mean temperatures is varied from 37°C in summer to 13°C in winter and mean annual precipitation is 130cm. The soil of this area is underlain by very fine sand (0.05cm) to small pebbles (1.5cm). This area contains the plantation of *Casuarina* with an average density of 3500 trees/ha. Both the soils were alike preceding to the establishment of the plantations in year 1993-95. To prevent degradation of mobile sand dunes, thirty-forty rows of *Casuarina* plantation are made in this infertile coastal land. Along with fixing of sand dunes, *Casuarina equisetifolia* L. is an actinorhizal nitrogen-fixing species with good wind and salt resistance and is often used for the establishment of protection forests in coastal sandy areas of Odisha. The *C. equisetifolia* plantations in Ganjam district play an important role in local environment improvement and ecological security maintenance.

Two soils representative (barren coastal sand dune and sand dune with 6–8 years old uniculture plantation of *Casuarina*) were selected for the study. The barren coastal sand dune was located on the seashore of the Bay of Bengal without any vegetation. The sand dune with the intensive plantation of *Casuarina* was situated about 1km apart from the first one. Soils were sampled for a period of two years. The soil samples were collected in sterilized test tubes (in triplicate) from the upper layer of 0-0.5 cm in depth at monthly interval and brought to the laboratory for further analysis. The soil microorganisms were isolated by serial dilution (Waksman, 1927), and soil plate method (Warcup, 1950). The isolation of bacteria and microfungi from soil samples was initiated by taking 10 g freshly collected soil in 250 ml flasks containing 100 ml distilled water and shaken for 15 min on a horizontal mechanical shaker. The suspension was further diluted to 10^2 and then to 10^3 using sterile distilled water. 1 ml aliquot of 10^2 dilutions for microfungi and 10^3 dilutions for bacteria was inoculated on potato dextrose agar media. The microbial colonies were studied after 3-7 days of incubation. A soil thermometer was used to determine soil temperature. The soil moisture was measured by gravimetric method (oven dry at 105°C till constant weight). Soil total organic carbon was determined by Walkley–Black method (Jackson 1967).

Soil CO_2 flux (mg CO_2 m^{-2} h^{-1}) was estimated by alkali absorption method (Witcamp 1966). Open-ended aluminium cylinders, 10 cm diameter and 15 cm height, were inserted into the soil up to 5 cm depth. Five cylinders each of the same size were used in each of the study sites, i.e. barren sand dune, and sand dune with *Casuarina* plantation, of which one cylinder each was used as blank in all the two study sites. Next 20 ml of 0.1 N KOH solution was kept in plastic vials and the cylinder was made airtight with anchor grip and placed for 6 h to absorb the CO_2 released. The carbon dioxide absorbed was then determined by titrating the KOH solution with 0.1 N standard dilute HCl solution using phenolphthalein as an indicator.

RESULTS

The month-wise fluctuation of soil respiration, bacterial population, fungal population (*Absidia glauca, Aspergillus awamori, Cladosporium cladosporioides, Curvularia eragrostidis, Drechslera australiensis, Fusarium oxysporum, Paecilomyces varioti, Penicillim citrinum, Rhizopus nigricans, Trichoderma viride*), soil temperature and moisture among the two sites was depicted in figure 1. Soil respiration rates represented marked variations between the two soils (Fig. 1). Both the soils exhibited a similar pattern with a peak occurring during September/October and minimum during May. Soil respiration was highest during the wet season compared to the dry one. The amount of soil carbon dioxide flux ranged from 93 to 248 mg CO_2 m^{-2} hr^{-1} with an average of 169 mg CO_2 in plantation site; and 71 to 181 mg CO_2 m^{-2} hr^{-1} an average of 145 mg CO_2 in virgin sand dune. Anova showed a significant difference ($F= 10.08$ $p< 0.01$) in soil CO_2 flux between different seasons. The monthly fluctuation of fungal and bacterial population exhibited similar trend to the output of CO_2. A linear regression between soil edaphic factors and soil respiration; fungal population and soil respiration analysis was performed (Fig. 2 and 3). Results showed that soil respiration had significant relationships with fungal population ($R^2 = 0.497$ $p<0.05$ for barren sand dune; $R^2= 0.598$ $p<0.05$ for sand dune with plantation). Over the study period, the bacterial

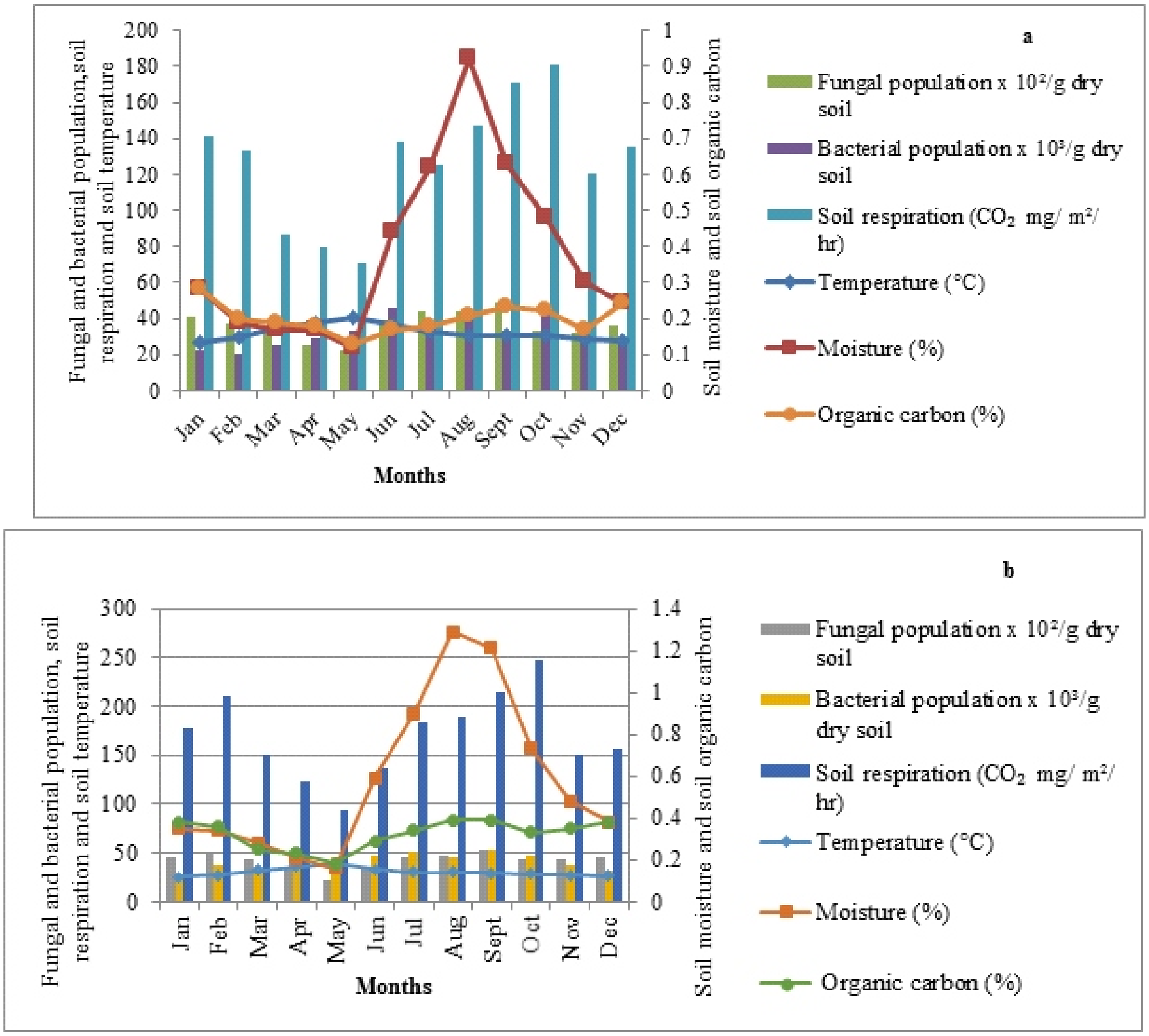

Figure 1. Seasonal variation of fungal and bacterial population, soil respiration and edaphic variables in (a) coastal sand dunes without any plantation and (b) coastal sand dunes with monoculture plantation of *Casuarina* of southern Odisha. Data are monthly averages.

population was constantly higher than the fungal population. Significant but negative correlation was found between soil CO_2 efflux and soil temperature ($R^2 = 0.446$ $p<0.05$ for barren sand dune; $R^2= 0.435$ $p<0.05$ for sand dune with plantation). During the present investigation, soil moisture increased significantly from June to September and accordingly higher soil respiration at both sites. Soil moisture showed a positive significant correlation with CO_2 efflux ($R^2 = 0.395$ $p<0.05$ for barren sand dune; $R^2= 0.345$ $p<0.05$ for sand dune with plantation). The monthly variation of soil organic carbon exhibited positive correlation with soil respiration ($R^2 = 0.395$ $p<0.05$ for barren sand dune; $R^2= 0.501$ $p<0.05$ for sand dune with plantation). In addition, no correlation was observed between soil moisture and soil temperature in both the sites ($R^2 = 0.056$ for barren sand dune; $R^2= 0.047$ for sand dune with plantation).

DISCUSSION

Rates of soil-CO_2 evolution on the sites reported here (71 to 248 mg $CO_2/m^2/hr$) is comparable to different ecosystems of the world – temperate forest (Laishram *et al.* 2002; Oishi *et al.* 2013), tropical forest (Takahashi *et*

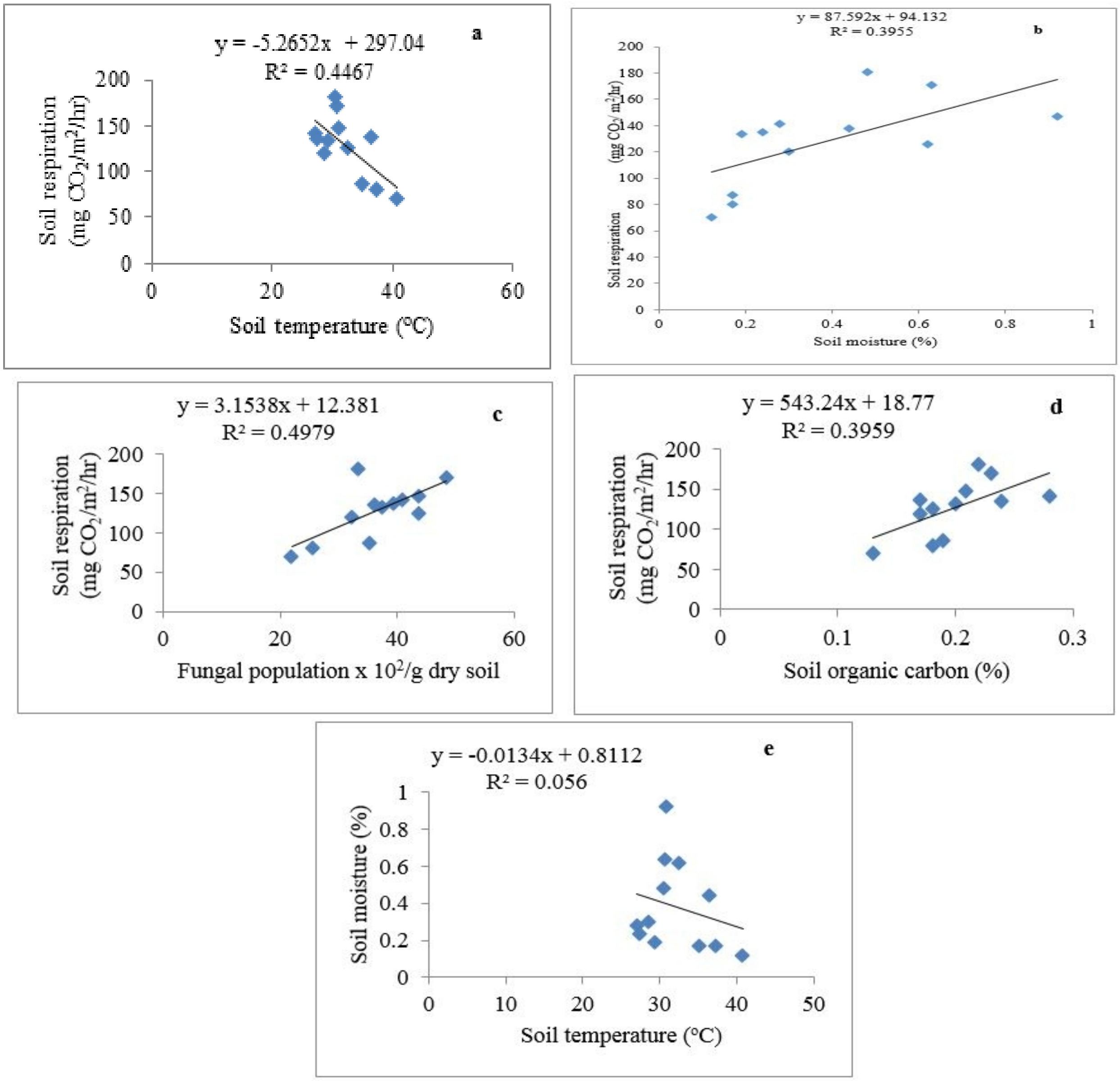

Figure 2. Regression analysis of soil respiration a) soil temperature, b) soil moisture c) fungal population, d) soil organic carbon and e) between soil temperature and soil moisture in coastal sand dune with *Casuarina* plantation of southern Odisha.

al. 2011), agricultural ecosystem (Han 2007), subtropical montane forest (Chang *et al.* 2008), subtropical forest (Devi and Yadava 2009; Wang *et al.* 2011), Mediterranean ecosystems (Almagro *et al.* 2009), steppe semi-arid ecosystem (Rey *et al.* 2011), boreal forest (Laganiere et al. 2012), tropical savannas (Richards *et al.* 2012), mixed forest(Chen *et al.* 2013), bamboo forest ecosystems (Duking *et al.* 2011; Yashiro *et al.* 2012), grassland ecosystem (Mielnick *et al.* 2000) and *Dipterocarpus* forest (Hanpattanakit *et al.* 2009). In the present study, significant seasonal variations of soil respiration corroborate the findings of Saraswathi *et al.* (2008). More amount of soil CO_2 flux during rainy season and early part of winter results from gush mineralization of the soil organic matter that has accumulated during the dry period of the soil (Devi and Yadava 2009). The moist soil condition promoted the physiological activities of soil microbes and root respiration

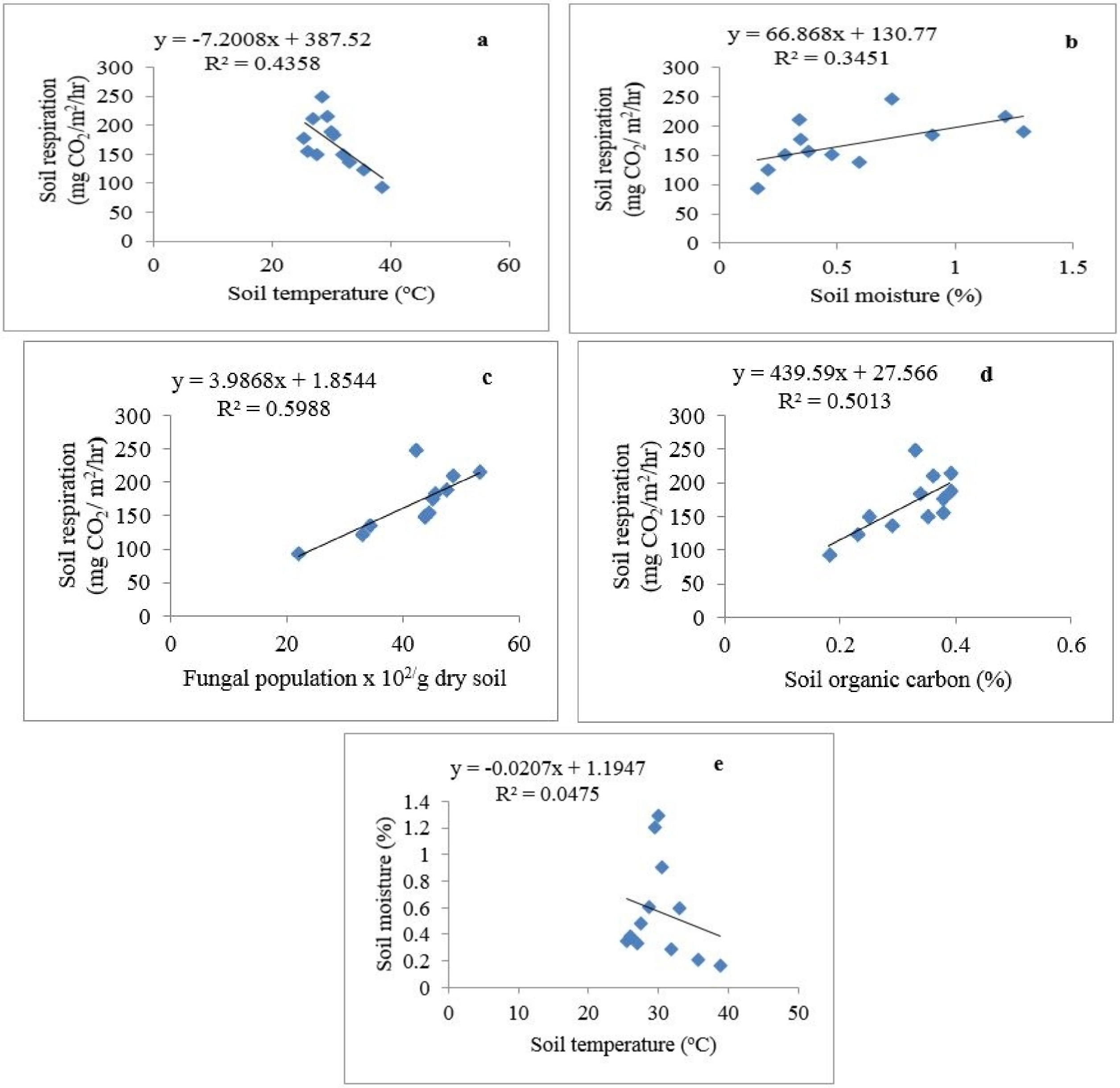

Figure 3. Regression analysis of soil respiration a) soil temperature, b) soil moisture c) fungal population, d) soil organic carbon and e) between soil temperature and soil moisture in coastal sand dune with *Casuarina* plantation of southern Odisha.

resulted in the increase of soil CO_2 flux. This is in agreement with the studies in temperate forest (Laishram *et al.* 2002), subtropical forest (Devi and Yadava 2009), warm temperate forest (Mo *et al.* 2005), tropical rainforest (Kosugi *et al*. 2007), mixed forests (Takahashi *et al.* 2011; Chen *et al.* 2013), Afromontane forest (Yohannes *et al.* 2011) and bamboo plantation (Liu *et al.* 2011). Minimum rate of soil respiration in the month of May may be due to low moisture content, high temperature and decreased microbial population. The higher amount of CO_2 evolution from plantation site may be attributed to the high microbial activities and soil organic matter than the barren sand dune. Thus higher soil resources in the soil component also resulted in greater rate of soil CO_2 flux in coastal sand dune with monoculture *Casuarina* plantation.

Soil respiration varies significantly

with vegetation (Wang *et al.* 2011, Wang *et al.* 2013, Chen *et al.* 2014) and also among major biome types (Singh and Gupta 1977, Raich and Schlesinger 1992). Different tree species can have different dominant factors such as soil temperatures and soil moisture which affect the soil microbiota and their activities via root litter and exudates (Niemi *et al.* 2007) and have diverse effects on specific microbial populations (Hartmann et al. *et al.* abundant taxa of soil fungi and bacteria have differing capacities to degrade available and complex forms of plant-derived C resources (Waring *et al.* 2013). For instance, fungi are thought to express a broader suite of enzymes capable of transforming and stabilizing inputs (McGuire *et al.* 2010), carbon turnover rate is slow due to more incorporation of soil C into biomass (Rousk and Baath 2011) and significantly higher C use efficiency (Six *et al.* 2006) than bacteria. This clearly indicates that changes in the relative abundance and activity of bacteria and fungi may significantly affect C cycling, and in particular to changing plant-C inputs (Waring *et al.* 2013). The higher respiration rate beneath *Casuarina* plant cover can be correlated to the detritus composition and native soil carbon pool, which feeds soil organisms (Paul *et al.* 1999; Conant et al. 2000). Such results show that surface vegetation type is a significant predicting factor of carbon dioxide efflux rate, and therefore aboveground vegetation may profoundly affect the dynamics of soil respiration by influencing soil microclimate and the production and transfer of aboveground photosynthate to belowground.

The observed trend of a negative coherence between soil respiration and temperature is also reported by Cindy et al. (2018). Carey *et al.* (2016) stated that depending on the biome, respiration rates increase with temperature up to ~25 °C (23–34°C), above which respiration rates level off and decrease. The negative correlation observed in the present study is likely due, in part, interaction with other factor (s). Moisture in soils was essential for both plant growth and soil microbial activity, hence affecting carbon inputs including the litter decomposition and soil organic matter, and consequently, respiration and carbon outputs (Moyano *et al.* 2013). Generally, soil respiration increases from low to medium soil moisture, reaches a plateau at optimum moisture, and declines at high soil moisture. Previous studies suggest that soil respiration increases following increased prolonged wet season, and decreases with drought (Meir *et al.* 2015, Liu *et al.* 2016). Water saturation limits aeration and low soil moisture leads to desiccation, reduced substrate access or diffusion and blocking CO_2 transport, which restricts microbial metabolism (Davidson et al. 2000; Ding et al. 2010; Talmon *et al.* 2011, Gong *et al.* 2014). The optimum soil water content is usually somewhere near the field capacity, when macropore spaces are mostly air-filled, thus facilitating O_2 diffusion. When micropore spaces are mostly water-filled, the diffusion of soluble substrates is facilitated (Zhang *et al.* 2010). In agreement with other studies (Ding *et al.* 2010, Gong *et al.* 2014; Wang *et al.* 2014; Bao *et al.* 2016), soil water content in the present study has apparent influence on soil respiration. In the present study as soil moisture and soil temperature were dependent-variables, one affecting the other, i.e. extremes of temperature coincided with lowest soil moisture.

Soil organic carbon (SOC) is one part of the much larger global carbon cycle that involves the cycling of carbon through the soil, vegetation, ocean, and the atmosphere. Soil carbon stocks express a balance between organic inputs and their stepwise decomposition by soil biota. An increasing trend of soil respiration was observed in response to increasing in SOC. This study showed that SOC was a foremost influencing factor for the fate of soil respiration between different sites. Therefore, a significant positive correlation between soil respiration and SOC was derived because respiration is a result of the mineralization of SOC that is stored in large stocks (Fang *et al.* 2005, Knorr *et al.* 2005).

CONCLUSION

I found that the variation in soil properties and microbial activity between barren sand dunes and sand dune with monoculture plantation of *Casuarina equisetifolia* caused the major difference in annual soil respiration efflux among these two sites. Soil moisture is the dominant factor that influenced soil respiration. In contrast to the case for many other ecosystems, the relation of soil moisture and soil temperature failed to explain variations in respiration among the studied sites. The results suggest that vegetation cover may alter the soil C balance in this ecosystem.

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