Cantaurus, Vol. 17, 17-19, May 2009 © McPherson College Division of Science and Technology

The Effect of Temperature on Aerobic Methane Production by the Fiddle Leaf Fig and the Norfolk Island Pine

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ABSTRACT

Due to the growing concern of global warming, the emissions of gases are closely monitored. Methane is known to be produced from anaerobic bacterial process from energy production, biomass burning, coal mining, landfills, wetlands, ruminant animals and rice paddies. Frankenberg and his colleagues (2005) observed a surprisingly high amount of methane over tropical forest regions during the dry seasons using SCIAMACHY (Scanning Imaging Absorption Spectrometer for Atmospheric Chartography) (Frankenberg, 2005). Keppler and his colleagues (2006) followed this discovery with the findings that certain plants are a source of aerobic methane production. Because methane is a greenhouse gas, these studies may be important for the prevention of global warming. I studied methane emissions from the leaf of the Fiddle Leaf Fig (*Ficus lyrata*) a plant native to western Africa, and the Norfolk Island Pine (*Araucaria heterophylla*) indigenous to a small island between New Zealand and Australia know as Norfolk Island, using gas chromatography with a flame-ionization detector. Leaves of both plants were incubated in 22 mL glass vials in the dark at 20°C, 30°C, and 40°C. Gas was extracted from the vials and methane was measured at T=0 and T=24 hours. The experiments were analyzed using the paired t-test. The results imply that there is not a statically significant increase in methane in the Fiddle Leaf Fig at 20℃ and 30℃ or in the Norfolk Island Pine at 20°C, 30°C, and 40°C. Results did show a statistically significant increase of methane emissions in the Fiddle Leaf Fig at 40℃. My results suggest that aerobic methane production is not common in all plants and at all temperatures.

Keywords: *methane, greenhouse gas, global methane budget, Ficus lyrata, Araucaria heterophylla*

INTRODUCTION

Air pollution due to human activities has accelerated the amount of greenhouse gases released into the atmosphere over the last few decades. Due to greenhouse gases in the atmosphere, the average temperature of the Earth has increased. Methane gas (CH_4) is one of the more abundant greenhouse gases in the atmosphere with a mixing ratio of 1.8 parts per million (p.p.m.). Methane is also important because it has a strong absorption of infrared radiation and an atmospheric half-life of seven years, which is longer than the other gases, including carbon dioxide (Keppler, 2006). Currently, between 500 and 600 Tg CH_4 y^{-1} is emitted according to the global methane budget (Butenhoff, 2007). Previously it was thought that methane emission was solely an anaerobic bacterial process from landfills, wetlands, ruminant animals and rice paddies. According to Christopher L. Butenhoff and M. Aslam Khan Khalil (2007), bacterially released methane accounts for 70% of methane emissions.

Recently, researchers found that an unknown source of methane within the global methane budget has not been accounted for. This concept was confirmed by Frankenberg and his colleagues (2005), when they noticed an unusually larger amount of methane over tropical evergreen forests. Frankenberg used a SCIAMACHY (scanning imaging absorption spectrometer for atmospheric chartography) during the dry season. Frankenberg, et al (2005) examined the methane emissions in tropical evergreen forests in South America, Africa, and Indonesia, and found a larger amount of methane emission than expected.

Scientists have suggested that methane released into the atmosphere may also be produced by terrestrial plants. In 2006, Keppler and his colleagues form the Max-Planck Institute for Nuclear Physics in Heidelberg, Germany, found that aerobic methanogenesis occurs in leaves of plants such as ash (*Fraxinus excelsior*), sweet vernal grass (*Anthoxanthum odoratum*), maize (*Zea mays*), beech (*Fagus sylvatica*), and wheat (*Triticum aestivum*). Though the plant processes are unknown, it is thought that plant pectin plays a role in the formation of CH_4 in plants. It is uncertain however if all plants are capable of methane production. Keppler's results show the amounts of methane emitted by these plants are dependent on the amount of sunlight to which the plants were exposed and the temperature in the external environment. From his findings, Keppler (2006) estimated that plants produce between 62 and 236 Tg CH_4 y^{-1} with the main sources being from temporal grasslands and tropical forests.

Dueck *et al* (2007) prepared an experiment similar to that of Keppler et al. They found their methane emissions to not be statistically significant from zero. They also found plants to not be temperature and sunlight dependant in methane production. Compared to study from Keppler *et al*. in 2006, Dueck et al. found their emission results to be six to eighteen times less. A recent study (Ferretti *et al*., 2007) confirmed results of methane produced by plants to be an estimated 80% lower than that of Keppler et al.

Foliar biomass is converted using the leaf area index, which is the ratio between the one-sided area of the leaf and the area it projects on the ground. It is a possibility that the foliar biomass estimation is an overestimate because the specific leaf area relation has a bias toward needle-bearing evergreens, which tend to have a lower specific leaf area than the broadleaf evergreens (Butenhoff, 2007). Butenhoff (2007) found plants to be very temperature dependant, thought the functional relation between plants and methane is unknown.

According to Evans (2007), researchers such as Dueck *et al*. (2007), were unable to verify the original findings of Keppler and colleagues of statistically significant aerobic methane production by terrestrial plants. Because of this failure, the requirement for specialist aerobic methane detection equipment, and the difficulty of publishing negative findings, it is unlikely that many others will attempt this experiment (Evans, 2007).

I studied the methane emissions from the leaf of the Fiddle Leaf Fig (*Ficus lyrata*) and the needle leaves of the Norfolk Island Pine (*Araucaria heterophylla*) at temperatures from 20°C to 40°C using a gas chromatograph with flame ionization detector.

MATERIALS AND METHODS

I used a known concentration of 50 p.p.m. Methane gas (Scott Specialty Gas, Plumsteadville, PA), using injections from 1µL to 100µL to find the standard methane curves. My r^2 values were 0.969, 0.949, and 0.945. I used these to compare the methane gas emissions levels from the plants to the pure methane. Methane emission rates were detected in the leaves of the Fiddle Leaf Fig (*Ficus lyrata*) and the needle leaves of the Norfolk Island Pine (*Araucaria heterophylla*) due to temperature variation without sunlight.

I collected 30 samples of each plant, making 60 total samples, and prepared the samples using a method similar to that of Keppler (2006), and Rodriguez (2007). Using sterile scissors, I cut the plants into samples of approximately 0.2g per piece. Then, using sterile tweezers, I cleaned any potential bacteria from the plants by rinsing them in a 5% bleach solution, followed by a rinse in deionized water. I placed the samples into sterile, properly marked 22mL vials and closed them with gastight, rubber septum containing tops. I then incubated 10 samples of each plant in the dark for 24 hours at 20°C, 30°C, and 40°C. I used a gas tight syringe to take measurements of 500µL (1/44 of the total volume) before and after incubation (T=0 and T=24). The injection was made into the Clarus 500 Gas Chromatography with flame-ionizing detector (FID). With the temperature of column set at 45°C, and the flow rate of the N_2 (Nitrogen) at 30ml/min the cycle following the injections ran for 3 minutes.

Finally I dried each plant sample in an oven set at 105°C for 24 hours and weighed them individually to determine the dry weight of each sample. This step is to insure that the plant samples were of the same size to better assess the rate and emission levels of the methane in plants ($ngCH_4g^{-1}h^{-1}$).

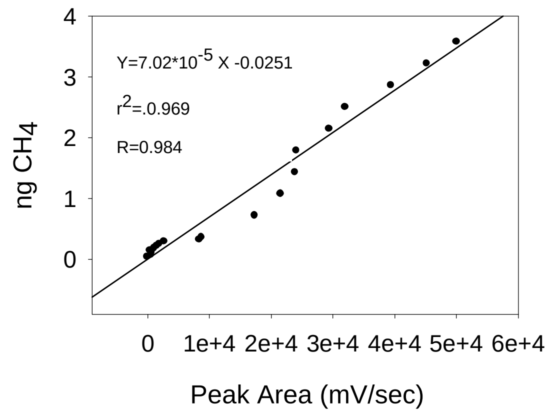

Figure 1. Methane standard curve. Sample CH_4 concentrations fell between 1 and 100 µl.

RESULTS

Using the paired t-test, the data from each plant was analyzed at each temperature variation. It showed that the Norfolk Island Pine leaves did not show a significant aerobic methane production at any of the temperatures. (P=0.424 for incubation at 20°C, P= 0.127 for incubation at 30°C, and P=0.177 for incubation at 40°C).

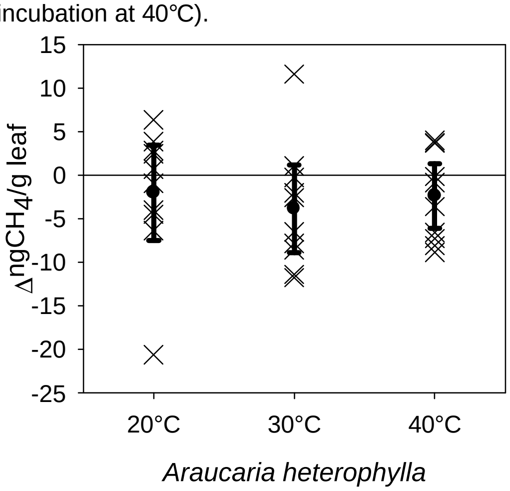

Figure 2. Change of CH_4 Concentration in the Norfolk Island Pine per gram of dry leaf per hour for plant samples incubated at 20°C, 30°C, and 40°C. The mean of each sample is indicated by the solid circle and the bars indicate the 95% confidence levels.

The Fiddle Leaf Fig leaves incubated at 20°C and 30°C also did not show a significant aerobic methane production (P=0.672 for incubation at 20°C and P=0.213 for incubation at 30°C). The Fiddle Leaf Fig samples incubated at 40°C did show a significant increase in methane concentration (P=0.025).

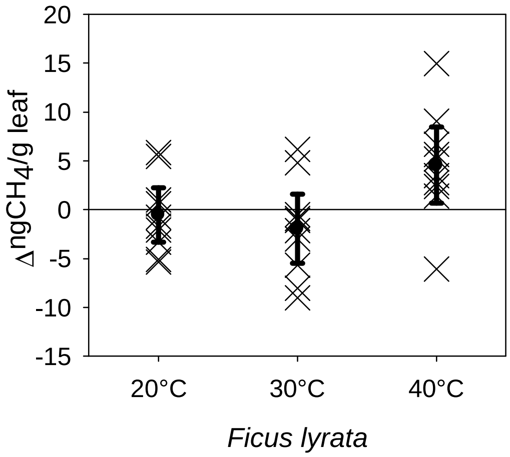

Figure 3. Change of CH_4 Concentration in the Fiddle Leaf Fig per gram of dry leaf per hour for plant samples incubated at 20°C, 30°C, and 40°C. The mean of each sample is indicated by the solid circle and the bars indicate the 95% confidence levels.

DISCUSSION

My initial objective was to note the difference in methane production in a broadleaf evergreen plant and in needle bearing evergreen plant because of the obvious difference in specific leaf area such as Butenhoff (2007) noted. My plan was to use a plant that I knew produced methane and was available such as the banana (Musa acuminate) from the study of Rodriguez (2007). Because of an unfortunate event, I was forced to find another plant, and the readily available plant was the Fiddle Leaf Fig (*Ficus lyrata*). Fiddle Leaf Fig (*Ficus lyrata*) plant is a significant source of methane production under 40℃. The Fiddle Leaf Fig does not appear to be a significant source of methane emissions under 20℃ or 30℃. The Norfolk Island Pine is not a significant source of methane production under the variation of temperatures. Because of this, no conclusions can be made as to the difference in methane production due to specific leaf area.

Because there are so many species of plants that have not yet been tested for aerobic methane emissions, future studies may be conducted on many more species of plants with the goal of determining the missing figures in the Global Methane Budget. Also, once methane emissions have been determined in plants of the broadleaf nature and the needle bearing nature, they may be compared to one another to identify the amounts of methane released due to specific leaf area.

ACKNOWLEDGEMENTS

I would like to thank Dr. Jonathan Frye for all of his assistance during my research. I would also like to thank McPherson College and the Science Department for the use of facilities, equipment and supplies.

LITERATURE CITED

Butenhoff, C.L., M.A. Khan Khalil. 2007. Global Methane Emissions from Terrestrial Plants. Environmental Science and Technology. 41:4032-4037.

Dueck, Tom A., Ries de Visser, et al. 2007. No evidence for substantial aerobic methane emissions by terrestrial plants: a C-labeling approach. New Phytologist 175:29-35.

Evans, John R. 2007. Resolving methane fluxes. New Phytologist. 175:1-4.

Ferretti, D.F., J.B. Miller, J.W.C. White, K.R Lassey, D.C. Lowe, and D.M. Etheridge. 2007. Stable isotopes provide revised global limits of aerobic methane emissions from plants. Atmospheric Chemistry and Physics 7:237-241.

Frankenberg, C., J.F. Meirink, M. van Weele, U. Platt, and T. Manger. 2005. Assessing Methane Emissions from Global Space-Borne Observations. Science 308:1010-1014.

Keppler, F., J.T.G. Hamilton, M. Brass, and T. Röckmann. 2006. Methane emissions from terrestrial plants under aerobic conditions. Nature 429:187-189.

Rodriguez, J. 2007. Aerobic methane production by Banana plant. Cantaurus 15:21-23.