Omonrice 16: 100-106 (2008)

EFFECTS OF SOIL ENVIRONMENT AND FERTILIZER ON SOIL MICROORGANISM POPULATION AND RESPIRATION IN THE INTENSIVE RICE MONOCULTURE AREA

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ABSTRACT

The population and respiration of soil microorganisms were assessed from soil samples collected from the long-term experiment of chemical fertilizer application for 20 years in alluvial soil of the lower section of land river area, combination of inorganic and organic fertilizer for 6 years, and alluvial soil of the upper section of land river area. The assessment showed that the population of soil microorganism was generally high in the soil with fertilizer supply because of high content of organic carbon. It was lower in the soil without fertilizer application. The population of microorganism in soil with only N supply was lower than soil supplied with NK, NP and NPK. The population of microorganism in soil with only P supply was higher than soil supplied with NK, NP, and NPK. It was the highest in the soil applied PK.

In rice monoculture, the population of bacteria was the highest, followed by actinomycetes, and algae. Fungus population was the lowest.

The respiration was reflected by CO_2 content released by soil microorganism. Soil samples after treated were incubated at 1, 3, 7 and 14 days. Under the assistance of NaOH solution, the content of CO_2 was measured and we found that it increased by time of incubation. The content of CO_2 in soil of lower section of river land area increased until 14 days after incubation meanwhile it increased until 7 days after incubation in soil of upper section of river land.

INTRODUTION

Soil microorganism population and their interactions within the cropping system are the basis for assessment of the relationship soil fertility and cropping system. An effective method can be selected to increase useful microorganism population and their biomass production to improve soil fertility. Suitable cropping system can activate the growth of microorganisms to improve soil fertility, crop yield, and economic efficiency in agricultural production. Soil CO_2 concentration reflects microorganism population and their respiration in soil.

The general objective of this study is to know the roles microorganisms in soil mineralization to supply nutrients for crop plant.

The specific objectives are:

-To determine population of soil micro-organisms as fungus, algae, bacterium and actinomycetes,

-To know the respiration of micro-organisms in alluvium soil with rice monoculture in the lower section of river land area (long term experiment of N, P, K in Cuu Long Rice Research Institute) and the upper section of river land area (Binh My village, Chau Phu district, An Giang province) in South Vietnam.

METHODS

Examine soil microorganism population

Soil samples used to examine microorganism population.

Alluvial soils experienced 2 rice crops per year applied only chemical fertilizer from long term experiments for 20 years (with 8 treatments of no fertilizer application, N, K, NK, P, PK, NP and NPK) and combination of chemical fertilizer with decomposed organic matters for 6 years (NPK + decomposed organic matter) were collected before

beginning the following dry crop season. The soil samples were collected from the plots of eight treatments with chemical fertilizer and one treatment with combination of NPK + decomposed organic matter. This long term N, P, K experiment was conducted at experimental field of Cuu Long Delta Rice Research Institute. With eight treatments of chemical fertilizer only, the rice straws and stubbles were removed after harvesting of both dry and wet rice seasons to exclude the effect of organic matters. The chemical fertilizers of N, P, K used were from the sources of urea, super phosphate, and clorur kali, and each was applied at two dosages. Nitrogen was used at 0 and 80 kgN/ha in both wet and dry seasons. Phosphorous was used at 0 and 40 kgP_2O_5/ha, clorur kali at 0 and 90 kgK_2O/ha.

Treatment number 9 was a combination of organic and chemical fertilizers (50-15-15 kg N-P_2O_5-K_2O/ha + 5 tons organic fertilizer in dry season, 40-15-15kg N-P_2O_5-K_2O/ha + 5 tons organic fertilizer in wet season). Soil samples were analyzed from this treatment during 6 years of experiment (since the year of 2000). Source of organic fertilizer was from decomposed rice straws and stubbles by treated with *Trichoderma* and incubated for 4 weeks.

Soil sampling and microorganism density measurements were conducted in December 2006.

Methods to examine soil microorganism population.

Soil samples were collected within 0-20cm from soil surface at 10 days before planting the following dry rice season and kept in refrigerator. These soil samples were processed within 72 hours.

Soil microorganism densities were assessed by method of plate counting by using the following media (Subba Rao 1977):

1 Nutrient agar-agar to count bacteria.
2 Media of Kenknight and Munaier to count actinomycetes.
3 Media of Bristol to count algae.
4 Media of RBA to count fungi.

The unit used to assess microorganism densities was colony formed unit per 1 gram dried soil (CFU/g dried soil).

Examine respiration of soil microorganism.

Soil samples used to examine respiration of soil microorganism.

Soil samples were also from the above eight chemical fertilizer treatments of long term N, P, K experiment and were collected before planting the 35th wet rice crop. This soil was as alluvial soil at the lower section of river land area. For comparison, alluvial soils at the upper section of river land area were collected at Binh My village, Chau Phu district, An Giang province (this was coded as CPAG)

The measurement of respiration of soil microorganism was conducted during November and December 2006.

Methods of respiration measurement

Soil samples were dried under air temperature and finely ground. This processed soil was divided into many small parts of 10g each. Each 10 g of this soil was put into the small plastic bottle (the bottle volume of 35ml). The moisture of 60% was created (saturated moisture). These soil bottles together with one glassed bottle contained 10 ml NaOH 0,1N were placed inside a large plastic box with cover. These were incubated for 1, 3, 7 and 14 days (called as four treatments). Blank sample without soil was conducted synchronically. NaOH solution will absorb CO_2 released from soil due to microorganism respiration. The bottles with NaOH inside plastic boxes were took out and covered immediately at 1, 3, 7 and 14 days after incubation. After that, they were poured into beakers for standardization with H_2SO_4 0.1 N to determine excess of NaOH. Before standardization, each beaker was added 1 ml $BaCl_2$. $2H_2O$ 3N and 3 drops of Phenol phthalein 0.1%. From this, CO_2 concentration was estimated from each treatment (Anderson, 1982).

RESULTS AND DISCUSSION

Microorganism densities

According to Penamareva (1961, quoted by Do Anh, 2002), the nature of soil fertility is the energic level in soil organic matter, of which micro-organism was indirect agent, that means micro-organism used organic matter as energy source for its activities. Dung (2000, quoted by Do

Anh 2002) reported that organic matters and microorganism activities were strongly correlated in alluvial soil of Red River Delta. The soil with the cropping system of Spring peanut- Summer soybean- Winter maize contained highest microorganism population of $80*10^6$CFU/g dried soil meanwhile it was only $28*10^6$CFU/g dried soil in Hoang Lien Son soil planted with tea crop.

Microorganism population was significantly different among cropping systems and species of microorganism (Table 1). In this study on soil with rice monoculture, the highest population was bacteria, followed by actinomycetes, then algae, and the lowest one was fungus. The microorganism population in this study was very high and it ranged $2.155*10^8$ – $38.722*10^8$ CFU/g dried soil. The average population was $10.051*10^8$ CFU/g dried soil.

Table 1. Microorganism population in rice soil samples collected before 2007 dry rice season

Micro-organism population counted in diluted solution at 10^6						Population $*10^8$ CFU/g dried soil
Treatment	Fungus	Algae	Bacteria	Actinomycetes	Total	
Control	0.052	7.488	156.000	52.000	215.540	2.155
N	0.000	6.490	309.056	103.019	418.566	4.186
K	0.000	7.636	324.942	487.413	819.990	8.200
NK	0.053	6.867	581.056	52.823	640.799	6.408
P	0.051	15.357	354.785	354.785	724.978	7.250
PK	0.105	7.312	3,760.282	104.452	3,872.150	38.722
NP	0.055	6.483	332.436	387.842	726.815	7.268
NPK	0.049	4.677	292.263	487.105	784.093	7.841
NKP + organic fertilizer	0.725	13.617	414.207	414.207	842.756	8.428
Mean	0.121	8.432	725.003	271.516	1,005.076	10.051
SD	0.229	3.568	1,143.760	189.153	1,094.617	10.946

Note: CFU: Colony formed unit

Fungus population

Fungus population was lowest, and it ranged from 0.000 – $0.725*10^6$ CFU/g dried soil. Fungus was not seen in the plots treated with N and/or with K. In the plots added organic fertilizer, fungus population was the highest ($0.725*10^6$CFU/g dried soil). Fungus densities were very low in the other treated plots, such as control treatment ($0.052*10^6$CFU/g dried soil), NK ($0.053*10^6$CFU/g dried soil), NP ($0.055*10^6$CFU/g dried soil), P ($0.051*10^6$CFU/g dried soil), NPK ($0.049*10^6$CFU/g dried soil), PK ($0.105*10^6$CFU/g dried soil). Fungus population was low because it is aerobic organism and usually settles in soil surface layer. Soil before planting dry rice crop was still submerged, and it was drained out for crop starting. Under in-aerobic condition, fungus population was low.

Algae population

Algae population was higher those of fungus. However, it was lower than those of bacteria and Actinomycetes. Algae population ranged from $4.677*10^6$ to $5.357*10^6$CFU/g dried soil. Algae population was lowest in soil applied with NPK ($4.677*10^6$/g dried soil), and highest in soil with P treatment ($15.357*10^6$CFU/g dried soil). Algae population in soil added organic fertilizer aside from inorganic fertilizer was also high. Algae population in the rest treatments varied from $6.483*10^6$ to $7.636*10^6$CFU/g dried soil.

Bacteria population

Bacteria population was highest among microorganisms. It ranged from $156*10^6$ to $3760.282 *10^6$CFU/g dried soil. It was lowest in control treatment ($156*10^6$CFU/g dried soil), and highest in PK treatment ($3760.282*10^6$CFU/g dried soil). It varied from $292.263 *10^6$ to $581.056*10^6$CFU/g dried soil in the rest treatment.

Actinomycete population

Actinomycete population was relatively high among microorganisms. It ranged from $52.000*10^6$ to $487.413*10^6$ CFU/g dried soil. This

was very low in the control treatment ($52.000*10^6$ CFU/g dried soil) and NK treatment ($52.823*10^6$ CFU/g dried soil). The population was highest in K treatment ($487.413*10^6$ CFU/g dried soil). The population was high in NPK treatment ($487.105*10^6$ CFU/g died soil) and the treatment of inorganic and organic combination ($414.207*10^6$ CFU/g dried soil).

In general, microorganism population was lowest in the control treatments ($2.155*10^8$ CFU/g dried soil). The microorganism population was low in all treatments exception of PK treatment ($38.722*10^8$ CFU/g dried soil).

According to Phung (1994), the differences in microorganism population was due to the differences in cropping systems and ranged from $0.3*10^6$ to $13*10^6$ CFU/g dried soil. The higher population was mostly found in crop rotation systems (about $2*10^6$ to $10*10^6$ CFU/g dried soil). The population was lower about $1*10^6$ CFU/g dried soil in the soil with mono crop cultivation or fallow. Phung (1994) reported that alluvial soil in the Mekong Delta was the most fertile soil and it hosted highest microorganism population of $13*10^6$ CFU/g dried soil. This soil was rich in microorganisms. There were many new microorganisms in the cropping system of rice-legume on alluvial soil. Microorganism population in the alluvial soil without silt deposit and continues rice monoculture was lower than the soil with silt deposit and crop rotation in the Mekong Delta.

Microorganism population in the acid sulphate soil was lower 2-4 folds as compared to grey soil, and 15-30 folds as compared to alluvial soil. Total population of microorganism in this soil was less than $1*10^6$ CFU/g dried soil and was classified into "very poor micro-organism soil". The limitation of microorganism development in this soil may be due to low pH, toxicity from Fe^{3+} and Al^{3+}, and acidity in soil. The more microorganism population was found in the soil with more organic matters. Though the rice straw and stubbles were removed after harvesting rice in the treatments with inorganic fertilizers, the soil organic carbon, total N and available N at the form of $N\text{-}NH_4^+$ in experimental field at Cuu Long Rice Research Institute was still high after 20 years cultivating rice. Similarly, the contents of organic carbon, total N and available N in the form of $N\text{-}NH_4^+$ in the treatment of inorganic and organic combination were also high (Table 2). Low ratio of C/N proved that there was strong activities from microorganisms and their capacities in decompose of organic matter was high.

Table 2. Organic carbon, total N, available N and C/N ratio

Treatment	Organic Carbon (%)	Total N (%)	$N\text{-}NH_4^+$ (ppm)	C/N ratio
Control	2.66	0.248 a	26.0	10.7
N	2.77	0.266 ab	29.3	10.4
K	2.63	0.249 a	23.8	10.6
NK	2.60	0.284 bc	32.0	9.2
P	2.81	0.297 bc	27.8	9.5
PK	2.71	0.289 bc	30.8	9.4
NP	2.85	0.303 c	28.2	9.4
NPK	2.75	0.278 abc	25.2	9.9
NPK+organic fertilizer	3.13	0.292	32.6	10.7
Mean	2.77	0.278	28.4	10.0
SD	0.16	0.020	3.1	0.6

Respiration of microorganisms within soil surface layer (0-20 cm)

Soils are said to respire because of their ability to take up oxigen and lose carbon dioxide. The oxigen is used primarily by the bacteria (other soil microorganisms are involved too) and by plant roots. Bacteria other than autotrops utilize soil organic materials as a source of energy for their metabolism and in the process oxidize the organic compounds to CO_2. Plant roots also utilize organic compounds as energy sources but in this case, the compounds are synthesized in the leaves of the plants and translocated to the roots where they are oxidized to CO_2, the energy being used for root growth and the uptake of nutrients by the roots. In both cases, therefore, O_2 is used and CO_2 is produced.

The rate at which respiration occurs depends on many factors but prime importances are: (a) amount of organic matter available for oxidation- this is the food supply- and the general fertility of the soil since it controls root growth; (b) soil temperature, because the rate of respiration approximately doubles for every 10^0C rise in temperature; (c) soil moisture for the bacteria and roots need water as part of their environment; and (d) oxygen concentration in the soil air. Respiration rates remain constant until the O_2 concentration reaches low levels; at concentrations of about one hundredth of value for atmosphere respiration declines rapidly and ultimately stops. This is what happens in a waterlogged soil and is one of the reasons why good soil drainage is necessary for plant growth. The most active respiration therefore occurs in organic-rich, moist, well- structured soils (Rowell 1981).

Contents of CO_2 was increased by duration 1, 3, 7 and 14 days after incubation in treatment number 9 presented in table 3 and figure 1. This indicates that the contents of CO_2 released from all treatments increased by duration from 1 to 14 days after incubation. Exception of treatment number 9 (soil from mono rice culture in Chau Phu district, An Giang -CPAG), CO_2 did not increased at 14 days after incubation.

As compared to the study with 9 treatments on heavy acid sulphate soil conducted in Tan Tuyen Commune of Tri Ton district in An Giang province by Minh (2003), the CO_2 concentrations accumulated by time of incubation on alluvial soil in Chau Phu district of An Giang province and in Co Do district of Can Tho province were higher. Minh reported that at 1 day after incubation (DAI), CO_2 concentrations ranged from 345-669 $mgCO_2$/kg dried soil; at 3 DAI from 1.243-2.035 $mgCO_2$/kg dried soil; at 7 DAI from 1.617-2.750 $mgCO_2$/kg dried soil and at 14 DAI from 1.885-2.867 $mgCO_2$/kg dried soil. Though CO_2 concentrations on heavy acid sulphate soil increased by time of incubation, it were lower than those in alluvial soils. This is due to low pH, high concentrations of Fe^{2+} and SO_4^{2-} in heavy acid sulphate soil.

Table 3. Contents of CO_2 ($mgCO_2$/kg) distributed by duration of incubation

Treatment	Days after incubation			
	1	3	7	14
Control	1,238 b	2,365 b	5,396 bc	8,690 b
N	2,283 c	2,255 b	4,593 abc	9,213 b
K	2,283 c	2,200 b	3,163 a	9,295 b
NK	1,733 bc	2,585 b	4,730 bc	10,368 b
P	1,403 b	3,025 bc	4,950 bc	10,395 b
PK	1,733 bc	2,860 bc	5,528 c	9,488 b
NP	1,733 bc	3,465 c	5,418 bc	9,790 b
NPK	1,788 bc	2,750 bc	5,583 c	9,350 b
Soil at CPAG	523 a	1,350 a	3,865 ab	3,845 a
F	**	**	*	**
CV (%)	24.0	20.3	20.6	12.7

At 1 day after incubation, CO_2 in soil at CPAG was the lowest (523 $mgCO_2/kg$). It reached the highest in N and K treatments (2,283 $mgCO_2/kg$) and was not statistically and significantly different with other treatments NK, P, PK, NP and NPK.

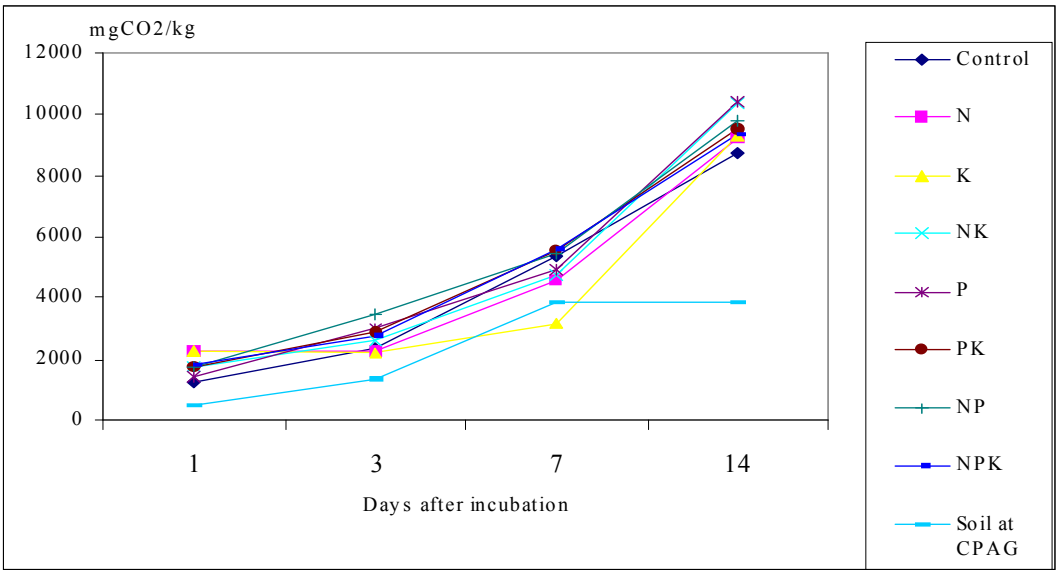

Figure1. Respiration of soil organism ($mgCO_2/kg$)

At 3 days after incubation, CO_2 in soil at CPAG was lowest (1,350 $mgCO_2/kg$). It was the highest in NP treatment (3,465 $mgCO_2/kg$) and was not significantly different with the treatments P, PK and NPK. CO_2 contents among control treatment, N, K and NK treatments were not significantly different.

At 7 days after incubation, CO_2 in K treatment was lowest (3,165 $mgCO_2/kg$) and was not significantly different with N treatment and soil at CPAG. It was highest in NPK treatment (5,583 $mgCO_2/kg$) and was not significantly different with PK treatment (5,528 $mgCO_2/kg$). CO_2 contents among control, N, NK, P and NP treatments were not different.

At 14 days after incubation, CO_2 content in soil at CPAG was lowest (3,845 $mgCO_2/kg$). It was not different among the rest treatments. This indicates that the activities of microorganism at 14 days after incubation were strongest and CO_2 content was two folds higher than that at 7 days after incubation. Except the soil at CPAG, CO_2 was not increased at 14 days after incubation as compared with 7 days after incubation.

CONCLUSION

In general, the population of soil microorganism was high in all treatments because of organic carbon content was high. It was lower in the soil without fertilizer application. The population of microorganism in soil with only N supply was lower than the soil supplied with NK, NP and NPK. The population of microorganism in soil with only P supply was higher than the soil supplied with NK, NP, and NPK). It was highest in the soil applied PK.

In rice monoculture, the population of bacteria was the highest, followed by actinomycetes, and algae. Fungus population was lowest.

The content of CO_2 increased by time of incubation. The content of CO_2 in soil of lower section of river land area increased until 14 days after incubation meanwhile in the soil of upper section of river land area (CPAG) it increased by time from 1, 3 and 7 days after incubation but not increased at 14 days after incubation.

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Ảnh hưởng của môi trường đất và phân bón đến mật số và hô hấp của vi sinh vật đất ở vùng độc canh lúa cao sản

Mật số và hô hấp của vi sinh vật đất được khảo sát từ đất thu thập ở thí nghiệm dài hạn bón phân hoá học cơ trong 20 năm (đất phù sa cuối nguồn ở Viện Lúa, Cờ Đỏ, TP Cần Thơ), bón kết hợp phân vô cơ với hữu cơ trong 6 năm, và đất phù sa đầu nguồn ở Châu Phú, An Giang. Một cách tổng quát, mật số vi sinh vật cao ở đất có bón phân vì hàm lượng carbon hữu cơ cao. Mật số thấp ở đất không bón phân. Ở đất chỉ bón N mật số vi sinh vật thấp hơn đất bón NK, NP và NPK. Mật số vi sinh vật ở đất bón P cao hơn ở đất bón NK, NP và NPK. Mật số cao nhất ở đất có bón PK.

Ở hệ thống độc canh lúa, mật số vi khuẩn cao nhất, kế đến là actinomycetes và tảo. Nấm có mật số thấp nhất.

Sự hô hấp của vi sinh vật được phản ánh bằng hàm lượng CO_2 thải ra từ chúng. Mẫu đất sau khi xử lý được ủ ở 1, 3, 7 và 14 ngày. Dưới sự hấp thu của dung dịch NaOH, lượng CO_2 được đo lường và thấy nó tăng theo thời gian ủ. Lượng CO_2 do vi sinh vật ở đất cuối nguồn tăng dần tới 14 ngày sau khi ủ, trong khi đó ở đất đầu nguồn chỉ tăng tới 7 ngày sau khi ủ.