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UNIVERSITY OF CENTRAL FLORIDA

ETHYLENE DYNAMICS IN THE CELSS BIOMASS PRODUCTION CHAMBER

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ABSTRACT

A material balance model for ethylene was developed and applied retrospectively to data obtained in the Biomass Production Chamber of CELSS in order to calculate true plant production rates of ethylene. Four crops were analyzed ; wheat, lettuce, soybean, and potato. The model represents an effort to account for each and every source and sink for ethylene in the system. The major source of ethylene is the plant biomass and the major sink is leakage to the surroundings. The results , expressed in the units of ppb/day, were converted to nl of ethylene per gram of plant dry mass per hour and compare favorably with recent glasshouse to belljar experiments.

SUMMARY

During crop production (lettuce, wheat, soybean and potato) in the CELSS Biomass Production Chamber (BPC) ethylene can accumulate in the air space and affect plant viability. The chief source of ethylene is the plants which reside in plastic trays containing circulating nutrient solution and the main sink is chamber leakage. There are a variety of other sources and sinks, however , they are small in comparison with the plants and leakage. Accordingly, a material balance model was developed and applied to historical data from the BPC. The data consists of chamber concentration in ppb plotted against time in days. Each plot was curve fitted to obtain an expression for concentration as a function of time. The first derivative was used in the model as the accumulation rate. Production rates in ppb/day were then calculated by adding time averaged leakage rates to the accumulation rate. The time averaging of leakage rates poses a problem in calculating a continuous production rate because some of the leakage is discrete, e.g., door openings and closings. For lettuce the peak production rate was approximately 30 ppb/day whereas wheat had a peak rate of about 80 ppb/day. These numbers were converted to nl/gram-dry mass/hr (46 for lettuce and 15.6 for wheat) and compare favorably with recent glasshouse to bell jar experiments for lettuce and wheat. In addition a dosing experiment was conducted under static conditions wherein 60 ml of pure ethylene was injected into the upper chamber and ethylene concentration was tracked for 18 hours . The ethylene level varied considerably in the 400-700 ppb range. For perfect mixing a level of 1000 ppb was expected. Several explanations can be put forth to account for the lower than expected value. One in particular should be further investigated, i.e., dispersion rates in the BPC under both static and dynamic conditions could shed light on the existence of concentration gradients in the system and the validity of the perfect mixing assumption.

TABLE OF CONTENTS

LIST OF ILLUSTRATIONS

I. Introduction

Ethylene is an important plant hormone which is produced and possibly consumed by plants during their growth cycle. There has been concern over its potential detrimental effect on plant health in closed systems where appreciable levels can build up in the air space of a plant production chamber. For example, parts per billion to parts per million levels can cause one or more of the following: leaf epinasty, flower abortion , stem swelling, adventitious roots, leaf chlorosis, or fruit ripening. Should NASA decide to control ethylene levels in future CELSS (Closed Ecological Life Support Systems) development efforts, or attempt to measure plant production rates of ethylene in closed systems, it will be imperative to account for all ethylene flows in the system. In other words a material balance model is of paramount importance to ethylene quantification. The work described herein is an attempt to develop a working model to include all sources and sinks in the BPC (Biomass Production Chamber).

The material balance is based on the principle of conservation of mass and is applied to the air space in the BPC to compute the transient concentration of ethylene assuming a well mixed chamber. The main sources and sinks in the chamber are the plants, solid materials, and any leakage from or to the chamber. The solid materials can be both sources and sinks, since ethylene can be adsorbed on surfaces as well as outgassed from plastics and sealants. Leakage from or to the chamber has three basic routes: door openings, incomplete sealing of the chamber, and HVAC seals. Experiments were conducted to establish leakage rates for these three modes and the results are employed in the model calculations. In addition, a literature search on off-gassing and binding characteristics of construction materials was initiated to generate rates for the model calculations. Experiments were also conducted in the BPC during which ethylene was measured using gas chromatography. In these experiments there were no plants present in the chamber in order to determine ethylene-material interactions. This information is useful in building the materials component of the model.

With a good understanding of leakage and material interactions, the model was used to analyze previous data obtained in the BPC for lettuce, potato, wheat, and soybean tests. The results are interpreted and discussed in view of ethylenes role in plant physiology.

II. Material Balance Model

A typical plant growth experiment conducted in the BPC includes the measurement (Gas Chromatography with Photoionization Detector through sampling port) of ethylene concentration (ppb) in the chamber air space at daily intervals. Although concentration gradients can exist in the chamber , during these experiments the air handling units were operational and good circulation was maintained (~ 400 cubic meters/min for 113 cubic meters of total BPC volume). Therefore, it is assumed that the chamber air space is perfectly mixed. This means that any ethylene produced is instantaneously distributed throughout the air space. It is also assumed that there are no chemical reactions taking place within the air space itself which could either produce or consume ethylene. If one can account for all sources and sinks of ethylene in the BPC other than from or to the plants, and if the accumulation rate is measured (from concentration vs. time data), then the plant production rate can be obtained by difference.

The general starting equation in words is : Rate of Accumulation = Rate of Input - Rate of Output+ Rate of Generation - Rate of Consumption. If the right hand side is negative then Decumulation occurs and the ethylene concentration decreases with time. Since there is no generation or consumption within the air space the word equation reduces to Rate of Accumulation= Rate of Input - Rate of Output. The input and output terms are the various sources and sinks in the BPC and the accumulation term using the perfect mixing assumption is:

$$\text{Accumulation} = V\frac{dC}{dt}$$

where V is the volume of the chamber air, C is the concentration of ethylene in ppb, and t is time in days. In the following section expressions are developed for the various input and output terms in order to complete the balance so that the production rate can be calculated.

III. Ethylene Sources and Sinks

The sources and sinks are depicted in Figure 1 and are described in detail in this section.

3.1 Plants

The production of ethylene by plants and other microorganisms was first observed sixty years ago(1). However, there is a dearth of data on whole, unadulterated plants in closed chambers. Corey et al.(2) recently measured ethylene evolution by plants grown in closed environments. Lettuce and wheat were grown in a glasshouse and then transferred to sealed bell jars for measurement of ethylene. For lettuce the production rate ranged from 20 to 200 nl/g dm/hr, whereas wheat ranged from 5 to 30 nl/g/hr. Care must be taken in comparing these results with the BPC, since the plant mass to head volume ratio in a bell jar is different from the ratio in the BPC. (The BPC has a high volume to plant area ratio of ~5.6:1)

3.2 Materials

There are two mechanisms for material exchange; outgassing and sorption. In a Lockheed study (3) the steady-state outgassing rate of ethylene for all Space Station equipment was estimated to be 1.2 mg/day. On a per mass of non-metallic material basis this rate is 0.000214 mg ethylene/kg/day; however, it is based in part on tests conducted at 322 degrees Kelvin which is about 30 degrees higher than BPC temperatures. Even if the BPC materials outgassed at this rate it would be less than 1 ppb/day. According to Johnson (4) , however, there should be negligible outgassing in the BPC since the major plastic components are PVC and ABS, not polyethylene. Also, Johnson claims that ethylene is outgassed in significant amounts from polymers only at highly elevated temperatures (with other degradation products). Therefore, in the material balance for the BPC (substantial plant mass and minimal outgassing conditions) outgassing will be neglected.

With regard to sorption , an experiment was conducted in the BPC which involved the measurement of ethylene concentration in the head space , with no plants present and the air handler in the off mode. The leakage rate under these conditions was determined to be 1% per day (see Figure 2 and section 3.3), therefore, the exchange of ethylene between the air space and any material surfaces in the biochamber could be ascertained. Over an 8-hour period under the static conditions of the chamber the ethylene concentration remained fairly constant. This suggests that the material-gas interface may have been saturated with ethylene. In a related experiment conducted by Dubay (5) 7500 ng of ethylene were injected into a 65-liter chamber consisting of a PVC tray covered with plexiglass. At 6 hours an equilibrium was established between the air space and surfaces of the chamber such that the concentration of ethylene in the air space was 47 ppb. The difference between the amount injected and the amount in the air space divided by the estimated surface area of the chamber yields 3237 ng ethylene/ square meter of surface adsorbed at equilibrium. In order to establish whether the surface could have adsorbed additional molecules, it would have been necessary to inject more ethylene into the chamber after the 47 ppb equilibrium. However, this result is a good upper limit for the amount of ethylene adsorbed by the plastic in the BPC since 47 ppb is sufficiently higher than the average concentration in the BPC during a typical plant cycle. Also, based on the work of Kamiya et al. (6), lower surface concentrations can be calculated for ethylene sorbed by polybutadiene.

Using a value of 70 square meters for the total plastic air interface in the upper chamber the surface can sorb about 3 ppb. Since a typical plant growth cycle is a minimum of 30 days, the amount adsorbed averaged over the entire plant cycle is less than 0.1 ppb/day. Thus sorption will be neglected in this study since ethylene production rates are subsequently shown to range from a few ppb/day all the way up to 80 ppb/day.

3.3 Ambient Air

There are three conditions that exist in the BPC which have a major bearing on leakage of air from the system when plant growth occurs. In the normal mode of operation during plant growth, the chamber door is closed and sealed and sealant is applied to all necessary areas such that the only leakage route is most probably through the air handling unit. The other two modes are a closed, unsealed chamber and an open chamber resulting from an open door. In an average plant growth cycle the normal mode is maintained for 98% of the time. The closed , unsealed state is estimated to exist for 1.6% ot the cycle and the door is in the open state 0.4% of the time. In order to measure the leakage rate for each of these modes experiments were conducted to track the concentration of injected carbon dioxide over time. For a well mixed air space the rate of carbon dioxide decline should equate with the leakage rate. The carbon dioxide decay (Figures 3 and 4) yields the following rates: closed and sealed-10% of the chamber volume per day; closed and unsealed-77%; and open chamber-13,100%. Linear combination of these rates results in an overall leakage rate of 63% of the chamber volume per day. It is this rate which will be used in the material balance model even though it is an average, continuous rate, i.e., leakage occurs in a non-continuous manner during 2% of the cycle.

3.4 Gas Cylinders

Carbon dioxide is metered into the BPC on a daily basis to maintain a constant level for plant growth. A typical rate is approximately 200 liters per day from a gas cylinder. According to Eastwell et al. (7), carbon dioxide cylinders contain 20 ppb of ethylene although no ethylene was found in cylinders used in the BPC at the 10 ppb detection limit. Twenty ppb would introduce about 0.1 ppb/day of ethylene to the BPC upper chamber. Therefore, this source shall be neglected in this study.

3.5 Nutrient Solution

Ethylene is soluble in water so the nutrient solution that is circulated through the plant trays represents a sink for ethylene. The Henry's Law constant for ethylene at 0 degrees C is 5,520 atm/mole fraction. If the concentration of ethylene in the air space were 100ppb then the equilibrium mole fraction of ethylene in water would be 0.00000000002. For 250 liters of nutrient solution (14,000 gmoles water) the water would hold 28 x 10^{-8} gmoles ethylene. At STP this corresponds to 627x10^{-8} liters of ethylene. The chamber volume is 56,000 liters,therefore, the total sink would be 0.1 ppb. Since the nutrient solution (contains 100 liters of condensate) is recycled and has a lower capacity for ethylene than pure water it represents a negligible sink for ethylene and is ignored in this study.

3.6 Removal Systems

There are a variety of contaminant control methods such as catalytic convertors, activated carbon adsorption and bypass configurations. Currently, potassium permanganate is used to remove ethylene from the BPC, however, during the experiments to measure ethylene production rates, it was not employed so removal is not included in the material balance calculations. If a removal system is employed during the growth cycle it can be incorporated into the material balance if a rate expression is available for the method employed. Removal would then behave as a sink in the balance equation.

3.7 Dosing

The upper chamber of the BPC was maintained in a closed, sealed state with the air handlers off after 60ml of pure ethylene was injected by hand into the air space. A small circulating fan was operated to disperse the gas. Ethylene level was then measured by GC with photoionization detector. Ethylene fluctuated between 400 and 700 ppb during an 18 hour period (Figure 5). If all the ethylene injected were uniformly dispersed in the BPC a concentration of 1000 ppb would be expected (even higher levels could be expected if the injected ethylene didn't reach the ducts of the air handler units).Perhaps some ethylene escaped in exiting the BPC after injecting the ethylene and/or the fan was inadequate for dispersion purposes resulting in stagnant pockets of undiluted ethylene. Based on the previous calculation for the sorption sink, it is unlikely that the missing ethylene sorbed to the surfaces inside the BPC.

Dosing did not occur during the experiments (historical data) presented in Figure 6 so it is not included in the material balance calculation in this study. It could be included in future experiments and calculations as a spike, or instantaneous change in the chamber concentration if the dose instantaneously disperses throughout the chamber. Then the type of analysis outlined in the next section can be applied to plant response.

3.8 Sampling

The chamber air is continuously sampled for carbon dioxide. The sampling rate is .75 liters per minute and the ethylene , in all likelihood , is completely consumed as it hits the platinum catalyst within the sampler at high temperature. This rate is equivalent to 1.9% leakage and will affect the production rate by about 0.4 ppb/day on average. It is not included in the analysis , but could easily be incorporated by adjusting the leakage rate.

IV. Analysis

The objective is to calculate ethylene production rates for different crops in the BPC. The first step in the calculation involves a curve-fit of the concentration vs. time data for different crops in order to develop an expression for the accumulation rate (proportional to the derivative of the curve). The leakage rate can then be added to the accumulation rate to yield the production rate. The data to be analyzed is presented in Figure 6.

The first step in the analysis involved an eyeball smoothing of the data in Fig.6 followed by an equation fit to the smoothed curve using Sigma Plot. An expression for the first derivative of the equation was then calculated and used to compute the accumulation rate for different times. Then the leakage rate expression was added to the accumulation rate to give the instantaneous production rate during the plant growth cycle. The following is an example for lettuce:

For lettuce the production rate is assumed to be first order in C(ppb), i.e., production rate (P) is equal to a constant times the concentration of ethylene at any time, t(days) , i.e., P=k C/V. The leakage rate (L) is also first order, i.e., L=.63 C, where V=56,000 liters (upper chamber volume). The material balance equation for lettuce then becomes

$$V\frac{dC}{dt} = kC - .63VC$$

or , dividing both sides by V:

$$\frac{dC}{dt} = (k/V - .63)\,C$$

If we let $a=(k/V-.63)$ and if at time zero C=1 ppb then $C=\exp(a\,t)$.

This last equation is used to fit the data with a= 0.1235 giving the best fit(Fig.7). The value of a from the curve fit can now be used to solve for the production rate constant, k. Then at any time t the production rate in ppb/day can be computed simply by multiplying k/V by the concentration at t. The results for four different times are

t, days	P, ppb/day	L, ppb/day
0	.75	.63
10	.85	.71
20	8.9	7.5
30	30.5	25.5

and are plotted in Figure 8.

Similar computations were made for the three other crops and the results for these rates are presented in figures 9 through 14. For any crop as long as the production rate is greater than the leakage rate accumulation occurs and the concentration rises in the chamber. When P is equal to L the concentration remains constant and when L becomes larger than P the concentration declines.

V. Discussion of Results

It is important to keep in mind that the leakage rate was taken to be continuous, i.e, any sudden leakage (e.g., door opening) was distributed uniformly over the entire growth cycle. Sudden leakage, such as a door opening for one minute, can cause a 10% drop in chamber concentration and may explain some of the variation in the concentration verses time data , especially for wheat. Therefore, at certain times during the cycle the production rate may have been higher or lower (perhaps even negative, i.e., the plants could have consumed ethylene during a period of declining concentration or may even have sorbed ethylene) than the calculated values. On the other hand, if ethylene production is not dependent on concentration in the BPC, i.e., if it correlates with plant growth in an independent manner , then the leakage rate will not affect the production rate. For this reason it would be desirable to conduct an experiment at different continuous leakage rates , to determine any dependency. Also, it is desirable to avoid door openings for as long a period as possible (e.g., 7-10 days for wheat as the peak is approached) to reduce concentration variations.

The results for ethylene production can be converted to peak rates per dry biomass and compared with other data on ethylene production. Figure 15 shows the dry biomass for the different crops. The peak value for lettuce is about 3 kg of dry mass for the total of upper and lower biochambers. Therefore the upper value is taken to be 1.5 kg of drymass, i.e., one half of the total. At the peak lettuce is producing 30 ppb/day or 1,680,000 nl/day. This yields a value of 46 nl/g dm /hr , which is in the range of Corey's (2) data. Similarly, for wheat a value of 15.6 nl/g/hr is obtained which also falls within Corey's range. Since Corey found rates as high as 200 nl/dryg/hr for lettuce , this might suggest that the glasshouse to bell jar experiments were more stressful to the crop and more ethylene was produced accordingly.

VI Conclusions and Recommendations

The ethylene production rates calculated here are in good agreement with the limited closed system data in the literature. Whether ethylene production correlates best with plant growth and is independent of concentration in the BPC or whether a concentration dependent kinetic model (e.g., first order) best describes the system remains an open question. This question might be resolved by conducting experiments at different continuous leakage rates as previously mentioned or by dosing the chamber. In a dose experiment, concentration independence (plant biomass correlation) would be established if the production rate is unaltered by injection of ethylene. However, in the dose experiment where 60 ml of pure ethylene was injected into the BPC, there were problems in accounting for all the ethylene injected. Therefore, it would be desirable to perform dispersion experiments, perhaps using solid carbon dioxide, to establish diffusional patterns in the BPC to determine whether concentration gradients exist. This could be done in both the static and dynamic modes (air handlers on) to confirm the ideal mixing assumption of the model. If ethylene can be injected and accounted for in the dynamic mode then a dose experiment could resolve the concentration dependency question.

VII References

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4) Johnson, H. Personal Communication from NASA WSTF. July, 1994.

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6) Kamiya, Y, K. Terada , K. Mizoguchi, and Y. Naito. Sorption and Partial Molar Volumes of Organic Gases in Rubbery Polymers. Macromolecules. Volume 25. 1992. pp4321-4324

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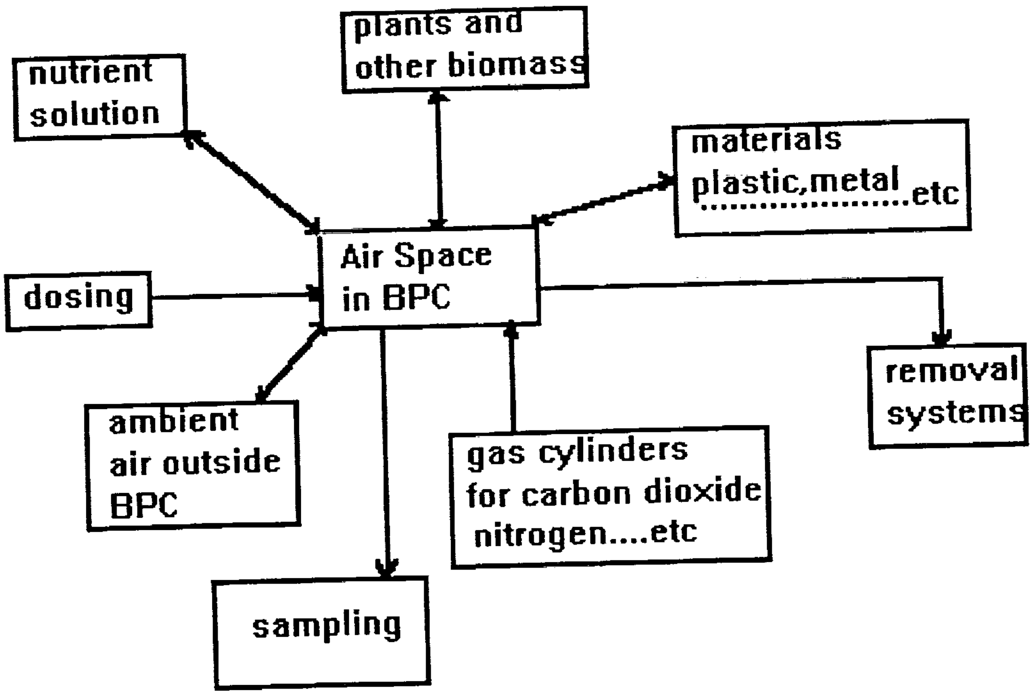


Ethylene Sources and Sinks

Figure 1

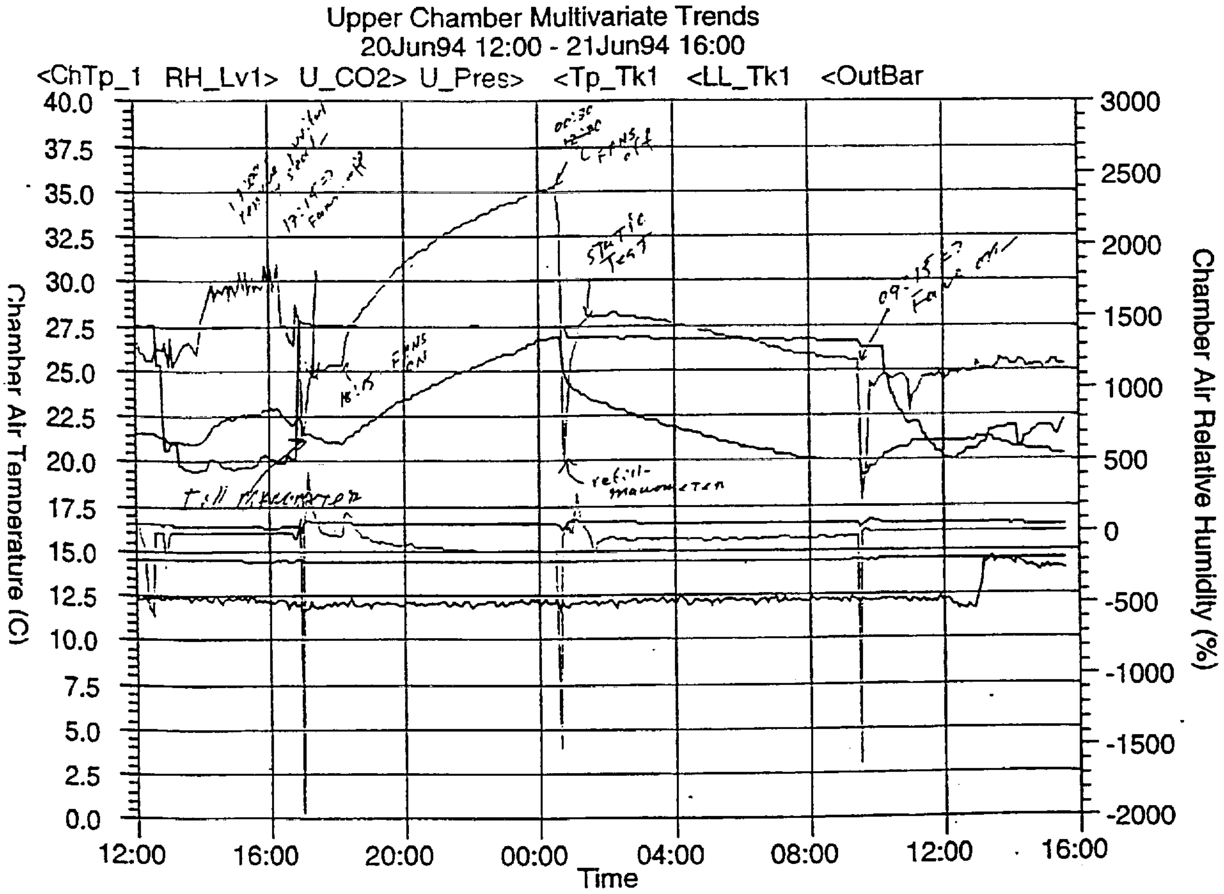

Upper Chamber Multivariate Trends
20Jun94 12:00 - 21Jun94 16:00
<ChTp_1 RH_Lv1> U_CO2> U_Pres> <Tp_Tk1 <LL_Tk1 <OutBar
Chamber Air Temperature (C)
40.0
37.5
35.0
32.5
30.0
27.5
25.0
22.5
20.0
17.5
15.0
12.5
10.0
7.5
5.0
2.5
0.0
Chamber Air Relative Humidity (%)
3000
2500
2000
1500
1000
500
0
-500
-1000
-1500
-2000
12:00
16:00
20:00
00:00
04:00
08:00
12:00
16:00
Time
17:14 => Fans off
18:15
FANS ON
00:30
~~12:30~~
FANS off
STATIC TEST
refill manometer
fill manometer
09:15 => FANS ON

Figure 2

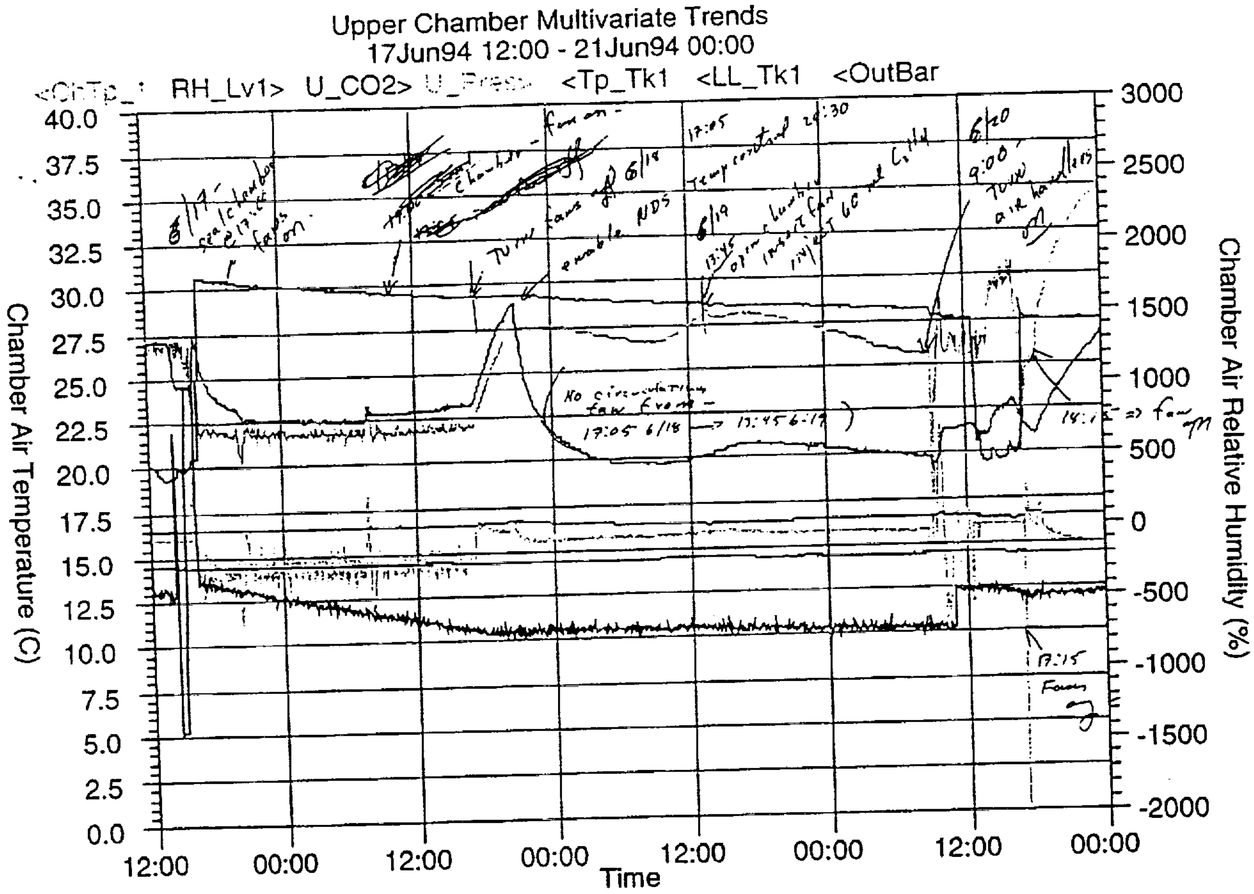

Upper Chamber Multivariate Trends
17Jun94 12:00 - 21Jun94 00:00
<ChTp_1 RH_Lv1> U_CO2> U_Press> <Tp_Tk1 <LL_Tk1 <OutBar
Chamber Air Temperature (C)
40.0
37.5
35.0
32.5
30.0
27.5
25.0
22.5
20.0
17.5
15.0
12.5
10.0
7.5
5.0
2.5
0.0
Chamber Air Relative Humidity (%)
3000
2500
2000
1500
1000
500
0
-500
-1000
-1500
-2000
12:00
00:00
12:00
00:00
12:00
00:00
12:00
00:00
Time
6/17 - seal chamber @ 17:00 fans on
~~17:00 chamber - fans off~~
Turn fans off 6/18
fans on -
enable NDS
17:05
Temp control 20:30
6/19
13:45 open chamber insert fan inject 60 ml CH4
6/20
9:00 - Turn air handler on
No circulating fan from 17:05 6/18 → 13:45 6-19
14:1 => fan off
17:15 Fans off

Figure 3

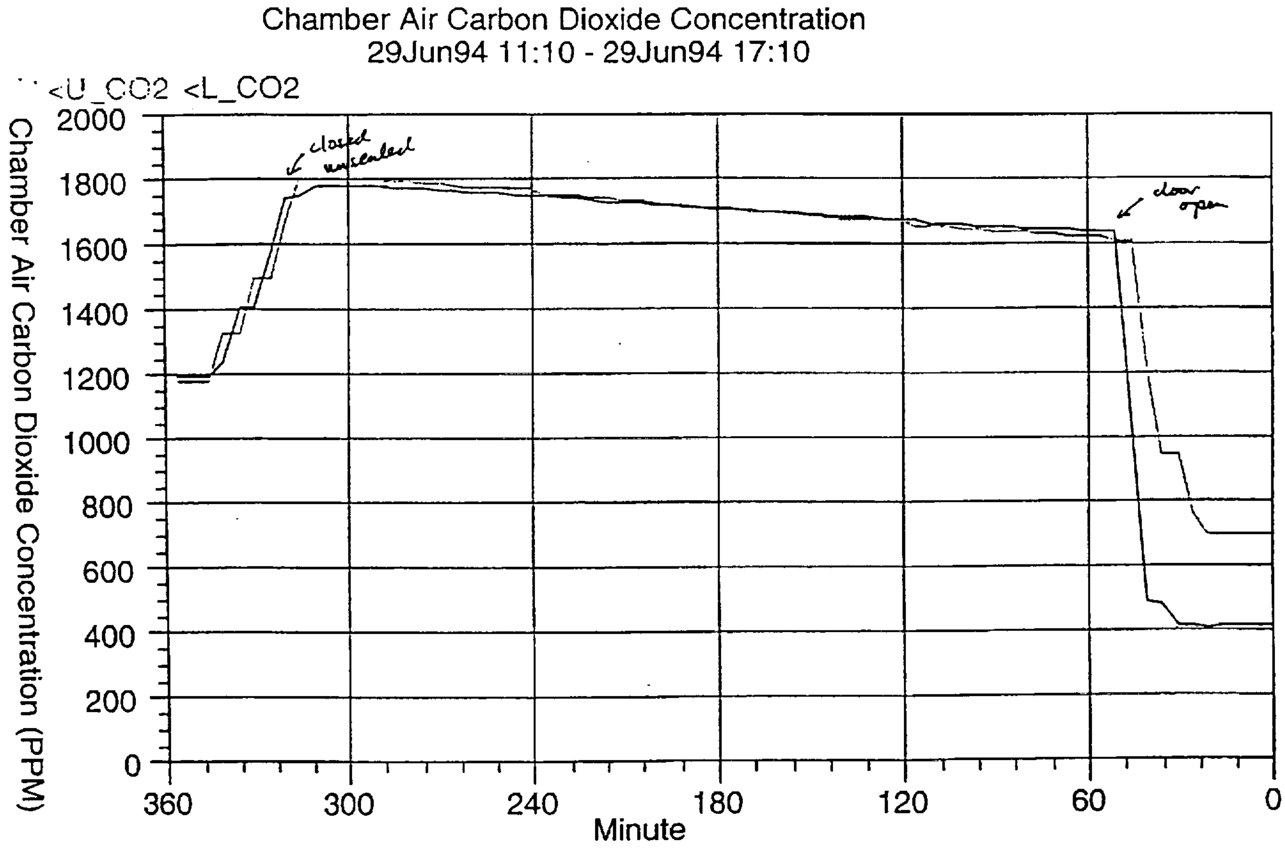

Chamber Air Carbon Dioxide Concentration
29Jun94 11:10 - 29Jun94 17:10
<U_CO2 <L_CO2
Chamber Air Carbon Dioxide Concentration (PPM)
2000
1800
1600
1400
1200
1000
800
600
400
200
0
360
300
240
180
120
60
0
Minute
closed unsealed
door open

Figure 4

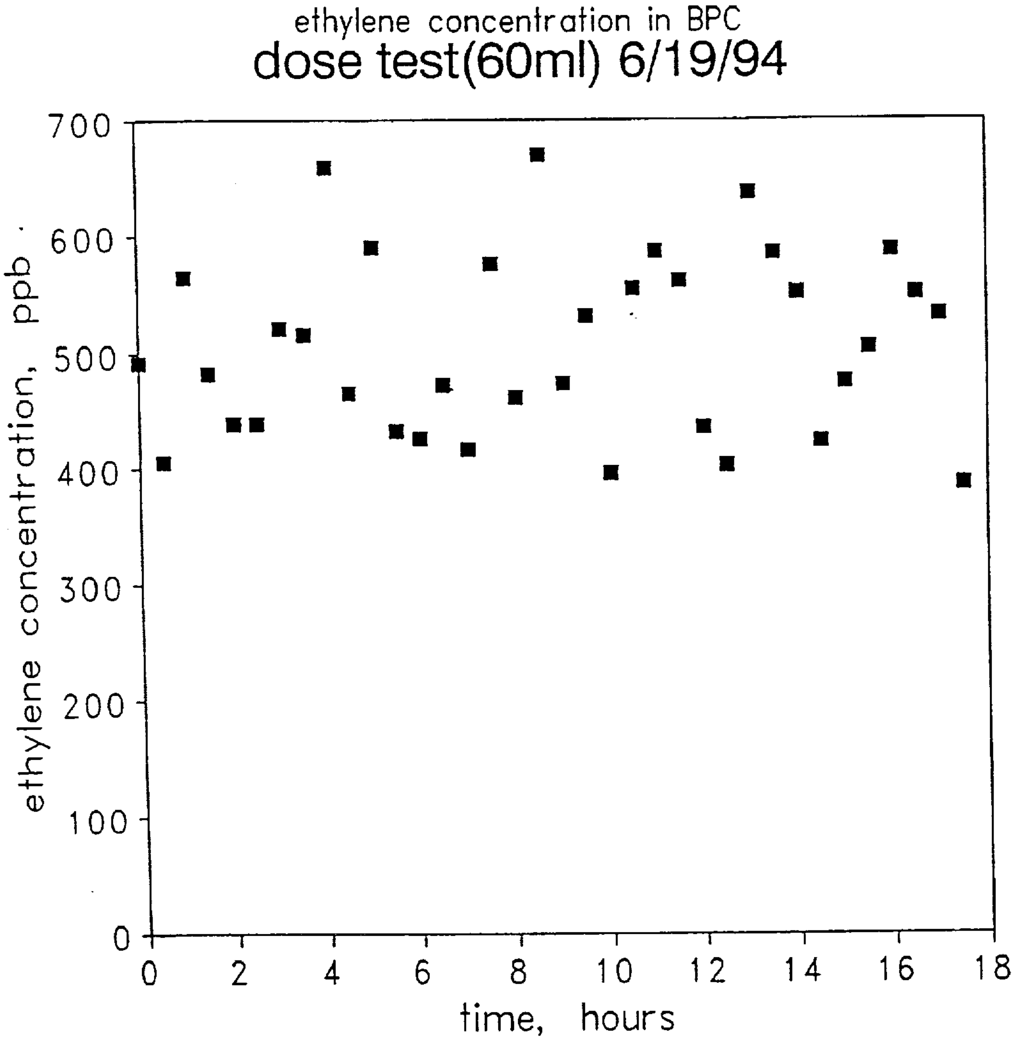

ethylene concentration in BPC
dose test(60ml) 6/19/94
ethylene concentration, ppb
700
600
500
400
300
200
100
0
0
2
4
6
8
10
12
14
16
18
time, hours

Figure 5

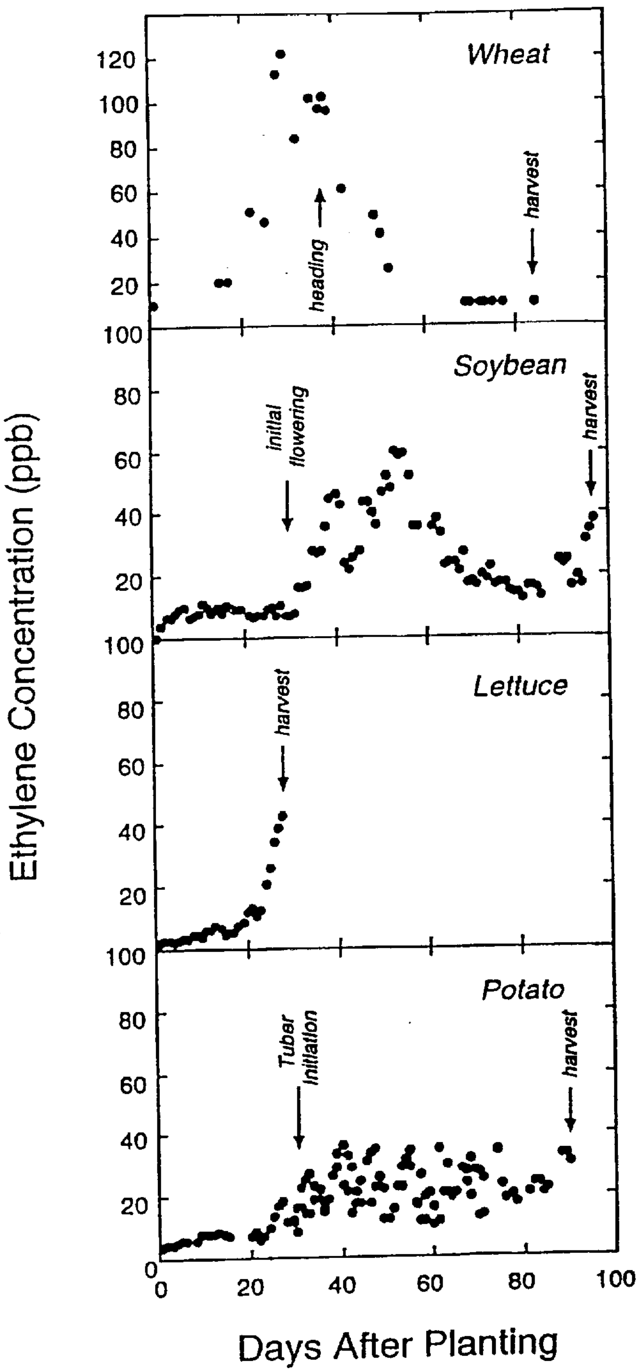

Wheat
heading
harvest
Soybean
Initial flowering
harvest
Lettuce
harvest
Potato
Tuber Initiation
harvest
Ethylene Concentration (ppb)
120
100
80
60
40
20
0
20
40
60
80
100
Days After Planting

Figure 6

Ethylene Concentration in BPC (Lettuce)

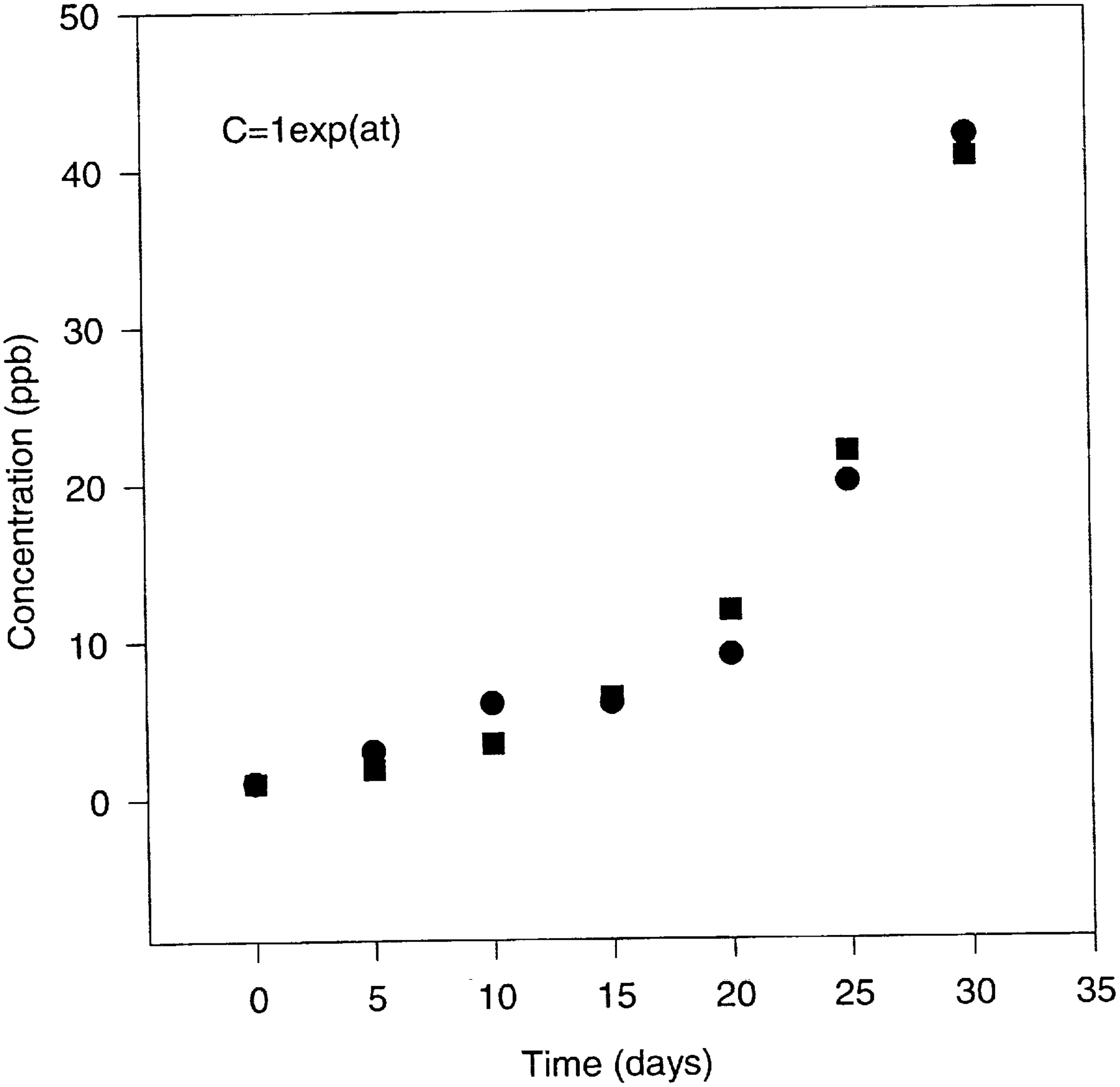


Figure 7

Ethylene Production Rate (Lettuce)

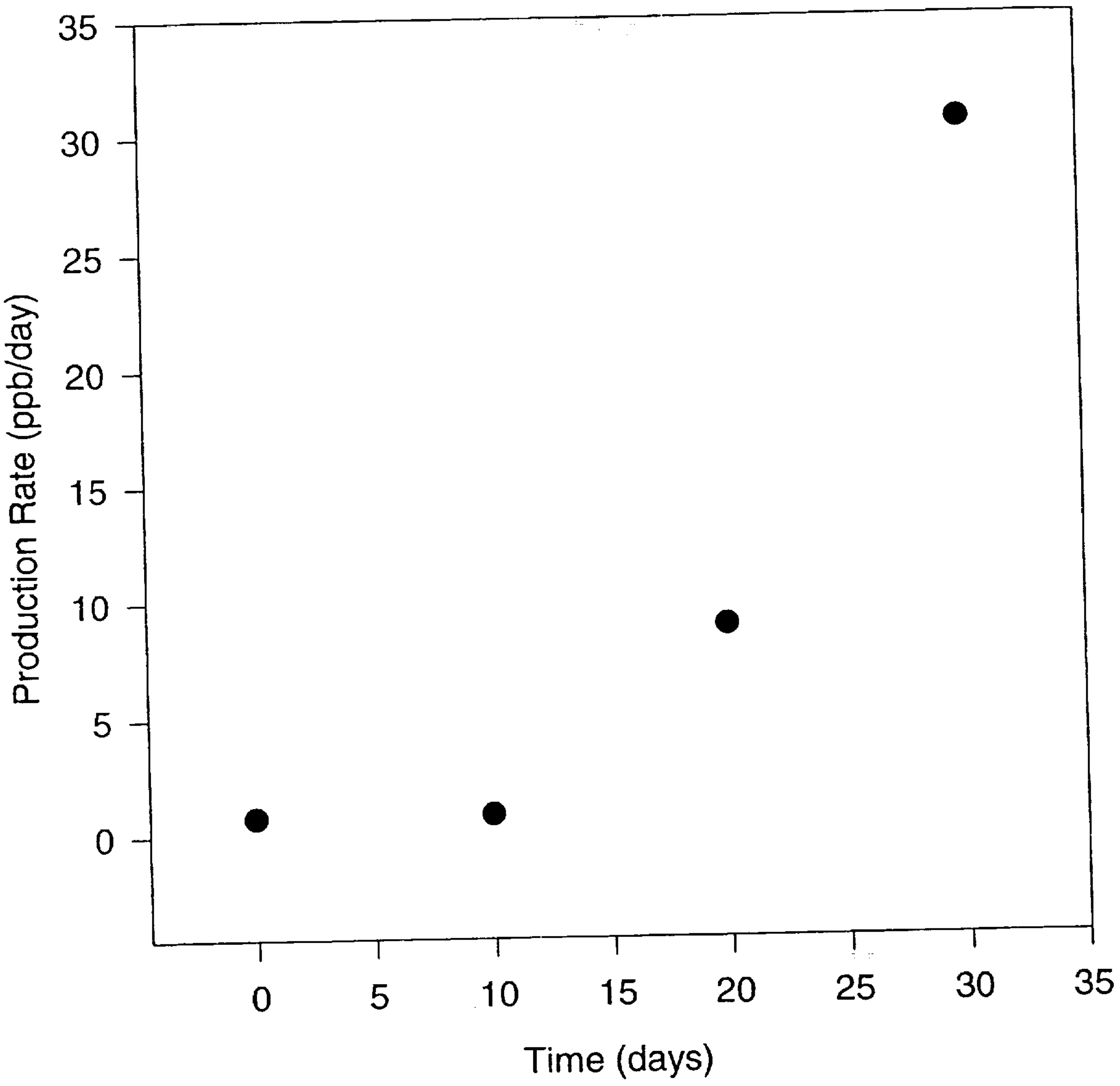


Figure 8

Ethylene Concentration in BPC (Wheat)

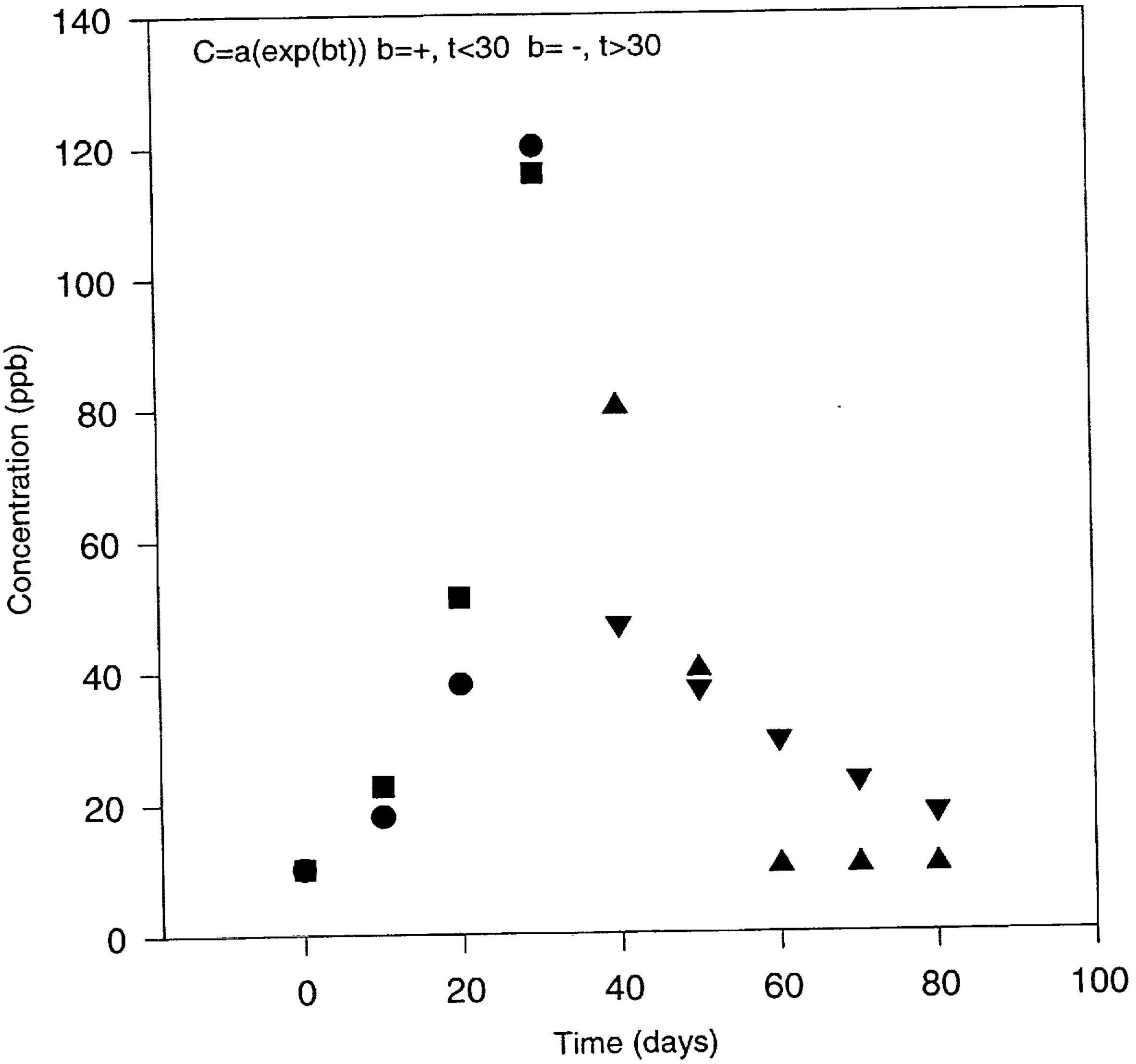


Figure 9

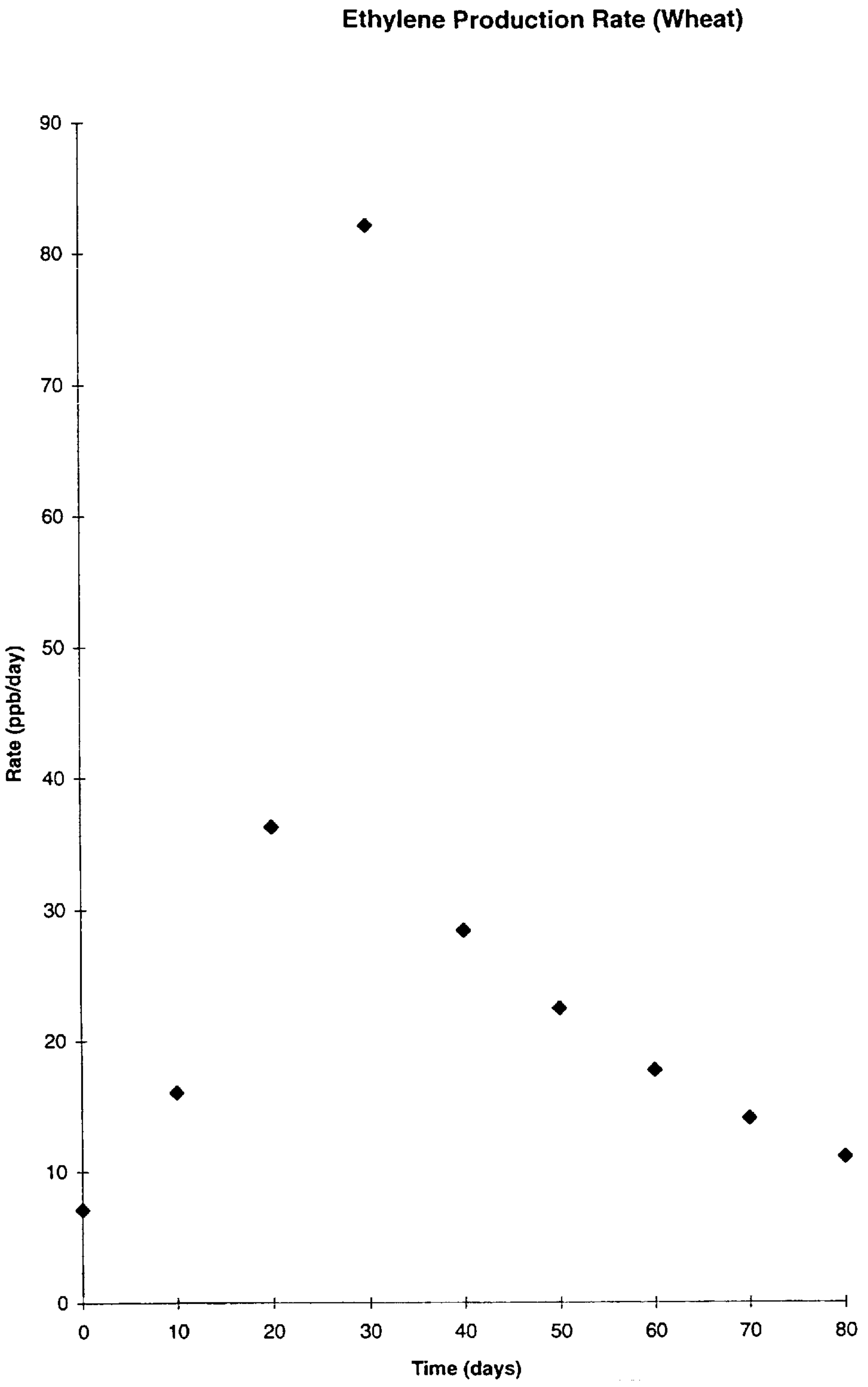

Ethylene Production Rate (Wheat)
Rate (ppb/day)
90
80
70
60
50
40
30
20
10
0
0
10
20
30
40
50
60
70
80
Time (days)

Figure 10

Ethylene Concentration in BPC (Potatoes)

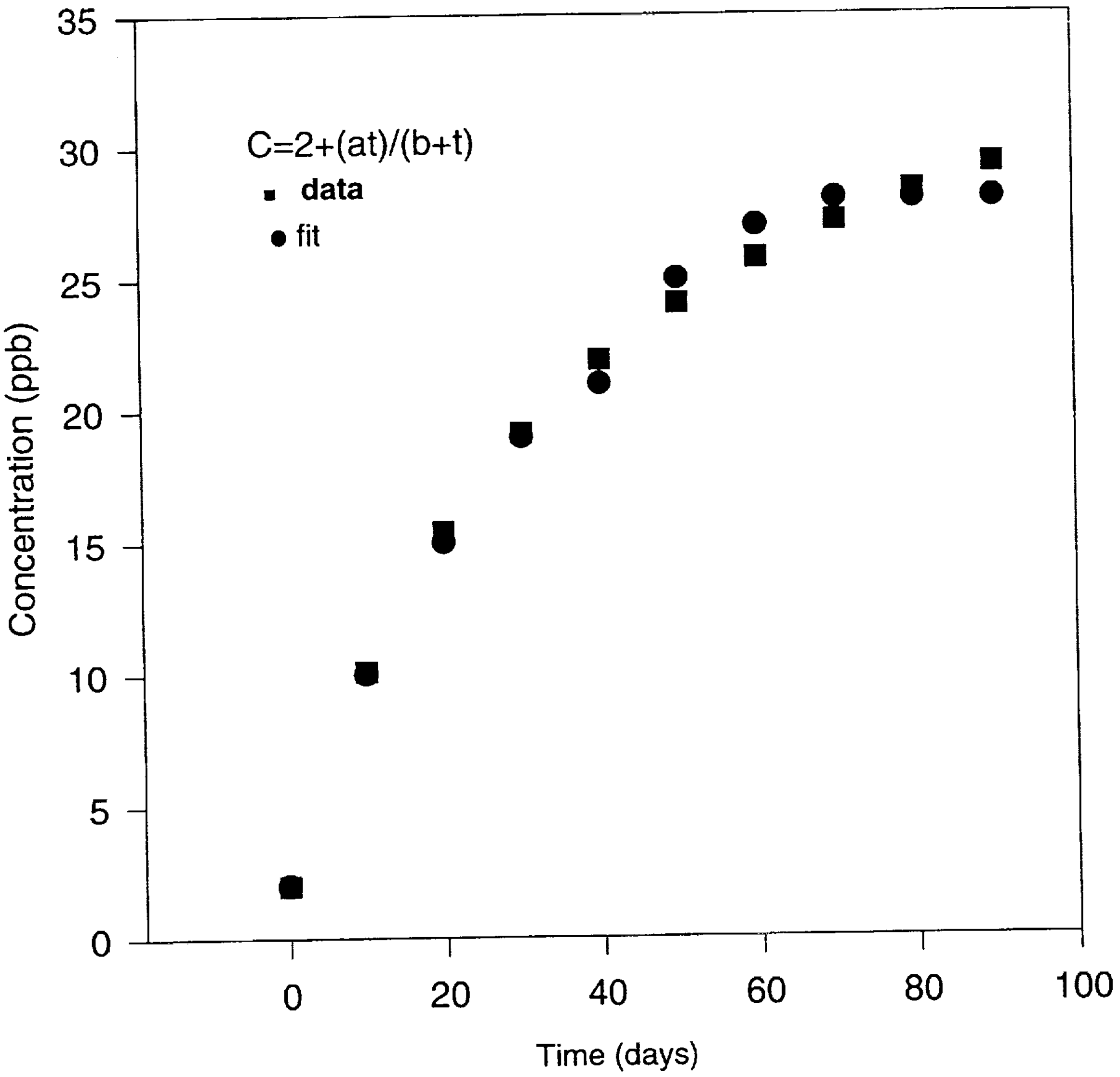
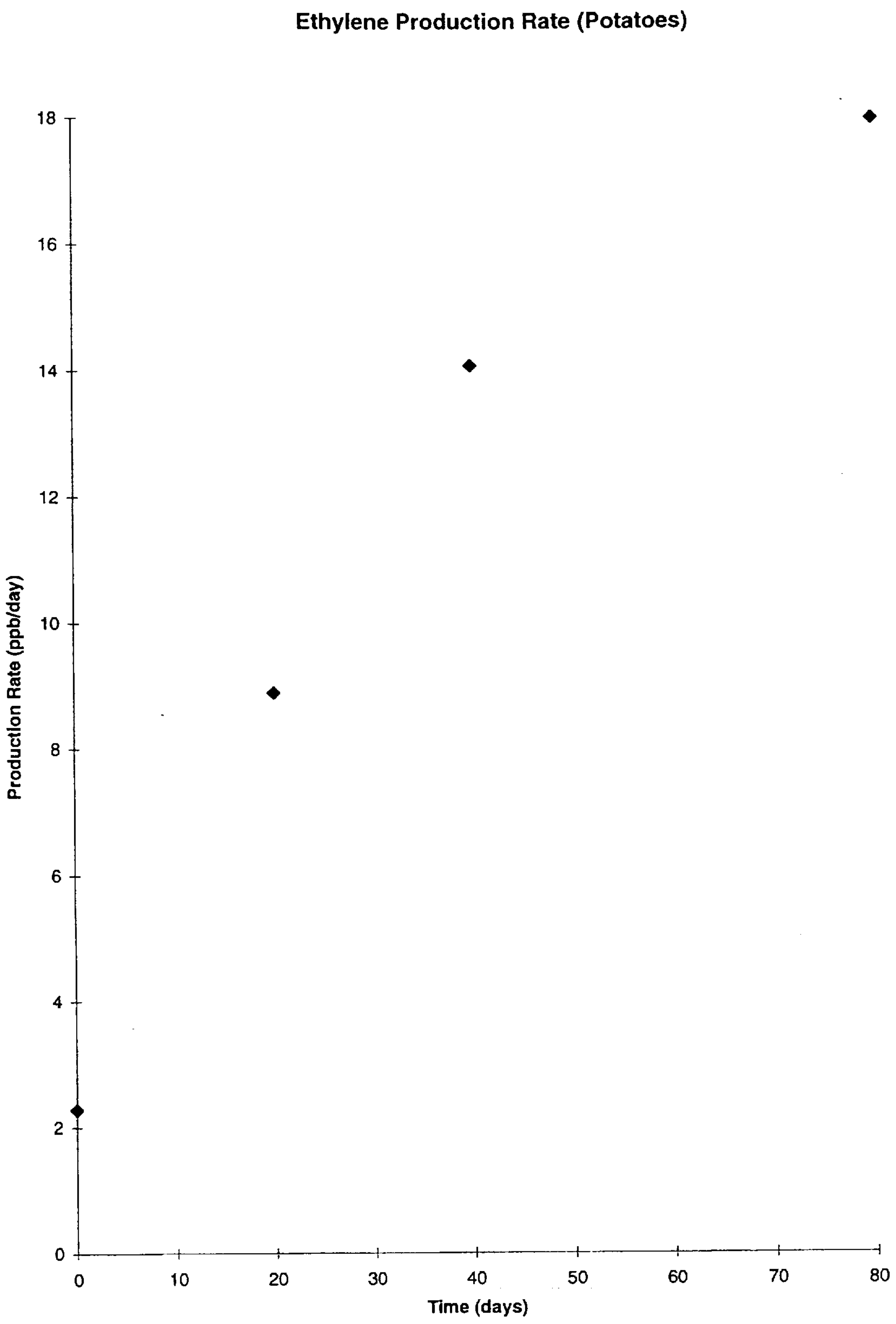


Figure 11



Ethylene Production Rate (Potatoes)
18
16
14
12
10
8
6
4
2
0
Production Rate (ppb/day)
0
10
20
30
40
50
60
70
80
Time (days)

Figure 12

Ethylene Concentration in BPC (Soybean)

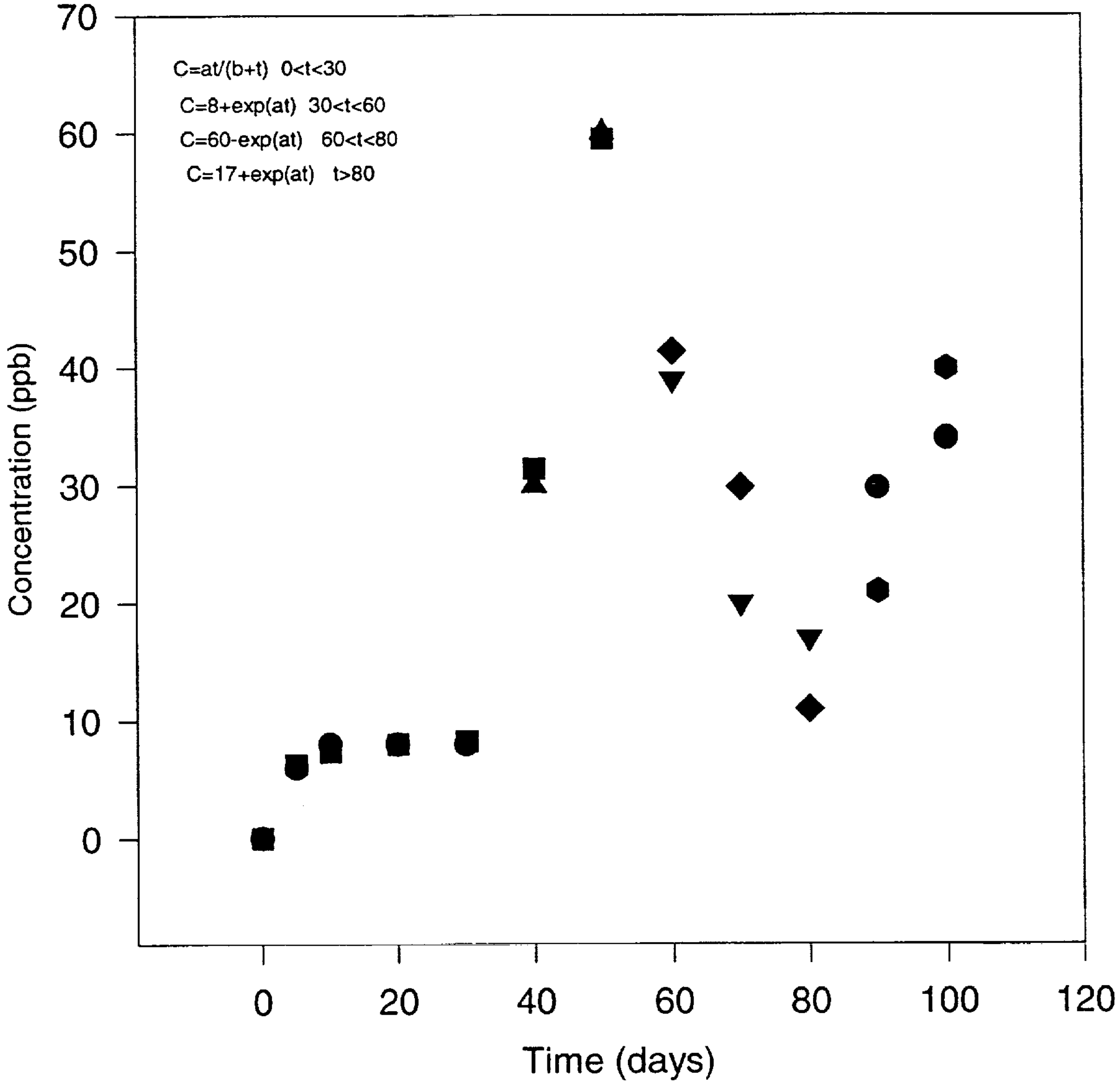


Figure 13

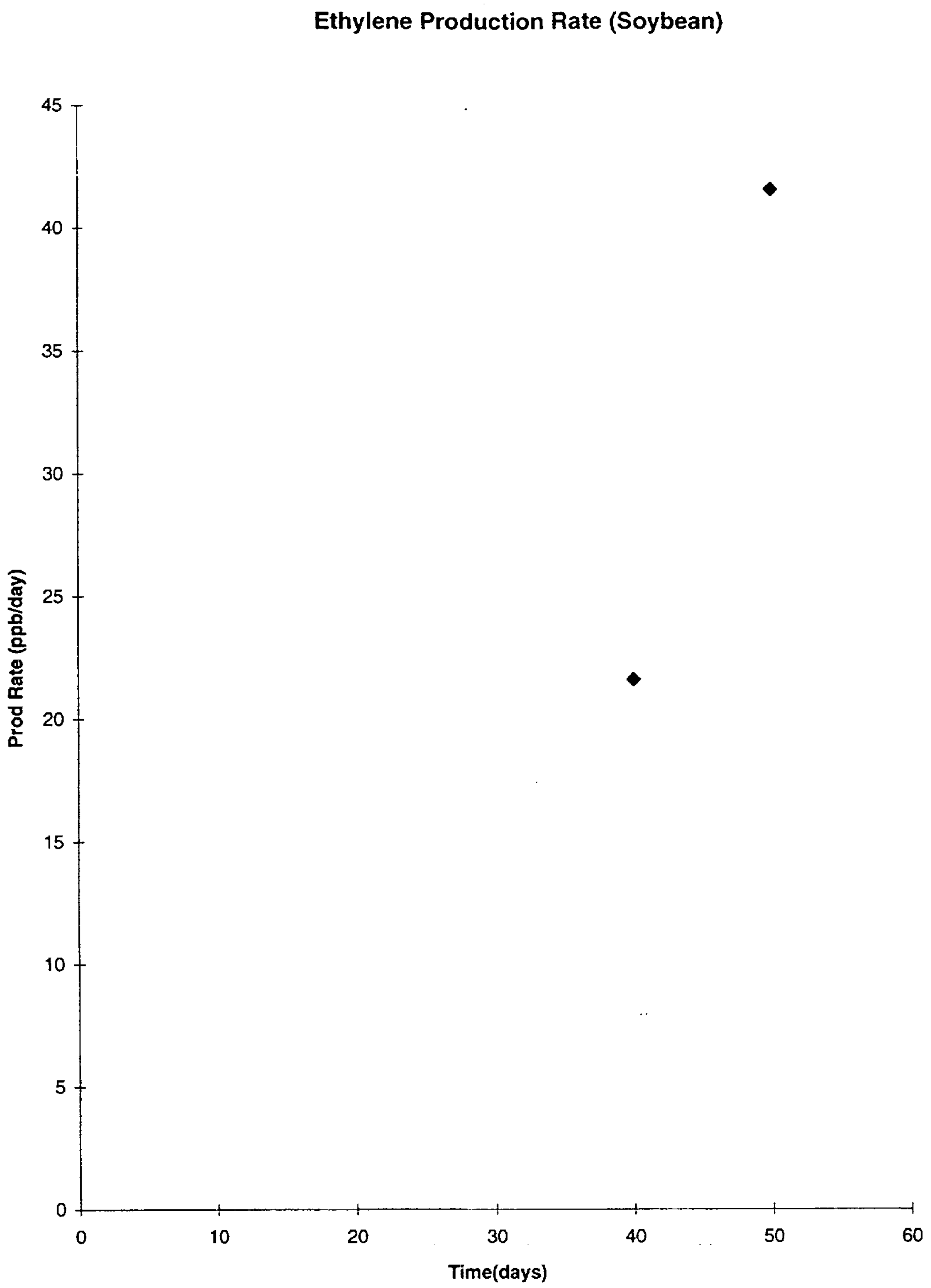

Ethylene Production Rate (Soybean)
45
40
35
30
25
20
15
10
5
0
Prod Rate (ppb/day)
0
10
20
30
40
50
60
Time(days)

Figure 14

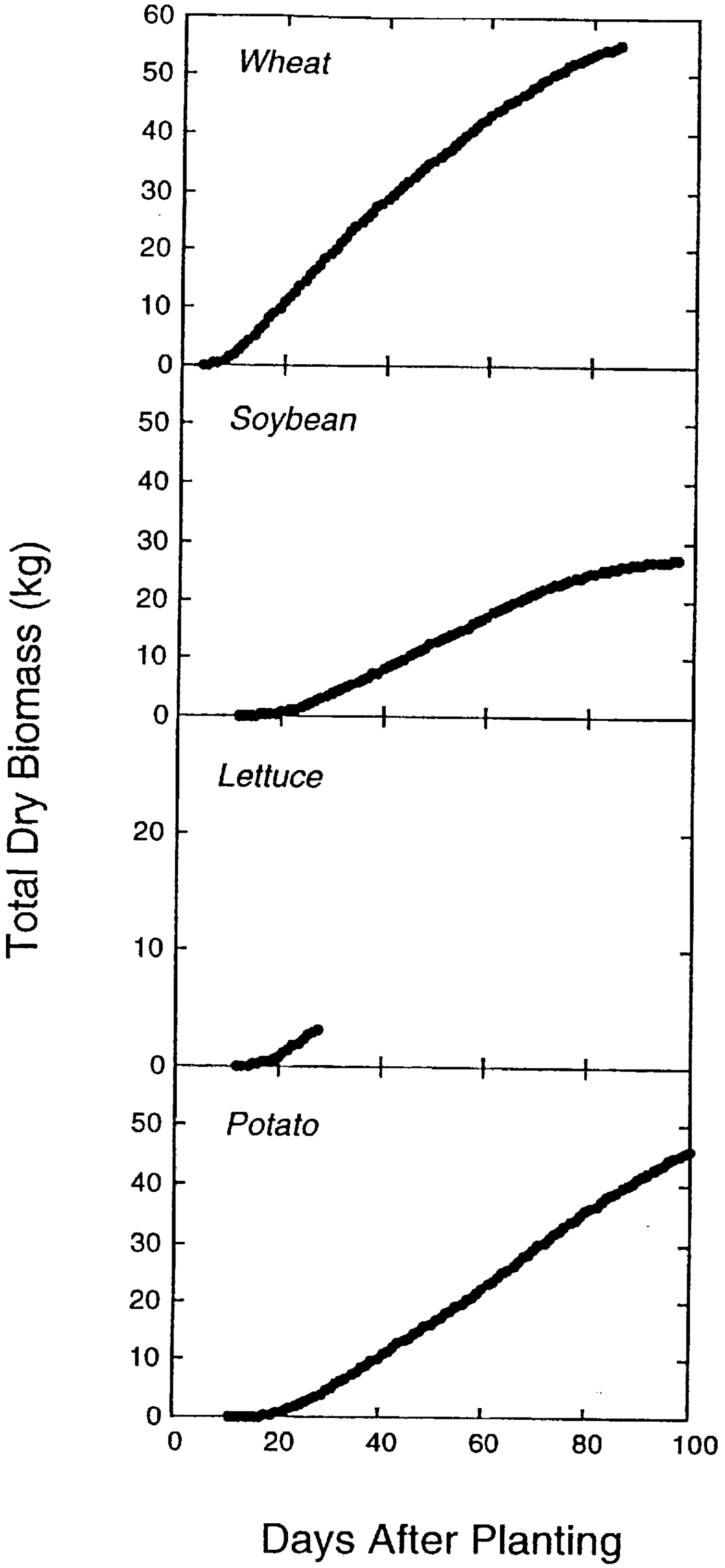

Total Dry Biomass (kg)
Wheat
60
50
40
30
20
10
0
Soybean
50
40
30
20
10
0
Lettuce
20
10
0
Potato
50
40
30
20
10
0
0
20
40
60
80
100
Days After Planting

Figure 15