

## NOx Gas Absorption in the Green Tobacco

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*(Received February 24, 2004)*

**ABSTRACT** : On-going research has recently documented the certain tobacco specific nitrosamines (TSNAs) are formed during the curing process by an interaction of nitrogen oxides (NOx) contained in combustion gases and naturally occurring compounds in the tobacco leaves. Although the role of TSNAs in human health have been extensively investigated, little research has been conducted on the physical and chemical phenomena relating to their formation during curing. In this paper, we developed a mathematical model for describing NOx absorption into green (uncured) tobacco. We found considerable uptake of NOx by green tobacco with variations related to several factors. Specifically, tobacco from the lower stalk positions (bottom one third) absorbs more NOx gas per unit weight than tobacco from upper stalk positions. Additionally, the green tobacco packed with a density of 0.103 g/cm<sup>3</sup> absorbs more NOx gas per unit weight than either 0.0443 g/cm<sup>3</sup> or 0.0739 g/cm<sup>3</sup>. Further, the NOx absorption increases proportionally with temperature with the maximum absorption point around 40°C.

**Key words** : tobacco, TSNAs, NOx, mathematical model

### Notation

|                |   |
|----------------|---|
| $c$            | concentration (ppb)                                       |
| $D_{AB}$       | binary diffusivity for system A, B (cm <sup>2</sup> /min) |
| $k$            | constant (min <sup>-1</sup> )                             |
| $k'$           | reaction rate constant (min <sup>-1</sup> )               |
| $K_n$          | equilibrium rate constant for reaction n                  |
| $R$            | rate of accumulation (ppb/min)                            |
| $\bar{U}$      | mass average velocity (cm/min)                            |
| $\bar{\nabla}$ | the del operator  |
| $t$            | time (min)  |

### Subscripts

|     |                                |
|-----|--------------------------------|
| $0$ | initial NOx gas in the chamber |
| $A$ | component A of NOx gas         |
| $n$ | number                         |
| $S$ | equilibrium state              |

### Introduction

Since the first mention in the scientific literature in 1962, many investigators have tried to discover the role and characteristics of tobacco specific nitrosamines (TSNAs) (Dong et al., 1978; Johnstone et al., 1962; Neurath and Ehmke, 1964). To date, most research has centered on the role of TSNAs in human health and the qualitative analysis of TSNAs in leaf tobacco. Recently, it has become known that green (uncured) tobacco leaves contain little to no TSNAs and the TSNAs found in cured leaf may be formed by several different mechanisms during curing and storage. On-going research has therefore become more

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focused on the understanding these mechanisms and how they may be controlled. Peele and Gentry (1999) have demonstrated that nitrogen oxides (NO<sub>x</sub>) gas, a product of combustion found in all direct fired curing barns, reacts with certain alkaloids in green tobacco leaves to form TSNA's. In air-cured burley tobacco, Andersen et al. (1987) has reported that various factors may influence TSNA's formation including curing environment, harvest dates and stalk position. Others have conducted research on the effect of fertilization rates, genotype and specific curing conditions on level of TSNA's in air-cured tobacco (Long et al., 1999; Wahlberg et al., 1999). Although some works on TSNA's formation in flue-cured tobacco during the process of curing have been published (Boyette and Hamm, 2001; Nestor et al., 2003), the actual dynamics on the movement of NO<sub>x</sub> into the leaf is poorly understood. Considering the serious effects on human health, it is important to research NO<sub>x</sub> absorption in curing leaves as it relates to TSNA's formation. By understanding the mechanisms of diffusion, adsorption and reaction in the green leaf, we stand a much better chance of developing strategies to limit levels of TSNA's in cured leaf. In this study, we have investigated the temporal change on NO<sub>x</sub> levels in a closed chamber containing green tobacco as a way to understand the dynamics of NO<sub>x</sub> absorption. From the data, we suggest a mathematical relationship describing NO<sub>x</sub> absorption.

## Materials and methods

An experimental apparatus like the one shown schematically in Fig. 1 was used to determine NO<sub>x</sub> absorption in green tobacco. Two kinds of NO<sub>x</sub> gases were used for this experiment. One of them was a mixture of nitric oxide (NO) of 4.07 ppm and nitrogen dioxide (NO<sub>2</sub>) of 0.39 ppm in nitrogen (NO<sub>x</sub> of 4.46 ppm). Another one was a

mixture of NO of 5.17 ppm and NO<sub>2</sub> of 0.18 ppm in nitrogen (NO<sub>x</sub> of 5.35 ppm). Each NO<sub>x</sub> gas at a known concentration was metered into a closed thermostatically controlled chamber (Vacuum Oven, Model 48, Fisher Scientific Co.). The inside dimensions of the oven is 25.0 by 29.0 by 25.0 cm. The outlet gas flow rate was measured with a mass flow meter (Model FL-202, Omega Engineering) at a rate of 3,000 ml/min. The actual gas composition was determined by a chemiluminescence analyzer (Model 42, Thermo Environmental Instruments, Inc.). The resulting data was recorded automatically into the memory of a laptop computer.

For this study, samples of green tobacco leaves (flue-cured variety K-326) were harvested from three stalk positions (lower, middle and upper third) from the Central Crops Research Station, Clayton NC. The leaves were stored for one day at 13°C and 89% relative humidity. Just prior to the tests, the leaves were cut into four pieces, approximately x by y cm. The average moisture content of the green leaves was 84.9%, (wet basis).

For the tests, the temperature inside the chamber was maintained at three different levels of either 28, 40 and 52°C. In order to maintain a consistent and known density, the tobacco leaves were placed inside a mesh metal container of dimensions 10.2 by 28.0 by 23.7 cm. This container was then placed inside the vacuum oven. In all, 12 tests were performed as shown in the table below:

After each green tobacco sample was exposed to NO<sub>x</sub> gas for 40 minutes, it was removed and subsequently analyzed for levels of total TSNA's by a commercial testing laboratory (Southern Testing and Research Laboratories, Inc., Wilson N.C. 27896, USA).

In order to develop the mathematical models that describe NO<sub>x</sub> gas absorption in green

| Test number | Chamber <sup>1)</sup> | Temperature (°C) | Tobacco                              |                | NOx (ppm) |
|-------------|-----------------------|------------------|--------------------------------------|----------------|-----------|
|             |                       |                  | Packing density (g/cm <sup>3</sup> ) | Stalk position |           |
| 1           | A                     | 28               | -                                    | -              | 4.46      |
| 2           | A                     | 40               | -                                    | -              | 4.46      |
| 3           | A                     | 52               | -                                    | -              | 4.46      |
| 4           | A                     | 28               | -                                    | -              | 5.35      |
| 5           | B                     | 28               | 0.0739                               | upper          | 4.46      |
| 6           | B                     | 28               | 0.0739                               | upper          | 5.35      |
| 7           | B                     | 28               | 0.0739                               | middle         | 4.46      |
| 8           | B                     | 28               | 0.0739                               | lower          | 4.46      |
| 9           | B                     | 28               | 0.103                                | upper          | 4.46      |
| 10          | B                     | 28               | 0.0443                               | upper          | 4.46      |
| 11          | B                     | 40               | 0.0739                               | upper          | 4.46      |
| 12          | B                     | 52               | 0.0739                               | upper          | 4.46      |

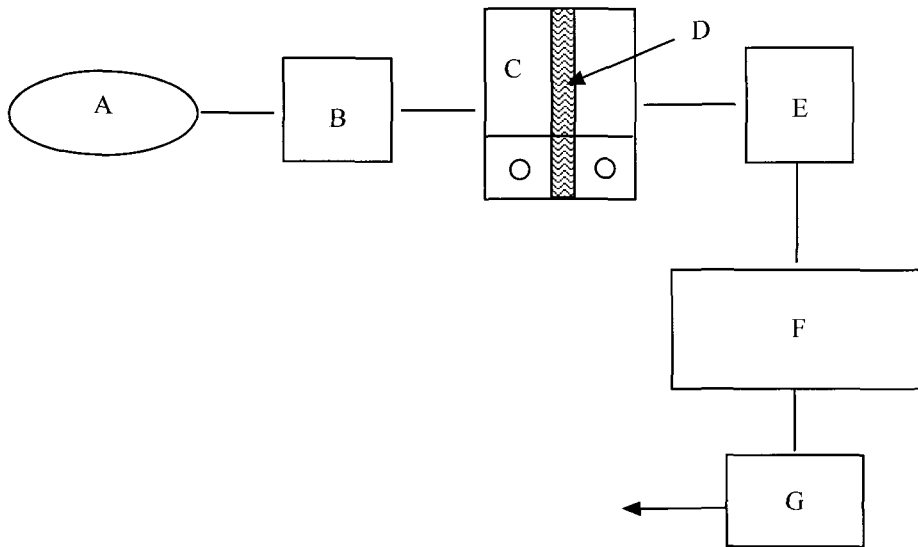
<sup>1)</sup> A: empty chamber; B: chamber filled with green tobacco leaves.

tobacco in the isothermal system shown in Fig. 1, we assumed a constant flow pressure. We further assumed that the gas flow rate inside the chamber is low enough to neglect the pressure drop through the sample or piping. Additionally, since the concentration of NOx gas is relatively low in the mixture, the binary diffusivity was assumed to be constant inside the chamber permitting the use of the equation of continuity for binary mixtures (Eqn (1)).

$$\frac{\partial c_A}{\partial t} + (\vec{U} \cdot \vec{\nabla} c_A) = D_{AB} \nabla^2 c_A + R_A \quad (1)$$

The small size of the test chamber allows us to neglect as insignificant the resistance due to the diffusion of NOx gas in the chamber to therefore assume no difference in concentration with respect to position within the chamber during the test. Assuming no positional differences in concentration allows Eqn (1) to be reduced and rewritten as Eqn (2), below.

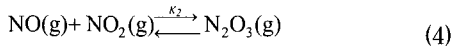
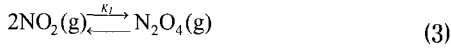
$$\frac{dc_A}{dt} = R_A \quad (2)$$



A: NOx/N<sub>2</sub> gas, B: mass flow controller, C: chamber, D: container for the green tobacco, E: mass flow meter, F: NOx analyzer, G: recorder

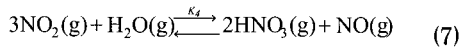
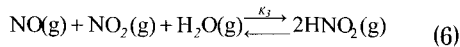
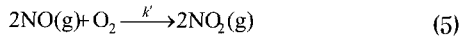
Fig. 1. Schematic diagram of a NOx gas absorption measurement system.

Using Eqn (2), we develop mathematical models separately for the empty chamber and with the chamber filled with green tobacco leaves. Absorption of NOx gases in green tobacco occurs simultaneously accompanied by several chemical reactions. The NOx gas in the supply gas



actually a mixture of several compounds including NO, NO<sub>3</sub>, N<sub>2</sub>O<sub>3</sub>, N<sub>2</sub>O<sub>4</sub>, etc. Inside the supply tank, two reversible reactions, described by Eqn (3) and Eqn (4), prevail.

Upon delivery to the test chamber, in addition to a continuation of Eqns (3) and (4), the tank mixture has several chemical reactions with the O<sub>2</sub> and water vapor in the ambient air. These



reactions are described by Eqns (5), (6) and (7), where  $k'$  is the reaction rate constant and  $K_1$  through  $K_4$  are equilibrium constants. The forward and reverse rates of Eqns (3), (4), (6) and (7) have been reported to be very rapid. Therefore all the NOx components are considered to be a state of equilibrium (Pradhan and Joshi, 2000). We opted to neglect the nitric oxide formation (Eqn (5)) because the supply of oxygen was limited in the vacuum chamber used for this this investigation.

### Mathematical Model I - Empty Chamber

A concentration proportional to the difference

$$\frac{dc_A}{dt} = k_{A1}(c_{AS1} - c_A) \quad (8)$$

between the input and output concentration will be accumulated continually in the empty chamber,

(Eqn (8)). Further, the NOx concentration will also be influenced by Eqns (3) through (7) as described above.

The solution to Eqn (8) is shown by Eqn (9).

$$\frac{c_A - c_{AS1}}{c_{A0} - c_{AS1}} = \exp(-k_{A1}t) \quad (9)$$

### Mathematical Model II - Chamber Filled with Tobacco Leaves

In this model, (Eqn (10)) an amount proportional to differences between output concentration and the equilibrium of the supply gas will be accumulated simultaneously on the tobacco leaves in addition to NOx level changes described by Model I, above.

$$\frac{dc_A}{dt} = k_{A1}(c_{AS1} - c_A) + k_{A2}(c_{AS2} - c_A) \quad (10)$$

The solution of Eqn (10) is shown by Eqn (11).

$$\frac{k_{A1}(c_A - c_{AS1}) + k_{A2}(c_A - c_{AS2})}{k_{A1}(c_{A0} - c_{AS1}) + k_{A2}(c_{A0} - c_{AS2})} = \exp\{-(k_{A1} + k_{A2})t\} \quad (11)$$

### Mathematical Model III - Chamber Filled with Tobacco leaves

In model II, the absorption of NOx gas on the tobacco leaves is regarded as a reversible reaction. In model III (Eqn (12)), the reaction is assumed to be irreversible.

$$\frac{dc_A}{dt} = k_{A1}(c_{AS1} - c_A) - k_{A2}c_A \quad (12)$$

The solution of Eqn (12) is shown below in Eqn (13).

$$\frac{k_{A1}(c_A - c_{AS1}) + k_{A2}c_A}{k_{A1}(c_{A0} - c_{AS1}) + k_{A2}c_{A0}} = \exp\{-(k_{A1} + k_{A2})t\} \quad (13)$$

To obtain the working constants for the above equations, we employed a nonlinear regressive method (Levenberg Marquardt algorithm) of SPSS software. The initial values for getting  $k_{A1}$  and  $k_{A2}$  were both 0.0001 and the values for  $c_{AS1}$  and  $c_{AS2}$  were both 3000 ppb.

## Results and discussion

Fig. 2 shows the temporal changes in NO and NOx level in an empty chamber under condition of 40 °C and NOx input level of 4.46 ppm. Fig. 3 shows the temporal changes in NO and NOx levels in a test chamber filled with green tobacco leaves from the upper stalk position also under conditions of 40 °C and NOx level of 4.46 ppm. The packing density of the green tobacco was 0.0739 g/cm<sup>3</sup>. Note the difference between Fig. 2 and Fig. 3. In Fig. 2, after 30 minutes, the NOx concentration is approximately 4,000 ppb whereas after 30 minutes in Fig. 3, the NOx concentration is approximately 3,300 ppb. This difference is assumed to be because of absorption into the green tobacco.

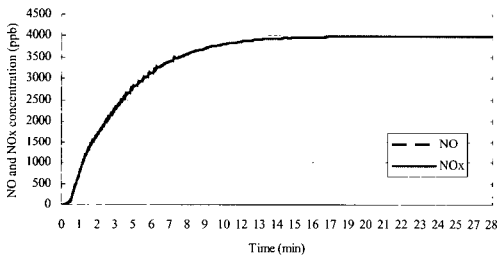


Fig. 2. Temporal changes in NO and NOx levels in an empty chamber under conditions of 40°C, and NOx level of 4.46 ppm.

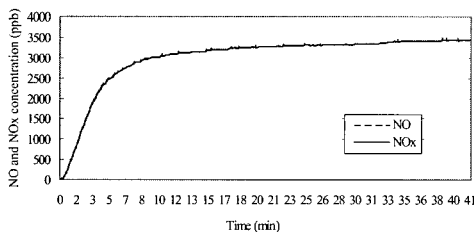


Fig. 3. Temporal changes in NO and NOx levels in a chamber filled with green tobacco leaves from the upper stalk position under conditions of 40°C, NOx level of 4.46 ppm, and tobacco packing density of 0.0739 g/cm<sup>3</sup>.

Note that the concentration curve of NO coincides well with that of NOx in both the empty and filled chamber conditions. This is because the NO<sub>2</sub> component rapidly disappears as shown by the sharp peak near the beginning in Fig. 4. This phenomenon is considered to occur due to the conversion of NO<sub>2</sub> (Eqns (3)-(7)) or NO<sub>2</sub> absorption in the green tobacco under conditions of very low initial concentration of NO<sub>2</sub> gas. Therefore, in this study, we found it useful to work only with NO gas absorption.

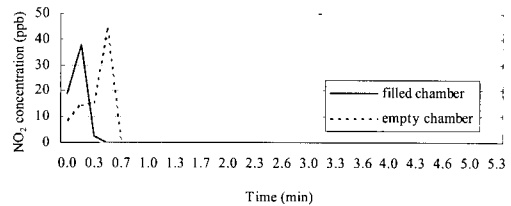


Fig. 4. Temporal changes in NO<sub>2</sub> levels in a chamber filled with green tobacco leaves, and empty one under NOx level of 4.46 ppm.

After getting a nonlinear regressive equation like Eqn (9) from data measured in the empty chamber, we show the NO concentration curve of calculated data in Fig. 5. In this case each constant  $k_{A1}$  and  $C_{AS1}$  is 0.255 min<sup>-1</sup> and 4,029 ppb ( $R^2 = 0.994$ ) respectively. Even though the NO concentration of the input gas is 4,070 ppb, the curve reaches the equilibrium concentration of

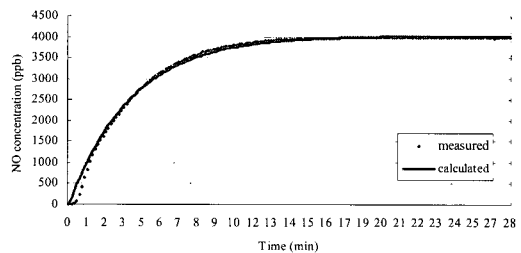


Fig. 5. Temporal changes in NO levels in an empty chamber under conditions of 40°C, and NOx level of 4.46 ppm.

Table 1. Results of nonlinear regressive curve fitting about NO changes in an empty chamber and a chamber filled with green tobacco leaves

| Temperature (°C) | Empty chamber                 |                 |       | Filled chamber <sup>1)</sup>  |       |
|------------------|-------------------------------|-----------------|-------|-------------------------------|-------|
|                  | $k_{A1}$ (min <sup>-1</sup> ) | $c_{AS1}$ (ppb) | $R^2$ | $k_{A2}$ (min <sup>-1</sup> ) | $R^2$ |
| 28               | 0.182                         | 3,930           | 0.990 | 0.0143                        | 0.964 |
| 40               | 0.255                         | 4,029           | 0.994 | 0.0565                        | 0.967 |
| 52               | 0.251                         | 4,091           | 0.991 | 0.0577                        | 0.985 |

<sup>1)</sup> from upper stalk position according to various temperatures under fixed conditions of NOx level of 4.46 ppm and packing density of 0.0739 g/cm<sup>3</sup>.

Table 2. Results of nonlinear regressive curve fitting about NO changes in an empty chamber and a chamber filled with green tobacco leaves

| NOx gas level (ppm) | Empty chamber                 |                 |       | Filled chamber <sup>1)</sup>  |       |
|---------------------|-------------------------------|-----------------|-------|-------------------------------|-------|
|                     | $k_{A1}$ (min <sup>-1</sup> ) | $c_{AS1}$ (ppb) | $R^2$ | $k_{A2}$ (min <sup>-1</sup> ) | $R^2$ |
| 4.46                | 0.182                         | 3,930           | 0.990 | 0.0143                        | 0.964 |
| 5.35                | 0.232                         | 5,336           | 0.994 | 0.0446                        | 0.988 |

<sup>1)</sup> from upper stalk position according to two levels of input NOx gas under fixed conditions of 28°C and packing density of 0.0739 g/cm<sup>3</sup>.

Table 3. Results of nonlinear regressive curve fitting about NO changes in a chamber filled with green tobacco leaves according to various other conditions under fixed conditions of 28 °C and NOx level of 4.46 ppm

|                               | Stalk position <sup>1)</sup> |        |        | Packing density (g/cm <sup>3</sup> ) <sup>2)</sup> |        |        |
|-------------------------------|------------------------------|--------|--------|--|--------|--------|
|                               | lower                        | middle | upper  | 0.0443   | 0.0739 | 0.103  |
| $k_{A2}$ (min <sup>-1</sup> ) | 0.0310                       | 0.0190 | 0.0143 | 0.0120   | 0.0143 | 0.0235 |
| $R^2$                         | 0.973                        | 0.947  | 0.964  | 0.960  | 0.964  | 0.949  |

<sup>1)</sup> Packing density is fixed as 0.0739 g/cm<sup>3</sup>.

<sup>2)</sup> Stalk position is fixed as upper.

4,029 ppb. This may be explained by Eqns (3) through (7). Even with no green tobacco in the chamber, the molecules react with the ambient oxygen and water vapor in the chamber.

In the chamber with the green tobacco, we tried to get a nonlinear regressive equation like Eqn (11) but the equilibrium concentration,  $c_{AS2}$

assumed a negative value. Since a negative concentration value has no physical meaning, model II fails to adequately explain NOx absorption. Model II was developed under the hypothesis that NOx absorption in green tobacco was a reversible reaction. Therefore, by substituting model III with irreversible absorption

phenomena, we were able to get the results shown in Fig. 6. The constant  $k_{A2}$  of Eqn (13) was  $0.0565 \text{ min}^{-1}$  ( $R^2 = 0.967$ ). The other constants used in Eqn (13) were the same as calculated from Eqn (9) since the same conditions prevailed except for the green tobacco. After obtaining the nonlinear regressive equation from each experimental result, they are shown in tables 1-3.

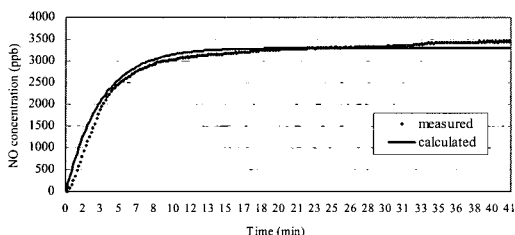


Fig. 6. Temporal changes in NO levels in a chamber filled with green tobacco leaves from the upper stalk position under conditions of  $4 \text{ }^{\circ}\text{C}$ ,  $\text{NO}_x$  level of  $4.46 \text{ ppm}$ , and tobacco packing density of  $0.0739 \text{ g/cm}^3$ .

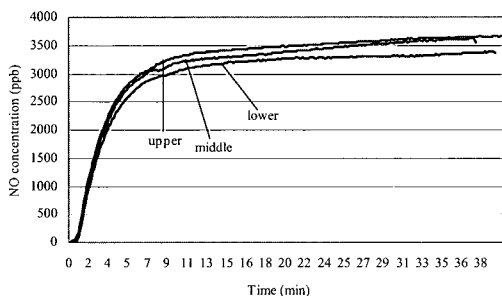


Fig. 7. Temporal changes in NO levels in a chamber filled with green tobacco leaves from various stalk positions under conditions of  $28 \text{ }^{\circ}\text{C}$ ,  $\text{NO}_x$  level of  $4.46 \text{ ppm}$ , and tobacco packing density of  $0.0739 \text{ g/cm}^3$ .

Fig. 7 shows the temporal changes of NO with regard to stalk position with other conditions altered as before. The equilibrium concentration of NO with upper stalk tobacco is higher than that of lower stalk which means that upper stalk tobacco absorbs less NO gas under the same

conditions as lower stalk tobacco. In table 3, note that the constant  $k_{A2}$  of the lower stalk position tobacco is greater than the other two stalk positions. This fact also strongly suggests that lower stalk tobacco absorbs NO gas more than upper stalk tobacco.

Fig. 8 shows the temporal changes of NO concentration with regard to differences in packing density. The concentration curves of packing density of  $0.0443 \text{ g/cm}^3$  essentially coincide with that of  $0.0739 \text{ g/cm}^3$  but with density of  $0.103$  the lowest. Further, Fig. 9 shows the temporal changes of NO concentration at two  $\text{NO}_x$  input concentrations.

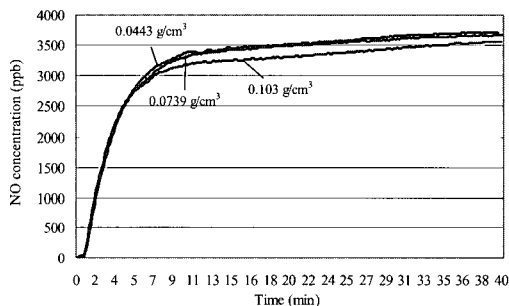


Fig. 8. Temporal changes in NO levels in a chamber filled with green tobacco leaves from the upper stalk position under conditions of  $28 \text{ }^{\circ}\text{C}$ , and  $\text{NO}_x$  level of  $4.46 \text{ ppm}$  with regard to various levels of tobacco packing density.

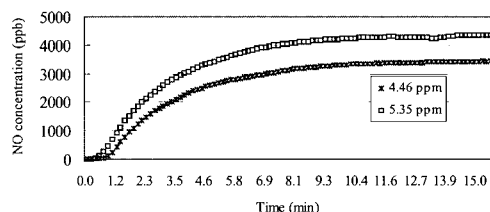


Fig. 9. Temporal changes in NO levels in a chamber filled with green tobacco leaves from the upper stalk position under conditions of  $28 \text{ }^{\circ}\text{C}$ , and tobacco packing density of  $0.0739 \text{ g/cm}^3$  with regard to various levels of input  $\text{NO}_x$  gas.

Fig. 10 shows the temporal changes in NO concentration at different temperature conditions. Although the graphs for 40°C closely coincide with the one for 52°C, the equilibrium concentration of NO at 28°C is greatest. This suggests that the amount of NO absorption in green tobacco increase proportionally with temperature. This fact is also shown to a lesser degree in Fig. 11, which exhibits a smaller difference between constants ( $k_{A2}$ ) with temperatures above 40 °C. Those data were applied to the Arrhenius equation, but we were unable to get the proper curve fit for that equation. From this fact we assume that constants ( $k_{A2}$ ) from NOx absorption in green tobacco did not result from the effect of a single mechanism but from a complicated combination of perhaps physical adsorption and various chemical reactions.

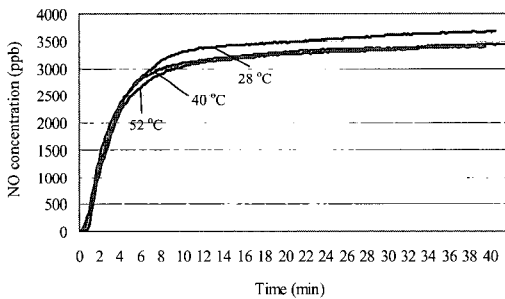


Fig. 10. Temporal changes in NO levels in a chamber filled with green tobacco leaves from the upper stalk position under conditions of NOx level of 4.46 ppm, and tobacco packing density of 0.0739 g/cm<sup>3</sup> with regard to various temperatures.

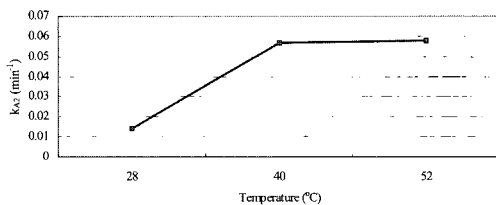


Fig. 11. Changes in constant ( $k_{A2}$ ) with temperatures.

Table 4. Results of TSNA's analysis about green tobacco leaves from upper stalk position treated with 4.46 ppm NOx gas for 40 minutes under the packing density of 0.0739 g/cm<sup>3</sup>

|             | 28 °C             | 40 °C | 52 °C |
|-------------|-------------------|-------|-------|
| NNN (ppm)   | BDL <sup>1)</sup> | BDL   | BDL   |
| NAT (ppm)   | BDL               | BDL   | BDL   |
| NAB (ppm)   | BDL               | BDL   | 0.769 |
| NNK (ppm)   | BDL               | BDL   | 0.859 |
| TSNAs (ppm) | BDL               | BDL   | 1.63  |

<sup>1)</sup> BDL = Below detection Limit (Detection limit is 0.1 ppm).

<sup>2)</sup> TSNAs results are reported on a dry weight basis.

Table 4 shows the results of TSNA's analysis of green tobacco of the upper stalk position exposed to 4.46 ppm NOx gas for 40 minutes with a packing density of 0.0739 g/cm<sup>3</sup>. In this study, TSNAs formation was below detection limit except for NAB and NNK of the sample treated at 52 °C. This agrees with the fact that green tobacco has minimal TSNAs and that the TSNAs found in cured leaf are formed during curing and storage (3).

## Conclusions

We successfully developed a mathematical model for describing NOx absorption into green (uncured) tobacco. We found that NOx gases were considerably absorbed by green tobacco with variations related to several factors. Specifically, tobacco from the lower stalk positions (bottom one third) absorbs more NOx gas per unit weight than tobacco from upper stalk positions. Additionally, the green tobacco packed with a density of 0.103 g/cm<sup>3</sup> absorbs more NOx gas per unit weight than either 0.0443 g/cm<sup>3</sup> or 0.0739 g/cm<sup>3</sup>. Further, the NOx absorption



increases proportionally with temperature with the maximum absorption point around 40°C.

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