Journal of Chromatography, 107 (1975) 351–357

CHROM. 8088

PRINCIPLE OF OPERATION OF THE THERMAL ENERGY ANALYZER FOR THE TRACE ANALYSIS OF VOLATILE AND NON-VOLATILE N-NITROSO COMPOUNDS

D. H. FINE, D. LIEB and F. RUFEH
Thermo Electron Corporation, 85 First Avenue, Waltham, Mass. 02154 (U.S.A.)
(Received November 12th, 1974)

SUMMARY

The theoretical basis for the thermal energy analyzer is discussed. Using the principles outlined, the feasibility of selectivity detecting μg/kg levels of volatile and non-volatile N-nitroso compounds is established.

INTRODUCTION

A prerequisite for investigating whether N-nitroso compounds have a role in human cancer is the availability of selective methods for the trace analysis of both volatile and non-volatile N-nitroso compounds in foods, beverages, and complex biological mixtures. A new detector, called the thermal energy analyzer (TEA) or thermoluminous analyzer, which could meet these goals, was reported recently[1,2]. In the present paper, the underlying physical principles of the technique are discussed. The competing chemical reactions occurring in each step are outlined, and quantitative inferences are made as to their relative importance. Theoretical limitations on temperature, pressure and residence time are examined so that design conditions may be optimized to achieve maximum sensitivity with minimum possible interference.

METHOD OF OPERATION

The TEA is a gas phase technique with the essential chemical sequences occurring in the gas phase, and/or at the solid–gas interface. The method of operation may be understood with reference to the simplified schematic of Fig. 1. The N-nitroso compound, dissolved in a suitable solvent, is introduced into a flash catalytic heater or pyrolyser, where the N–NO bond is ruptured, with the release of the nitrosyl radical (·NO). The nitrosyl radical is then oxidized with ozone in an evacuated reaction chamber, to give electronically excited nitrogen dioxide (NO_2^*). The excited nitrogen dioxide decays back to its ground state with the emission of characteristic radiation. The intensity of the emission is detected by means of a sensitive photomultiplier tube and is proportional to the nitrosyl radical concentration, and hence to the N-nitroso compound concentration. Because of the unique series of chemical reactions

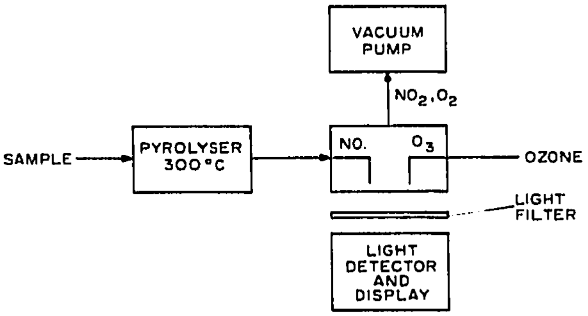

Fig. 1. Simplified schematic of the thermal energy analyzer.

involved, the analyzer is sufficiently selective to be used for screening purposes. It can also be used as a detector for either gas chromatography or for high-performance liquid chromatography.

Pyrolysis of N-nitroso compounds

The weak nature of the N–NO bond in volatile N-nitrosamines has been suspected since the pioneering work of Bamford[3] on the UV vapor photolysis of dimethyl- and diethylnitrosamine. The strength of the N–NO bond in a particular N-nitroso compound under standard equilibrium conditions of 25° and 1 atm pressure may be calculated from a knowledge of the standard heat of combustion of the parent compound, together with the standard heats of formation of the relevant fragment radicals. For dimethylnitrosamine enough information is available to allow a calculation to be made[4]

$$D[(CH_3)_2N\text{–}NO] = \Delta H_f^0[\cdot NO] + \Delta H_f^0[(CH_3)_2N\cdot] - \Delta H_f^0[(CH_3)_2NNO]$$

from the reaction:

$$(CH_3)_2NNO = (CH_3)_2N\cdot + \cdot NO$$

giving $D[(CH_3)_2N\text{–}NO] = 52.1$ kcal mole^{-1}. Because of the paucity of experimental data on the standard heats of formation of the parent N-nitroso compound, and/or the radical fragments, bond dissociation energies are available for only a few N-nitroso compounds[4,5]:

$$\begin{aligned}
&D[(CH_3)_2N\text{–}NO] &&= 52 \text{ kcal mole}^{-1}\\
&D[\{(CH_3)_2CHCH_2\}N\text{–}NO] &&= 54 \text{ kcal mole}^{-1}\\
&D[\{CH_3C(NO_2)_2CH_2\}_2N\text{–}NO] &&= 43 \text{ kcal mole}^{-1}\\
&D[(C_6H_5)_2N\text{–}NO] &&= 11 \text{ kcal mole}^{-1}
\end{aligned}$$

The N–NO bond strengths shown above are small when compared with values of 70–90 kcal mole^{-1} typical of the C–N, C–C and C–H bonds in most organic molecules. The N–NO bond is therefore likely to be the weakest bond in the N-nitroso compounds.

In acid solution, UV photolysis of N-nitrosamines yields nitrite, possibly by rupture of the N–NO bond; nitrite is then assayed by means of the Griess Illosvay reagent[6]. In solution, and even in the vapor phase, the UV photolysis of N-nitroso-amides and N-nitroso-ureas does not yield the nitrosyl radical[7,8]. For example, in solution, the N-nitroso-amides rearrange on heating by migration of the acyl group from N to O to form an unstable diazo ester; the diazo ester then decomposes with the elimination of gaseous nitrogen. However, in the gas phase, where rearrangement and disproportionation reactions occur only rarely, the reaction sequence is considerably simplified. When a compound is suddenly sublimed or vaporized, the sudden increase in thermal energy pyrolyses the molecule, causing it to fall apart at its weakest bond. In N-nitroso compounds the weakest bond is the N–NO bond, so that flash heating should result in the release of the nitrosyl radical. The nitrosyl radical which is split off is itself a stable gas at normal temperatures and pressures. The organic fragment is unstable and must either decompose further or suffer rearrangement to a more stable species; in either event it is unlikely to form the nitrosyl radical and it is therefore not likely to be a source of potential interference.

A number of catalysts were investigated for cleaving the N–NO bond[1,2,5]. As expected from bond energy considerations, many catalysts successfully cleaved the N–NO bond. However, in most cases the reaction was incomplete, was not reproducible and was strongly temperature dependent. Depending on the temperature history, the catalyst often absorbed a fraction of the nitroso compounds and subsequently released them at random intervals. Catalysts which were used included WO_3, $W_{20}O_{58}$, acid-passivated stainless steel, and nickel oxide and nickel-based alloys; in addition, the purely pyrolytic mode was also investigated. We found that a mixture of WO_3 and $W_{20}O_{58}$ was most effective for selective and reproducible cleavage of the N–NO bond. Pyrolytic decomposition is inherently a simpler process, but requires higher temperatures and may therefore not be as selective. It is important, however, to design and construct the chamber in such a way as not to trap the nitrosyl radical once it has been formed.

Detection of the nitrosyl radical

The reaction of the nitrosyl radical with ozone occurs according to the following two competing reactions:

$$NO + O_3 = NO_2^* + O_2 \tag{1a}$$

$$NO + O_3 = NO_2 + O_2 \tag{1b}$$

Once formed, the electronically excited NO_2^* decays back to its ground state by one of the following three possible paths.

$$NO_2^* = NO_2 + h\nu \tag{2a}$$

$$NO_2^* + M = NO_2 + M \tag{2b}$$

$$NO_2^* + M + M = NO_2 + M + M \tag{2c}$$

The rate constants of the two primary reactions, 1a and 1b, are well established[9]: $k_{1a} = 7.6 \times 10^{11} \exp(-4180/RT)\,\mathrm{cm^3\,mole^{-1}\,sec^{-1}}$ and $k_{1b} = 4.3 \times 10^{11} \exp(-2330/RT)\,\mathrm{cm^3\,mole^{-1}\,sec^{-1}}$. The fraction of NO which is converted to an electronically excited state is given by F^*, where:

$$F^* = k_{1a}/(k_{1a} + k_{1b}) = \frac{1}{1 + \dfrac{k_{1b}}{k_{1a}}} \tag{3}$$

Fig. 2 shows the variation of F^* with temperature, calculated from eqn. 3, from which it may be seen that at ambient temperatures less than 8% of the available nitrosyl radicals are electronically excited.

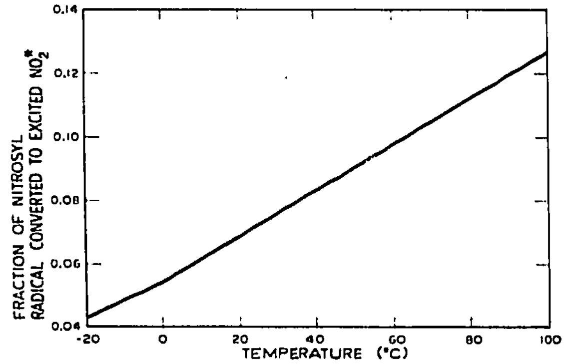

Fig. 2. The effect of temperature on the fraction of nitrosyl radicals which are converted to excited NO_2^*.

Excited NO_2^* can lose energy by emission of light (reaction 2a), by two body collisions (reaction 2b) or by three body collisions (reaction 2c). At pressures much below one atmosphere, the contribution to deactivation by three body collisions can usually be neglected. The rate of decay of excited NO_2^* is therefore given by:

$$\frac{d\,[NO_2^*]}{dt} = k_{2a}\,[NO_2^*] + k_{2b}\,[NO_2^*]\,[M]$$

The fraction of NO which decays to its ground state with the emission of light is given by:

$$\begin{aligned} F(NO) &= F^* \frac{k_{2a}}{k_{2a} + k_{2b}}\,[M] \\ &= \frac{F^*}{1 + \dfrac{k_{2b}}{k_{2a}}}\,[M] \end{aligned} \tag{4}$$

Using Clough and Thrush's value[9] of $k_{2a}/k_{2b} = 4.5 \times 10^{-10}$ mole cm^{-3}, the variation of F (NO) with pressure (at a temperature of 20°) is shown in Fig. 3. The overall ozone–nitrosyl radical–light emission reaction is seen to be exceedingly inefficient. At

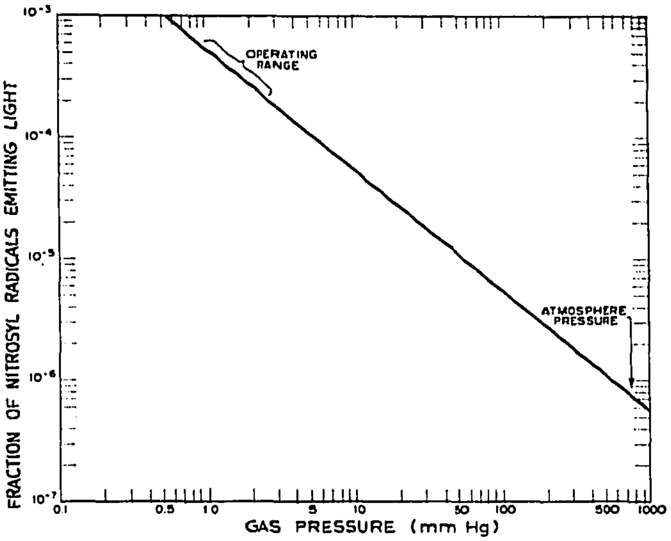

Fig. 3. The effect of pressure on the fraction of nitrosyl radicals which emit light.

1 atm pressure, the fraction of nitrosyl radicals producing light quanta is less than 1×10^{-6}, and even at 2 mm Hg pressure the fraction of nitrosyl radicals contributing to the light emission is only 3×10^{-4}. The number of useful light quanta is further reduced by losses due to poor reaction chamber geometry (typical efficiencies are about 30%), photomultiplier tube collection characteristics (about 80% efficient) and the quantum efficiency of the photomultiplier tube (less than 7% efficient). The TEA is preferably operated at low pressures because low pressure reduces the importance of variations in the second body effect. Provided that the mole throughput remains constant, reducing the reaction chamber pressure will increase the intensity of the light emitted. Reactor volume must also be considered, for if the pressure is reduced too far at constant mole throughput, the residence time of the species in the reactor may become so short that the decay of the excited NO_2^* species may occur after the species have left the reaction chamber[10].

Selectivity

The selectivity of the TEA relies upon the breakdown of the N-nitroso compound to release the nitrosyl radical, and the subsequent luminescent reaction of the nitrosyl radical with ozone. The distribution of the emission in the near IR region of the spectrum between 0.6 and 3.0 μ has been mapped out[9]. Because of the low resolution of the experimental technique which was used, the emission appears to be continuous (see Fig. 4). A red filter is used to eliminate light of wavelengths shorter than 0.6 μ which may be present from such potential interfering chemiluminescent reactions of ozone with carbon monoxide, ethylene, and other compounds. Because of the poor spectral response characteristics of photomultiplier tubes in this wavelength region, only a small part of the distribution out to 0.8 μ is useful. Thus, by exploiting a combination of a red cut-off filter and poor photomultiplier tube characteristics, only the narrow region between 0.6 and 0.8 μ is monitored.

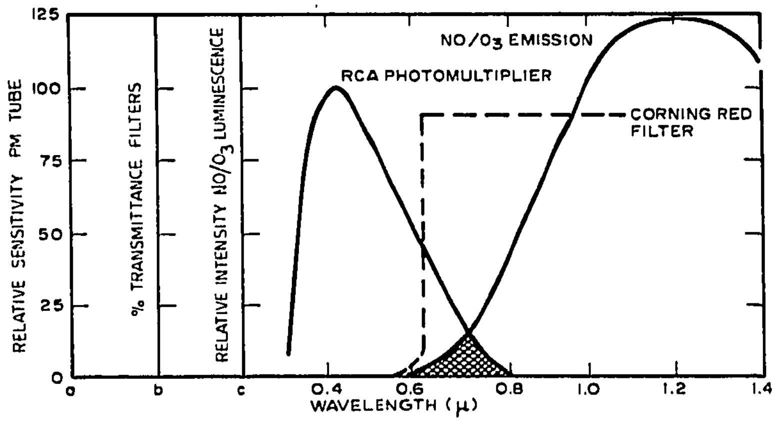

Fig. 4. Distribution of emission in the near IR, showing red-filter cutoff point and responsivity of photomultiplier tube.

If bonds in other compounds rupture or if other bonds in the N-nitroso compound itself rupture, the TEA response remains unaffected unless the nitrosyl radical is released. All nitrogen-containing compounds are suspect, but provided that the temperature is not too high, and provided that oxygen gas is excluded, molecular fragments will not be converted into the nitrosyl radical. Nitro (NO_2) compounds can give nitrosyl radicals under certain conditions, and care must be taken in order to prevent either of the following two reactions from occurring:

$$2NO_2 = 2NO + O_2 \qquad (5)$$

$$NO_2 + C = CO + NO \qquad (6)$$

These two reactions are discussed below.

The thermodynamic equilibrium for the system N_2–O_2–NO–NO_2 can be calculated as a function of temperature if it is assumed that the only reaction which is allowed to proceed is reaction 5 (see Table I). The equilibrium value describes the maximum possible conversion under ideal conditions. In practice, the kinetics of the reaction are so slow that even at 400° no detectable conversion of NO_2 to NO occurs

TABLE I

EQUILIBRIUM PER CENT NO IN THE SYSTEM N_2-O_2-NO-NO_2 AS A FUNCTION OF TEMPERATURE

Pressure 1 atm.

Temperature (°C)	*Conversion (%)*
300	10
400	40
500	72
600	86
700	94

within the time frame (seconds) of the TEA technique. The reduction of nitrogen dioxide to nitric oxide by reaction 6 is more important. The reaction is exothermic and can proceed extremely rapidly even at 200° in the presence of metal catalysts such as gold[11] and molybdenum[12]. In order to minimize the reduction of nitrogen dioxide, precautions must be taken to both eliminate active forms of carbon, and also to avoid contact with potential metal catalysts.

CONCLUSION

The theoretical basis for the TEA has been discussed. Practical devices using the principle developed will be described in subsequent publications.

ACKNOWLEDGEMENTS

The authors thank William Zolner for many useful discussions. The most recent work upon which this publication is based was performed pursuant to Contract N01CP45623 with the National Cancer Institute, U.S. Department of Health, Education and Welfare.

REFERENCES

1 D. H. Fine, F. Rufeh and B. Gunther, *Anal. Lett.*, 6 (1973) 731.
2 D. H. Fine, F. Rufeh and D. Lieb, *Nature (London)*, 247 (1974) 309.
3 C. H. Bamford, *J. Chem. Soc.*, (1939) 12.
4 V. I. Pepekin, R. G. Gafurov, Ya. A. Lebedev, L. T. Eremenko, E. M. Sogomonyan and A. Ya. Apin, *Izv. Akad. Nauk SSSR, Ser. Khim.*, (1973) 318.
5 D. H. Fine and F. Rufeh, *N-Nitroso Compounds Analysis and Formation*, International Agency for Research on Cancer, Lyon, 1975.
6 P. Issenberg and S. R. Tannenbaum, *N-Nitroso Compounds Analysis and Formation*, International Agency for Research on Cancer, Lyon, Publication No. 3, 1972, p. 31.
7 E. H. White and R. J. Baumgarten, *J. Org. Chem.*, 29 (1964) 2070; 29 (1964) 3636.
8 E. H. White and C. A. Aufdermarsh, *J. Amer. Chem. Soc.*, 83 (1961) 1174, 1179.
9 P. N. Clough and B. A. Thrush, *Trans. Faraday Soc.*, 63 (1967) 915.
10 D. M. Steffenson and D. H. Stedman, *Anal. Chem.*, 46 (1974) 1704.
11 J. A. Hodgeson, J. P. Bell, K. A. Rehme, K. J. Krost and R. K. Stevens, *Joint Conference on Sensing of Environmental Pollutants*, Amer. Inst. of Aeronautics and Astronautics, Paper No. 71-1067, November 1971.
12 J. N. Pitts, 1974, personal communication.