# Interactions of Sulfur Dioxide and Acrolein as Sensory Irritants

LAUREL E. KANE[1] AND YVES ALARIE[2]

*Department of Industrial Environmental Health Sciences, Graduate School of Public Health, University of Pittsburgh, Pittsburgh, Pennsylvania 15261*

Interactions of Sulfur Dioxide and Acrolein as Sensory Irritants. KANE, L., AND ALARIE, Y. (1979). *Toxicol. Appl. Pharmacol.* **48**, 305–315. Sulfur dioxide/acrolein atmospheres were evaluated for their sensory irritation potential by monitoring the respiratory rate of groups of mice during 5-min control, 10-min exposure, and 5-min postexposure periods. The results demonstrated that depending upon the concentration ratio of these sensory irritants, either irritant can alter, or completely block, the effect of the other during the exposure. Following exposure to these mixtures, recovery was extremely slow, unlike the rapid recovery observed with each irritant alone.

Both industrial and environmental atmospheres tend to be complex mixtures containing numerous pollutants. Studying the health effects of these pollutant mixtures has many difficulties: the presence of numerous chemical species, the wide range of physiological and pathological responses that may be evoked, and the possibility of complicated chemical interactions.

In previous investigations, the sensory irritation response to formaldehyde/acrolein mixtures was evaluated in mice (Kane and Alarie, 1978a) and the results were consistent with the hypothesis that the two chemicals acted at the same receptor site and exhibited competitive agonism as sensory irritants.

It has been shown that sensory irritation can be evaluated by monitoring the decrease in respiratory rate in mice that are exposed to sensory irritants (Alarie, 1966, 1973a; Alarie et al., 1973; Kane and Alarie, 1977; Ulrich et al., 1972). Sensory irritation in man is usually associated with a burning sensation in

the eyes, nose, and throat of an exposed individual. However, airborne sensory irritants also evoke numerous physiological reflexes, including a decrease in respiratory rate. This reflex is provoked by stimulation of free nerve endings of the afferent trigeminal nerve located in the nasal mucosa. There is a characteristic breathing pattern associated with this response, in which the inspiratory phase remains rapid, but there is a pause before exhalation, resulting in a lowered respiratory rate. In mice, the percentage decrease in respiratory rate, from preexposure control levels, has proved to be a quantitative index of sensory irritation, and shows a linear relationship with the logarithm of the exposure concentration (Alarie, 1966, 1973a; Alarie et al., 1973; Kane and Alarie, 1977).

It has been shown that when mice are exposed to sulfur dioxide alone, there is an immediate, rapid decrease in respiratory rate (Alarie et al., 1973). Within 2 min of the onset of exposure, the respiratory rate begins to return toward control values, although the mice are still being exposed to the sulfur

[1] Present address: Foster D. Snell, Inc., Florham Park, N.J. 07932.

[2] To whom all correspondence should be sent.

dioxide. The rate of this accommodation or desensitization phase depends upon the concentration of sulfur dioxide present. With acrolein, the onset of the response is slower, but once the mice have attained their maximum response, usually within 5 min, it is maintained at this plateau level throughout the exposure (Kane and Alarie, 1977).

Thus, when evaluating a sensory irritant, it is necessary to consider both the level of response and the time pattern of the response. Since the time pattern of response for sulfur dioxide varied widely from the one observed with acrolein, as noted above, mixtures of these two sensory irritants were investigated and the results are reported here.

## METHODS

*Animals.* The animals used in this study were outbred Specific Pathogen Free male Swiss–Webster mice, weighing between 22 and 30 g, obtained from Hilltop Laboratories. New groups of four mice were used for each exposure.

*Acrolein and Sulfur dioxide.* High pressure gas cylinders of acrolein in nitrogen were obtained from Linde Speciality Gases. The concentrations were determined by chemical analysis (NIOSH, 1974). High pressure gas cylinders of sulfur dioxide in nitrogen were obtained from Air Products. The concentrations were determined by a modification of the West and Gaeke (1956) method, and confirmed by infrared analysis.

In order to obtain the desired exposure concentrations, each generation system was tested without animals in the chamber. If the analytical concentration was within 12% of the desired concentration, the exposure was then conducted and analysis for exposure concentration was performed. Therefore, all exposure concentrations reported are ±12% and usually within 5%.

*Generation of test atmospheres.* The test atmospheres were generated by metering a known rate of each sensory irritant through a glass flowmeter, and then diluting by controlling the airflow through the exposure chamber.

*Exposure chamber.* The exposure chamber has been described in detail (Barrow *et al.*, 1977; Kane and Alarie, 1978a). Briefly, it consists of an all-glass chamber of approximately 2.3 liters with four side-arms. Each side arm can hold one mouse so that only the head of the animal is exposed to the test atmosphere. Airflow through the chamber was achieved by a suction pump and regulated (by a flowmeter) at

15 liters/min or at higher flow rates, up to 50 liters/min, for the lower exposure concentrations.

*Measurement and evaluation of response.* The response parameter in these studies was the percentage decrease in respiratory rate of four mice, simultaneously exposed to the same atmosphere. The method for measuring this has been described in detail in a previous publication (Kane and Alarie, 1977). In summary, each side arm of the exposure chamber functioned as a plethysmograph, allowing the changes in pressure caused by the breathing of the mouse to be sensed by a pressure transducer. These signals were processed and displayed as the averaged respiratory rate of the four mice. Typical responses for sulfur dioxide and acrolein are shown in Figs. 1 and 2, respectively.

In order to evaluate the response to the acrolein/sulfur dioxide mixtures, it was necessary to know the expected response at each cencentration for each chemical alone. These concentration–response relationships for sulfur dioxide or acrolein have been previously developed (Alarie *et al.*, 1973; Kane and Alarie, 1977). For each, the maximum percentage decrease in respiratory rate was found to be a linear function of the logarithm of the exposure concentration. For sulfur dioxide, this relationship was:

$$y = -47.1 + 47.0 \log x,$$

and for acrolein:

$$y = 41.2 + 39.4 \log x,$$

where $y$ represents the maximum percentage decrease in respiratory rate, and $x$ represents the exposure concentration in parts per million. The expected responses for each exposure concentration used in these experiments were calculated from these equations and these relationships were confirmed during this series of experiments, as indicated below.

*Experimental protocol.* Each experiment consisted of a control period, during which the respiratory rate was measured while the animals were breathing room air. Then the irritants were introduced into the exposure chamber for 10 min. Following this exposure period, the respiratory rates were measured for an additional 5 min with the animals breathing room air.

A typical set of inhalation exposures consisted of two experiments in which two different groups of four mice were exposed to a given concentration of acrolein alone, two experiments using sulfur dioxide alone, and two with a mixture of acrolein and sulfur dioxide, using the same concentrations that had been used for the individual exposures but using different groups of mice for each exposure. Two experiments were conducted for each exposure condition. In this way, the previously developed concentration–response relationships (Alarie *et al.*, 1973; Kane and Alarie, 1977) were also confirmed.
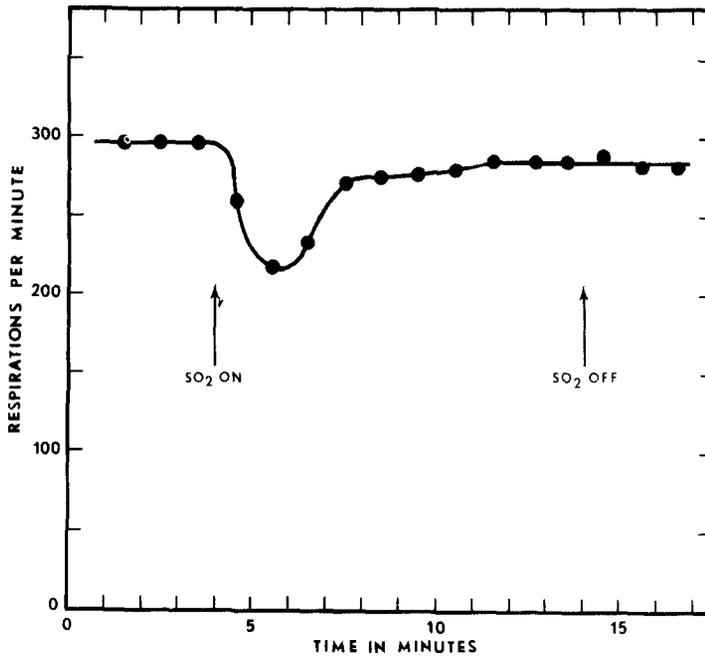
Initially, three series of experiments were conducted.

FIG. 1. Time–response pattern during a 10-min exposure to 40 ppm sulfur dioxide. This represents the average respiratory rate of four mice exposed simultaneously. The exposure period immediately follows a 5-min control period, during which the mice are breathing room air.
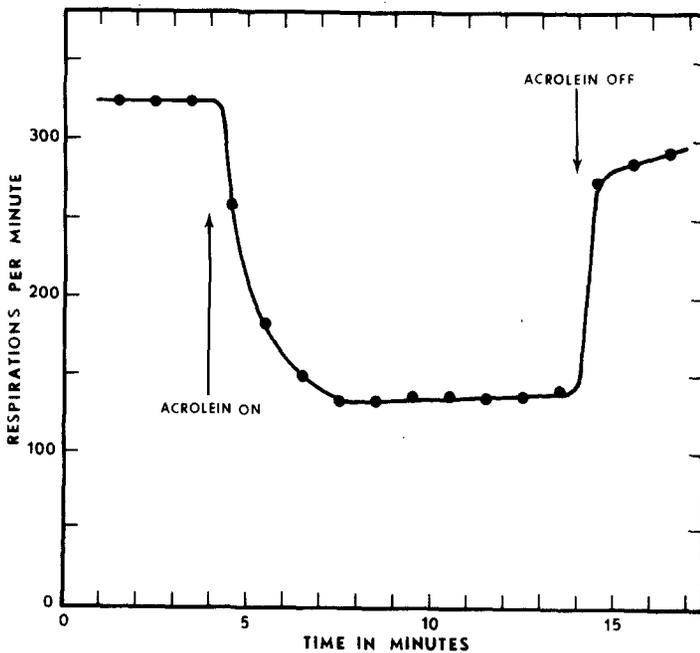


FIG. 2. Time–response pattern during a 10-min exposure to 3.4 ppm acrolein, following a 5-min control period, during which the mice are breathing room air.

In Series I, each set used a constant ratio of sulfur dioxide/acrolein. Three sets were run with sulfur dioxide/acrolein ratios of 41/1, 21/1, and 10/1. In Series II the concentration of acrolein was kept constant, and the concentration of sulfur dioxide was varied. In Series III, the sulfur dioxide concentration was kept constant, and the acrolein concentration was varied. The concentrations used in each series are shown in Tables 1–3.

For each exposure, the maximum percentage decrease in respiratory rate from control values was calculated. In addition, the patterns of the response during the exposure and recovery periods were evaluated.

After the results of the first three series were evaluated, an additional series was conducted. Series IV consisted of combined sulfur dioxide/acrolein exposures lasting from 1 to 5 min. The sulfur dioxide and acrolein concentrations were held constant in this series, since the purpose of these experiments was specifically to determine if the duration of the exposure period affected the respiratory pattern during the recovery period.

## RESULTS

A typical set of results consists of data from two runs with sulfur dioxide alone, two with acrolein alone, and two runs with the combined exposures. For example, with 55 ppm sulfur dioxide, the two observed maximum percentage decreases in respiratory rate were 36.1 and 31.3%. The predicted response is 35% (Alarie et al., 1973). For acrolein, the observed data were 51.6 and 50.8%. The predicted response is 50% at 1.65 ppm (Kane and Alarie, 1977).

Each observed response for all the single exposures in this study was well within 15% of the expected value. It has previously been reported that this is to be expected (Kane and Alarie, 1977). This earlier work has shown that for acrolein at 1.65 ppm (SD = 0.18), the observed responses for 11 experiments were 51.7% (SD = 3.5). At 0.68 ppm (SD = 0.25), the observed responses for eight experiments were 31.1% (SD = 3.3).

For all the experiments, the results consisted of the observed maximum percentage decrease in respiratory rate, the response pattern (e.g., rapidity of onset, duration, accommodation), and the respiratory pattern

during the recovery phase. The response pattern was keyed as a sulfur dioxide pattern (S) if it was similar to that shown in Figs. 1 and 3, or an acrolein pattern (A) if similar to that in Figs. 2 and 3. If the response showed characteristics of both patterns, it was keyed as mixed (S/A) response. Two such patterns are depicted in Fig. 3.

### Series I: Constant Sulfur Dioxide/Acrolein

The results of Series I are shown in Table 1. When the sulfur dioxide/acrolein ratio was 41/1, the pattern of the responses during the exposure was typical of sulfur dioxide (S) at the highest concentrations, and was mixed (S/A) at the lower concentrations. When the sulfur dioxide/acrolein ratio was 21/1, or 10/1, the response patterns were always that typical of acrolein (A).

The magnitude of the response also showed the same trend, as the data in Table 1 show. The magnitude was very close to that expected
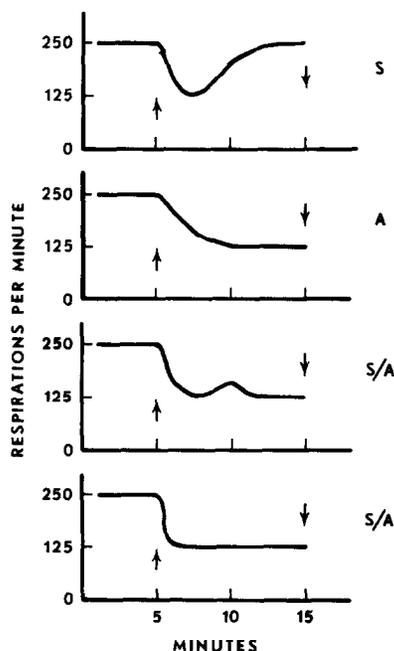


FIG. 3. Time–response patterns during exposure to sulfur dioxide/acrolein mixtures: S, typical of sulfur dioxide alone; A, typical of acrolein alone; S/A, rapid decrease typical of sulfur dioxide, and plateau response typical of acrolein.

TABLE 1

RESULTS FOR SERIES I: CONSTANT SULFUR DIOXIDE/ACROLEIN CONCENTRATION RATIO EXPOSURES.
RESPIRATORY RATE IN MICE

| Sulfur dioxide | | Acrolein | | | Observed response | | Expected response if effects are additive. |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Concentration (ppm) | . Expected decrease in respiratory rate[a] (%) | Concentration (ppm) | Expected decrease in respiratory rate[b] (%) | Sulfur dioxide acrolein Concentration ratio | Pattern observed | Decrease in respiratory rate (%) | Decrease in respiratory rate (%) |
| 140 | 54 | 3.4 | 62 | 42/1 | S | 53 | 85[c] |
| 52 | 33 | 1.27 | 45 | | S/A | 32 | 78 |
| 35 | 23 | 0.85 | 38 | | S/A | 18 | 61 |
| 70 | 42 | 3.4 | 62 | 21/1 | A | 62 | 85[c] |
| 35 | 23 | 1.7 | 50 | | A | 52 | 73 |
| 18 | 12 | 0.85 | 38 | | A | 19 | 50 |
| 35 | 23 | 3.4 | 62 | 10/1 | A | 54 | 85 |
| 18 | 12 | 1.7 | 50 | | A | 52 | 62 |
| 9 | 10 | 0.85 | 38 | | A | 17 | 48 |

[a] From sulfur dioxide linear regression $y = -47.1 + 47.0 \log x$.

[b] From acrolein linear regression $y = 41.2 + 39.4 \log x$.

[c] Eighty-five percent is the greatest response experimentally observed, even at very high acrolein concentrations (Kane and Alarie, 1978).

for sulfur dioxide alone at the high sulfur dioxide/acrolein ratio, but closer to that expected from acrolein alone at the lower sulfur dioxide/acrolein ratios. For all ratios tested, the response at the lowest concentrations for each ratio was definitely less than expected for acrolein alone and the mangitude of the response was always less for all concentrations tested and at all ratios, than would be expected from adding the response for each sensory irritant.

In each experiment the recovery phase always showed a characteristic pattern in which the respiratory rate initially increased when the exposure ended, and then decreased, as shown in Fig. 4. This response pattern during recovery has never been previously observed in this laboratory with single or repeated exposures to individual irritants or mixtures (Alarie, 1966; 1973a,b; Barrow et al., 1977; Barrow et al., 1978; Kane and Alarie, 1977, 178a,b). It was quite different from the recovery phase following

acrolein or sulfur dioxide alone (Figs. 1 and 2).

*Series II: Constant Acrolein Concentration*

The results obtained during the second series of experiments are shown in Table 2. Within each set at high sulfur dioxide concentrations, the response pattern was that typical of sulfur dioxide. With low sulfur dioxide concentrations, the response was typical of acrolein. At intermediate sulfur dioxide concentrations, mixed (S/A) responses were seen.

The maximum percentage decrease in respiratory rate was always lower than that expected for acrolein alone, except when the sulfur dixoide concentration was very low. An interesting finding was that as the sulfur dioxide concentration decreased within each constant/acrolein set, the magnitude of the response decreased slightly, and then increased. This was most notable in the second group, with a constant acrolein concentration
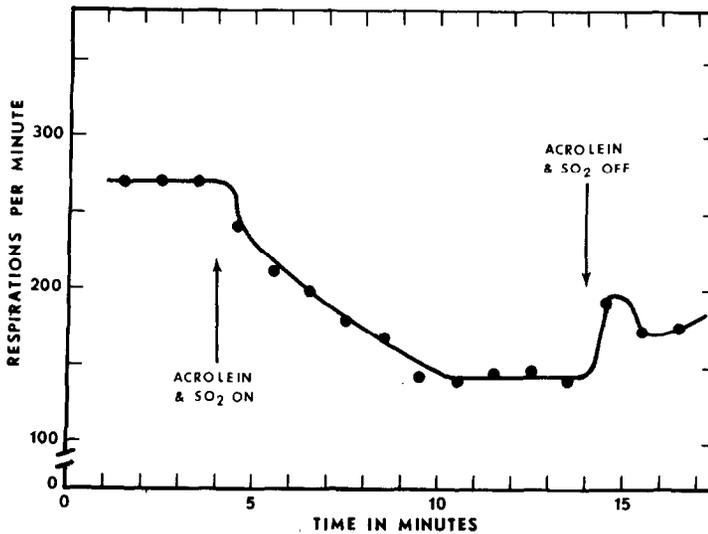
FIG. 4. Time pattern during joint sulfur dioxide/acrolein exposure showing an acrolein type (A) of response during the exposure to 55 ppm sulfur dioxide and 1.7 ppm acrolein. Note the decrease in respiratory rate after the acrolein and sulfur dioxide are turned off.

TABLE 2

RESULTS FOR SERIES II: CONSTANT ACROLEIN CONCENTRATION. RESPIRATORY RATE IN MICE.

| Acrolein | | Sulfur dioxide | | Sulfur dioxide acrolein Concen- tration ratio. | Observed response | | Expected response if effects are additive. Decrease in respiratory rate (%) |
|---|---|---|---|---|---|---|---|
| Concen- tration (ppm) | Expected decrease in respiratory rate[a] (%) | Concen- tration (ppm) | Expected decrease in respiratory rate[b] (%) | | Pattern observed | Decrease in respiratory rate (%) | |
| 3.4 | 62 | 140 | 54 | 42/1 | S | 53 | 85[c] |
| 3.4 | 62 | 55 | 35 | 16/1 | S/A | 41 | 85[c] |
| 3.4 | 62 | 32 | 23 | 9/1 | A | 58 | 85 |
| 3.4 | 62 | 23 | 17 | 7/1 | A | 54 | 79 |
| 1.7 | 50 | 55 | 35 | 32/1 | S | 34 | 85 |
| 1.7 | 50 | 51 | 33 | 30/1 | S | 22 | 83 |
| 1.7 | 50 | 37 | 26 | 22/1 | S | 38 | 76 |
| 1.7 | 50 | 23 | 17 | 14/1 | S/A | 30 | 67 |
| 1.7 | 50 | 18 | 12 | 11/1 | A | 37 | 62 |
| 1.7 | 50 | 10 | 10 | 6/1 | A[d] | 50 | 60 |
| 0.85 | 38 | 55 | 35 | 65/1 | S | 24 | 73 |
| 0.85 | 38 | 23 | 17 | 27/1 | S | 12 | 55 |
| 0.85 | 38 | 10 | 10 | 12/1 | A | 17 | 48 |

[a] From acrolein linear regression $y = 41.2 + 39.4 \log x$.

[b] From sulfur dioxide linear regression $y = -47.1 + 47.0 \log x$.

[c] Eighty-five percent is the greatest response experimentally observed even at very high acrolein concentrations (Kane and Alarie, 1978).

[d] No respiratory decrease seen during recovery phase.

of 1.7 ppm. If no sulfur dioxide was present, a 50% decrease in respiratory rate would be expected. When the sulfur dioxide concentration was 55 ppm, the respiratory decrease was 34%; when the sulfur dioxide concentration was lowered to 10 ppm, the respiratory decrease was 50%, which is what would be expected for acrolein alone. Thus, it appears that the higher concentrations of sulfur dioxide allowed for some inactivation of the acrolein, a finding similar to the results presented in Table 1. In each group, the lowest responses (smallest decreases in respiratory rate) occurred at intermediate sulfur dioxide levels, and not the highest, as would be expected if the two irritants acted antagonistically.

As in Series I, the postexposure phase showed a characteristic pattern with an increase, and then *decrease* in respiratory rate. This pattern was seen in all the experiments, except at low sulfur dioxide (10 ppm) concentrations.

## Series III: Constant Sulfur Dioxide Concentration

This series does not include any new experiments; rather it is a new grouping of the results. These results presented in Table 3 show that for a constant sulfur dioxide exposure concentration, the pattern of the response changed from an acrolein-type (A) response at high acrolein concentrations to a mixed (S/A) response at intermediate acrolein concentrations, and sulfur dioxide (S) pattern at very low acrolein concentrations.

The results concerning the magnitude of the response and pattern during the postexposure phase have been presented for Series I and II.

## Series IV

Since a postexposure decrease in respiratory rate has never been previously observed the purpose of this series of experiments was to determine if the duration of the combined sulfur dioxide/acrolein exposure affected the

TABLE 3

RESULTS FOR SERIES III: CONSTANT SULFUR DIOXIDE CONCENTRATION. RESPIRATORY RATE IN MICE

| Sulfur dioxide | | Acrolein | | Sulfur dioxide acrolein Concentration ratio | Observed response | | Expected response if effects are additive. |
|---|---|---|---|---|---|---|---|
| Concentration (ppm) | Expected decrease in respiratory rate[a] (%) | Concentration (ppm) | Expected decrease in respiratory rate[b] (%) | | Pattern observed | Decrease in respiratory rate (%) | Decrease in respiratory rate (%) |
| 55 | 35 | 3.4 | 62 | 16/1 | A | 62 | 85[c] |
| 55 | 35 | 1.7 | 50 | 32/1 | A | 49 | 85 |
| 55 | 35 | 0.85 | 38 | 65/1 | S/A | 29 | 73 |
| 35 | 23 | 3.4 | 62 | 10/1 | A | 54 | 85 |
| 35 | 23 | 1.7 | 50 | 21/1 | A | 52 | 73 |
| 35 | 23 | 0.85 | 38 | 42/1 | S/A | 18 | 61 |
| 23 | 17 | 3.4 | 62 | 7/1 | A | 54 | 79 |
| 23 | 17 | 1.7 | 50 | 14/1 | S/A | 30 | 67 |
| 23 | 17 | 0.85 | 38 | 21/1 | S | 12 | 55 |

[a] From sulfur dioxide linear regression $y = -47.1 + 47.0 \log x$.

[b] From acrolein linear regression $y = 41.2 + 39.4 \log x$.

[c] Eighty-five percent is the greatest response experimentally observed even at very high acrolein concentrations (Kane and Alarie, 1978).

recovery pattern using 55 ppm of sulfur dioxide and 1.7 ppm acrolein. It was found that if the exposure lasted less than 3 min, there was no decrease in respiratory rate during the postexposure phase. If the exposure lasted 4 min or more, the postexposure decrease in respiratory rate was always seen as in the series of experiments described above.
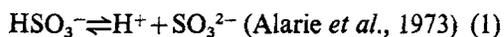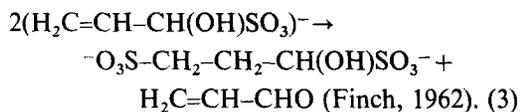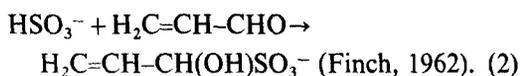
## DISCUSSION

This work followed previous experiments (Kane and Alarie, 1978a) in which it was shown that the respiratory rate decrease caused by inhalation of acrolein/formaldehyde mixtures was consistent with the hypothesis that the two sensory irritants acted at the same receptor site and exhibited competitive agonism. The results of the sulfur dioxide/acrolein exposures, showed that three distinct time–response patterns were observed, and that each pattern depended mainly upon the sulfur dioxide/acrolein concentration ratio, and not the individual concentrations *per se*. Because of this difference in time response pattern and the fact that the maximum response occurred more rapidly with sulfur dioxide than with acrolein, it was, unfortunately, not possible to use the more general approach of data analysis for interactions used in the previous study with acrolein/formaldehyde mixtures (Kane and Alarie, 1978a). Thus, the results were analyzed in terms of calculating the expected responses and comparing with the observed responses regardless of the time at which the maximum occurred. It should be noted that there was no indication of competitive agonism or synergism between acrolein and sulfur dioxide. At the highest sulfur/dioxide acrolein concentrations, 140/3.4, the magnitude of the observed response was what would be expected from sulfur dioxide alone (53 vs 54%). The presence of the acrolein had no apparent effect on the level of the response. Acrolein alone at 3.4 ppm would be

expected to induce about a 62% decrease in respiratory rate and thus, definite antagonism would be suggested.

The experiments presented in this paper all showed a consistent finding of a decrease in respiratory rate *after* the exposure had ended and thus, a delay in recovery toward preexposure levels. When mice were exposed to acrolein alone, the response during the postexposure period was always a rapid return to the preexposure respiratory rate (Kane and Alarie, 1977). With sulfur dioxide alone, the mice recovered toward preexppoure respiratory rate during the exposure (Alarie et al., 1973). Numerous other sensory irritants have been evaluated in this laboratory individually (Alarie, 1966, 1973b; Barrow et al., 1977; Kane and Alarie, 1977) and in mixtures (Barrow et al., 1978; Kane and Alarie, 1978a,b), and at no time has a decrease in respiratory rate been observed *after* the end of the exposure.

This observation may be of significance because it implies that a second sensory irritation episode might occur following removal from a contaminated environment. To explain this phenomenon, it can be postulated that during exposure, the presence of the sulfur dioxide results in some deactivation of the acrolein by the formation of an acrolein bisulfite adduct in the nasal mucosa. This postulate is consistent with the finding that the level of response to the mixture was always less than predicted from additive effects of each component. Formation of acrolein bisulfite adduct has been demonstrated *in vitro* (Finch, 1962) and it is rapid in aqueous media above pH 5.2. With the termination of the exposure, the adduct formed would then disproportionate to release free acrolein and, thus, immediate recovery would be prevented. This series of reactions could occur as follows in the water phase of the nasal mucosa, since both sulfur dioxide and acrolein are water soluble.

$$SO_2 + H_2O \rightleftharpoons H_2SO_3 \rightleftharpoons H^+ +$$

$$HSO_3^- \rightleftharpoons H^+ + SO_3^{2-} \text{ (Alarie et al., 1973) (1)}$$

$$HSO_3^- + H_2C=CH-CHO \rightarrow$$
$$H_2C=CH-CH(OH)SO_3^- \text{ (Finch, 1962). (2)}$$

$$2(H_2C=CH-CH(OH)SO_3)^- \rightarrow$$
$$^-O_3S-CH_2-CH_2-CH(OH)SO_3^- +$$
$$H_2C=CH-CHO \text{ (Finch, 1962). (3)}$$

Reactions (1) and (2) would require a constant source of sulfur dioxide. At the end of the exposure, with the removal of the source of sulfur dioxide, reaction (3) would occur releasing free acrolein, thus allowing a sensory irritation response to occur.

According to this postulate, if a combined sulfur dioxide/acrolein exposure were begun, and then the sulfur dioxide alone was stopped, an increase in response (i.e., greater decrease in respiratory rate) should occur. This would be because the removal of the sulfur dioxide would allow disproportionation of the acrolein adduct formed, resulting in a higher acrolein concentration near the receptor sites. When the acrolein exposure ended, no respiratory rate decrease should be seen in the postexposure phase, since there would be no sulfur dioxide/acrolein adduct left to disproportionate and immediate recovery would be expected.

Conversely, if an experiment were conducted in which initially only acrolein was present, and then sulfur dioxide was introduced, the response would decrease as the sulfur dioxide would inactivate some of the acrolein by adduct formation. If both were stopped simultaneously, a decrease in respiratory rate should occur as the sulfur dioxide/acrolein adduct disproportionated to release free acrolein.

To test such predictions, sulfur dioxide was presented alone for the first 2 min, and then the acrolein exposure was also started. This was done to eliminate the possibility of ambiguous results, due to the S/A type of response pattern that would otherwise occur. After a definite plateau response to the combined presence of sulfur dioxide and acrolein had been established, the sulfur dioxide was turned off. Immediately following the removal of the sulfur dioxide, there was an increase in the response (i.e., greater decrease in respiratory rate) as shown in Fig. 5. When the acrolein was turned off, as also
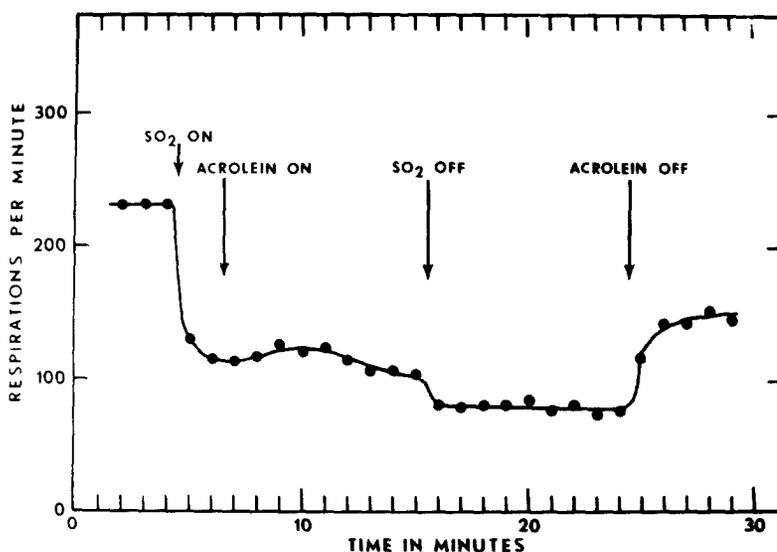


FIG. 5. Response pattern for exposure to sulfur dioxide (120 ppm) for 2 min, immediately followed by concurrent exposure to acrolein (1.7 ppm) for the next 8 min. The sulfur dioxide was then turned off, and the acrolein exposure continued for another 10 min. When the acrolein exposure ended, there was no decrease in respiratory rate during the postexposure period.
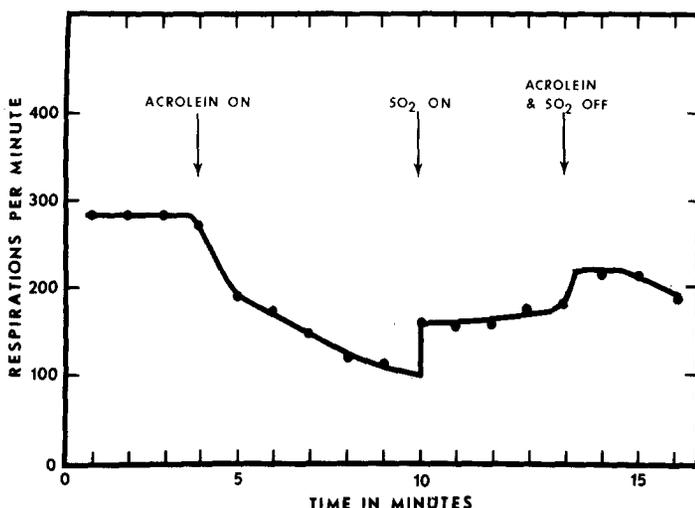
FIG. 6. Response pattern for exposure to acrolein (1.7 ppm) alone for 6 min, immediately followed by concurrent exposure to sulf dioxide (50 ppm). Note the decrease in response when the sulfur dioxide administration began, and the decrease in respiratory rate after the exposure to both acrolein and sulfur dioxide ended.

shown in Fig. 5, the response pattern showed only the increase in respiratory rate, as would be predicted with acrolein alone, with the immediate recovery as expected.

In the next experiment (Fig. 6), first, only acrolein was present. After the response to acrolein had reached a plateau, the sulfur dioxide was introduced and there was an immediate decrease in response (i.e., increase in respiratory rate) thus, showing inactivation ˉof the effect of acrolein. When both irritants were turned off, a decrease in respiratory rate was observed as in all experiments with sulfur dioxide/acrolein mixtures and opposite to the expected effect of each irritant alone.

These results support the fact that sulfur dioxide can inactivate the effects of acrolein, possibly by forming the adduct as postulated. The second sensory irritation reaction occurring at termination of exposure would be due to the acrolein formed from disproportionation of the adduct.

In conclusion, it has been shown that two sensory irritants, sulfur dioxide and acrolein can exhibit an antagonistic effect that is ratiodependent. In addition, this combined exposure can lead to a secondary sensory irritation effect after the exposure is terminated.

## REFERENCES

ALARIE, Y. (1966). Irritating properties of airborne materials to the upper respiratory tract. Arch. Environ. Health 13, 433–449.

ALARIE, Y. (1973a). Sensory irritation by airborne chemicals. CRC Crit. Rev. Toxicol. 2, 299–363.

ALARIE, Y. (1973b). Sensory irritation to the upper airways by airborne chemicals. Toxicol. Appl. Pharmacol. 24, 279–297.

ALARIE, Y., WAKISAKA, I., AND OKA, S. (1973). Sensory irritation by sulfur dioxide and ˙chlorobenzylidene malononitrile. Environ. Physiol. Biochem. 3, 53–64.

BARROW, C., ALARIE, Y., WARRICK, J., AND STOCK, M. (1977). Comparison of the sensory irritation response in mice to chlorine and hydrogen chloride. Arch. Environ. Health 32, 68–76.

BARROW, C., ALARIE, Y., AND STOCK, M. (1978). Sensory irritation and incapacitation evoked by thermal decomposition products of polymers and comparisons with known sensory irritants. *Arch. Environ. Health* **33**, 79–88.

FINCH, H. (1962). Bisulfite adducts of acrolein. *J. Org. Chem.* **27**, 649–651.

KANE, L., AND ALARIE, Y. (1977). Sensory irritation to formaldehyde and acrolein during single and repeated exposures in mice. *Amer. Ind. Hyg. Ass. J.* **38**, 509–522.

KANE, L., AND ALARIE, Y. (1978a). Evaluation of sensory irritation from acrolein–formaldehyde mixtures. *Amer. Ind. Hyg. Ass. J.* **39**, 270–274.

KANE, L., AND ALARIE, Y. (1978b). Sensory irritation of some select experimental photochemical oxidants. *Arch. Environ. Health* **33**, 244–250.

NIOSH (1974). *NIOSH Manual of Analytical Methods.* U.S. Department of HEW, Public Health Service, pp. 118.1–118.6.

ULRICH, C., HADDOCK, M., AND ALARIE, Y. (1972). Airborne chemical irritants; role of the trigeminal nerve. *Arch. Environ. Health* **24**, 37–42.

WEST, P., AND GAEKE, G. (1956). Fixation of sulfur dioxide as disulfitomercurate (II) and subsequent colorimetric estimation. *Anal. Chem.* **28**, 1816–1819.