

## Paraquinone Secretion by Confused Flour Beetles After Carbon Dioxide or Nitrogen Anesthesia<sup>1</sup>

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The existence of paraquinone secretions from confused flour beetles, *Tribolium confusum* Jacquelin duVal, and certain functions of these compounds have been known for a considerable period. Chapman (1926) first demonstrated that paraquinone secretions induced physical abnormalities in immature *Tribolium* sp. It has been variously suggested that paraquinone toxicity may be 1 reason why this species, given a normal respiratory gas environment and nonconditioned media, is unable to increase its population density in cultures beyond a certain point. The objective of this study was to determine if para-

quinones were secreted during and/or after anesthesia and appraise these secretions as possible biasing factors in toxicological situations.

PROCEDURES.—Six 60-ml respirometric flasks were partly immersed in a 30°C water bath. Each flask contained 100 2-week-old confused flour beetles. One of the following gases—dry or humidified N<sub>2</sub>, CO<sub>2</sub>, or preanalyzed air (21.5% O<sub>2</sub>; 0.03% CO<sub>2</sub>; and 78.47% N<sub>2</sub>)—was released into each flask at a flow rate of 100 cc/min. The treatment gas flowed past a given lot of beetles for 2 hr. After this, each lot was permitted a 2-hr recovery period in which the atmosphere consisted of moistened air flowing at 100 cc/min. Each gas or air stream passed over the insects and was directed by glass tubing to the bottom of an Erlenmeyer flask containing a 2-in.-diam potassium iodide (KI) starch-paper disk dampened with 1 ml of 1% H<sub>2</sub>SO<sub>4</sub>. The

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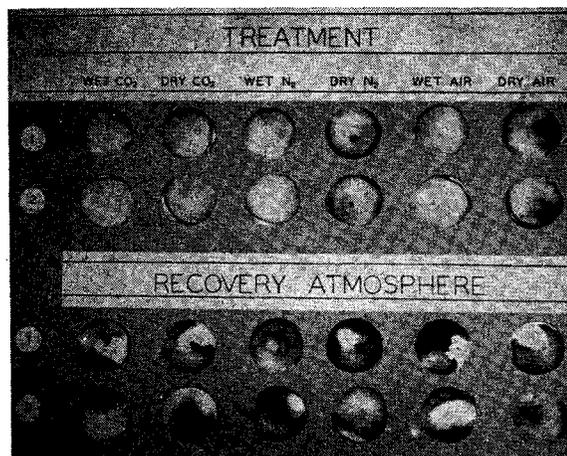


FIG. 1.—Quinone secretion of confused flour beetle adults during CO<sub>2</sub> and N<sub>2</sub> anesthesia and air exposure and in subsequent recovery. Dark areas on disks indicate quinone reaction.

glass tube conveyed the gas with insect effluent to a point  $\frac{1}{4}$  in. above the center of the disk. Another glass tube in the Erlenmeyer flask's rubber stopper directed the exiting air to a flowmeter. The disks were changed every hour and the color development was noted. In a duplicate test, the air purge rate during the recovery period was reduced to 5 cc/min. No paraquinone assay was performed in this series.

The secretion of *T. confusum* adults was later collected after the manner of Alexander and Barton (1943). Separation and identification of these substances was by a Wilkins Aerograph<sup>8</sup> gas chromatograph with a hydrogen flame ionization detector. The column was  $5 \times \frac{1}{8}$  in. packed with 1% FFAP on 100/120 mesh AW-DMCS-treated Chromosorb G. Both paraquinones were eluted at 225°C. Hydrogen flow rate was 30 cc/min.

**RESULTS.**—A qualitative estimate was secured of paraquinone production by confused flour beetles in humidified or dry CO<sub>2</sub>, N<sub>2</sub>, or preanalyzed air. Acid-treated KI

<sup>8</sup> The use of proprietary names is for identification only, and does not necessarily constitute endorsement by the U.S. Department of Agriculture.

starch paper reacting with paraquinone-laden gas issuing from the respiratory flasks developed a purple color caused by reduced KI (Fig. 1). On this evidence, no or negligible paraquinones were secreted when the beetles were in a humidified or dry CO<sub>2</sub>, or humidified N<sub>2</sub>, or air atmosphere during the 2-hr exposure period. Slight secretion was noted during the 1st and 2nd hours in the dry N<sub>2</sub> environment. Dry air evoked a similar but more pronounced secretion during this exposure period. Considerable secretion was noted in all treatments during the following 2-hr recovery period in moistened air. All beetles treated with fast-flowing (100 cc/min) CO<sub>2</sub> or N<sub>2</sub> died or became moribund during the succeeding low rate (5 cc/min) air purge.

The secretion as analyzed by gas chromatography consisted of 2-methyl-1,4 hydroquinone and 2-ethyl-1,4 hydroquinone.

**DISCUSSION.**—The study of paraquinone secretion under these circumstances was not by original intent. Other experiments required fumigation of the confused flour beetle after a recovery period from a CO<sub>2</sub> or N<sub>2</sub> exposure. Originally, the gas was purged from the chambers with pre-analyzed air at a high rate of speed (100 cc/min) during the recovery. It was known that a 2-hr, 100-cc/min air purge without previous treatment history usually caused 2 to 3% mortality. Later, a low airflow rate (5 cc/min) was substituted because this method was thought to cause even less stress to the insects. When the flask-ventilation rate was reduced, paraquinones concentrated until they became autotoxic to the insects. Had this fact not been recognized, mortality immediately after fumigation would have been ascribed to fumigation and/or the pre-fumigation anoxia. It would appear that paraquinones could exert a debilitating to actual mortal effect on the insects in later survival studies. To leave flour beetles without absorbent food or on an enclosed and slippery glass substrate for extended periods might introduce unnecessary variables into the experiment.

These data suggest also that paraquinone secretion by the confused flour beetle may be suppressed by anoxia. The expression of these compounds appears to be a response to the stress of anoxia and rapidly flowing air.

#### REFERENCES CITED

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