

# MICRONIZED FLUORESCENT DUSTS FOR MARKING *CULICOIDES VARIIPENNIS* ADULTS<sup>1, 2, 3</sup>

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**ABSTRACT.** Sixteen colors of micronized fluorescent dusts were evaluated as marking materials for *Culicoides variipennis* adults. The dusts were applied directly rather than incorporated with gum arabic. Only three pigments; U. S. Radium green (1953), yellow (2267), and green (3206), were found to be distinguishable

for the duration of the life of the flies, non-transferable from marked to unmarked individuals during copulation, and insoluble in 70% ethanol. These three colors also had no significant (0.05 level) effect on adult longevity.

## INTRODUCTION

A variety of materials have been used to mark insects for flight range and dispersal studies. A review of the various materials and application methods is given by Dobson (1962), Gangwere et al. (1964) and Southwood (1972). We evaluated 16 colors of micronized fluorescent dusts for marking *Culicoides variipennis* (Coquillett) (Diptera: Ceratopogonidae) adults. The durability of each color as a marker was

determined. Colors found to remain distinguishable for extended periods were evaluated further to determine the effect on adult longevity, the probability of transfer from marked to unmarked individuals during copulation, and the solubility/removal of the materials in 70% ethanol. Ethanol is commonly used as a killing agent on *C. variipennis* collection traps (McDonald 1970, Lillie et al. 1979); therefore, it is important to determine whether the fluorescent material would be removed by it.

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<sup>2</sup> The opinions and assertions contained herein are the private ones of the authors and are not to be construed as views, either official or unofficial, of the USAF.

<sup>3</sup> Mention of a proprietary product in this paper does not constitute an endorsement of the product by the USAF or the USDA.

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## MATERIALS AND METHODS

Adult *C. variipennis* from a colony at the USDA Agricultural Research Center in Denver, Colorado, were used for all tests. Six colors of dusts were obtained from United States Radium Corporation<sup>6</sup> and 10 colors from Day-Glo Color Corporation<sup>7</sup> (Table 1). Particle size, chemical base, and other properties of each color are given by the respective corporations (US Radium 1976, Day-Glo 1977a,b). The dusts were applied to adult flies in the manner described by Moth and Barker (1975). The marking procedure involved anesthetizing the specimens with CO<sub>2</sub>, placing them in a 60 ml wide mouth bottle, and applying the dust with a DeVilbiss number 119 insufflator. Three puffs of dust were administered. Each color was applied directly rather than incorporated with gum arabic.

Table 1. Fluorescent dusts tested as marking materials with colony flies. Three colors (\*) were found to remain distinguishable for the life of the flies, to be insoluble in 70% ethanol, to be nontransferable from marked to unmarked individuals during copulation, and to have no effect on the mortality of marked specimens.

Day-Glo	US Radium
Corona Magenta	*Green (1953)
Aurora Pink	Blue-White (2200)
Rocket Red	Red (2225)
Fire Orange	*Yellow (2267)
Blaze Orange	*Green (3206)
Arc Yellow	Orange (3336)
Neon Red	
Saturn Yellow	
Signal Green	
Horizon Blue	

The durability of each color as a marker was determined by marking approximately 200 adults with the material and periodically comparing the marked flies with an unmarked group. The probability of transferring dust from marked to unmarked individuals during copulation was determined by placing marked flies with unmarked flies of the opposite sex. The sexes were segregated in the pupal stage and allowed to emerge separately. Fifty-three females were marked and caged with 55 unmarked males. The same procedure was followed for a group of 47 marked males caged with 29 unmarked females. Both groups were allowed to blood feed and were kept caged for 7 days before being killed for examination.

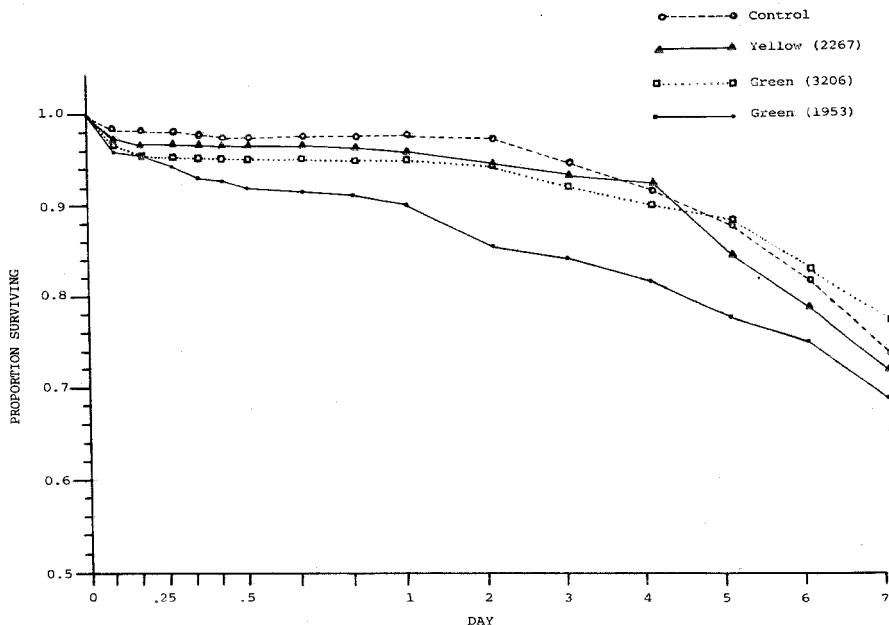


Fig. 1. Survival curves for colony flies marked with fluorescent dusts and for an unmarked control group. There was no significant (0.05 level) difference between the survival of marked and unmarked flies when the results were compared with a two-way analysis of variance.

The solubility/removal of the fluorescent dusts in 70% ethanol was determined by placing a number of marked flies, 24 hr after marking, in the ethanol solution. Periodically, over a 10-month period the container was agitated and the specimens were examined for fluorescent dust.

The effect of the marking material on adult longevity was examined by comparing the mortality of marked flies with that of an unmarked control group. The controls were anesthetized as often as each dusted group. Cages containing 200–300 marked or control flies were placed in an incubator with a constant setting of  $23 \pm 1^\circ\text{C}$  and 24 hr fluorescent light. A solution of 10% sucrose and water was provided as a carbohydrate source. Vials containing the solution were changed daily. Mortality was determined every 2 hr for the first 12 hr, every 4 hr for the following 12 hr, and then every 24 hr for 6 days. The test was replicated 3 times. The data were transformed to relative frequencies and a two-way analysis of variance was used to determine significance (Dunn and Clark 1974).

## RESULTS AND DISCUSSION

All Day-Glo dusts were unsatisfactory because they did not adhere to the cuticle of the flies (Table 1). Only 3 of the 6 US Radium dusts: green (1953), yellow (2267), and green (3206), were durable as marking materials (maximum observed, 30 days). Other researchers have incorporated the dusts with gum arabic (Sinsko and Craig 1979, Brust 1980), but that was not necessary for this study. Marked flies were able to remove some of the material with preening and wing movements but they were not able to remove particles from the thorax. The 3 durable colors were insoluble and would not wash off in 70% ethanol (maximum observed, 10 months). Ethanol can safely be used as a killing agent on collection traps for dispersal studies with these dusts.

Colors 1953, 2267, and 3206 were also nontransferable from marked to unmarked individuals during copulation.

Thus, there is a low probability of unmarked flies acquiring the dust upon contact with marked specimens. This ensures that specimens recovered in a mark/release/recapture study are those actually released. Most importantly, the 3 durable colors did not significantly (0.05 level) affect the longevity of marked flies when compared with an unmarked control group (Figure 1).

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