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Communications

Effect of Some Perfume Oils on Dehydrogenases in *Escherichia coli*

Sir:

Since the discovery that perfume oils inhibit the growth of bacteria (1, 2) we have investigated the antimicrobial properties of many such oils further (3, 4). The mode of action by which bacteria are killed by perfume oils remains unknown but Gal'perin and Dunaeva (5, 6) have reported that essential oils affect dehydrogenases in paramecia and helminths. Since essential oils are commonly used in perfume oil formulas it was thought that bacterial dehydrogenases might be affected by perfume oils.

We selected as the test organism *Escherichia coli* ATCC 11229. The bacteria were harvested after 24 hours from nutrient agar slants and washed three times with sterile physiological saline by centrifugation. A 2-ml. quantity of washed packed cells was added to 3 ml. of 1:1000 perfume oil solution (1% alcoholic). After 30 minutes of contact the cells were removed by centrifugation and washed three times with sterile saline and suspended in saline to 300 Klett units in the Klett-Summerson photoelectric colorimeter using the No. 42 filter with a spectral range of 400 to 465 m μ . This treated standard cell suspension was used for all subsequent experiments.

The dehydrogenase activity was determined by the Thunberg technique (7). The substrates used were glucose, succinic acid, and malic acid. The color intensity of the methylene blue in the Thunberg tubes was recorded every 4 minutes for a total of 32 minutes. All experiments were run at least in duplicate with controls and were replicated several days later.

Perfume oil of bouquet No. 821, sweet grass,

lilac water, and bouquet No. 21 completely inhibited glucose, succinic acid, and malic acid dehydrogenase. Plate counts of the standard cell suspension showed no reduction in cell number compared to controls. However, after 2-hour exposure of the test organism to each perfume oil, plate counts revealed a 10 to 50% reduction depending upon the oil used. This finding suggests that perhaps the reduction of dehydrogenases precedes the death of the cell.

E. coli was exposed to perfume oil of neroli (artificial), evergreen bouquet, osheana, and chypre 66D at a concentration of 1:2000 in the same manner as above and dehydrogenase activity measured. It was found that oil of neroli and evergreen bouquet completely inhibited all tested dehydrogenases, while oil of osheana and chypre 66D caused little effect on glucose dehydrogenase, more than 90% inhibition of succinic dehydrogenase, and complete suppression of malic dehydrogenase.

Essential oils and their components are presently being investigated for their effects on bacterial dehydrogenases.

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