

of n (4, 5, 6, 8 and 10) are not evident in their reported spectrum. Either they are buried in the noise or there is a reason for their absence.

If indeed there are harmonic emissions corresponding to 13.6 n eV, $n = 2-11$, these lines could be scattered inelastically by the He atoms that are abundant in the plasma, as illustrated in Figure 1 *b*. The most likely inelastic transition in He is $1s^2 \rightarrow 1s2p$ and this involves an energy difference of 21.21 eV. Therefore, the observed Raman (inelastic scattering of the harmonics shown in Figure 1 *a*) lines would be expected at $(13.6n - 21.21)$ eV, $n = 2-11$. For $n = 1$, the energy of the photon is much less than that required for the electronic excitation in He. For $n = 2$ and 3, the Raman lines would be expected at 206.7 and 63.31 nm, respectively and they fall outside the region of the EUV spectrum reported in refs 1, 2. For $n = 4$, the Raman line would be expected to occur at $\lambda = 37.4$ nm, and it is clearly seen as a prominent peak in figures 7, 10 and 12 of ref. 1. The other lines corresponding to $n = 6$ and 8 occur, as expected, at $\lambda = 20.5$ and 14.15 nm, respectively. The lines corresponding to $n = 5$ and 10 are clearly missing (or are not above the noise level).

Clearly there is a complementarity between the observed harmonics and the Raman lines. For $n = 2, 3, 9$ and 11, the harmonics are clearly noticeable and for $n = 4, 6$ and 8 the Raman lines are clearly noticeable. For some reason, lines corresponding to $n = 5$ and 10 are missing, on both counts.

There could also be emission resulting from the recombination of He^{2+} and an

electron to yield He^+ in its ground state, at $\lambda = 22.8$ nm and this could be inelastically scattered by He ($1s^2$), accounting for the line at $\lambda = 37.4$ nm, as illustrated in Figure 1 *c*. In addition, there could be the emission corresponding to the second harmonic of the (He^{2+} , e) recombination and concomitant Raman scattering by He ($1s^2$) resulting in emission at $\lambda = 14.15$ nm (see Figure 1 *c*). In principle, one could resolve the components of emission at $\lambda = 22.8$ nm and 14.15 nm arising from the harmonics of (H^+ , e) recombination and (He^{2+} , e) recombination, based on the differences in the reduced mass of the electron relative to the hydrogen nucleus and the helium nucleus. In practice, this is not possible at the present level of spectral resolution.

Mills and Ray^{1,2} have clearly pointed out that the emission lines mentioned above are not observed when pure He or H_2 is used in the discharge, or when other gases such as Ne, Ar, Kr, Xe, N_2 , O_2 and CO_2 are used along with H_2 .

Why only the helium-hydrogen (98/2%) plasma leads to EUV emission remains to be understood. One has to examine the origin of the harmonics emission. There could be some aspects of the plasma that make this possible.

One might argue about our usage of the expression 'Raman scattering', which is usually used in the context of vibrational/rotational inelastic transition. This is because the usually available visible light sources do not have sufficient energy to cause electronic transitions. There is no reason why electronic Raman cannot occur, if suitable light source is available and the corresponding selection rules are

obeyed³. It is not clear as to why emission lines corresponding to $n = 5$ and 10 are missing and also why there is a complementarity between the emission corresponding to $n = 2, 3, 7, 9$ and 11 and Raman scattering with emission corresponding to $n = 4, 6$ and 8.

These are clearly issues that need further investigation and we hope to take up some of them in the immediate future.

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A rapid method for measuring olfactory responses of *Drosophila* larva*

The olfactory responses of *Drosophila* larvae are measured by following its chemotactic movements on a petri dish. Several variants of this test have been employed. In the commonly used version of the experiment¹⁻³, diluted odorant is placed near the edge of the dish and a

spot of the diluent diametrically opposite. After 5 min, the larvae on the two halves of the dish, the odour side (*O*) and the control side (*C*) are counted, and the response index is computed as $O - C / O + C$. This method of measuring olfactory response suffers from certain disadvantages. The test is slow; since the odour spreads on the dish by diffusion, it takes a few minutes for the larvae on the farther side of the odour source to res-

pond. On the other hand, the larvae near the source respond early and, if the concentration of the odorant is high, they become desensitized and wander-off. We describe here a modification of the larval test, which enables us to assess the attraction response rapidly by measuring the initial rate of entry in a zone around the odour source.

CsBZ flies were grown on standard cornmeal yeast medium at 24°C and

*Dedicated to Prof. S. Ramaseshan on his 80th birthday.

maintained on a 12 h day/night cycle. Standard procedures were used in handling cultures^{4,5}. Larvae at a prescribed stage of development were separated from the cornmeal culture medium by floating on 30% polyethylene glycol and washed free of debris in tap water. A fixed number of larvae, between 30 and 50, were placed at the centre of a petri dish filled with 1% agar and allowed to spread out. As they reach an outer 3 cm ring, 10 μ l of diluted odorant ethyl acetate (EA) was placed in a small well at the centre (Figure 1). As the larvae sense the odour, they turn around and move towards the centre. The response is measured by the initial rate at which the larvae cross the inner 1 cm ring, marked on the dish. The response index may be defined as per cent of larvae entering the ring per minute.

The petri dish is photographed with a digital camera at short intervals of 15 s or less, and the larvae outside the ring are counted, either from a printout of time-lapse photographs or using a computer program written by one of the authors (V.A.). The automation of the experiment eliminates the errors in timing as well as counting, and greatly reduces experimenter's bias.

The response of third instar larvae to varying dilutions of EA in the range 10^{-7} to 10^{-4} is shown in Figure 2. It may be seen that attraction is proportional to concentration of the odorant over a hundred-fold change, but falls at concentrations greater than 10^{-5} , as repulsion sets in.

Tracks of individual larva can be reconstructed from the photographic record taken at intervals of 5 s. The larva turns towards the odour source gradually (Figure 1). The usual track is circular or spiral, although an occasional larva might make a sharp 180° turn. The curvature of the track shows considerable variation. Analysis of larval tracks provides valuable information about the mechanism of orientation⁶. The crawling speed increases with age and is correlated with an increase in body size (Figure 3). The average length of the first instar larva was 0.88 mm and its speed 1.71 cm/s. The third instar larva had a length of 3.58 mm and a crawling speed of 5.88 cm/s. The average speed in body-lengths thus remained unchanged. The larval speed also increased with the concentration of the odorant. For a tenfold increase in concentration, the speed in our experiment went up by 3.3 mm/min.

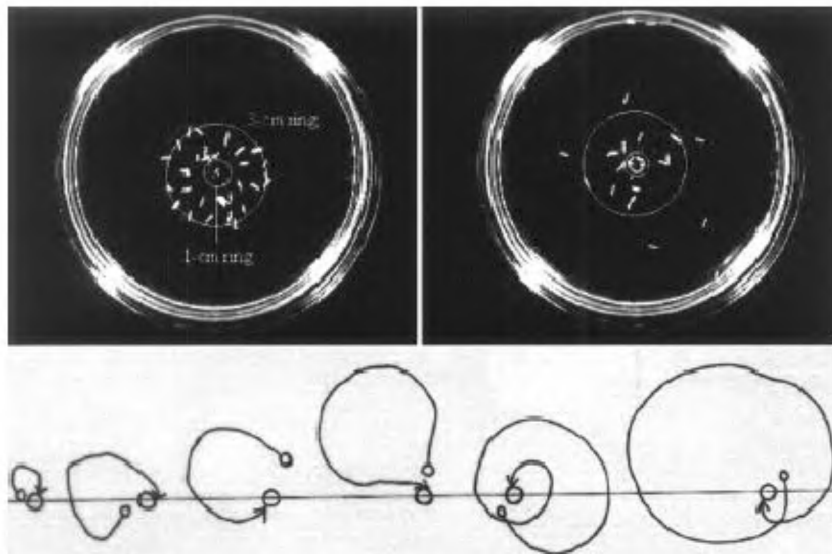


Figure 1. (Top) Test plate at 15 s (left) and 180 s (right). Tracks show larvae spiralling into the odour well. Response is measured by the rate at which they enter the inner 1 cm ring.

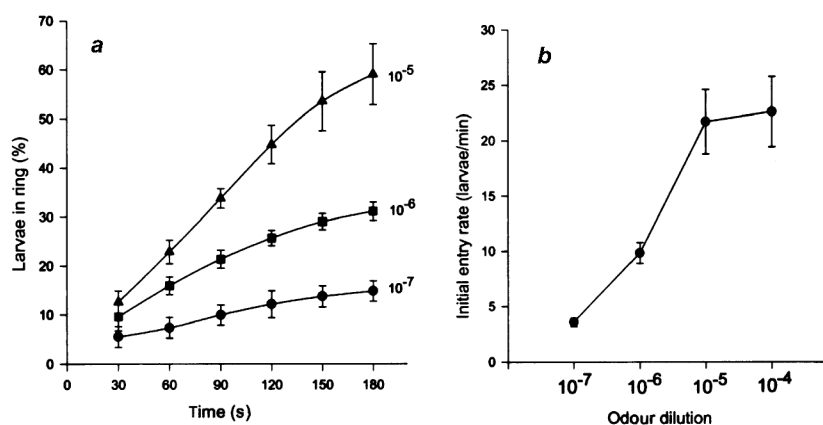


Figure 2. Entry curves showing response of larvae to increasing concentration of ethyl acetate (a). Initial rate of entry is proportional to odorant concentration in the range 10^{-7} and 10^{-5} . Attraction declines above 10^{-5} (b).

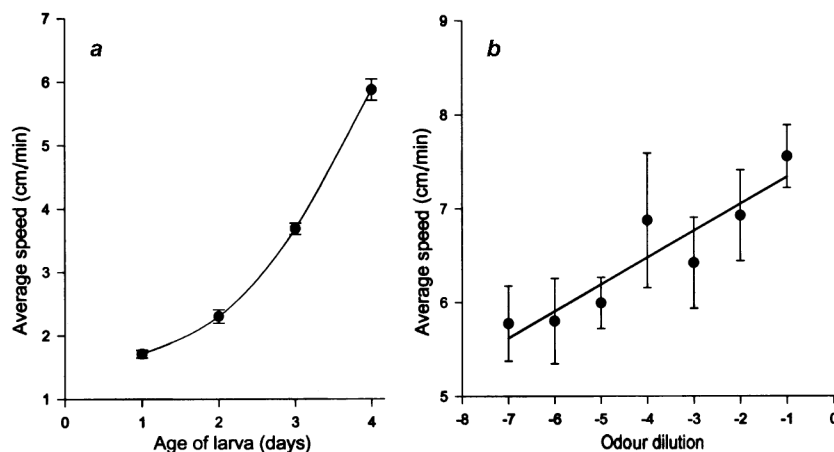


Figure 3. Crawling speed of larvae increases with age (a) and odorant concentration (b).

The chief merits of the method described above are its rapidity and accuracy. The initial rate of entry can be determined in less than 2 min and the standard error of the measurement can be easily brought down to less than 10%. The paradigm can be used for analysing the tracks (Figure 1) of individual larvae, and is specially suitable for studying olfactory learning and memory.

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