

A simple nail polish imprint technique for examination of external morphology of *Drosophila* eyes

In view of the ever-increasing genetic information and other experimental strategies, *Drosophila* continues to be a useful model for studying the various developmental pathways and genetic networks that regulate these events^{1,2}. In recent years, fly models for different human neurological and other genetic disorders have also been developed and these have been widely used for identifying interacting genes and for developing therapeutic strategies³. The compound eyes of flies have been particularly useful in this context because (i) the genetic pathways that determine eye development are better understood, (ii) the adult eyes contain both neuronal and non-neuronal cell types, (iii) they are dispensable for survival of the flies, and (iv) the phenotypic consequences of any perturbations in eye development are distinct and quantifiable⁴. The adult *Drosophila* eye contains about 800 ommatidia arranged in a highly ordered matrix with arrays of sensory bristles projecting out from the surface of each ommatidial unit⁵.

Most studies exploiting these features of the *Drosophila* eyes use scanning electron microscopy (SEM) to examine any disruption in the ordered arrays of the ommatidia in adult eyes. We describe here a novel, simple and inexpensive method that provides high quality images, comparable to those obtained by SEM, of the external surface of eyes of adult flies, but which does not require expensive facility and can be used in any laboratory with a good light microscope.

In this method, we use a transparent nail polish to create an exact replica of the external surface of the eye, which is subsequently examined using a light microscope. To obtain an imprint of adult eye, a small drop of transparent nail polish is placed on the surface of a clean glass slide. The fly to be examined for eye morphology is anaesthetized, placed on a dry area of the slide and decapitated with a sharp blade or needle. The decapitated head is held with forceps or needles and is briefly dipped in the still fluid drop of nail polish. The head is then placed in a clean and dry area of the same slide and the nail polish layer on the eyes is allowed to dry at room temperature (24°C) for 5–10 min. The dried layer of nail polish can

be easily peeled-off from the eye with the help of fine dissecting needles. The separated peel, being an exact replica of the eye surface, assumes a goblet-shaped appearance. This peel is carefully placed on another clean glass slide with the imprint side facing upright. This nail polish imprint can be directly examined and photographed under a stereobinocular microscope to provide a low magnification image of the eye surface (Figure 1 *a*). For higher magnification and better-resolved image of the eye surface, the peel is carefully flattened by gently placing a cover slip over it and carefully applying a slight pressure. The eye imprint is then examined under a microscope using

10X (Figure 1 *b, g, j*), 20X (Figure 1 *d, h, k*) or 40X (Figure 1 *e, i, l*) differential interference contrast (DIC) objective. These can be seen with bright field optics as well, but the DIC optics provide a better image. As may be noted from Figure 1, details of eye surface are generally comparable with those obtained by SEM (Figure 1 *c, f*). As shown in Figure 1 *g–l*, the nail polish imprints also easily reveal the characteristic defects due to different mutants affecting eye development. This method can be applied to study surface details of compound eyes of any insect and possibly other cuticular structures from which the peel of nail polish can be easily removed.

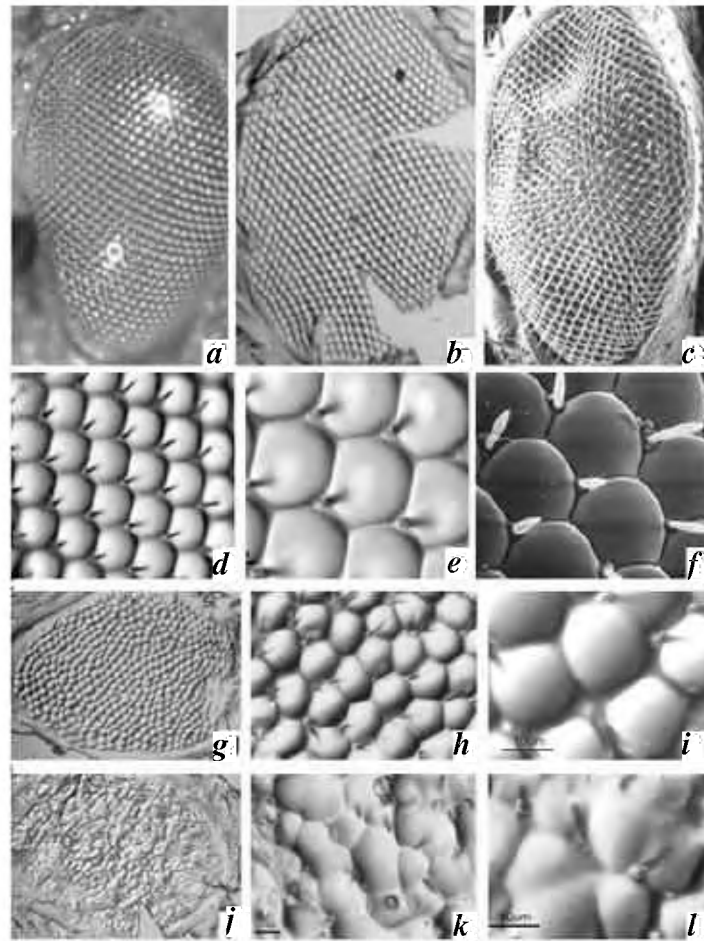


Figure 1. Nail polish imprints (*a, b, d, e, g–l*) and SEM images (*c, f*) of eyes of adult *Drosophila*. Images in (*a–f*) are from wild type, in (*g–i*) from *Elp-B1/CyO* and in (*j–l*) from *GMR-argos* mutant flies. The image in (*a*) was taken with a stereobinocular microscope (Zeiss Stemi SV6) using a 5X objective; in (*b*), (*g*) and (*j*) with a 10X objective; in (*d*), (*h*) and (*k*) with a 20X, and in (*e*), (*i*) and (*l*) with a 40X objective using a Nikon Ellipse 800 DIC microscope.

1. Flannery, C. M., *Am. Biol. Teach.*, 1997, **59**, 244–248.
2. Rubin, G. M., *Science*, 2000, **287**, 2204–2215.
3. Fortini, M. E. and Bonini, N. M., *Trends Genet.*, 2000, **16**, 161–167.
4. Sang, T. K. and Jackson G. R., *Neuro Rx.*, 2005, **2**, 438–446.
5. Ready, D. F., Hanson, T. E. and Benzer, S., *Dev. Biol.*, 1976, **53**, 217–240.

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Imprints of Chilika Lake in the offshore region – a geomorphologic evidence

Chilika Lake is the largest lagoon in the subcontinent along the east coast of India, situated between lat. 19°28' and 19°54'N, and long. 85°05' and 85°38'E. It is a shallow, brackish-water lake formed due to the silting action of the Mahanadi River, which drains into the northern end of the lake, and the northerly currents in the Bay of Bengal, which have formed a sandbar along the eastern shore leading to the formation of a shallow lagoon. During the Pleistocene, the coastline was at the western shores of Chilika¹, and the entire northeastern region above Chilika was under the sea. Since then, the coastline has moved considerably eastward due to sea-level regression. Studies^{2–12} related to the occurrence of topographic highs and relict sediments on the eastern continental shelf of India indicate the presence of different still stands of lower sea levels during late Pleistocene–Holocene due to sea-level oscillations. Three prominent still stands¹³ of the sea level were identified at depths of 30, 100 and 120 m. In particular, the regions off Gopalpur, Orissa coast have revealed^{2,3} the occurrence of some still stands up to a water depth of 120 m. However, the lowest strandlines around 130 m water depth off Mahabalipuram¹² and Visakhapatnam¹¹ are reported to have been formed during the Pleistocene. Many lagoons seen today were formed as a result of a worldwide rise in sea levels over the last 6000–8000 years. There was a pause in the rise in sea level about 7000 years ago, when a sandy beach might have formed near the coast at the southern sector. As the sea rose further, this sand beach grew gradually and progressed northeast, to form what is now the spit of Chilika. A shell of *Ostrea Virginiana* from the southwestern edge of

the spit has been dated at 3750 ± 200 years BP by the radiocarbon method, which indicates the age of the lagoon¹. The spit of Chilika is constantly changing. Due to widening of the sand bar, the position of the mouth has been constantly shifting, moving generally towards the northeast. The mouth connecting the channel to the sea is close to the northeastern end of the lagoon. A 32 km long, narrow outer channel connects the main lagoon to the Bay of Bengal.

The inlet to a lagoon is a product of interaction between waves, currents and sediments. The tidal fluctuation in Chilika Lake is about 0.2 m during non-monsoon periods, driven by a tide in the Bay of Bengal ranging from 0.9 to 2.4 m. Chilika

Lake also receives the benefit of the Mahanadi River flow in keeping the tidal inlet open.

In order to study the morphology of the seafloor over the shelf region between Gopalpur and Paradip, bathymetry data were collected along 12 coast perpendicular traverses within water depths of 40–2300 m (Figure 1), on-board the research vessel *Gaveshani* by National Institute of Oceanography, Visakhapatnam. The echo-sounding records have been digitized at close interval in order to resolve minor topographic variations. The digitized data have been plotted along the survey traverses and a stacked bathymetry profile map has been prepared (Figure 1). Further, the data have been contoured at

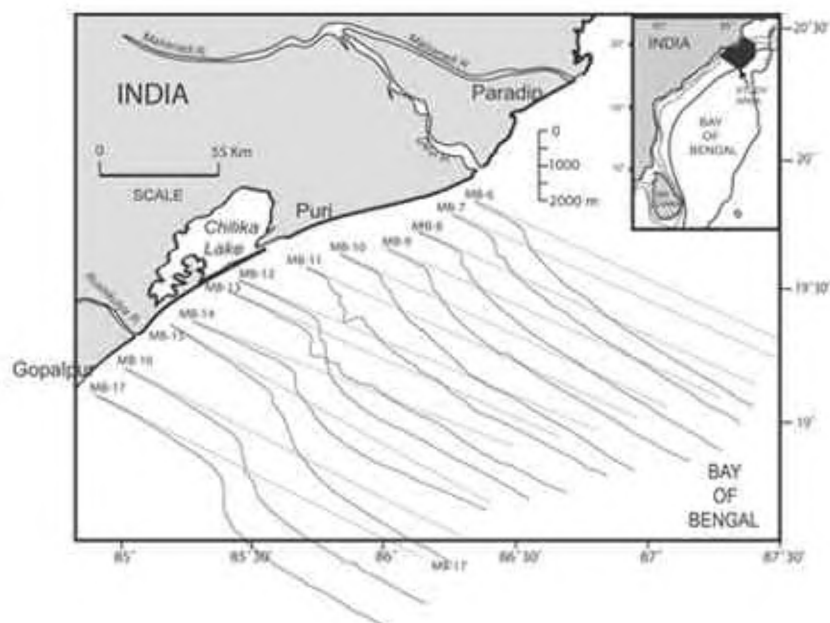


Figure 1. Map showing survey traverses. Bathymetry profiles are shown along the traverses.