

or water) securely anchored in a rock-locked steel-reinforced concrete base-plate will induce a shallow freshwater reservoir to quickly accumulate<sup>5-8</sup>. Subsequently, ~ 100% of the river's flow can be diverted (during appropriate times or season) directly to the pressure tunnel's head-gate. Nearly abandoned, the steep deep-canyon stream's bed may then be transformed into a topographically descending series of stepped runoff and erosion control sediment-trapping basins; its appearance then will resemble the 12 June–25 November 1969 dewatering of the Niagara River's American Falls Channel – a period which encompassed the First Moon Landing – and its future anthropogeomorphological state may be as identically iffy as that which Shailer S. Philbrick (1908–1994) projected for an unrestored Niagara Falls landscape region<sup>9</sup>. Water flow fluctuation means the pressure tunnel must be very well designed, dug and defended by a strong impermeable lining. An impoundment with a shallow pool depth ought not increase significantly the local seismicity and rock-slide hazard.

Imagine an integrated hydropower-generation and super-conducting electric power delivery grid netted throughout the Himalayas, serving the geopolitical and

economic development interests of India, China and other inter-linked national ecosystems! The more vociferous Greens beg or bully India and China not to burn their affordable existing coal resources – to delay the onset, or to reduce the final overall impact of, a commonly alleged 'enhanced global atmospheric warming' – the more self-righteous India and China may become when they do jointly harvest the TAR's hydropower for further separate industrialization; India and China are space-faring UNO members which remain bedeviled by down-to-earth widespread low human standard of living levels. Cooperation of these two major players as guides for Tibet's future economic development may ensure their own 21st century peace and prosperity!

Tibetans ought to have an important role in this proposed flood-mitigation and developmental macroproject's planning as well as its post-construction operation, especially if they can tolerate a small alteration in their sacred river's altitude (i.e. its flowing level) or its temporary capture for industry's use. In Tibet the major environmental impact should occur in the low-flow river within the valley north, east and south of Namjagbarwa Feng. However, the

inflatable dam can be deflated, or removed forever, at any time! This macroproject might be undertaken during 2002, which was declared (on 10 November 1998) by the United Nations Organization to be the 'International Year of the Mountains'<sup>10</sup>.

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## SCIENTIFIC CORRESPONDENCE

### Temperature-dependent cell transformation in a calcium-binding protein from *Entamoeba histolytica*

Temperature is a principle state variable which governs the structural, physical and functional properties of proteins to a large extent. We have observed an unusual temperature-dependent structural transition while attempting to collect X-ray diffraction data at room temperature (20°C) and at 4°C. This temperature-dependent cell transformation was observed in a calcium-binding protein (CaBP) from *Entamoeba histolytica*<sup>1,2</sup>. This CaBP is very similar to the structurally well-studied archetypal calmodulins (CAMs)<sup>3</sup> with two calcium-binding domains separated by a central linker. It is, however, smaller than CAM (134 residues) and has a two-residue insertion

in the linker region. As a control, similar studies have also been carried out on small proteins like ribonuclease and lysozyme. These studies suggest that this kind of cell transformation is not a general phenomenon.

Crystals of native CaBP were obtained in 63% MPD, 10 mM CaCl<sub>2</sub> and 100 mM sodium cacodylate buffer (pH 4.3) using protein at a concentration of 10 mg/ml (ref. 2). Typical crystal dimensions were 0.6 × 0.2 × 0.2 mM. The crystals belonged to the space group P6<sub>3</sub>22. Diffraction data collection was carried out using a CuK $\alpha$  X-ray source provided by a Rigaku RU200 rotating-anode generator equipped with a 0.2 mM focusing cup

and operating at 40 kV, 56 mA. Data were collected on a MAR Research imaging plate system at a crystal to film distance of 150 mm. The oscillation angle was set to 1°. Constant temperature was maintained using an Oxford Cryo-system Cryocooler. Each frame was exposed for 15 min and a total of five frames were collected at each temperature. The data set was processed using the DENZO/SCALEPACK suite of programs and the cell parameters were determined for each temperature.

The cell parameters determined for each of the eight different temperatures are shown in Table 1. As can be seen, the volume of the unit cell shows a

Table 1. Change in unit cell parameters with temperature

Temperature (K)	Unit cell parameters (Å)		Volume × 10 <sup>5</sup> (Å <sup>3</sup> )
	a = b	c	
110	95.80	63.09	5.01
120	95.72	62.94	4.99
130	96.22	63.06	5.05
140	96.34	63.01	5.06
277	96.34	62.67	5.04
283	98.96	64.50	5.47
288	98.41	64.67	5.42
293	99.26	64.62	5.51

significant increase of 8% between 4 and 9°C. Similar studies carried out on ribonuclease and lysozyme show no apparent structural changes ( $\leq 0.2\%$ , data not shown) within the same small, low temperature range.

This study thus indicates that temperature-dependent cell changes, even for small temperature alterations, cannot be altogether ignored. It is a common crystallographic practice to collect diffraction data at low temperature as it enhances the crystal's lifetime in the beam and thereby increases the quality of the measure-

ments. The data collected at different temperatures are often merged with the basic presumption that the structure remains largely unchanged and the data are isomorphous. Although true in most cases, this may not be universal as observed in the present case. Therefore, a degree of caution is necessary while merging data collected at different temperatures. These observations also underline the special features of the CaBP molecule wherein functional viability critically depends on large conformational changes.

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## Role of carboxylesterase in relation to pyrethroid resistance towards *Helicoverpa armigera* (Hubner) (Lepidoptera: Noctuidae) in Tamil Nadu

*Helicoverpa armigera* is a serious pest of many crops in India including cotton. In India, large-scale failure to control this pest with insecticides was first recorded in the major cotton-growing region of Andhra Pradesh in 1987 (ref. 1). Synthetic pyrethroid is one of the latest chemical insecticides in the series of insecticide molecules released in the Indian market in 1982. Appearance of pyrethroid resistance in *H. armigera* has been reported in Andhra Pradesh<sup>1-3</sup>, Tamil Nadu<sup>2-4</sup>, Delhi, Punjab and Haryana<sup>3,5</sup>. Three types of pyrethroid resistance mechanisms were identified in *H. armigera*<sup>6,7</sup>, i.e. reduced cuticular penetration, decreased nerve sensitivity and enhanced metabolic detoxification. Among these, esterases-mediated metabolic detoxification plays an important role in the detoxification of pyrethroid insecticides in *H. armigera*<sup>8</sup> and other insect pests<sup>9-12</sup>. However, the relation-

ship between pyrethroid resistance and carboxylesterase in the Tamil Nadu populations was not known and this prompted the present investigation. This information could be relevant to management of *H. armigera* control programmes and might help in the development of strategies to overcome resistance to pyrethroids.

Four different populations of *H. armigera*, namely pyrethroid susceptible, fenvalerate-resistant, cypermethrin-resistant and deltamethrin-resistant were reared on semisynthetic diets<sup>13</sup>. The resistant individuals were collected from insecticidal selection pressure areas of Tamil Nadu under cotton cultivation. Pyrethroid-susceptible larvae were obtained from Tamil Nadu Agricultural University, Coimbatore. The F<sub>1</sub> generation larvae were used for insecticide bioassay and carboxylesterase enzyme studies.

Bioassays were conducted on third instar larvae (30–40 mg weight/larva) using a procedure based on the standard *Heliothis* susceptibility test recommended by the Entomological Society of America<sup>14</sup>. Serial dilutions of technical grade pyrethroid insecticides (fenvalerate, cypermethrin and deltamethrin from Spic Science Foundation, Chennai) in analytical grade acetone were prepared and a 1.0 µl drop dispensed on to the thoracic dorsal of individual larvae using a Hamilton repeating dispenser fitted with a 50 µl syringe. Control larvae were treated with acetone alone. In each bioassay, 36 larvae were treated with different concentrations of insecticides. In assays including the synergist DEF was applied as 1.0 µl drops to the mesothorax 15–20 min prior to the insecticide treatment at a rate of 50 µg/larva. After treatment, each test larva was held individually at 25 ± 2°C with adequate