

**Results:** The preliminary results showed that the patients with acute cholera had significantly higher faecal concentrations of TBARS than had the healthy volunteers ( $9.56 \pm 4.41 \mu\text{mol/l}$  for cholera patients vs.  $4.03 \pm 1.86 \mu\text{mol/l}$  for the controls). This observation indicates that patients with active cholera may be associated with varying degrees of oxidative stress probably due to toxin-induced alteration of mucosal metabolism involving xanthine production leading to loss of fluid and electrolytes in the diarrhoeal stool.

**Conclusions:** Further studies will be required to characterize the metabolic abnormalities in cholera patients which may have important therapeutic implications.



## Survival Potential of Non-Culturable *Vibrio Cholerae* O1 by Laboratory Microcosms Using Polymerase Chain Reaction and Fluorescent Antibody Methods

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**Objective:** Assess the survival potential of *V. cholerae* O1 using conventional cultural, fluorescent antibody and recently developed polymerase chain reaction techniques. In Bangladesh, cholera is endemic in certain areas and flares into seasonal epidemics. *V. cholerae* O1 is one of the causative agents of cholera. The survival potential of *V. cholerae* O1 was carried out in various environmental samples by using conventional techniques.

**Methods:** The strain *V. cholerae* O1 biotype El Tor serotype Ogawa was used in this study carried out at the ICDDR,B's laboratories in Dhaka, Bangladesh. A measured inoculum of about  $10^5$  *V. cholerae* O1 per ml was added to 100 ml autoclaved pond water in a 500-ml conical flask, mixed and stored at room temperature. Culturable cells were counted on gelatin agar (GA) and taurocholate tellurite gelatin agar at various time intervals until the bacteria were no longer culturable.

**Results:** The non-culturable *V. cholerae* O1 was detected by fluorescent antibody and PCR techniques. The culturable *V. cholerae* O1 was isolated up to 44 days from the pond water microcosms. The non-culturable *V. cholerae* O1 was detected up to 7 weeks by FA and PCR techniques after they lost their culturability.

**Conclusions:** The non-culturable stage reported here for *V. cholerae* O1 is significant for understanding the epidemiology of cholera because the non-culturable state of *V. cholerae* O1 may pose health problems. Volunteer studies have shown that non-culturable *V. cholerae* O1 became culturable in volunteers intestine. This study demonstrated the survival of non-culturable *V. cholerae* O1 in surface water which may be important from the view point of public health.



## Cholera Toxin Stimulates Absorption of D-Glucose from the Adult Rabbit Small Intestine in Vivo

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**Objective:** Determine the effects of purified cholera toxin (CT) on intestinal absorption of glucose. Glucose is known to stimulate intestinal sodium absorption which provides the basis for the glucose-containing oral rehydration solution for the treatment of diarrhoea. Although this physiologic mechanism is well-preserved during severe cholera, the effects of purified cholera toxin on intestinal absorption of glucose itself has not been evaluated.

**Methods:** In this study, carried out in the facilities of the ICDDR,B's Dhaka-based Animal Resources Branch, the effects of CT on the absorption of glucose from the small intestine of anaesthetized rabbits were evaluated. In the duodenum, 1-2 microgram of purified CT (Sigma) was added after laparotomy and was incubated for 18 hours. After the incubation period, the segment was flushed with phosphate-buffered solution; 10 ml of the solution containing 0.25 g of D-glucose was injected into the duodenum, and peripheral blood glucose was monitored every 10-15 minutes using specific method for detection of D-glucose.

**Results:** The results indicate that there was a progressive increase in the concentrations of glucose in the CT-treated rabbits over a period of 120 minutes, and the concentrations were significantly higher ( $p < 0.001$ ) than that in the control rabbits treated with glucose-free saline (blood glucose:  $103 \pm 3.5$ ,  $260 \pm 13.9$ ,  $274 \pm 18.0$ ,  $317 \pm 18.8$ ,  $415 \pm 38.6$ ,  $375 \pm 22.5$ ,  $350 \pm 18.4$ ,  $327 \pm 7.1$  mmol/l (mean  $\pm$  SD) for CT-treated rabbits vs.  $109 \pm 5.5$ ,  $156 \pm 4.9$ ,  $187 \pm 4.7$ ,  $224 \pm 4.6$ ,  $260 \pm 6.9$ ,  $257 \pm 5.7$ ,  $243 \pm 4.8$ ,  $228 \pm 5.7$  mmol/l for the control rabbits across all periods). A higher dose of CT (1  $\mu$ g vs. 2  $\mu$ g) significantly produced better stimulation ( $p < 0.01$ ) of intestinal glucose absorption. The CT-specific rises in glucose concentrations ranged between 41.5% and 66.6% during the 120 minutes.

**Conclusions:** These preliminary results indicate that CT induces dose-dependent glucose absorption from the small intestine of rabbits. To explain these effects of CT, it is speculated that this action may be mediated through the stimulation of glucose transport mechanism in apical membrane, involving the carrier proteins. Further studies will be needed to explore these hypotheses.



## Trends in and Determinants of Infant Mortality in Rural Bangladesh

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**Objective:** Study the trends in and determinants of infant mortality in rural Bangladesh.

**Methods:** About 48,000 infants born during 1984-1990 were followed up for one year in the Demographic Surveillance Systems of three ICDDR,B field sites (Matlab, Abhoynagar, and Sirajgonj). Logistic regression was used in modelling mortality risks during infancy, evaluating deaths during the first 28 days of life (neonatal mortality) and during the rest of the first year (post-neonatal mortality).

**Results:** Neonatal and post-neonatal mortality declined in the Matlab treatment area (receiving improved health and family planning services) and the Matlab comparison area. The mortality decline was concentrated between 4 and 14 days for neonates, and 1 and 5 months for post-neonates, which were the ages when the treatment area had lower mortality than the comparison area. There was no significant decline in neonatal mortality, and a modest reduction in post-neonatal mortality was observed at Sirajgonj, a more remote and traditional area. There was significant decline in post-neonatal mortality in Abhoynagar, although neonatal mortality did not decline during this period. In Matlab, risk factors for neonatal and post-neonatal mortality include lack of mothers' formal education and a short preceding birth interval. Additional risk factors include age of mother (young and old), religion (Hindu), sex (male), and birth order (2+) for neonates; and size of dwelling house (small) for post-neonates.

**Conclusions:** Reductions in neonatal mortality in three field sites have been modest except in the Matlab treatment area, where maternal tetanus immunization has improved. Reductions in post-neonatal mortality have, however, been more substantial and widespread.

