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Transforming Growth Factor- β 1 Enhances Expression of Brain-Derived Neurotrophic Factor and Its Receptor, TrkB, in Neurons Cultured From Rat Cerebral Cortex.

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TGF- β 1 on expression of BDNF and its high-affinity receptor, TrkB, in neurons cultured from the cerebral cortex of 18-day-old embryonic rats were examined. BDNF mRNA was significantly increased from 24-48 hr after the TGF- β 1 treatment over 20 ng/ml. Accumulation of BDNF protein in the culture medium was also potentiated by TGF- β 1, although the intracellular content of BDNF was nearly unchanged. The enhancement of BDNF mRNA expression was suppressed by the co-presence of decorin, a small TGF-β-binding proteoglycan that inhibits the biological activities of TGF- β s. mRNA expression of full-length TrkB was also upregulated after treatment with TGF- β 1. These observations suggest that: 1) TGF- β 1 potentiates BDNF/TrkB autocrine or local paracrine system; and 2) the neurotrophic activity of TGF- β 1 is partly responsible for the BDNF induced by TGF- β 1 itself. To test this latter possibility, we examined the neuronal survival activity of TGF- β 1 with or without K252a, a selective inhibitor of Trk family tyrosine kinases. TGF- β 1 significantly enhanced neuronal survival, but the co-presence of K252a completely suppressed the activity. These results seem to be in line with recent findings that some neurotrophic factors including BDNF require TGF- β s as a cofactor to exert their neurotrophic activities.

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Purification and Characterization of Two Lectins from a Toxic Moray, *Gymnothrax javanicus*.

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Two lectins, *Gymnothrax javanicus* lectin-I (GJL-I) and *Gymnothrax javanicus* lectin-II (GJL-II) were isolated from the stomach and intestine, and the liver, respectively, of a toxic moray eel, *Gymnothrax javanicus*. GJL-I is a polymer of two heterogeneous subunits of 67 and 51 kDa. In a hemagglutination inhibition assay, it had sugar-binding specificity toward lactose and lactulose among the mono- or oligo-saccharides and BSM among the glycoproteins tested. The lectin stimulated NGF synthesis by astroglial cells. GJL-II was a polymer of subunit of 41 kDa. This lectin had N-acetyllactosamine binding specificity.

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Nucleotide sequence analysis of Shiga (-like) toxins from an enterohemorrhagic *Escherichia coli* isolated from the Gifu, Japan, outbreak.

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Two Shiga toxin genes (*stx-1* and *stx-2*) were cloned and sequenced from a strain of enterohemorragic *E. coli* O157:H7, a clinical isolate (GPU96MM) during an outbreak in Gifu, Japan, in 1996. The 1,257 bp of *stx-1* and 1,321 bp of *stx-2* genes were analyzed, and each gene contained two open reading frames (ORFs), 948 or 960 bp of A and 270 bp of B subunits, respectively. Although the nucleotide sequences of the *stx-1* ORFs had several nucleotide alterations, the deduced amino acid sequence was identical to that of *stx-1* from *E.coli* O157:H7 isolated in Canada, and *stx* of *Shigella dysenteriae* type 1. The nucleotide sequences of the *stx*-2 ORFs were identical to those of *stx-2* in two *E.coli* (O157:H7) strains, KNIH317 isolated from Korea and 933.

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Construction of deletion mutants of Shiga (-like) toxin genes (*stx-1* and/or *stx-2*) on enterohemorrhagic *Escherichia coli* (O157:H7).

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We constructed isogenic *stx-1* and *stx-2* gene deletion mutants of enterohemorrhagic *E. coli* (EHEC) GPU96MM (O157:H7). A vector with temperature-sensitive replication origin was used for the construction. The parts of *stx-1* and *stx-2* on the GPU96MM genome were replaced with kanamycin and chloramphenicol resistance genes, respectively. The mutants deficient in *stx-1*, *stx-2* and both of them were named GPU993, GPU994 and GPU995, respectively. Each mutation was confirmed by the polymerase chain reaction, enzyme-linked immunosorbent assay, and the cytotoxic activity of the bacterial culture supernatants toward HeLa cells. These results indicate that GPU993 and GPU994 lack productivity for the respective toxins and GPU995, for both of them.