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Liposome-Mediated Delivery of Antibodies into Animal Cells with the Aid of Sendai Virus (HVJ)

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The efficient introduction of antibodies from the lipid vesicles into the animal cell was studied. This approach is to develop an efficient delivery system of macromolecules into the intact animal cells using fusion-competent liposomes. Liposome was prepared by the reverse-phase evaporation using phosphatidyl choline, phosphatic acid, and cholesterol (PC:PA:CHOL=7:2:1, in molar ratio). Cell fusion-inducing HVJ (Hemagglutinating Virus of Japan) was used as a fusogen of the liposome for interacting with the plasma membrane. Encapsulation of antibodies into liposomes was confirmed by the western blot analysis and immunofluorescence microscopy. After liposomes was incubated with HVJ at 37°C, the fusogenic HVJ-Liposomes was confirmed by the sucrose gradient centrifugation. Obtained fusion-competent HVJ-Liposome was incubated with the animal cells and the efficient introduction into the animal cells was confirmed by the immunofluorescence microscopy. Our results suggest that the envelope glycoprotein F of HVJ on the Liposome-HVJ are able to induce the efficient fusion of Liposome-HVJ with the animal cells. We conclude that HVJ-Liposome can be a very useful system for the functional delivery of various macromolecules including antibodies.

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Central Pathway of Motor Nuclei Innervating the Tongue in Rat Brain

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Bartha strain of pseudorabies virus (PRV-Ba) was utilized as a tracer to identify the neural axis of rat tongue muscles; intrinsic and extrinsic muscles. After injection of 10 μl PRV-Ba into tongue muscles, rats were perfused with 4% paraformaldehyde lysine periodate at 48–96 hr survival times and brains were removed. PRV-Ba were localized in neural circuits by in situ hybridization method and immunohistochemistry. PRV-Ba immunoreactive cells appeared early in hypoglossal nucleus and motor trigeminal nucleus. Later, raphe nucleus, prepositus hypoglossal nucleus, spinal trigeminal nucleus, A1, A5, and facial nucleus of rhombencephalon showed immunoreactivity. There were positive neurons in parabrachial nucleus, locus ceruleus and mesencephalic trigeminal nucleus of mesencephalon, and paraventricular nucleus, suprachiasmatic nucleus, organum vasculosum of lamina terminalis of diencephalon. Subsequently the viral antigens were found in forebrain cell groups, bed nucleus of stria terminalis, lateral hypothalamic area, and primary motor cortex in frontal lobe bilaterally at 80–90 hrs postinjection. These results demonstrate that PRV-Ba were detected in the circuit specific connection of several cell groups along the time sequence, and localized in the neural axis of the tongue muscles. These data imply that PRV-Ba were proved as an excellent neurotracer in the tract-tracing researches.