The function and survival of the brain depends on the regulation of nervous tissue volume. Brain volume can change when the size of one of the three brain compartments – intracellular, extracellular or, intravascular – changes. These changes can result from a whole series of pathological events. For example, enlargement of the intracellular and extracellular space owing to the accumulation of water leads to brain oedema (Pappius 1974). If water accumulates intracellularly, it causes cytotoxic oedema (Klatzo 1967). Impairment of the blood-brain barrier and the accumulation of extracellular fluid rich in proteins leads to vasogenic oedema. Dynamic, reversible changes in the size of glial cells (and probably neurones and nerve fibres as well) also occur during neuronal activity, even under physiological conditions. Enlargement of cells is compensated by a decrease in the size of the ECS, without any increase in the total amount of water in the nervous tissue. These processes alter the size of the brain compartments simply as a result of a water shift from one compartment to another, so that brain oedema does not develop.

Using the optic method, Lipton (1973) observed reversible enlargement of the cells in cerebral cortical slices 1 s after applying a train of excitatory impulses. In the frog brain, Van Harreveld and Trubatch (1974) and Trubatch et al. (1977) used measurements made in the electron microscope to show that, after depolarization with KCl solution, water shifted from the extracellular to the intracellular space; the size of postsynaptic structures increased from 4.8% to 14.2% of tissue volume. Using a highly sensitive piezoelectric sensor, Iwasa et al. (1980) and Tasaki and Iwasa (1982) recorded an increase in the size of crab nerve axones and squid giant axones during action potentials, while Tasaki and Burne (1983) found swelling of the cells in the dorsal ganglia and enlargement of neuronal elements in the spinal cord of the frog during sciatic nerve stimulation. These studies show that volume changes occur in the elements and ECS of the nervous system during excitation, but they do not provide any information on the size or dynamics of these changes.

The method of determining the volume of the ECS using K⁺-ISMs allowed dynamic measurement of changes during electrical and adequate stimulation.

### 7.1 Measurement of Changes in Size of the ECS by Means of K⁺-ISMs

ISMs allow the monitoring of ion concentrations in real time and at a specific tissue site, and thereby act as indicators of ion diffusion in tissue. In the CNS, diffusion in the ECS is influenced by its size (α) and by its tortuosity (λ) – factors described in Sect. 3.3.3. Using ISMs, Phillips and Nicholson (1979)
Dynamic changes in the ECS demonstrate changes in the ECS during SD. During neuronal activity after electrical stimulation, ECS changes were found in the cat sensorimotor cortex (Dietzel et al. 1980, 1982), the rat optic nerve (Ransom et al. 1985a,b), the isolated frog spinal cord (Syková 1987; Syková et al. 1987a), and the rat spinal cord in vivo (Svoboda et al. 1989, 1990; Svoboda and Syková 1989, 1991). A decrease in the size of the ECS was also observed in the rat spinal cord during chronic nociceptive stimulation (see Sect. 7.3).

Dynamic changes in the size of the ECS are studied by means of iontophoretic administration of ions which do not cross the cell membranes and therefore remain in the ECS. Their concentration in the ECS is in inverse proportion to the size of the space (Nicholson et al. 1979). The ion exchanger for K$^+$ is highly sensitive to tetraalkylammonium ions and choline, which, in small concentrations, are not toxic and do not cross cell membranes until after at least 60 s contact (Phillips and Nicholson 1978; Kříž and Syková 1981). Choline and tetraethylammonium (TEA$^+$) or tetramethylammonium (TMA$^+$) ions are therefore used for testing changes in the size of the ECS. Figure 81 represents the principle employed in the measurement of relative dynamic changes in the size of the ECS in the CNS. Using a current of 10–100 nA, the ions are administered for 5–60 s into the ECS with a single or multichannel iontophoretic pipette. The tip of the K$^+$-ISM which records ion concentration changes during administration is 40–200 $\mu$m distant from the tip of the iontophoretic pipette. If this distance needs to be known exactly – for instance, to determine changes in $\lambda$ or $\alpha$ of ECS – the two electrodes must be glued together with dental