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SHORT COMMUNICATIONS

PIEZOELECTRIC DRIVE FOR STEP-BY-STEP MICROELECTRODE ADVANCEMENT

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It is a common experience in working with glass-microelectrodes that penetration of a cell is performed with greatest ease if a single thrust of high velocity is applied to either the electrode or the preparation. In most cases this is accomplished by tapping on the base of the experimental set-up, or on the micromanipulator carrying the electrode. However, it is often difficult to impale small cells with this method. Various attempts have been made to overcome this difficulty by the use of electromechanical drives of high speed and spatial resolution. These include electromagnetic (Fish *et al.* 1971; Tomita, 1969), stepping motor (Brown & Flaming, 1977) and piezoelectric devices. Potentially, piezoelectric transducers are most suitable for these applications because of their high speed. Single elements, however, are hampered by the fact that only small excursions can be obtained at high voltages. To obtain a larger working range it is necessary to use either devices composed of many elements stacked in series or piezoelectric bender elements. There are several reports on microelectrode drives incorporating bender-elements (Lassen & Sten-Knudsen, 1968; Prazma, 1978; Tupper & Rikmenspoel, 1969) but the mechanical response to an applied electrical signal has not been documented.

We initially tested a piezoelectric drive of the stacked elements type (Physik Instrumente, D-7517 Waldbronn, W. Germany; type P 172, Excursion 20 μm/1000 V). But although it was very much faster than one employing bender elements, it was inferior to the latter with respect to ease of penetration of a cell in our preparations. Thus, above a certain level, velocity of advance does not seem to be of critical importance.

In the following we describe a simple and robust piezoelectric microelectrode drive together with a control circuit. Only 40 V are required for full excursion, which amounts to ca 140 μm.

(1) *Piezoelectric drive*

Circular piezoelectric bender elements were used (Motorola, 32 mm diameter, 150 nF capacity) which have recently been developed for loudspeaker applications (Bost, 1978; Schafft, 1976). Briefly, one element functions as follows: It consists of two discs of piezoelectric ceramic coupled mechanically to each other by a common centre electrode. Upon application of a suitable voltage one disc tries to expand while the other contracts. In consequence the whole element dishes out and the centre of the element performs a rectilinear motion which can be transferred to the electrode

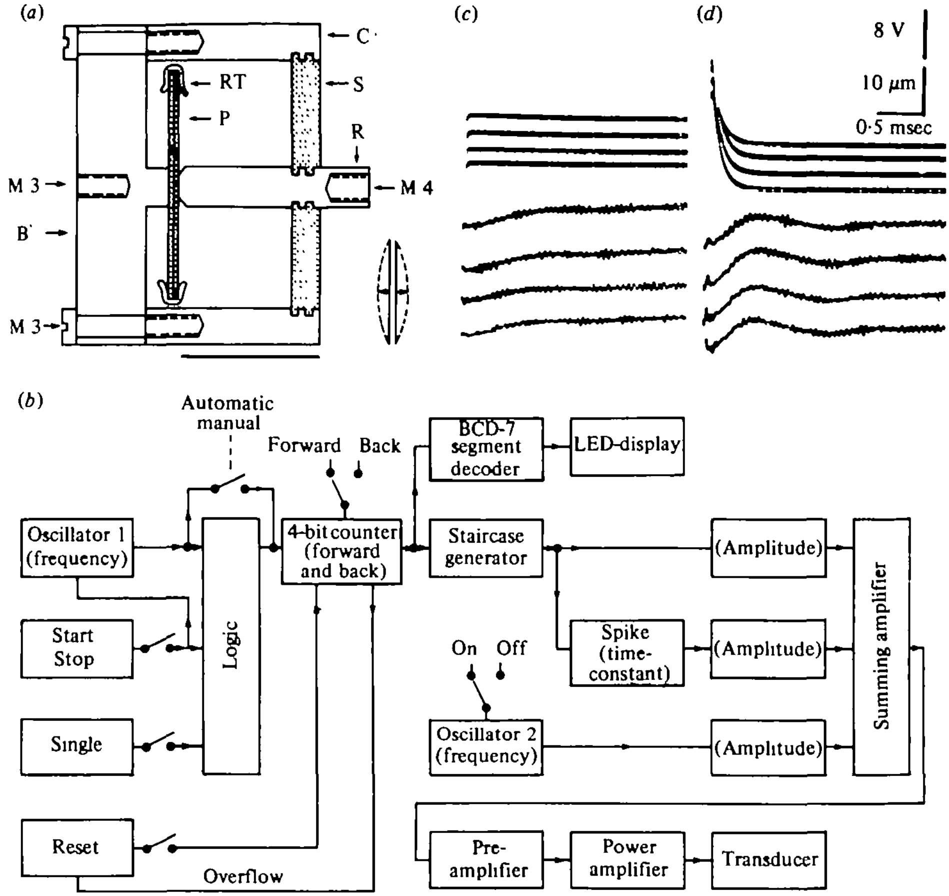

Fig. 1 (*a*). Longitudinal section through piezoelectric drive. Parts machined from plexiglass unless otherwise stated. Two circular piezoelectric bender elements (P) are clamped together with a tightly fitting piece of rubber tube (RT). One element is glued to the base plate (B) which may be fastened to a micromanipulator. The other one is glued to a rod (R) which may take up a suitable electrode holder on its free end. A silicon-gum spring (S) carried by a cylinder (C) prevents lateral movements of the rod. M3, 4: metric screw and thread respectively. Bar: 2 cm. The inset schematically shows the bowling motion of both elements (stippled) upon application of a suitable voltage. (*b*) Block diagram of electronic control (adjustable parameters in brackets). Three types of signals can be applied to the microelectrode drive: (1) 15 rectangular voltage steps (step number may be increased by simply extending the counter), (2) voltage steps with a superimposed needle-shaped impulse (spike), (3) recurrent square wave pulses (oscillator 2). (*c*) Mechanical response of the drive to voltage steps: upper trace, control voltage; lower trace, excursion of the drive. The oscilloscope was triggered with the control-signal. (*d*) Same as in (*c*) but with needle-shaped impulse superimposed upon each voltage step to increase speed of advancement.

relatively easily. A longitudinal section through the drive is shown on Fig. 1 (*a*). To extend available excursion, two elements working in opposite directions are used, which are held together by a tightly fitting piece of rubber tube. Bender configurations with and without an applied voltage are indicated on the inset of Fig. 1 (*a*). The centre of one element is glued to a socket on the baseplate. The centre of the other one is glued to a rod which can take up a suitable electrode holder on its free end. To mini-

Fig. 2. Penetration of muscle cells in the pharynx-levator muscle of the snail, *Helix pomatia* by step-like advance of the microelectrode. Stable impalements indicated by closed arrows. Zero level, open arrow; upper trace, control voltage; lower trace, membrane potential.

mize lateral movements of the rod it is supported by a disc-shaped spring cast from silicon gum (Sylgard, Dow Corning).

Mechanical behaviour of the advancer was measured with a lightbarrier arrangement like that described by Tomita (1969). The sensitivity of the device was found to be *ca.* 3·5 μm/V.

(2) *Electronic control circuit* (Fig. 1 *b*)

This essentially consists of a staircase generator and a suitable amplifier (25 Ω, 50 V output). The generator delivers steps of adjustable amplitude. The response of the drive to voltage steps is shown in Fig. 1 (*c*). As can be seen, the velocity is about 1 cm sec^{-1} with steps of 10 μm. To improve speed even further a needle-shaped impulse (spike) can be superimposed upon the rising phase of each step. This results in an excursion which is nearly twice as fast (Fig. 1 *d*) and an increased yield of successful penetrations.

For some applications (e.g. impalement of nerve cells covered with connective tissue) vibration of the electrode turned out to be more advantageous than advance step by step. A separate squarewave generator was added for this purpose.

The device has been used in investigations on snail muscle and nerve cells. An example of impalements of muscle cells (diameters ranging between 5 and 10 μm) is shown in Fig. 2. Several stable penetrations were obtained in the course of microelectrode advancement. Electrodes in this case were drawn from filament capillaries on a D. Kopf vertical puller modified for air-cooling of the electrode during the second, strong pull. They were filled with 5-M-K-acetate. Tip-diameters were about 0·1 μm as judged from electron microscopic observation.

It has to be mentioned finally that, although the microelectrode drive described above greatly facilitated penetration of cells in our preparations, microelectrode dimensions remained to be the limiting factor for the successful impalements.

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REFERENCES

BOST, J. R. (1978). A new piezoelectric driver enhances horn performance. Audio Engineering Society, preprint no. 1374 (D-6).

BROWN, K. T. & FLAMING, D. G. (1977). New microelectrode techniques for intracellular work in small cells. *Neuroscience* **2**, 813–827.

FISH, R. M., BRYAN, J. S., MCREYNOLDS, J. S. & RIES, J. J. (1971). A mechanical microelectrode pulsing device to facilitate the penetration of small cells. *IEEE Transactions on Biomedical Engineering* **18**, 240–241.

LASSEN, U. V. & STEN-KNUDSEN, O. (1968). Direct measurements of membrane potential and membrane resistance of human red cells. *J. Physiol, Lond.* **195**, 681–696.

PRAZMA, J. (1978). Penetration of cells membrane by the piezoelectric driver. *Experientia* **34**, 1387–1388.

SCHAFFT, H. (1976). Wide range audio transducer using piezoelectric ceramic. *Ferroelectrics* **10**, 121–124.

TOMITA, T. (1969). Single and coaxial microelectrodes in the study of the retina. In Glass Microelectrodes (ed. M. Lavallée, O. F. Schanne and N. C. Hébert), pp. 124–153. New York: J. Wiley.

TUPPER, J. T. & RIKMENSPOEL, R. (1969). Piezoelectric driving device for glass capillary microelectrodes. *Rev. Scientific Instruments* **40**, 851–852.