Die Anordnung des Dehnungsmessstreifens ist aus der Figur 4 ersichtlich. Der Sender wird auf den Bauch des Säuglings gelegt, leicht angedrückt und das Haften der Klebestreifen überprüft. Durch leichtes Dehnen der Haut mit dem Finger kann man die Funktion kontrollieren. Die Signale werden mit einem handelsüblichen Nachrichtenempfänger empfangen, der über das entsprechende Frequenzband verfügt. Die Aufzeichnung kann mit jedem beliebigen Tonbandgerät, die Auswertung entweder durch Registrieren des Pegels mittels Effektivwertgleichrichters und Streifenschreibers oder im einfachsten Fall durch Abzählen der Impulse pro Zeiteinheit[3,8] geschehen. Die Figur 5 zeigt einen Ausschnitt aus einer Registrierung.

Abschliessend ist zu bemerken, dass das Verfahren nicht nur an Säuglingen, sondern auch an erwachsenen Personen anwendbar ist. Hier ist die Befestigung auf der Brust empfehlenswert. Ebenso können anstelle des Dehnungsmessstreifens Temperatur oder Druckfühler mit den entsprechenden elektrischen Werten eingesetzt werden. Systembedingte Schwierigkeiten ergeben sich allerdings bei Messungen, in denen sich die Parameter sehr langsam verändern. In diesen Fällen ist ein anderes Auswertverfahren anzuwenden[3].

[3] B. Wolf, Diplomarbeit, Universität Freiburg i.Brg. Biolog. Instiut (1973), p. 1.
[4] Siemens AG, Datenbuch Tu 10-Tu 110.
[5] W. C. Lin and W. H. Ko, Med. biol. Engng. *6*, 309 (1968).
[6] G. Kessel, A. Ottmann and H. N. Toussaint, Nachrtech. Z. *13*, 114 (1960).
[7] An Stelle des Dehnungsmessstreifens können an diesem Punkt der Schaltung auch Fühler für andere physikalische Grössen (Druck, Temperatur, Strömung) angeschlossen werden, sofern sie auf ohmscher Basis arbeiten und den entsprechenden Innenwiderstand besitzen. Eine eingehende Betrachtung dieser Anwendungsmöglichkeiten, deren Applikation selbstverständlich nicht nur auf Messungen an Säuglingen beschränkt wäre, erfolgt an anderer Stelle.
[8] R. E. Smith, IEE Transact. Biomed. Engng. *17*, 151 (1970).

A Method for Determining Cross-Measurements of Motor Neurons

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Summary. This new triangulation technique allows the investigator to make cross-measurements of individual cells quickly and easily with a high degree of accuracy and reliability.

Morphological investigations often involve cross-area or volume measurements of cellular constituents and tissue organ components. Several techniques are available for determining cross-area measurements which employ an ocular micrometer[1] or polar-planimeter[2,3]. These techniques tend to be tedious, time-consuming and irritating to the eyes. With this in mind, a simple, accurate method was devised to determine the cross-measurement (diameter) of most cellular constituents or tissue component. As a model, cross-measurements were made on the soma of motor neurons in the albino rat. The motor neurons were stained with Luxol Fast Blue and counterstained with Cresyl-echt Violet.

A two-dimensional structure (Figure 1) was permanently constructed on white paper and attached to a flat vertical surface. Using a Prado microprojector, the motor neurons were projected (×1000, magnification was calibrated by a stage micrometer) onto the white paper. The intersection of the 4-equal-angle lines was placed on the nucleolus (Figure 2). Measurements of the distance across the soma were made along each of the 4 lines (Figure 2) using a millimeter rule (1 mm = 1 μm). The mean diameter of the soma was calculated from the four cross-measurements.

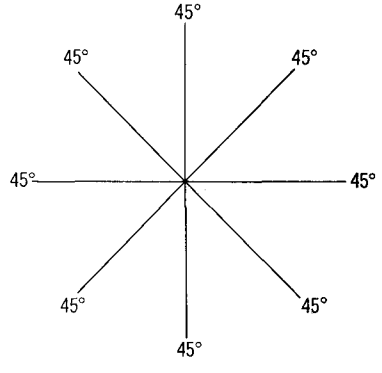

Fig. 1. Four intersecting lines at equal angles constitute the triangulation unit.

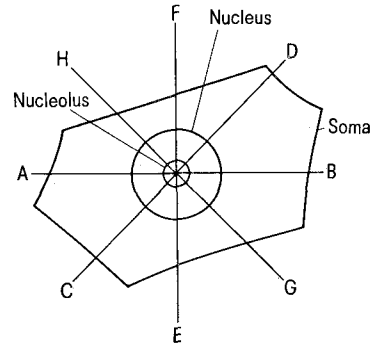

Fig. 2. Triangulation showing 4 measurements of the soma (AB, CD, EF, GH).

[1] A. Aherne, J. Neurol. Sci. *7*, 519 (1968).
[2] E. J. Dornfield, D. W. Slater and H. Schegge, Anat. Rec. *82*, 255 (1942).
[3] B. W. Königsmark, U. P. Kalayanaran, P. Corey and E. A. Murphy, Johns Hopkins med. J. *125*, 146 (1969).

This technique was employed to determine cross-measurements of motor neurons in lumbar spinal cord segments of the albino rat[4]. Upon completion of the study, measurements were repeated on 10% of the cells (r = 0.92). Using an ocular micrometer, another 10% of the cells were remeasured (r = 0.88). Thus, the technique is both reliable (r = 0.92) and valid (r = 0.88)[5–7].

Several advantages exist with this technique: 1. Any quantity of measurements can be made in less time than using an ocular micrometer or polar-planimeter for the same measurements; 2. tracing a projected image is not necessary; and 3. the mean cross-measurement can be used to determine the cross-sectional area of the cell. Disadvantages are minimal in that: 1. a mean diameter is calculated from 4-cross-measurements; and 2. two people are sometimes required depending upon the magnification of the projected image.

Thus, this technique allows the investigator to make crossmeasurements of individual cells quickly and easily with a high degree of accuracy and reliability.

[4] T. GILLIAM, unpublished Ph. D. thesis (1973).
[5] H. ELIAS, A. HENNIG and D. SCHWARTZ, Physiol. Rev. *51*, 158 (1971).
[6] E. R. WEIBEL and ELIAS, *Quantitative Methods in Morphology* (Springer Verlag, Berlin 1965).
[7] E. R. WEIBEL, G. S. KISTLER and W. F. SCHERLE, J. Cell Biol. *30*, 23 (1966).

A Simple Staining Technique to Demonstrate Chromosomal DNA Replication

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Summary. With the BrdU technique here described, Giemsa stained metaphases with either early or late replicational patterns can be obtained within 1 day after cell harvesting, showing a better resolution than ^{3}H-thymidine autoradiography.

The detection of chromosomal DNA synthesis by ^{3}H-thymidine labelling can be replaced by the use of its non-radioactive analogue 5-bromodeoxyuridine (BrdU) which leads to different coiling[1,2] and fluorescence-staining in unifilarly and bifilarly substituted DNA[3–5]. Recently a modification of the latter technique producing permanent preparations was developed[6–10].

Here we describe a simple and less time-consuming technique for the detection of early and late replicational patterns of mammalian chromosomes in Giemsa-stained preparations by combining the BrdU technique of LATT[3] with the staining procedure of KORENBERG and FREEDLENDER[7].

Material and methods. Various normal and abnormal human cell lines and a permanent line of *Microtus agrestis* were grown in Eagle's MEM supplemented with 20% and 10% fetal calf serum, respectively. Blood cultures were set up according to the method of ARAKAKI and SPARKES[11].

In one experimental series the cells were cultivated for most of their S-phase in the presence of 100 μM BrdU (Serva) and 0.4 μM FdU (to block endogenous thymidine synthesis). Similar results were obtained if 200 μM BrdU without FdU was used. Thus, the agent(s) were added 17 h before harvesting; 12 h later the medium was removed, the cultures were washed twice with a balanced salt solution and then reincubated in normal medium containing 0.2 mM TdR.

In another series 100 μM BrdU and 0.4 μM FdU were administered only for the last 5 h. In both protocols, Colcemid (Ciba) was present during the last 2 h. The cells were harvested as usual, treated with 0.075 M KCl for 12 min and fixed in methanol/acetic acid (3:1). The fixed cells were dropped on cold, wet slides and air-dried.

After storage for at least 1 day at room temperature, the slides were incubated at 88 °C for about 20 min in 1 M NaH_2PO_4 which was adjusted with solid NaOH to pH 8. Longer storage of slides usually requires longer incubation times. In case of poor chromosome morphology, older slides should be used. Then the slides were briefly rinsed in distilled water and stained with 2% Giemsa (Merck) diluted in phosphate buffer/aqua dest (1:9) for 7 min.

Results and discussion. With the simple procedure here described, those chromosomal regions which have incorporated BrdU during S-phase become only faintly

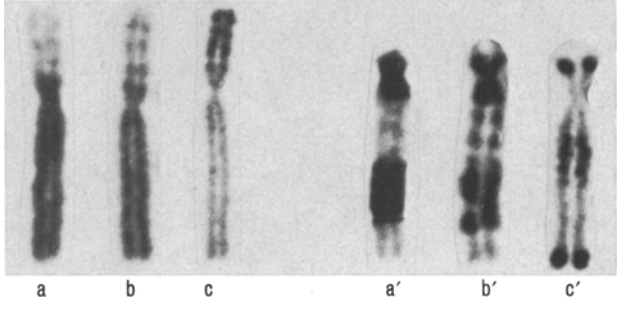

Fig. 1. X-chromosomes (left) and derived Y-chromosomes of an established cell line of *Microtus agrestis* after C-banding (a, a') and after incorporation of BrdU at early (b, b') and late S-phase (c, c').

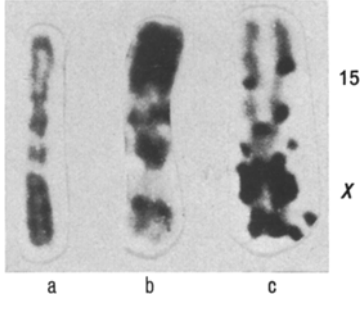

Fig. 2. Human X-autosome translocation: tdic (X; 15) (p21; p12) with regularly late replicating X. After BrdU incorporation at early (a) or late (b) S-phase the dark or the faint bands correspond with the silver grain patterns of the ^{3}H-TdR autoradiography (c).