

ments of both parents may be easily distinguished (Fig. 1a). The assembly of both chromosome complements together at metaphase has been observed frequently in this study and this we claim must be due to fusion. Generally mitosis in heterokaryocytes was observed to occur synchronously, even when the nuclei were not fused. Here, continuous division of these unfused nuclei led to the formation of chimerical colonies. In addition to our analysis of metaphases in the heterokaryocytes,

we also observed heterokaryocytes at anaphase. At this stage we could distinguish adjacent sets of *Petunia* and *Atropa* chromosomes which are presumably a result of the formation of a common or a split spindle (Fig. 1b).

These observations provide evidence in support of further division of the two chromosome complements with a common spindle mechanism which subsequently leads to hybrid callus and ultimately hybrid plants.

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1. Miller, R.A., et al.: Can. J. Genet. Cytol. 13, 347 (1971)
2. Constabel, F., et al.: Can. J. Bot. 53, 2092 (1975)
3. Kao, K.N., et al.: Planta (Berl.) 120, 215 (1974)

Reflexionskontrast, ein neues mikroskopisches Verfahren

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Extrem dünne Objekte, wie insbesondere Zellausläufer und Flüssigkeitsmembranen zwischen Zelle und Kulturkammer, waren mit den bisher bekannten lichtmikroskopischen Verfahren (Phasenkontrast, Interferenz) weder befriedigend auflösbar noch morphologisch interpretierbar. Derartige Objekte wurden daher bislang nur in fixiertem Zustand mit elektronenmikroskopischen Methoden dargestellt.

Reflexionskontrast, ein neues mikroskopisches Verfahren (in Kürze von der Firma Ernst Leitz GmbH lieferbar), ermöglicht jetzt die Darstellung dünnster lebender Objektstrukturen. Das Prinzip ist einfach und das gleiche wie bei der Entstehung von Newtonschen Ringen in glasgerahmten Diapositiven: Bei Auflichtbeleuchtung wird das Licht sowohl an der Obergrenze wie an der Untergrenze dieser Schichten reflektiert. Es kommt dabei zur Interferenz, wobei sich je nach Dicke der Schicht Dunkelheit oder Helligkeit ergeben kann. Bei variabler Dicke zeigt das Objekt dann dunkle und helle Streifen (Fig. 1), die somit Objektstellen gleicher Dicke verbinden.

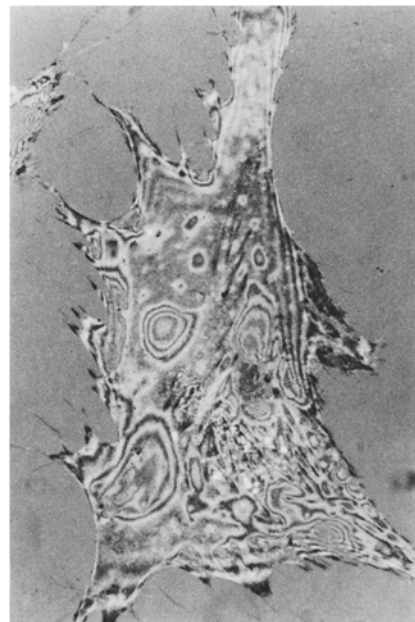


Fig. 1. Menschlicher Hautfibroblast, aufgenommen mit dem Reflexionskontrast-Verfahren. Die Interferenzstreifen verbinden Strukturen gleicher Dicke

Mit konventioneller Auflichtbeleuchtung sind diese Interferenzerscheinungen an feinsten Zellstrukturen nicht beobachtbar. Das ist einerseits auf das außergewöhnlich geringe Reflexionsvermögen dieser Grenzschichten, andererseits auf Störreflexionen der Linsenoberflächen zurückzuführen. Durch polarisationsoptische Hilfsmittel (Immersionkontrast-Verfahren) und eine Zentralblende im Beleuchtungsstrahlengang werden bei dem neuen Verfahren alle kontrastmindernden Reflexionen der Glasoberflächen ausgeschaltet, so daß schließlich die extrem schwachen Interferenzphänomene der Objekte sichtbar werden [1–3].

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1. Abercrombie, M., Dunn, G.A.: Exp. Cell Res. 92, 57 (1975)
2. Curtis, A.S.G.: J. Cell Biol. 20, 199 (1964)
3. Ploem, J.S., in: Mononuclear Phagocytes in Immunity, Infection and Pathology (ed. R. v. Furth). Oxford-London-Edinburgh-Melbourne: Blackwell 1975

Nonspecific Binding of Irrelevant Antibodies to the Complexes of Proteinic Antigen and its Antibody

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We reported the phenomenon that undesired antibodies present in the reaction mixture are non-specifically adsorbed to specifically antibody-coated red cells [1]. Much attention has been paid to the phenomenon especially in the field of blood group serology. We have given many examples of the phenomenon using syphilitic reagin, cold agglutinin, anti-egg white antiserum and so

on [2–5]. Allen et al. [6] also showed clearly that irrelevant antibody in an antiserum can bind non-specifically to the red cells coated with a specific antibody and they named the phenomenon The Matuhasi-Ogata phenomenon. Recently we could demonstrate the non-specific binding of irrelevant antibodies to the complexes of protein and its antibody by solubilizing

the complexes in the antigen excess zone. The procedures and results will be briefly reported here.

The optimum antigen-antibody ratio to form precipitate between human serum albumin (HSA, Hyland Lab. Co.) and horse antiserum to human whole serum (anti-HS, Behringwerke, Co.) was previously determined by the precipitin test. One ml of

the anti-HS was mixed with 0.1 ml of 1% HSA solution and the mixture was kept at 37 °C for 1 h followed by the incubation at 4 °C for 2 d. The resulting immune precipitate was washed according to Kabat and Mayer [7]. A small amount of 25% HSA solution was added stepwise to the precipitate, until the precipitate disappeared. The total amount of the HSA added was 30 mg. The solubilized immune complexes were further incubated at 37 °C for 2 h. In order to detect the presence of non-specific antibody in the solubilized complexes, the immunoelectrophoretic analysis was carried out according to the traditional method in our laboratory. After human serum and the HSA solution were electrophoretically developed on agar gel, the anti-HS or the solubilized immune complexes were placed in a trough parallel to the migration path. The agar plate was incubated in a moisture chamber overnight at room temperature. After the incubation, a precipitin line of β -lipoprotein and of α_2 -macroglobulin appeared against the solubilized immune complexes. In the reac-

tion between the developed HSA and the anti-HS, no other precipitin line was observed in addition to the HSA line, indicating no contamination of β -lipoprotein and α_2 -macroglobulin in the HSA solution. These findings suggest that the antibodies to β -lipoprotein and α_2 -macroglobulin in the solubilized complexes were not derived from the precipitates simultaneously formed between contaminated proteins and their antibodies, but from the complexes of HSA and anti-HSA to which the antibodies had adhered non-specifically. Three lots of rabbit antiserum to human whole serum were also subjected to the similar experiment, but only one of the resulting HSA-anti-HSA complexes was solubilized in the antigen excess zone. The immunoelectrophoretic analysis of the solubilized immune complexes revealed the presence of non-specifically involved antibody to transferrin in the solubilized immune complexes.

The above observations indicate that undesired antibodies present in an antiserum can be non-specifically attached to the

immune complexes of a proteinic antigen and its specific antibody. The mechanism of the Matuhasi-Ogata phenomenon has not been well clarified, however, it could be speculated that 1) a factor like anti-antibody might be present in the antiserum, especially in a hyperimmune serum, and the factor might fix non-specific antibodies to the specific antibodies conjugating with antigens; 2) since the electrostatic charge of immunoglobulin is weakly negative, the immune complexes may adsorb the immunoglobulin non-specifically while the complexes are reducing in their charge to form precipitates.

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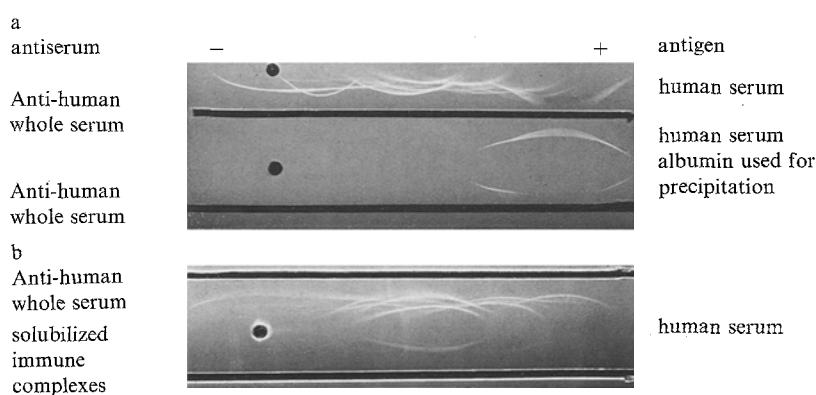


Fig. 1. Immunoelectrophoretic analysis of human serum albumin (HSA) used for forming immune complexes and of the immune complexes solubilized in antigen excess (see text). (a) No contamination of β -lipoprotein and α_2 -macroglobulin is evident in the HSA solution. (b) The presence of unexpected antibodies to β -lipoprotein and α_2 -macroglobulin in the solubilized complexes is clearly demonstrated

1. Matuhasi, T.: Proc. 15th Gen. Assembly Japan. med. Congr., 1959. Tokyo. 4, 80 (1959) (in Japanese)
2. Matuhasi, T., Kumazawa, H., Usui, M.: J. Jap. Soc. Blood Transf. 6, 295 (1960) (in Japanese). Matuhasi, T.: Bibl. haemat. 29, 378 (1968). Matuhasi, T., Usui, M., Nariuchi, H.: Protides of the biological fluids. Proc. 18th Colloquium, Bruges 1970, 341 (1971)
3. Ogata, T., Matuhasi, T.: Proc. 8th Congr. int. Soc. Blood Transf. 208 (1962). Proc. 9th Congr. int. Soc. Blood Transf. 528 (1964)
4. Usui, M., Matuhasi, T.: Proc. 1st Asian Congr. Blood Transf. 288 (1963)
5. Nakano, K., Oikawa, R., Matuhasi, T.: Proc. 19th Congr. Jap. Soc. Clin. Path. Sapporo, 1969, 469 (1969) (in Japanese). Oikawa, R., Nakano, K., Matuhasi, T.: Proc. 19th Congr. Jap. Soc. Clin. Path., Sapporo, 1969, 470 (1969) (in Japanese)
6. Allen, F.H., Jr., et al.: Vox Sang. 16, 47 (1969)
7. Kabat, E.A., Mayer, M.M.: Experimental Immunochimistry. Charles C. Thomas 1964

Buchbesprechungen

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Vorliegende Biographie ist ein begrüßens- und lesenswerter Beitrag zu der Reihe „Große Naturforscher“. Sie stellt die gekürzte und popularisierte Ausgabe einer wissenschaftshistorischen Dissertation dar, die

der Autor 1973 eingereicht hat und die dadurch ermöglicht wurde, daß Dr. Ernst Sommerfeld dem Stuttgarter „Lehrstuhl für Geschichte der Naturwissenschaften und Technik“ den gesamten wissenschaftlichen Nachlaß seines Vaters für mehrere Jahre zur Auswertung überließ. Die Wirksamkeit Sommerfelds als Lehrer und Forscher — er war ja Begründer und bis zum politischen

Umsturz 1933 Leiter einer der bedeutendsten Pflanzstätten der theoretischen Physik — wird lebendig geschildert. Auch gelingt es dem Autor, etwas von der wissenschaftlichen und menschlichen Atmosphäre jener nun schon so weit zurückliegenden Zeit fühlbar zu machen.

H. Mayer-Kaupp (Hinterzarten)