

From the Pathological Institute of the University of Bonn/Germany

Remnants of Paraffin ("Paraffin-Inclusions") in Histological Sections*

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In two recent papers^{1, 2} EDNA H. TOMPKINS described lymphocytes with "atypical nuclei". These nuclei were reported as unusually refractile, and were said to contain "chromatin staining bodies" and "birefringent deposits." The author did not provide an explanation of the "deposits", which, for reasons to be explained in the following paragraphs, are to be regarded as "paraffin-inclusions" („Paraffineinschlüsse“).

This peculiar phenomenon was first described in all its details³ in the salivary glands. If deparaffinization of any histologic section is controlled microscopically under polarized light one may see the birefringent deposits of paraffin rapidly disappear due to the dissolving action of xylol. At some sites, however, the paraffin is less readily removed, as for example, where mucus is present in mucus secreting glands. In these places the paraffin deposits dissolve in xylol only after an hour-long treatment at 58° C. If, after a first brief attempt to remove the paraffin, the section is then brought into water or aqueous solutions of acids, alkali, salts or dye-stuffs, and treated subsequently again with xylol, or even better with carbolxylol, the remnants of paraffin promptly disappear. This behavior explains why "paraffin inclusions" are best seen in unstained sections which have been mounted in Canada balsam immediately after xylol deparaffinization. Further treatment of the sections with xylol after staining them with aqueous solutions removes most of the "paraffin inclusions". Usually, staining procedures are carried out with the paraffin inclusions still present. Differences

* Die vorliegende Mitteilung wurde Anfang November 1960 dem Herausgeber des American Journal of Pathology mit der Bitte um Veröffentlichung zugeleitet in der Absicht, den Lesern der in dieser Zeitschrift eben erschienenen Mitteilung von E. H. TOMPKINS eine Erklärung der dort beschriebenen eigentümlichen Einschlüsse in Zellkernen zu vermitteln und so unnötigen weiteren Bemühungen um diese Gebilde vorzubeugen. War doch anzunehmen, daß das in unseren wissenschaftlichen Zeitschriften so bewährte Verfahren, zu einer eben erschienenen Arbeit verbessernde, erklärende oder gegensätzliche Stimmen zu Worte kommen zu lassen, ja eventuell dem betroffenen Verfasser noch die Möglichkeit zu einem Schlußwort zu geben, auch in USA geübt würde. Das scheint nun zumindest beim American Journal of Pathology nicht der Fall zu sein, denn der Herausgeber lehnte den Abdruck dieser Mitteilung ab und schlug vor, sie in Form eines Briefes an Dr. TOMPKINS gelangen zu lassen. Damit wäre freilich dem oben erwähnten Zweck, nämlich der Unterrichtung der Leser der Zeitschrift nicht gedient gewesen. Außerdem meinte der Herausgeber, die vorliegende Mitteilung bezöge sich zum größten Teil auf bereits veröffentlichte Tatsachen. Das ist richtig. Wird aber eine einem ganzen Leserkreis — wie die neuesten Veröffentlichungen zeigen — unbekannte Tatsache deswegen weniger wichtig, weil sie vor einigen Jahrzehnten in Europa gefunden wurde? Der Abdruck der vorliegenden Mitteilung in englischer Sprache läßt hoffen, daß damit wenigstens ein Teil des Leserkreises des American Journal of Pathology angesprochen wird.

would be expected, therefore, if the paraffin inclusions were removed before staining by placing the sections first in water and then in xylol. Indeed, such differences become evident.

It is difficult to explain why the paraffin within the mucus should be so resistant to the "primary" action of the deparaffinizing agents, and why it yields to a "secondary" action of these solvents after the pretreatment with aqueous solutions. It seems unlikely that these different treatments alter the chemical structure of the deposits of paraffin. Most probably, the differences in the behavior of the deposits are the result of changes in the physical state of the paraffin deposits and of their milieu. The resistance to the immediate action of the deparaffinizing solvent is probably due to the peculiar physical structure of mucus and of the mucus-paraffin mixture. Pretreatment with aqueous solutions apparently alters this physical structure in such a way as to make the deparaffinizing action possible.

As some experiments have shown, these concepts may be applied not only to mucus but also to other structures having peculiar physical properties. In the case of the "atypical nuclei" of lymphocytes it is evident from the photomicrographs of Dr. TOMPKINS that the "birefringent particles" are morphologically identical with the "paraffin inclusions." In addition, these particles behave similarly to the paraffin deposits described above, especially in regards to their prominence in unstained sections.

The explanation of the birefringent particles as "artefacts" does not, of course, diminish their significance or the value of finding them in the nuclei of some lymphocytes. On the contrary, these particles indicate that the nuclear substance of lymphocytes varies, and in some instances responds to the primary solvent action of xylol in different ways.

Summary

The birefringent deposits in lymphocytes with "atypical nuclei", as described by E. H. TOMPKINS, represent remnants of undissolved paraffin ("paraffin-inclusions").

Zusammenfassung

Die doppeltbrechenden Ablagerungen in „atypischen Zellkernen“ von Lymphocyten, wie sie jüngst E. H. TOMPKINS beschrieben hat, stellen Reste von nicht aus den Schnitten herausgelösten Paraffinteilchen („Paraffineinschlüsse“) dar.

References

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- ³ HAMPERL, H.: Beiträge zur normalen und pathologischen Histologie menschlicher Speicheldrüsen. *Z. mikr. anat. Forsch.* **27**, 2—45 (1931).

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