Fine Structural Demonstration of Acid Phosphatase Activity in Auer Bodies

By JAMES G. WHITE

AUER RODS HAVE BEEN SUBJECTED to intensive study by many investigators since the aberrant cytoplasmic bodies were first recognized in leukemic cells 60 years ago. A basis for this continued interest in the unusual particles stems from the possibility that Auer rods reflect an abnormality of leukocyte physiology related to the genesis or perpetuation of the leukemic state.

Histochemical, and fine structural studies have defined a number of similarities between Auer rods and the primary dense particles of immature granulocytes, the azurophilic granules. The resemblance of the two types of cytoplasmic organelles has prompted the conclusion that Auer bodies are unusual azurophilic particles, or arise by fusion of azure granules.

One facet of information relating Auer rods to azure granules has been the light microscopic demonstration of the lysosomal enzyme, acid phosphatase, in the two particle types, suggesting that both are lysosomes. However, Freeman in a recent discussion of the origin of Auer bodies indicated that: "Since lysosomes are heterogenous, usually electron dense groups of structures with no constant internal structure, it is necessary to utilize histochemical techniques in electron microscopy for their [Auer rods] definitive identification."

In the present study the peripheral leukocytes of three patients with acute myelogenous leukemia were evaluated by combined electron microscopy and ultrastructural cytochemistry. Acid phosphatase activity was specifically localized to both the Auer rods and azurophilic granules, indicating that both cytoplasmic particles are lysosomes. The unusual distribution of enzyme reaction product in the two particle types, and the fusion of azure granules with each other and with Auer rods, provides additional evidence of the intimate relationship between them.

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Supported by grants from the USPHS #AM 02917-06 and #AM 05153, American Cancer Society, Minnesota Division, Cardiovascular Clinical Research Program Project and the Graduate School, University of Minnesota.

First submitted July 25, 1966; accepted for publication Oct. 13, 1966.

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Abbreviations Used in Illustrations

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<td>In. fix.</td>
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<td>Post fixative</td>
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<td>Glt.</td>
<td>Glutaraldehyde</td>
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Blood, Vol. 29, No. 4. Part II (April), 1967
Fig. 1.—Progranulocyte from peripheral blood of a patient with acute myelogenous leukemia. Separated leukocytes were initially fixed in glutaraldehyde, incubated in modified Gomori media for acid phosphatase activity, and postfixed in osmic acid. The cell cytoplasm contains several oval or round azurophilic granules which contain specific deposits of lead phosphate, the reaction product of acid phosphatase activity. The degree of enzyme staining is highly variable, and may be related to the maturity of the granules. The two particles indicated by arrows have a similar distribution of enzyme reaction product, and about the same electron density. The particle on the right (†) is an azurophilic granule, and that on the left is an Auer body (††). Although in close proximity, the two particles are not fused. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 29,600.

MATERIALS AND METHODS

Peripheral blood was obtained from two children and one adult with acute myelogenous leukemia for this study. The leukocytes were separated from whole blood and processed for electron microscopy by methods previously recorded. For routine electron microscopic evaluation, leukocyte pellets were fixed in cold (4 C.) 1 per cent Veronal buffered osmic acid, pH 7.3 for 1½ hours, then dehydrated and embedded. Additional samples of leukocytes, fixed initially in 3 per cent glutaraldehyde in 0.05 M cacodylate buffer with 1 per cent sucrose, pH 7.3, were incubated 90 minutes in modified Gomori medium containing β-glycerophosphate and lead nitrate. Control samples of leukemic cells were incubated in a similar medium from which substrate β-glycerophosphate had been deleted. Enzyme-stained samples were refixed in osmic acid, dehydrated and embedded in Vestopalm. The plastic-embedded leukocytes were sectioned on an LKB ultramicrotome and examined in the Phillips 200 electron microscope. Thin sections were poststained with uranyl acetate and lead citrate after enzyme localization had been ascertained on unstained sections.
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Fig. 2.—A progranulocyte from a sample prepared in a similar manner to the cell shown in the previous illustration. In this cell there are two unstained azure granules (†) and two particles stained by enzyme reaction product. The smaller stained particle (‡‡) has the shape of an azure granule, but the lamellar distribution of lead phosphate in its matrix is similar to the arrangement of reaction product between the lattices of the second stained granule (†) which is an Auer body. In. fix.: Ght.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 29,600.

RESULTS

The peripheral blood of the three patients studied contained significant numbers of immature granulocytes. Unusual cytoplasmic particles characteristic of Auer bodies were common in progranulocytes and occurred with decreasing frequency as cell maturity increased.

The appearance of Auer rods and azurophilic granules in progranulocytes incubated for acid phosphatase activity is shown in Figures 1 and 2. The large, almost round granules in the progranulocyte of Figure 1 have the size, shape and distribution of particles which stain an azure color with Romanowsky dyes in the light microscope. Deposits of lead phosphate indicating the presence of acid phosphatase activity obscure the fine internal structure of these particles. The intensity of the staining reaction is highly variable. Two adjacent particles, indicated by arrows, have a similar distribution of enzyme reaction product, and about the same electron density. The particle on the left (‡‡) is an Auer rod, and the organelle to its right (†) is an azurophilic granule.

The progranulocyte in Figure 2 contains two mature azurophilic granules unstained by lead phosphate (†), and two particles with heavy deposits of en-
Fig. 3.—The series of illustrations making up this figure reveal various distribution patterns of acid phosphatase reaction product in Auer rods of cells from all three patients. The arrangement of micrographs suggests stages in transformation from Auer bodies composed of loosely arranged plaques to solid types. However, no evidence of such a progression was elicited in this study.

Fig. 3A.—A large Auer rod composed of loosely grouped blocks of electron dense substance. Enzyme reaction product indicated by deposits of lead phosphate is non-specifically distributed between the solid plaques. Small leukocyte granules in the surrounding cytoplasm did not stain for acid phosphatase, although they may well contain the enzyme. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. x 46,000.
zyme reaction product. The smaller stained particle (↑↑) may be an Auer body or an azure granule, while the larger, diamond-shaped organelle (↓) is an Auer body. Enzyme reaction product is distributed between the lattices which make up this Auer rod.

The substructure of Auer rods and the distribution of enzyme reaction product were extremely variable within leukocytes of the same sample, as well as from sample to sample in the three patients. The micrographs in Figure 3 suggest progressive development of the Auer body from loosely arranged blocks of protein to highly concentrated, electron-dense masses. No evidence for such a progression was elicited in this study, however, and the arrangement was primarily established to show the variation in enzyme staining.

Some Auer bodies (Fig. 3A, B, C, D) appear to be composed of loosely associated plaques which may have separated from one another along lines of cleavage during preparation. The product of acid phosphatase activity is non-specifically distributed in such particles, in between, or at the edge of the protein blocks. The extremely fine lamellar appearance within the plaques is evident at higher magnifications (Fig. 3B [inset], C). The fine linear pattern usually runs in the long axis of the plaque or rod, but may on occasion be perpendicular to it (Fig. 3B [inset], E).

In Auer bodies in which the plaques or blocks are more closely associated (Fig. 3E, F, G) the lead phosphate appears within the substance of the plaques, as well as the cleavage cisternae. More compact Auer bodies undivided by cleavage lines (Fig. 3G, H, I) reveal a splintered or laminar distribution of acid phosphatase reaction product. The distribution of the lead phosphate in such particles suggests the presence of a crystalline substructure even though fine lines are not clearly visible within the mass of these Auer bodies.

The solid type of Auer rods, considered by some workers to be the mature

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Fig. 3B.—An Auer body resembling the particle in Figure 3A, and demonstrating the same enzyme distribution pattern. An adjacent azurophilic granule is also acid phosphatase positive. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. x 54,500.

Fig. 3B (inset).—An area of Figure 3B enlarged to show the fine linear pattern evident in the plaques making up this type of Auer body. The dark lines alternating with clear spaces usually run in the long axis of the particle, but may be perpendicular to it. Arrows (↑) indicate the orientation of fine lines in the periodically banded blocks. The periodic interval of the striations is 60 to 80 Å. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. x 140,000.

Fig. 3C.—An Auer body similar to those shown in Figures 3A and 3B. The fine linear pattern in the slabs making up the particle and the nonspecific distribution of enzyme reaction product are evident. The periodic interval of the fine linear pattern in this example is approximately 70 Å. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. x 140,000.

Fig. 3D.—An Auer rod with fewer separations than in the unusual granules in Figures 3A, B and C, but having a similar pattern of enzyme staining. Lead phosphate does not appear within the blocks making up the mass of the particle. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. x 69,000.
form, appeared to have a nonspecific distribution of lead phosphate within the particle mass (Fig. 3J, K). Only a few areas in these dense particles had a laminar distribution of reaction product (Fig. 3K), suggesting an ordered pattern of the matrix.

The bizarre substructural anatomy and unusual enzyme distribution patterns were not limited to the Auer rods in these cells. Numerous azurophilic granules were noted to have similar abnormalities (Fig. 4). Several azure particles present in the cytoplasm of the cell depicted in Figure 4A revealed the variety of structures that may be encountered. Two of the granules (†) are lightly stained by enzyme reaction product, and have a smooth matrix. The central azure granule has a similar substructure, but no evidence of enzyme activity. The azurophilic granule below it (††) contains lead phosphate, and in addition reveals a multilayered concentric internal structure suggesting a myelin-like configuration. This lamellar arrangement was very common in azure granules of the AML leukocytes (Fig. 4B, C, D, E). The particle in Figure 4B has several laminations near its surface, but the pattern appears to loop more deeply into the matrix. Whorls of membranous configurations occasionally make up the entire body of the azure particle. The distribution of lead phosphate is predominately nonspecific in the heavily stained granules shown in Figures 4C and D, but some reaction product does follow the lamellar pattern.

Occasional azure granules were observed in which a linear pattern occurred, as well as concentric lamellae (Fig. 4E). This appearance suggested the possibility of transitional forms between azure granules and Auer bodies. The particle shown in Figure 4F also demonstrates this possibility. Enzyme reaction product appears just under the continuous surface membrane of the granule. A spindle-shaped piece lying within its surface membrane has a structure markedly different from the mass of the azure particle. The perpendicular striations (†) suggest the more rigid linear pattern apparent in Auer bodies. This unusual granule, however, may have arisen through fusion of two particles, rather than by internal transformation of a single azure granule.

The fusion of azurophilic granules has been considered a possible mechanism of Auer rod formation. In Figure 5 several aspects suggesting granule fusion are depicted. The mass of acid phosphatase positive granular material in Figure 5A retains the appearance of at least two distinct azurophilic granules, and possibly a third (†). Figure 5B reveals an extremely large azurophilic granular mass similar in all respects to surrounding azure granules, except for its size. High magnification study of this particle revealed faint outlines of limiting membranes within its substructure, suggesting that it originated by fusion of several azure granules, rather than from a single granule focus.

The particle in Figure 5C has the size and shape consistent with an Auer body. The concentric lamellar construction at its one end (†), however, strongly suggests the appearance of azure granules as demonstrated in Figure 4. In Figure 5D a similar type of Auer rod is shown. A myelin configuration similar to that of the particle in Figure 5C is apparent in its matrix at one end (†). The unusual organelle stained for acid phosphatase in Figure 5E
Fig. 3E.—The cleavage cisternae of this Auer body do not separate the dense slabs of the particle as widely as in previous examples. Enzyme reaction product appears primarily in the spaces, but some is deposited in the dense plaques. The fine striations in this particle, not apparent at this magnification, run perpendicular to its long axis. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 37,900.

Fig. 3F.—In this Auer rod the enzyme reaction product occurs in fine deposits not only in lines of cleavage, but also between the fine striations. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 67,500.

Fig. 3G.—The enzyme stain in this Auer body appears nonspecific, but in the absence of distinct planes of cleavage the lead phosphate still suggests a linear pattern running in the long axis of the particle. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 85,800.

Fig. 3H.—The cleavage cisternae are not evident in this Auer body, but the discrete deposits of lead phosphate suggest the presence of a fine linear pattern. Deposits of reaction product occur in the clear spaces between the fine dense lines forming the periodic pattern of the Auer rods. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 65,500.
Fig. 3I.—A portion of an Auer body at high magnification revealing linear deposits of lead phosphate, suggesting the presence of a periodic substructure. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 171,000.

Fig. 3J.—A dense Auer rod with primarily nonspecific distribution of enzyme reaction product. The clumps of lead phosphate have a linear pattern at higher magnification, as suggested by the area indicated (†). In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 59,400.

Fig. 3K.—A solid form of Auer body. This particle does not have an apparent substructural pattern, and enzyme reaction product is nonspecifically distributed in its matrix. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 56,000.

may represent two distinct particles, rather than one. On the left side the organelle has the lamellar substructure and shape of an azure granule, while on the right the distinct characteristics of an Auer rod are apparent. Whether the mass represents two separate particles or the fusion of an Auer rod with an azure granule is uncertain, but enzyme reaction product is deposited across their line of junction. The particles shown in Figures 5F and 5G also suggest intimate association of Auer rods with azure granules. The concentric lamellar substructure of an azurophilic granule (†)
Fig. 4.—The azurophilic granules in the immature granulocytes of the three patients were abnormal. The substructural anomalies and unusual distribution of enzyme reaction product suggested possible transformation of some individual azure granules into Auer bodies.

Fig. 4A.—A section of progranulocyte cytoplasm revealing the variations in azure granule structure apparent in single cells. The two granules at the top of the illustration have a homogenous substructure and light, nonspecific deposits of enzyme reaction product. The center unmarked particle is homogenous and unstained. Below it is an azure granule with clumps of lead phosphate, and an unusual substructural appearance (††). The alternating dense lines and clear spaces form a lamellar pattern similar to repeating membranes, or myelin sheaths. This concentric lamellar arrangement was extremely common in leukemic immature granulocytes, and occasional azure granules appeared entirely composed of whorls of myelin-like configurations. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 89,000.

Fig. 4B.—An azure granule stained by enzyme reaction product. The concentric lamellar pattern is evident near the surface of the particle (†). In fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 120,000.

Fig. 4C.—The concentric lamellar pattern of this azure granule loops back deeply into the mass of the particle. Enzyme reaction product is heavily deposited in the unstructured matrix, but small amounts are also evident on the dense lines of the lamellae. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 139,500.

Fig. 4D.—A heavily stained azure granule whose substructure is almost obscured by reaction product. The concentric lamellae are evident within the matrix, and lead phosphate appears distributed on the dense lines. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 128,000.
Fig. 4E.—The periodicity of this azure granule is accentuated by deposits of lead phosphate on the dense lines. Both a concentric lamellar pattern and a more rigid laminar arrangement of dense lines can be recognized in this particle. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 80,000.

Fig. 4F.—An unusual granule with substructural characteristics suggestive of both an azurophilic particle and an Auer rod. Enzyme reaction product is deposited under the surface membrane, which is continuous over the superior aspect of the particle. The spindle-shaped piece of the granule (†) has a laminar pattern suggestive of an Auer rod. The rest of the particle mass, however, is typical of an azurophilic granule. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 100,000.

Fig. 5.—Fusion of azure granules has been considered a possible mechanism of Auer rod formation. The following sequence indicates intimate association of azurophilic particles with each other and Auer rods.

Fig. 5A.—The acid phosphatase positive mass in this progranulocyte reveals profiles of two distinct oval granules. A possible third particle in close apposition is indicated (†). The deposit of enzyme reaction product in and around the associated particles suggests that they may be forming a single fused mass of granular material. In. fix.: Glut.; Post. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 56,100.

Fig. 5B.—A huge particle typical of the inclusion bodies occasionally noted in A.M.L. granulocytes. The electron density of the particle is identical to that of surrounding azure granules. At very high magnification residua of membranes can be seen in the matrix, suggesting that the particle arose by fusion of azure granules. In. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 31,200.
Fig. 5C.—The Auer rod seen in this micrograph has an unusual structural pattern at one end (†). The concentric lamellar arrangement strongly resembles the appearance of azurophilic granules. In. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 84,200.

Fig. 5D.—An Auer body heavily stained by enzyme reaction product has a concentric lamellar pattern in its matrix (†) suggestive of the structure seen in azure particles. In. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 82,700.

Fig. 5E.—An interesting acid phosphatase positive mass with characteristics of both the Auer rod and an azure granule. The portion of the particle on the left is round and has a matrix composed of concentric and linearly arranged lamellae. The right side has the characteristic structure of an Auer rod. A portion of the membrane of the oval particle is evident between the two sides, but enzyme reaction product obscures the junction at its superior aspect. In. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 95,700.

Fig. 5F.—The portion of the Auer rod shown in this micrograph has a protuberance (†). The lamellar substructure of this mass is similar to that of the azurophilic granule though it is incorporated in the body of the Auer rod (††). In. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 114,000.
Fig. 5G.—An area of progranulocyte cytoplasm revealing several azure granules stained heavily by enzyme reaction product. One of the oval particles is in direct continuity with an acid phosphatase positive Auer rod. In. fix.: O.A.; Post Stain: U.A. and L.C.; Mag. × 67,600.

is apparent in Figure 5F, but it is incorporated into the structure of an Auer rod, only a portion of which is visible in this illustration (††). An azure granule heavily stained by enzyme reaction product is in direct continuity with the Auer rod shown in Figure 5G.

DISCUSSION

The Auer bodies in leukemic granulocytes have posed a particular challenge to investigators of leukemic states. Morphologic, biochemical, and histochemical investigations have attempted to define the nature of this unusual cytoplasmic particle in order to ascertain its role in the leukemic process.1-10 Histochmical technics have been particularly helpful in determining basic chemical components of Auer rods.2,4,9 In recent years electron microscopy has also contributed significantly to knowledge of Auer rods through analysis of the fine structure of the aberrant granular particles.4,6,8,15-20 Questions, however, concerning the origin, nature, and function of Auer bodies in leukemic cells remain incompletely resolved.

A principal objective of this investigation was to define the presence of the lysosomal enzyme, acid phosphatase, in Auer rods by ultrastructural cytochemical methods.14 Leukocytes from the three patients incubated in modified Gomori medium contained highly specific deposits of lead phosphate in membrane bound particles, indicating sites of acid phosphatase activity. Both azurophilic granules and Auer rods in these leukemic cells contained the enzyme reaction product. The presence of acid phosphatase in azure granules supports
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their lysosomal character. The enzyme activity in Auer rods strongly indicates that they are also lysosomes. Control cell samples incubated in identical medium, except for deletion of substrate β-glycerophosphate, yielded no evidence of lead phosphate deposition in the leukocytes.

Definition of Auer bodies as lysosomes by fine structural cytochemistry agrees with the interpretation of Goldberg who, on the basis of the light microscopic demonstration of acid phosphatase in Auer rods, first suggested that the unusual granules might be lysosomes. Establishment of the lysosomal character of the Auer bodies is important because it immediately relates the bizarre particles to the normal granulocyte lysosomes. That is, Auer bodies, if they are lysosomes, must occur as an aberration of azure granule or specific granule production, derive by the fusion of lysosomal granules, or represent a mechanism of lysosome development not evident in normal myeloid elements.

The possibility that Auer bodies arise as a variant of the azurophilic granules has been considered by many workers. Physical and histochemical similarities have suggested a close relationship between the two particle types. In the present study Auer rods were found most often in the cytoplasm of immature myelocytes. Such cells contained variable numbers of azure granules, and little or no evidence of specific particle formation. The frequency of Auer rods in progranulocytes and their infrequency in more mature cells closely paralleled the pattern of azure granule production and disappearance during granulocyte maturation.

Additional evidence supporting a close association between Auer rods and azurophilic particles was suggested by the unusual structure and enzyme staining of azure granules in leukemic granulocytes. The primary cytoplasmic lysosomes in leukemic cells were generally larger than similar particles in normal progranulocytes. Concentric lamellar arrangements were evident within the matrix of many large azurophilic particles, and similar myelin-like configurations were apparent inside membranes surrounding Auer rods. The multilayered linear pattern present in Auer bodies was occasionally noted within the substructure of azure granules. Enzyme reaction product of acid phosphatase activity appeared to be nonspecifically distributed within the matrix of most azure granules, but occasionally assumed a rigid linear pattern closely resembling the distribution of lead phosphate in Auer rods. Although structural and cytochemical similarities are merely suggestive, they do indicate that some Auer bodies may originate by transformation of abnormal azure granules, or represent a product of disordered azure granule development.

The small specific granules which characterize transformation of progranulocytes to neutrophil myelocytes have also been implicated in the origin of Auer bodies. Biochemical and histochemical studies have shown that specific granules are lysosomes and could be related to the Auer rods which contain lysosomal enzymes. A structural similarity of the two particle types is also apparent. Many specific granules are spindle-shaped like Auer rods, and improved methods have permitted demonstration of a fine linear substructure in specific particles of the type often seen in Auer bodies. However,
the appearance of Auer rods in progranulocytes at the peak of azure granule production when specific particles are not evident, and the declining frequency of Auer bodies with increasing cell maturity, mitigate against an association of definitive neutrophil granules and Auer rods. In addition, no evidence of transformation or fusion of specific granules to form Auer rods was evident in this investigation. Acid phosphatase activity present in abundance in azure granules and Auer rods was seldom demonstrated in specific granules by the method used for this study. Previous experience with this technic has confirmed its specificity for immature granules and abnormal lysosomes, and only special treatment appears to labilize specific neutrophil granules sufficiently to permit cytochemical demonstration of acid phosphatase in large numbers.

The fusion of azurophilic granules with one another has been implicated as a possible mechanism of Auer body formation. A close association of azure granules with each other and with Auer rods was commonly observed in the material forming the basis of this study. In some large granular masses outlines of individual azure granules could be identified. Other large particles, resembling inclusion bodies occasionally seen in leukemic granulocytes, were structurally identical to azure granules except for their massive size. Auer rods were frequently noted to form close associations with azurophilic granules. The azure particles were round or slightly oval, and often contained a concentric lamellar substructure or heavy deposits of acid phosphatase reaction product. When similar particles were found incorporated into the structure of Auer bodies, the appearance strongly suggested that the association had arisen as a result of azure granule fusion with the Auer rod. Occasional examples of this intimate relationship were found in which some evidence of a membrane separating the azure granule and Auer body remained. Acid phosphatase reaction product was distributed across the line of junction in these examples, however, indicating that membrane separation was not complete. These observations suggest that azure granules can fuse to form large cytoplasmic masses, or contribute to the size of already formed Auer rods.

The concept that Auer rods arise through a mechanism of lysosome formation completely unassociated with the normal process of azure and specific granule development seems unlikely. Secondary lysosomes, such as phagolysosomes, autophagic vacuoles, and residual bodies may occur in human leukocytes, but bear no resemblance to granular Auer rods. It would appear that Auer bodies must arise by some aberration of primary lysosome production, or by fusion of preformed lysosomal particles.

The combined technics of electron microscopy and ultrastructural cytochemistry have been useful in clarifying aspects of the nature and origin of the Auer rods. The specific localization of acid phosphatase in Auer bodies at the fine structural level indicates that the unusual cytoplasmic organelles are lysosomes. The establishment of the Auer rod as a variety of lysosome immediately suggests its relationship to other lysosomes in the same immature granulocytic cells. The structural similarity of Auer bodies and some azure granules has been confirmed in this study. In addition, the unusual distribution of enzyme reaction product in azure granules and Auer rods has indicated a close rela-
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tionship between them. The possible transformation of individual azure gran-
ules to Auer bodies, and the union of azurophilic granules with Auer rods,
were also noted. The results appear to substantiate the long-held concept
that Auer bodies derive from azure granules. The basic cause for transforma-
tion or fusion of azure granules to form Auer rods, however, remains obscure.

SUMMARY

The peripheral leukocytes of three patients with acute myelogenous leukemia
have been studied by combined electron microscopy and ultrastructural cyto-
chemistry. Auer bodies present in immature granulocytes were found to con-
tain specific evidence of acid phosphatase activity, indicating that Auer rods are
lysosomes. A number of observations were made which support the origin of
Auer rod lysosomes from azurophilic granules which are known to be lyso-
somes. The nature of the stimulus causing transformation or fusion of azure
granules to form Auer bodies, however, remains obscure.

SUMMARIO IN INTERLINGUA

Le leucocytos peripheric de tres patientes con acute leucemia myelogene
esseva studiate per microscopia electronic in combination con cytochimia ul-
trastructural. Esseva trovate que corpores de Auer presente in granulocytos
immatur contineva evidentia specific de activitate de phosphatase acide, in-
dicante que le baculos de Auer es lysosomas. Esseva facite un numero de
observationes in supporto del these que le lysosomas del typo baculo de
Auer ha lor origine in granulos azurophilic que cognoscitemente es lysosomas.
Le natura del stimulo que cause le transformation o le fusin de granulos
azurophilic resultante in le formation del corpores de Auer, remane obscur.

ACKNOWLEDGMENTS

The author wishes to thank Kathleen Hagert, Gerri Lucey, Marcy Krumwiede, and
Norma Wubbena for their essential technical services.

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leucemies et des myelomes au micro-
scope a' contrast de phase et par la
methode de l'ombrage (avec une
etuade particuliere des corps d'Auer
Fine Structural Demonstration of Acid Phosphatase Activity in Auer Bodies

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