Res. Bull. Obihiro Univ., **8** (*1973*): *1～19*.

Fine Structures in the Blood Cells of Japanese Quails

(*Coturnix coturnix japonica*)

II. Basophils

Junzo YAMADA, Tadayuki YAMASHITA and Mikio MISU
(*Department of Veterinary Anatomy, Obihiro Zootechnical University, Obihiro, Hokkaido, Japan*)
Received November 30, 1972

ニホンウズラ (*Coturnix coturnix japonica*) の
血液細胞の微細構造について
II. 好塩基球について

山田純三*・山下忠幸*・三須幹男*

Introduction

In studies of peripheral blood of fowls, the fine structures of blood cells have been reported by various investigators, but these reports were obtained solely from chickens. As far as the authors know, only three reports (ENBERGS & KRIESTEN 1968, DHINGRA, PARRISH & VENZKE 1969, MAXWELL & TREJO 1970) have been published on the fine structures of basophils in chicken peripheral blood. In the authors' previous paper, fine structures of the heterophils and the eosinophils in the peripheral blood of the Japanese quails were reported. In this paper, as a part of the series of electron microscopic studies on peripheral blood cells of Japanese quails, the fine structures of basophils in clinically healthy quails are described.

Materials and Methods

The materials and methods used in this paper were exactly the same as those of the previous paper (YAMADA, YAMASHITA & MISU 1973). The specific granules which were contained in the cytoplasm of basophils, however, presented artificial structures in the samples which were fixed solely with OsO_4. On the other hand, the basophil granules of the samples which were fixed with mixture of glutaraldehyde and OsO_4, or were fixed with glutaraldehyde and followed by OsO_4 were thought to appear in their normal structure. Therefore, the

* 帯広畜産大学家畜解剖学教室

basophils obtained by the former fixation were omitted in the present study.

Results

Basophils were infrequently observed in the peripheral blood on the cut plane of section. Only seventeen basophils which were thought to be cut off in the central portion or the adjacent area of the cells were observed in this study. They were easily differentiated from any other kinds of leukocytes by the presence of many specific granules in the cytoplasm. Basophils appeared as round or oval cells, and their size was 4.4 by 5.8 μ on the average.

Nucleus

A single nucleus was usually eccentric in position and never lobulated. The nucleus had a round or oval shape and the size was 3.0 by 3.8 μ on the average. The nuclei in the majority of basophils contained more euchromatin than heterochromatin. The heterochromatin was usually clustered at the periphery of the nucleus. Flakes of chromatins were often arranged in a pattern suggesting a clockface or a cartwheel. Some nuclei had so small an amount of heterochromatin that they showed a very clear appearance (Fig. 7). Occasionally, one or two, irregularly shaped nucleoli of varying sizes and densities were present in some cells.

Cytoplasm

The cytoplasm was delineated by a thin plasma membrane. The cytoplasm which was on the side opposite the nucleus was very wide, and its nuclear side was narrow. In the wide area of the cytoplasm, there were a number of specific granules, a few mitochondria, a Golgi complex and some other micro-organelles. By contrast, the cytoplasm on the nuclear side had no specific granules, a small number of vesiculated smooth-surfaced endoplasmic reticulum, and a few free ribosomes and polysomes.

Specific granules: The specific granules of basophils were distributed randomly in the cytoplasm except for a part of the Golgi complex and a narrow peri-nuclear area. They were surrounded by a unit membrane. On the cut planes, they varied considerably in size and shape depending upon the direction in which granules were sectioned. These granules were considerably variable in electron density. Almost all of the specific granules had some internal structures. However, some of them were homogeneous without any internal structures. On the basis of electron density, size and internal structure, the granules were divided into eight types as follows.

1) The 1st type (Figs. 1 & 2)

The granules of this type were homogeneously compact, having very high electron density without any internal structures. They were also polymorphous, that is, were horseshoe-like and reniform in the cut plane of cells. The size of these granules was on the average 308 by 412 mμ in diameter. These granules were found in 7 of 17 basophils (41.1%).

2) The 2nd type (Figs. 3, 4 & 9)
In general, the granules of this type were round or oval in shape, and contained a core which was highly or moderately electron dense. The core consisted of homogeneously compact or very fine granules. These granules had a clear space between the limiting membrane and the core. The mean size of these granules was 450 by 593 mμ in diameter, and their cores were 366 by 413 mμ. These granules were found in only 3 of 17 basophils (17.7%).

3) The 3rd type (Figs. 3, 4, 6, 9 & 10)
These granules were usually round or oval in shape, highly or moderately electron dense granules having a mottled appearance. They were composed of coarsely granular subunits and ground substance. Each subunit measured approximately 200–300 A in diameter. A quarter of the granules of this type had a clear space between the limiting membrane and the granular material. The size of this type granule was on the average 514 by 603 mμ in diameter. They were found in 11 of 17 basophils (64.7%).

4) The 4th type (Figs. 3, 4 & 9)
These granules appeared as elaborately reticular structures. The reticular structures showed densely ropy cords, about 200–300 A in diameter. In some granules, these networks were very compact, and in others were moderately or very loose. The size of these granules was 542 by 665 mμ in diameter on the average. They were found in 7 of 17 basophils (41.1%).

5) The 5th type (Figs. 5 & 11)
This type granule was round or oval in shape and showed moderate electron density. These granules were composed of a finely granular subunit and a ground substance. This subunit measured approximately 50–100 A in diameter. The size of this type granule was on the average 489 by 638 mμ in diameter. These granules were found in 9 of 17 basophils (52.9%).

6) The 6th type (Figs. 5, 7 & 13)
These granules were vacuolated granules containing varying amount of granular materials and had a light appearance as shown in Fig. 7. They were round or oval in shape, and their size was on the average 552 by 709 mμ in diameter. This type granule appeared to be unstable with a tendency to disintegrate (Fig. 13). Sometimes they ruptured with internal materials running out into the cytoplasm (Figs. 12 & 13). A myelin-like internal structure was observed in the one of this type granule (Fig. 12). These granules were found most frequently in the granules of all types (in 12 of 17 basophils showing a

percentage of 70.6%).

7) The 7th type (Figs. 8, 12 & 15)

These granules were vacuolated granules containing a small amount of finely fibrilar structure, which was sometimes arranged in a coarse network. The granules of this type appeared to be unstable with a tendency to disintegrate as did the 6th type (Figs. 12 & 14). These granules were round or oval in shape, and their size was on the average 779 by 1,002 mμ in diameter. The granules of this type were the largest of all the specific granules. These granules were found in 6 of 17 basophils (35.3%).

8) The 8th type (Fig. 6)

These were very small granules consisting of a limiting membrane and an inner dense granular material. Some of them had a clear, narrow hollow between the limiting membrane and the inner material. They were round or oval in shape and their size was about 100 mμ in diameter. These granules were found in 9 of 17 basophils (52.9%).

Occasionally, intermediate forms of specific granules between the 3rd type and the 4th type, the 6th type and the 7th type granules (Fig. 7) were observed. These eight types of granules were arranged in order of their sizes from large to small as follow; the 7th type, the 6th type, the 4th type, the 3rd type, the 5th type, the 2nd type, the 1st type and the 8th type. In the appearance rates of the specific granules of each types, the 6th type granules were observed most predominantly and the 3rd type granules were observed next most predominantly. The appearance rates of the 5th type and the 8th type granules were 52.9%. The granules of the other types were below 50% and the 2nd type granules were most infrequent (17.7%). The combination of these type granules were quite variable in the cut plane of sections. All of the basophils had combinations of from 2 to 5 types of granules. Releasing of the specific granules were observed rarely in granules of the 7th type (Fig. 14). These granules were opened to the free surface of the cell and their limiting membranes connected to the plasma membrane. A part of the internal materials of the granules extruded outside of the cytoplasm.

Golgi complex; A poorly developed Golgi complex which consisted of a few lamellar membranes and moderate numbers of vesicles was located opposite the nucleus with a centriole. Occasionally, coated vesicles were seen around the Golgi complex (Fig. 13).

Mitochondria; A small number of mitochondria were distributed ramdomly in the cytoplasm except for the narrow perinuclear area. The majority of mitochondria were round or oval in shape, only one or two of them being slightly elongated. The size of the mitochondria was 442 by 604 mμ in diameter on the average. Usually, mitochondria had a dark matrix with obscure cristae. The mean number of mitochondria which appeared in the cut plane of basophil was three.

Endoplasmic reticulum ; A small number of smooth-surfaced endoplasmic reticulum were seen ramdomly throughout the cytoplasm. The majority of them showed a vesiculated form. They were filled with a clear substance. A few rough-surfaced endoplasmic reticulum were observed in the cytoplasm of only 4 of 17 basophils (Fig. 2). They were short and thin, canalicular in shape.

Others ; A moderate numbers of free ribosomes and polysomes were scattered throughout the cytoplasm. The microtubules were arranged radiately around a centriole (Figs. 2, 4 & 10). The microfilaments also were seen in some of the basophils, although only rarely (Figs. 9 & 15).

Discussion

Scince basophils constitute only 1.3% of the total blood leukocyte count in the Japanese quails (ATWAL, *et al*., 1964), their presence in the ultrathin sections is infrequent. Only seventeen basophils with various specific granules, which were thought to be cut off in the central portion or the adjacent areas of cells, were observed in this study.

The nuclei of the basophils in the quails stain pale blue with Wright's methods and have a delicate chromatin network under the light microscope (ATWAL & MCFARLAND 1966). Under the electron microscope, the nuclei of the majority of cells contained more euchromatin than heterochromatin, and usually, showed a relatively clearer appearance than those of the other granulocytes.

Until recently, attempts to analyze the fine structure of the basophil granules have been thwarted by the inadequacy of fixatives and embedding materials. Most of the early descriptions failed to outline any detail in the granules, and it may be seriously doubted whether they dealt with the normal structure of the circulating basophils.

It has been well known that the internal structure of granules varies considerably according to the difference in animal species. The following granules have been observed ; those filled with uniform particles in humans (ZUCKER-FRANKLIN 1967), those with lamellated, network-like or honeycomb-like structure in guinea pigs (PEASE 1956, WINQUIST 1960, FEDORKO & HIRSH 1965), in sheep (YAMADA & SONODA 1972) and in chickens (MAXWELL & TREJO 1970), those with finely granular structure in dogs (SHIVELY, FERDT & DAVIS 1969), those with more or less fibrillar contents in chickens (DHINGRA, PARRISH &

VENZKE 1967), and homogeneous ones without any internal structure in cats (OSAKO 1959) and in sheep (YAMADA & SONODA 1972).

In the quail basophils observed by the authors, the specific granules were divided into eight types of granules on the basis of size, their electron densities and internal structure as follows. The 1st type were homogeneously compact and very high electron dense granules without any internal structure. The 2nd type were round or oval in shape, and contained a core which consisted of homogeneously compact or very fine granules. The 3rd type were highly or moderately electron dense granules with a mottled appearance. The 4th type were those with an elaborately reticular structure. The 5th type were moderately electron dense granules with finely granular subunits. The 6th type were vacuolated granules containing varying amounts of granular materials. The 7th type were vacuolated granules containing finely fibrilar structures. The 8th type were very small granules consisting of a densely granular material and a limiting membrane. This classification is provisional. At the present time, it is very difficult to classify exactly the specific granules of the quail basophils, because the specific granules will change their forms under certain conditions, for example, maturation and degranulation of granules, and technical procedures used in handring of specimens. As far as the authors know, there is no report on the formation, maturation and degranulation of basophil granules under electron microscopy. In the mast cells, these processes of specific granules have been well described in rats (COMBS 1966, RÖHLICH, ANDERSON & UVNÄS 1971) and in chickens (WIGHT 1970).

The relationship between mast cells and basophils has been a subject of debate since their discovery. Although both cells have metachromatic granules which are thought to contain histamine (GRAHAM *et al.*, 1952, RILEY 1953) and heparin (JORPES 1946, BEHREMS & TAUBERT 1952), the granules of mast cells are not water soluble, whereas basophil granules are water soluble. In the electron microscopic observations, basophil granules have some similarity to the granules of mast cells. At present, it is uncertain whether mast cells and basophils are the same original cells or not. There are insufficient data to comment on this controversy.

We shall discuss the relationships between each of the granules of the eight types in the quail basophils on the basis of our data and the data on mast cells (COMBS 1966, WIGHT 1970, RÖHLICH *et al.*, 1971). Firstly, it should be emphasized that, classifications of basophil granules are very subjective and variable, ie, in the chickens, DHINGRA *et al.* (1969) and MAXWELL & TREJO (1970) found four types of granules, and DHINGRA *et al.* stated that these granules were different stages of maturation. In the mast cells of rats, there is some evidence that the varying internal appearance of granules is associated with stages of maturity (COMBS 1966) and degranulation (RÖHLICH *et al.* 1971). The same analysis of the phenomena may be applied to the basophil granules of quails.

In the quails, the homogeneous and very highly electron dense granules without any internal structure (the 1st type) suggest that they are swelling, or are compacting from mottled granules (the 3rd type). The mottled appearance of the dense granules (the 3rd type) suggest that they are forming, or are formed from reticular granules (the 4th type), while the beading of the cords of reticulum (the 4th type) suggests that they are disintegrating into the fragments which are seen in vacuolated granules (the 6th type). The homogeneous and highly or moderately electron dense granules (the 2nd type) suggest materials (the 5th type), while finely granular materials (the 5th type) suggest that they are disintegrating into a finely fibrilar structure which are seen in vacuolated granules (the 7th type). COMBS (1966) has indicated concerning the rat mast cells that the immature granules with small strands, resembling the 3rd type granules in this study, became dense and homogeneous mature granules, which correspond to the 1st type granules in this study. The 1st and 2nd type granules may be mature granules of quail basophils judged on the basis of their appearances. Occasionally, the 6th and 7th type granules ruptured and their internal materials ran out into the cytoplasm. These phenomena indicate that these granules are in the process of degranulation. Furthermore, this aspect receives support from the following facts; their internal materials were quite variable, and the size of these granules was the largest of all the granules amoung the basophils. But, it is not negligible that these granules are artifactual. If the 6th and 7th type granules are degranulating granules, and the 1st and 2nd type are mature granules, gaps between the mature and the degranulating granules will be too wide. Thus, it seems likely that the 3rd, 4th and 5th type granules are transitional forms between the mature and the degranulating granules.

COMBS (1966) and WIGHT (1970) have shown electron micrographs of rat and chicken mast cells respectively, which show that the small, membrane-bound granules correspond to the immature granules or progranules. In the quail basophils, the small dense granules (the 8th type) were found in the basophils of half of the observed cells. Thus, it is resonable to conclude that the small dense granules are immature granules in the quail basophils.

The 3rd type had a coarsely granular subunit (about 200–300 A in diameter), while the 5th type had a finely granular subunit (about 50–100 A in diameter). Fundamentally, it may be that there are two kinds of specific granules in the basophils of Japanese quails.

As far as the authors know, no report has been published on the fine structure of releasing of the basophil granules. In this study, releasing of the specific granules was rarely observed in the 7th type. The releasing form of the granules applied to the reverse pinocytosis (eccrine).

In human basophils, some workers (ZUCKER-FRANKLIN 1967, WATANABE *et al.* 1967, AKASAKA 1971) observed granules with myelin-like internal structures. In this study, the myelin-like internal structure was only seen in one of the 6th type granules. WINQVIST (1960), FEDORKO & HIRSH (1965) and SUGIMURA (1972) have observed the granules with crystaline internal structures in the basophils, but such granules were not recognized in this study. Some investigators (ITO 1958, WATANABE *et al.* 1967, WETZEL *et al.* 1967) have observed granules with a clear space between unit membrane and granular materials. In this study, these clear spaces were observed in some of th 1st, 3rd, 4th and 5th type granules. All of the 2nd type had this structure.

Lastly, it should be emphasized that the classification of Japanese quail basophil granules is imperfect. It still remains to be established whether the ultrastructures of these specific granules correlates with the chemical constituents of the granules or only reflects their functional state. Moreover, it is possible that some of the ultrastructures of these granules are artifactural features. Further studies on these problems are necessary.

Summary

The fine structures of the basophils of the peripheral blood obtained from nine, clinically healthy and adult male Japanese quails were examined under an electron microscope. The results obtained are summarized as follows:

1) The basophils were uninucleated cells and their size was 4.4 by 5.8 μ on the average.

2) The basophil granules were divided into eight types of granules on the basis of their size, electron density and internal structure.

3) This classification is imperfect and it still remains to be established whether the ultrastructures of these granules correlates with the chemical constituents of the granules or only reflects their functional states.

4) Some granules were released with reverse pinocytosis (eccrine).

5) A small number of mitochondria, a poorly developed Golgi complex, a small number of vesiculated smooth-surfaced endoplasmic reticulum and a few tubular rough-surfaced endoplasmic reticulum were observed in the cytoplasm. Some other micro-organelles were observed also.

References

1) AKASAKA, K. (1971): Ultrastructure of human blood cell. The Cell, **3**: 2-22 (in Japanese).
2) ATWAL, O. S., McFARLAND, L. Z. and WILSON, W. C. (1964): Hematology of Coturnix from birth to maturity. Poultry Sci., **43**: 1392-1401.
3) ATWAL, O. S. and McFARLAND, L. Z. (1966): A morphologic and cytochemical study of erythrocytes and leukocytes of *Coturnix coturnix japonica*. Am J. vet Res., **27**: 1059-1065.
4) BEHRENS, M. and TAUBERT, M. (1952): Der Nachweiss von Heparin in der basophilen Leukozyten. Klin. Wschr., **30**: 76. Cited by ZUCKER-FRANKLIN, D. (1967).
5) COMBS, J. W. (1966): Maturation of rat mast cells. An electron microscope study. J. Cell Biol., **3**: 563-576.
6) DHINGRA, L. D., PARRISH, W. B. and VENZKE, W. G. (1969): Electron microscopy of granular leukocytes of chicken (*Gallus domesticus*). Am. J. vet. Res., **30**: 637-642.
7) ENBERGS, H. E. and KRIESTEN, K. (1968): Die weissen Blutzellen des haushuhns in elektronmikroskopischen Bild. Dtsch. tierärzt. Wschr., **74**: 271-274.
8) FEDORKO, M E. and HIRSCH, J. G. (1965): Crystalloid structure in granules of guinea pig basophils and human mast cells. J. Cell Biol., **26**: 973-976.
9) GRAHAM, G. S., WHEELWRIGHT, F. W., PARISH, H. H., MARKS, A. R. and LOWRY, O. H. (1953): Distribution of histamin among blood elements. Fed. Proc., **11**: 350. Cited by ZUCKER-FRANKLIN, D. (1967).
10) ITO, T. (1958): Electron microscopic observation on myeloid leukemic cells with special reference to the findings resembling to virus infection. Acta haemat. Jap., **21**: 631-642 (in Japanese with English summary)
11) JORPES, J. E., (1946): The site of formation of heparin. In: Heparin in the treatment of thrombosis. New York, Oxford Univ. Press, p. 57. Cited by ZUCKER-FRANKLIN, D. (1967).
12) MAXWELL, M. H. and TREJO, F. (1970): The ultrastructure of white blood cells and thrombocytes of the domestic fowl. Brit. vet. J., **126**: 583-592.
13) OSAKO, R. (1959): An electron microscopic observation on the specific granules of eosinophil leukocytes of vertebrates. Acta haemat. Jap., **22**: 134-150 (in Japanese with English summary).
14) PEASE, D. C. (1956): An electron microscopic study of red bone marrow. Blood, **11**: 501-526.
15) RILEY, J. F. (1953): Histamine in tissue mast cells. Science, **118**: 332. Cited by ZUCKER-FRANKLIN, D. (1967).
16) RöLICH, P., ANDERSON, P. and UVNÄS, B. (1971): Electron microscope observations on compound 48/80-induced degranulation in rat mast cells. Evidence for sequential exocytosis of storage granules. J. Cell Biol., **51**: 465-483.
17) SHIVERY, J. N., FELDT, C. and DAVIS, D. (1969): Fine structure of formed elements in canine blood. Am. J. vet. Res., **30**: 893-905.
18) SUGIMURA, M. (1972): Personal communication.
19) WETZEL, B. K., HORN, R. G. and SPICER, S. S. (1967): Fine structural studies on the development of heterophil, eosinophil, and basophil granulocytes in rabbits. Lab. Invest., **16**: 349-382.
20) WIGHT, P. A. L. (1970): The mast cells of *Gallus domesticus*. I. Distribution and ultrastructure. Acta Ant., **75**: 100-113.
21) WINQVIST, G. (1960): The ultrastructure of the granules of the basophil granulocyte. Z. Zellforsch. mikroskop. Anat., **52**: 475-481.
22) YAMADA, J., YAMASHITA, T. and MISU, M. (1973): Fine structures on the blood cells of the Japanese quails (*Coturnix coturnix japonica*). I. Heterophils and eosinophils. Res. Bull. Obihiro Univ., **7**: 539-560.
23) YAMADA, Y. and SONODA, M. (1972): Basophils of ovine peripheral blood in electron microscopy.

Jap. J. vet. Sci., **34** : 29–32.
24) ZUCKER-FRANKLIN, D. (1967): Electron microscopic study of human basophils. Blood, **29** : 878–890.

摘　　要

ニホンウズラ (*Coturnix coturnix japonica*) の末梢血から得た好塩基球の微細構造を電子顕微鏡的に観察した。細胞のほぼ中心部分が切られていると思われる 17 個の細胞について詳細に検討し，次の所見を得た。

1) 好塩基球は類円形の単核細胞として認められ，その大きさは平均 4.4×5.8 μ で，顆粒球の中で最小であった。

2) 核は偏在性で，他の顆粒球より一般に明調で，その大きさは平均 3.0×3.8 μ であった。

3) 特殊顆粒はその大多数が類円形で，単位膜につつまれていた。これらの顆粒をそれらの大きさ，電子密度および内部構造により下記のような 8 型に分類した。I 型 (平均 308×412 mμ) は多形で，均質，高電子密度の顆粒，II 型 (平均 366×413 mμ) は均質あるいは非常に微細な顆粒状，高あるいは中等度の電子密度で，限界膜と内部物質との間に明かるい間隙を有する顆粒，III 型 (平均 514×603 mμ) は均質でなく，直径約 200～300 A の高電子密度の微細顆粒が集合した顆粒，IV 型 (平均 542×665 mμ) は大きさ約 200～300 A の高電子密度の紐状構造物が網状に配列した顆粒，V 型 (平均 489×638 mμ) は中等度電子密度の直径約 50～100 A の微細顆粒が種々な程度に集合した顆粒，VI 型 (平均 552×709 mμ) は空胞状に拡張し，内部に中等量から極少量まで種々な程度に集合した微細顆粒状物質を有する顆粒，VII 型 (平均 779×1002 mμ) は空胞状で，内部に微細線維状物質を種々の程度に有する顆粒，VIII 型は直径約 100 mμ の非常に小型で，電子密度が高い顆粒であった。

4) これら 8 型の顆粒の出現頻度は III 型，VI 型が高く，II 型が最も低く，他の型は約半数の細胞に認められた。

5) 顆粒崩壊および顆粒放出と思われる像も観察された。

著者らが行なったこの分類はまだ不完全であり，これら 8 型の顆粒の形態的差は含有されている化学物質の差によるものか，機能相によるものか，または固定などによる人工産物であるのか，さらに検討しなければならない。

Explanation of Plates

Plate I

Fig. 1. This basophil has mainly the 1st type granules. These granules show polymorphous form. The roundish nucleus locates eccentrically. On the wide side of the cytoplasm, there are a small number of mitochondria, ill-developed Golgi complex, a centriole and other micro-organelles. Fixed with mixture of glutaraldehyde and OsO_4. × 20,000.

Fig. 2. This micrograph is a higher magnification of Fig. 1. An ill-developed Golgi complex and a centriole are observed. Mitochondria show dark matrix and well-developed cristae. Microtubules are found near the centriole. One of the 1st type granules shows a horseshoe-like form. One small dense granule is observed (short arrow). A rough-surfaced endoplasmic reticulum is found between mitochondria (long arrow). × 37,000.

Plate II

Fig. 3. This basophil has three kinds of granules. A Golgi complex and a centriole are seen. Outline of this cell is relatively irregular. Fixed with mixture of glutaraldehyde and OsO_4. × 15,000.

Fig. 4. The micrograph shows a higher magnification of Fig. 3. Three of the 2nd type granules are seen (II). The 4th type granules is found at the left of figure (IV). Remaining granules are of the 3rd type (III). × 37,000.

Plate III

Fig. 5. This cell contains the 5th type granules (V). Other granules are of the 6th type. Fixed with glutaraldehyde followed by OsO_4. × 25,000.

Fig. 6. In this basophil, the 8th type granules (VIII) as well as the 3rd type granules are observed. Fixed with glutaraldehyde followed by OsO_4. × 22,000.

Plate IV

Fig. 7. This basophil is round and has a very clear nucleus. This cell contains the 6th type granules only. Notice variable amount of the inner materials in these granules. Fixed with mixture of glutaraldehyde and OsO_4. × 20,000.

Fig. 8. This basophil has mainly the 7th type granules. Only one of the 6th type granules has a very small amount of inner granular materials. The 7th type granules have filamentous materials. Fixed with mixture of glutaraldehyde and OsO_4. × 15,000.

Plate V

Fig. 9. In this figure, various type granules are shown. The 2nd (II), 3rd (III) and 4th type (IV) granules are observed. An intermediate form between the 3rd and 4th type granules is found (arrow). The largest granules (?) cannot be classified. Fixed with mixture of glutaraldehyde and OsO_4. × 50,000.

Fig. 10. This figure shows various forms of the 3rd type granules. Fixed with mixture of glutaraldehyde and OsO_4. × 50,000.

Fig. 11. In this micrograph, the granule on the right is of the 5th type. This granule consists of finely granular subunits which measured approximately 50-100 A in diameter. The granule

on the left is a very coarse example of the 3rd type. This granule consists of coarsely granular subunits which measured approximately 200–300 A in diameter. Fixed with mixture of glutaraldehyde and OsO_4. × 50,000.

Plate VI

Fig. 12. This micrograph is a higher magnification of a part of the cytoplasm in another basophil. A myelin-like figure is observed in the 6th type granule (long arrow). Intra-granular materials of some of the granules which ran out into the cytoplasm (short arrows). Fixed with mixture of glutaraldehyde and OsO_4. × 37,000.

Fig. 13. This micrograph shows a higher magnification of Fig. 7. An ill-developed Golgi complex and a centriole are observed. Coated vesicles are observed in the Golgi complex. Intra-granular materials of the 6th type granule ran out into the cytoplasm (arrow). Fixed with mixture of glutaraldehyde and OsO_4. × 37,000.

Plate VII

Fig. 14. In this figure, releasing of the specific granules is observed (arrow). Fixed with mixture of glutaraldehyde and OsO_4. × 50,000.

Fig. 15. Microfilaments are found around the Golgi complex (arrows). Fixed with mixture of glutaraldehyde and OsO_4. × 50,000.

YAMADA, J. *et al.*

Plate I

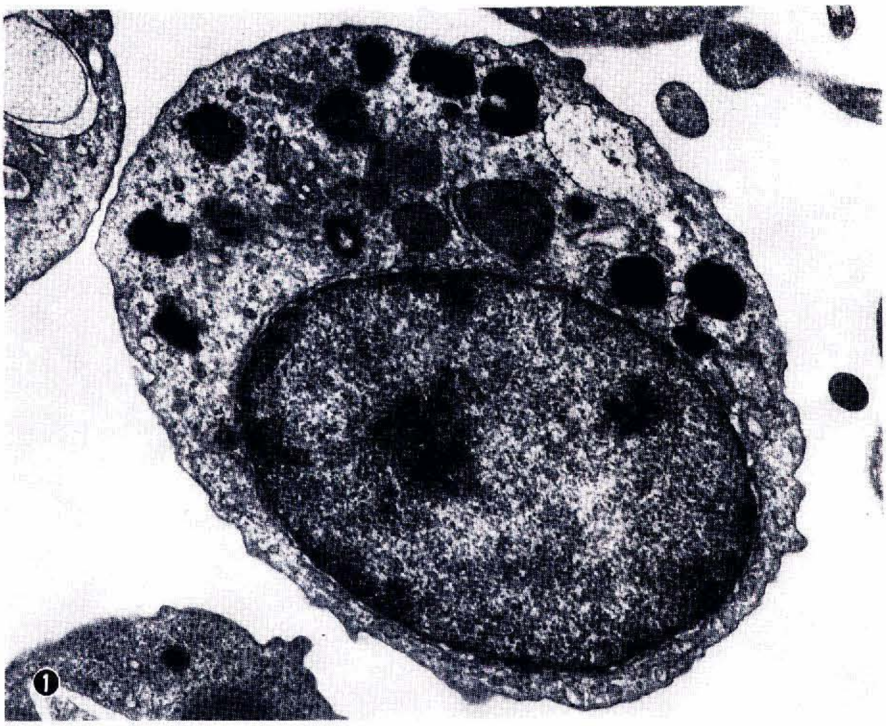
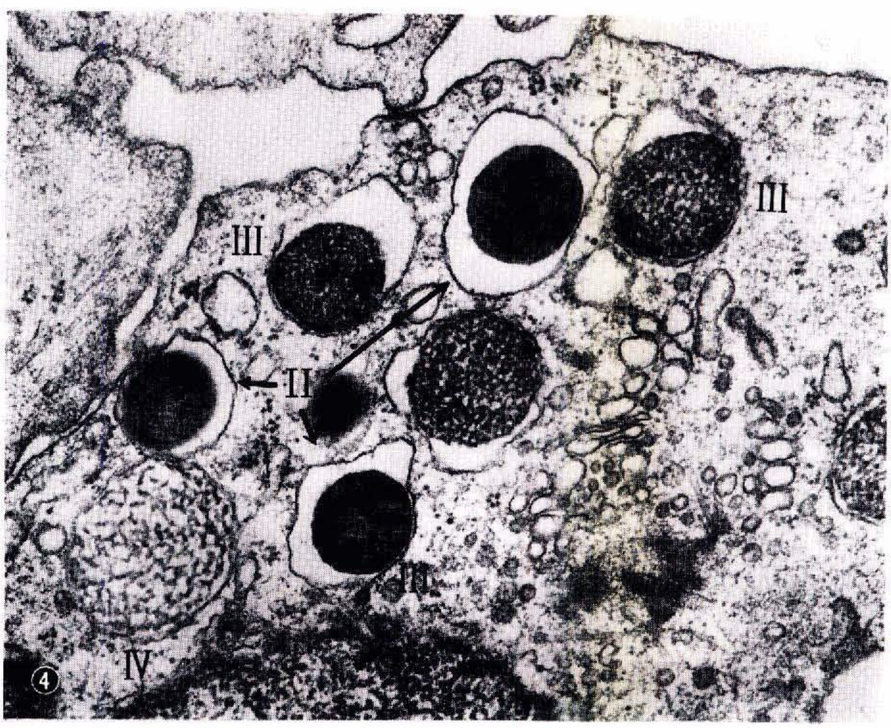
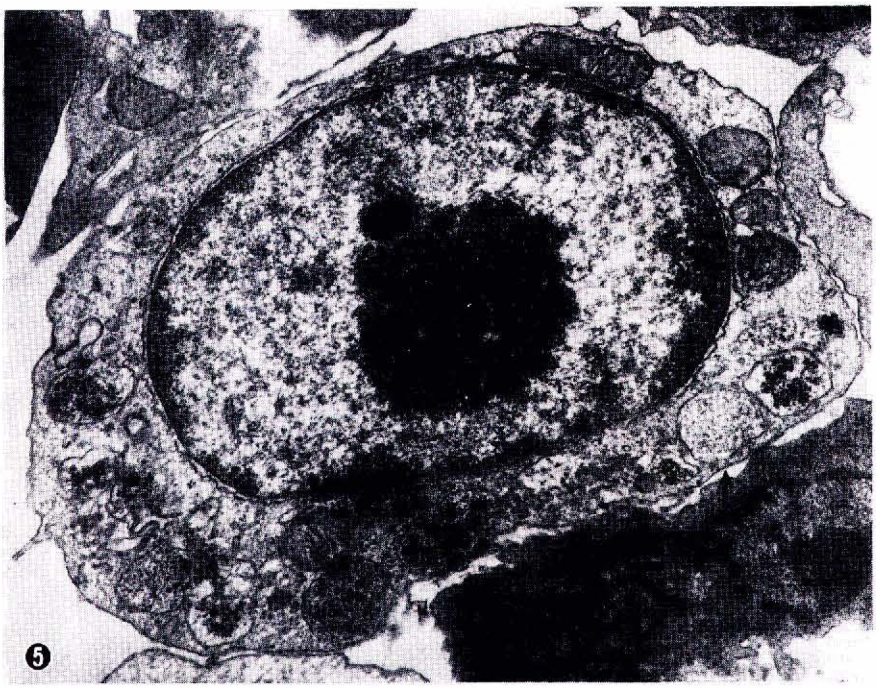


Plate II YAMADA, J. *et al.*



YAMADA, J. *et al.* Plate III



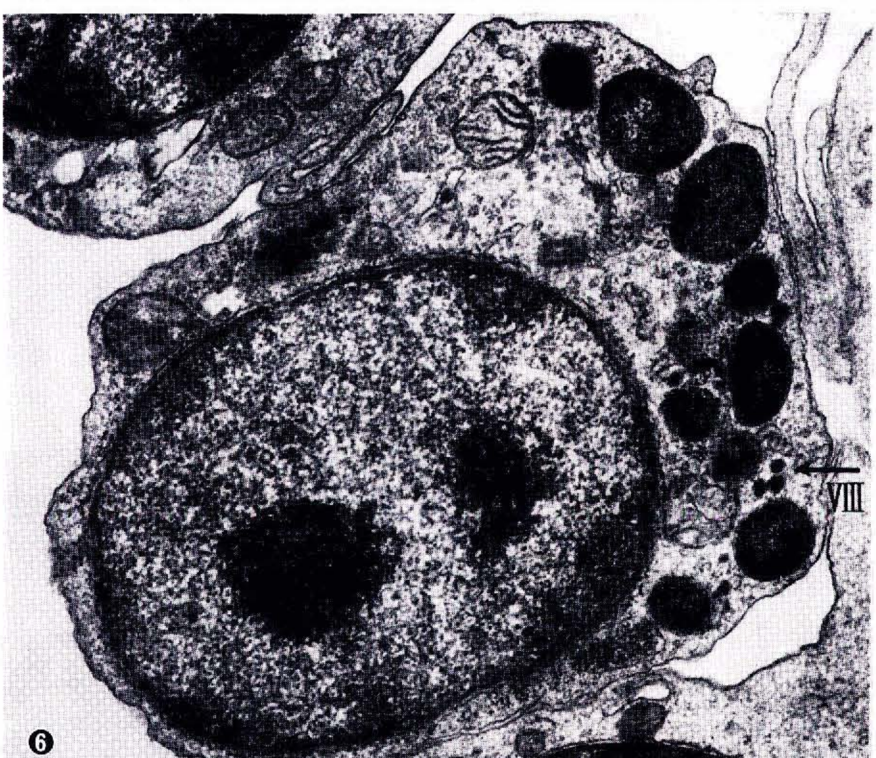
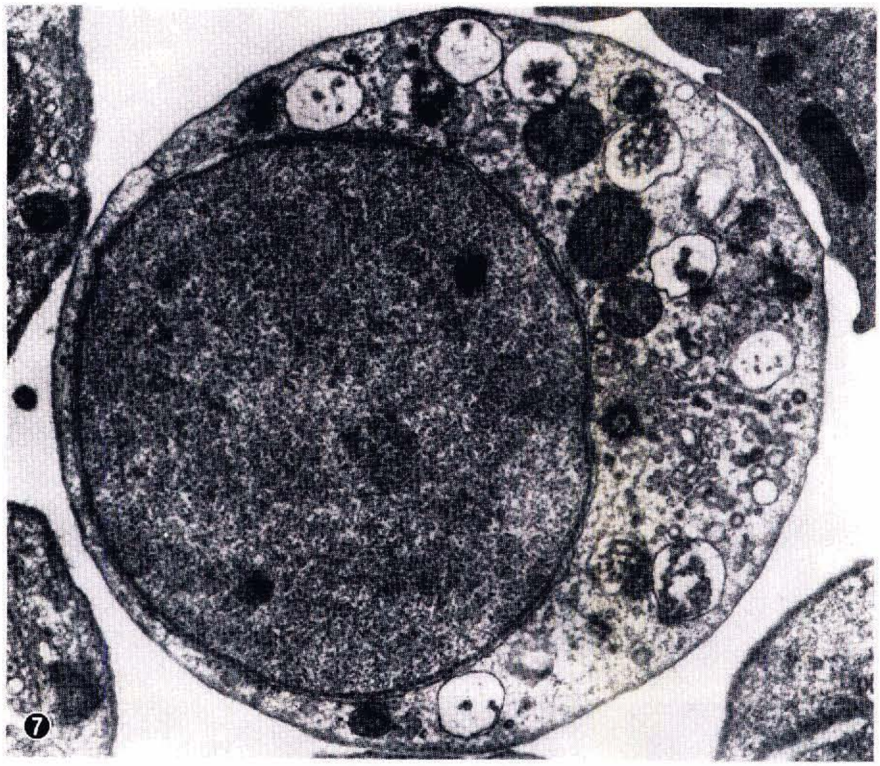
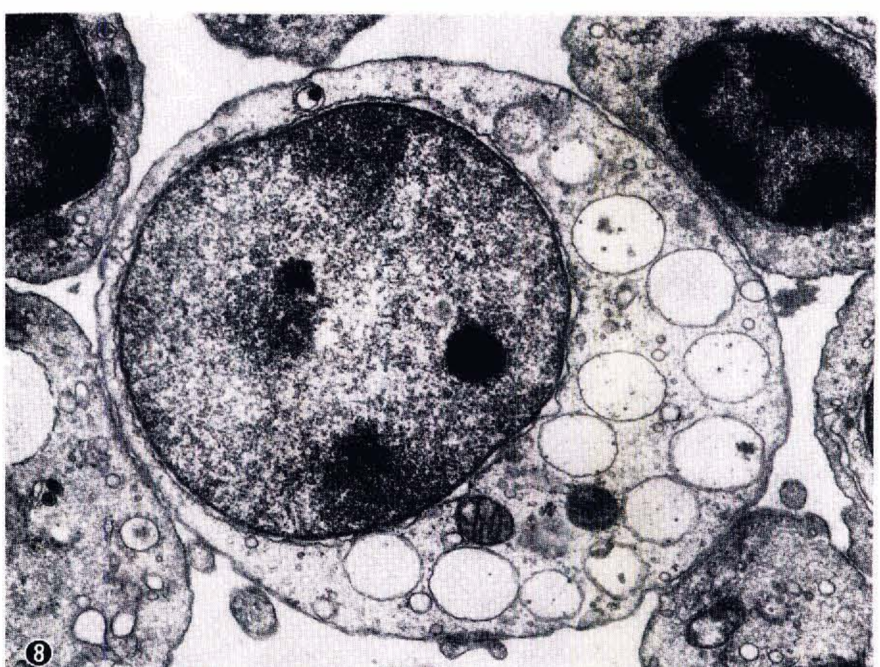
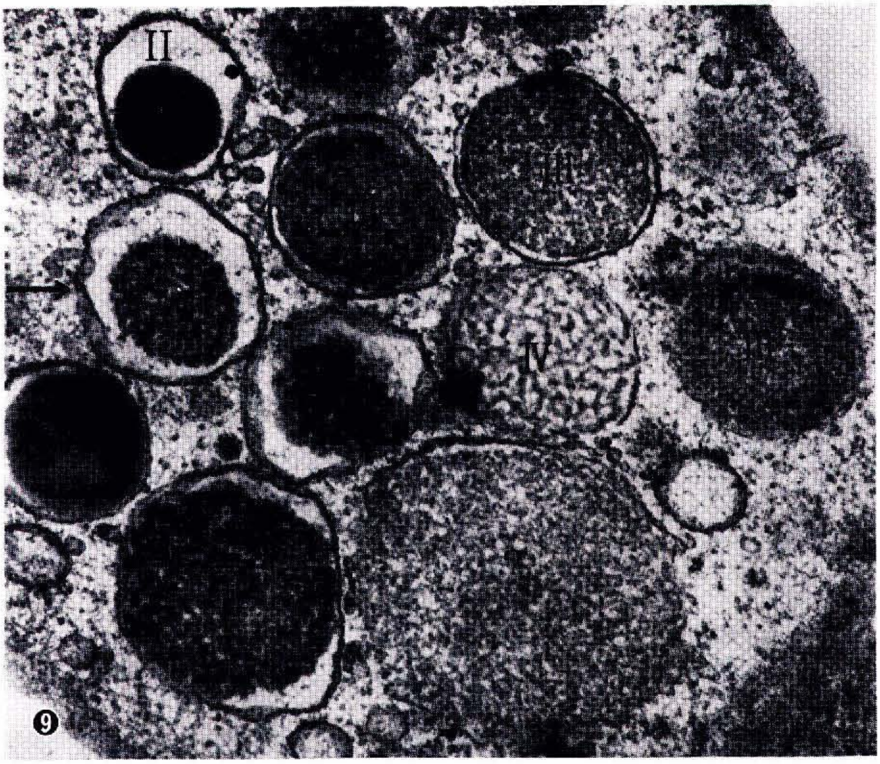


Plate IV

YAMADA, J. *et al.*






II
III
IV
9

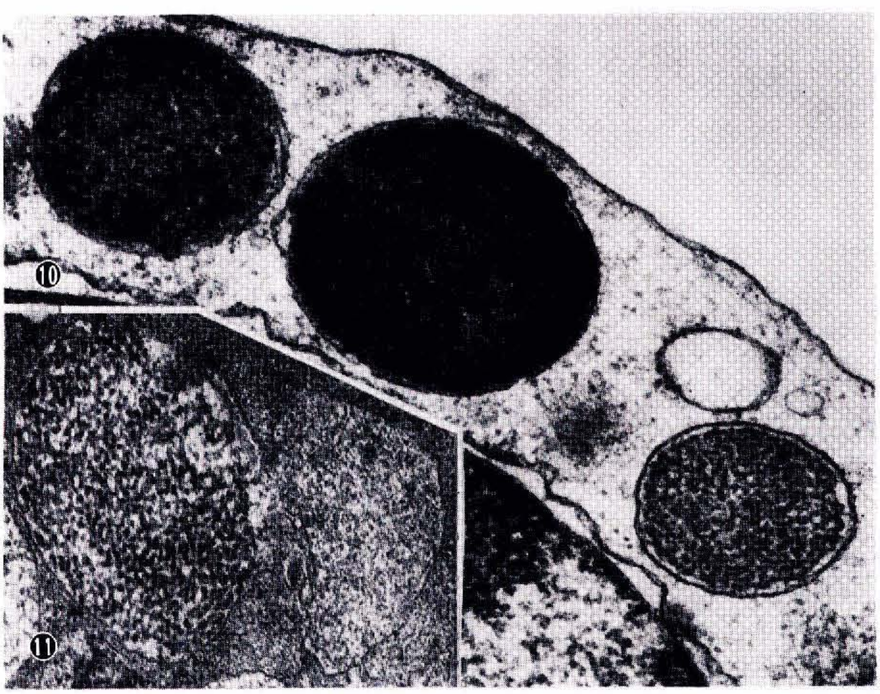
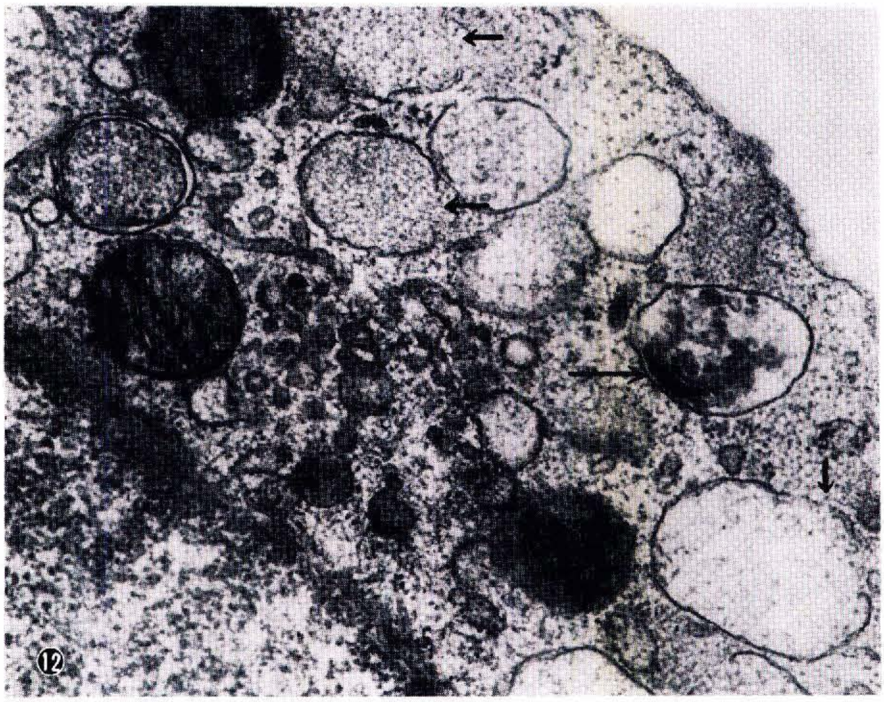
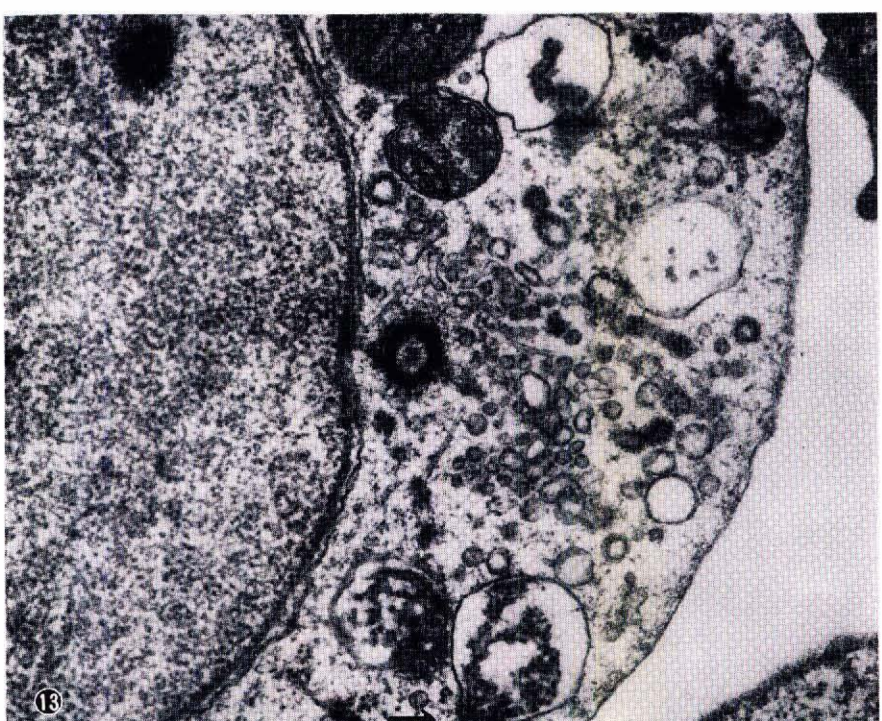

10
11

Plate VI YAMADA, J. *et al.*





YAMADA, J. *et al.* Plate VII