EDTA-Dependent Platelet Phagocytosis

A Cytochemical, Ultrastructural, and Functional Characterization

Kay A. Criswell, PhD, Michael A. Breider, PhD, and Michael R. Bleavins, PhD

Key Words: Platelet; Phagocytosis; EDTA; Satellitosis

Abstract

Platelet satellitosis of polymorphonuclear cells is a phenomenon induced or enhanced by the anticoagulant EDTA. In contrast with previously reported studies, the subject in the present case did not demonstrate platelet satellitism but was profoundly pseudothrombocytopenic owing to platelet phagocytosis. Virtually all polymorphonuclear leukocytes and monocytes contained numerous ingested platelets in contrast with previous cases in which phagocytosis was observed only rarely and involved ingestion of single cells. The phenomenon was documented by immunocytochemical staining and transmission electron microscopy. Autoantibodies were detected in EDTA-anticoagulated blood. However, neither platelet antibody nor phagocytosis was present when heparin, acid-citrate dextrose, or citrate was used as an alternative anticoagulant. The antibody was not temperature dependent. Mixing studies showed the transfer of the phagocytosis phenomenon to healthy donors. Although platelet function assays are typically normal in EDTA-dependent platelet satellitism, this subject showed no secondary aggregation wave in response to adenosine diphosphate and depressed adenosine triphosphate release with collagen, adenosine diphosphate, and arachidonic acid.

Platelet satellitism, or the binding of platelets to the periphery of neutrophils, was first reported in 1963[1] and has been reported subsequently by other investigators.[2-4] Binding occurs primarily to neutrophils, but also has been observed with monocytes. The phenomenon has been demonstrated only in vitro in the presence of EDTA. In addition, EDTA-dependent platelet satellitosis has been shown to be antibody mediated.[5,6] The condition has not been associated with any single disease or drug, and its greatest clinical relevance is spurious reporting that can lead to unnecessary treatment of thrombocytopenia.

In most cases of satellitism, platelet phagocytosis does not occur. However, phagocytosis of single platelets by neutrophils is seen as a rare event in isolated neutrophils after platelet satellitism.[7,8] We studied a case of EDTA-dependent pseudothrombocytopenia. Although platelet satellitism was not readily apparent in this person, surrounding neutrophils and monocytes had phagocytized nearly all circulating platelets. Ultrastructural, immunocytochemistry, direct platelet immunofluorescence, temperature dependence, and platelet function studies were conducted to document this case.

Case Report

A 22-year-old man was seen as a prospective candidate for a phase 1 clinical drug trial. Physical examination findings were unremarkable, and there was no history of surgery, transfusion, or bleeding disorders.

Laboratory findings showed an RBC count of $5.1 \cdot 10^6/\mu L$ ($5.1 \cdot 10^{12}/L$), a hemoglobin level of 15.5 g/dL (155 g/L), hematocrit of 45% (0.45), and normal RBC indices. A discrepancy between the impedance and optical methods used for the WBC count was seen on the hematology

analyzer (Cell Dyn 3500, Abbott, Abbott Park, IL). Manual estimation and hemocytometer determination of the WBC count confirmed the impedance WBC count of 8,600/μL ($8.6 \cdot 10^9$/L). The optical method showed a WBC count of approximately 30% higher. The platelet count was $35 \cdot 10^3$/μL ($35 \cdot 10^9$/L). The peripheral blood smear showed marked phagocytosis of platelets by neutrophils and monocytes with minimal numbers of free platelets.

Chemistry screening results were normal and included the following assays: glucose, cholesterol, bilirubin, protein, albumin, phosphorus, calcium, chloride, sodium, potassium, creatinine, aspartate aminotransferase, alanine aminotransferase, gamma-glutamyltransferase, alkaline phosphatase, creatine kinase, and urea nitrogen. Urinalysis screening results were negative.

In subsequent testing, the results for serum protein electrophoresis, quantitative immunoglobulins, direct and indirect antiglobulin tests, antinuclear antibody, antineutrophil antibody, rheumatoid factor, prothrombin time, activated partial thromboplastin time, thrombin clotting time, and fibrinogen were all within the reference ranges. Cryoglobulins and RBC cold agglutinins were not demonstrable. The candidate was not selected for the clinical trial owing to the pseudothrombocytopenic findings.

Materials and Methods

Examination of Peripheral Smear and Effects on Platelet Count

Duplicate EDTA samples were obtained. One sample was kept at room temperature and the other maintained at 37 C. Peripheral smears were prepared from both EDTA tubes within 2 minutes of collection and 15 and 60 minutes after collection. An aliquot of the room-temperature EDTA sample was refrigerated, and smears also were prepared at 15 minutes and 1 hour. Following the initial screening results, peripheral venous blood was collected in sodium heparin, sodium citrate (3.8%), and acid-citrate dextrose (ACD). Smears from alternative anticoagulants were prepared at 15 minutes only. Fingerstick smears also were obtained. Smears were Wright-stained and examined microscopically for the presence of platelet satellitism or phagocytosis. Platelet counts were performed on all samples using a Cell-Dyn 3500. Error flags showing significant differences between optical and impedance WBC counts occurred owing to interference by aggregated platelets. The percentage of neutrophils containing phagocytized platelets was determined during manual review of smears.

Antiplatelet Immunohistochemical Studies

Methanol-fixed blood smear slides were treated with normal horse diluent for 30 minutes at room temperature, mouse clone Y2/51 antiplatelet antibody (CD61; 1:25 dilution; DAKO, Carpinteria, CA) for 60 minutes at room temperature, horse antimouse IgG antibody linked to biotin (1:200 dilution; Vector, Burlingame, CA), the avidin-biotin complex kit (Vectastain ABC kit, Vector), and diaminobenzidine (Sigma, St Louis, MO). All sections were counterstained with hematoxylin. For negative controls, the primary antibody was omitted.

Transmission Electron Microscopy

One hundred microliters of EDTA- and ACD-anticoagulated buffy coat from the subject was pelleted, immersion fixed in 2.5% glutaraldehyde at room temperature for 2 to 3 hours, rinsed in sodium cacodylate buffer, postfixed in 1% osmium tetroxide for 3 hours, dehydrated through graded alcohols, and embedded in Epon resin. Specimens were examined with a CM100 BioTwin transmission electron microscope (Philips Electronics, Eindhoven, the Netherlands).

Flow Cytometric Direct Immunofluorescence Assay

Platelets were harvested from ACD-anticoagulated blood by centrifugation at 900*g* for 10 minutes. Platelet-rich plasma (PRP) was removed without disturbing the buffy coat RBC pellet and centrifuged for a second time at 900*g*. PRP again was removed and centrifuged at 2,000*g* to pellet platelets. The supernatant was discarded, and 1 mL of standard buffer (10 g powdered FA Buffer, Difco, Detroit, MI); 1 g NaN_3; and 10 mL heat-inactivated fetal calf serum) was added. This sample was divided into 2 aliquots and 10 mg EDTA added. PRP was not collected directly from an EDTA-anticoagulated tube since nearly all platelets were phagocytized under these conditions. The cells then were fixed by addition of 100 μL of 10% formalin while vigorously mixing. After 5 minutes at room temperature, platelets were washed and resuspended with standard buffer. The formalin-fixed platelets (50 μL) were stained for 20 minutes on ice with a cocktail of monoclonal antibodies (IgG, IgM, kappa, and lambda) that bound to immunoglobulin on the platelet surface. After washing, a second fluorescein-conjugated antibody, which reacts with the monoclonal antibody, was added. Unbound antibody was removed by additional washing, and the platelets were resuspended in standard buffer. Fluorescence emission at 488 nm was measured on an XL Flow Cytometer (Coulter, Hialeah, FL) and compared with a sample from a healthy donor (negative control) and a positive control sample prepared by incubating normal platelets with antibodies from commercially prepared positive serum.

Mixing Studies

Mixing studies were performed to determine whether the phagocytosis phenomenon could be transferred to ABO-compatible healthy control subjects (all group O). For

mixing studies, whole blood anticoagulated with EDTA or sodium citrate was centrifuged at 900*g* for 15 minutes to obtain PRP and 3,000*g* to obtain platelet-poor plasma (PPP). Washed platelets were prepared by 3 successive washes of platelets followed by centrifugation to pellet the platelets. Washed platelets were resuspended in sterile saline. Equal portions of PRP, PPP, or washed platelet suspension were mixed with equal portions of 2 donor whole blood samples. The following 4 conditions were tested: (1) EDTA-anticoagulated PPP from the subject plus whole blood from a healthy donor, (2) whole blood from a healthy donor plus washed platelets from citrated PRP from the subject, (3) citrated PRP from the subject plus normal citrated donor whole blood, and (4) citrated whole blood and washed platelets (citrated), both from the subject. Smears from mixing studies were prepared on samples kept at room temperature. Evaluations included enumeration of the percentage of neutrophils containing phagocytized platelets, platelet count, and analyzer-generated WBC error flags immediately after and at 15 minutes, 1 hour, 3 hours, and 5 hours after mixing.

Platelet Aggregation Studies

An aggregometer (Chrono-Log, Havertown, PA) was used to perform platelet aggregation on whole blood and PRP. Assays were replicated on 3 separate occasions during a period of 6 months. Specimens were anticoagulated with 3.8% sodium citrate and maintained at room temperature throughout testing. Testing was initiated within 30 minutes of venipuncture and completed within 2.5 hours. Simultaneous assessment of platelet adenosine triphosphate (ATP) release was measured using firefly luciferin-luciferase. Reagents were reconstituted with preservative-free, sterile saline, and a 1:2 dilution of whole blood was used in aggregation assays. The following platelet agonists also were purchased from Chrono-Log and used at the stated concentrations: collagen (1 mg/mL), arachidonic acid (50 mmol/L), and adenosine diphosphate (ADP; 1 mmol/L).

Results

Light-Microscopic Examination of Peripheral Blood

Examination of EDTA-anticoagulated whole blood smears revealed large accumulations of aggregated platelets within the phagosomes of neutrophils and monocytes. Relatively few free platelets were detected, consistent with the automated platelet count of 35 $\cdot 10^3/\mu L$ (35 $\cdot 10^9/L$). The degree of phagocytosis was marked at all collection times and could not be distinguished in the samples except for the advanced stage of platelet degeneration noted in the 1-hour and older samples. In most cases, individual platelets were engulfed in separate phagosomes, as shown in neutrophils **Image 1A** and a single monocyte **Image 1B**. In other instances, however, the entire cell was covered with platelets. Immunocytochemical staining using the platelet probe Y2/51 confirmed the identity of platelets and that aggregations of platelets were intracellular **Image 1C**. Repeated smears prepared from finger-stick samples or from samples containing sodium citrate, ACD, or heparin all revealed normal platelet morphologic features and distribution (ACD) **Image 1D** and absence of platelet phagocytosis or satellitism (ACD) **Image 1E**.

Electron-Microscopic Examination of Fixed Whole Blood

The ultrastructure of the platelet-neutrophil complex was examined by electron microscopy. With the exception of prominent pseudopodia and vacuole formation, neutrophils **Image 2A** and monocytes **Image 2B** showed no structural abnormalities. Platelet adherence to membrane was rare but could be identified (Image 2B). At higher magnification **Image 2C**), it could be discerned that the platelet within the monocyte vacuole had normal distributions of alpha-granules, dense bodies, mitochondria, and glycogen. Platelets were normal discoid shape, slightly irregular with blunt pseudopodia, or, more frequently, spheroid. In contrast with other ultrastructural studies of platelet satellitism, no long, projectile pseudopods were found on the platelets of this subject. The platelet adhering to the monocyte surface lacks a discrete, continuous cell membrane and is deficient in granules and dense bodies. Another internalized platelet within the same monocyte seems to be devoid of all internal structures (Image 2B). Platelets with deficient numbers of alpha-granules and dense bodies or general internal structure could be identified in approximately 50% of neutrophils and monocytes evaluated. However, in many instances, platelets were in an advanced stage of degeneration. Therefore, it was not possible to determine whether the structural defects were inherent to the platelet or were merely a by-product of phagocytic digestion or, more likely, whether degranulation had occurred following activation.

Anticoagulant-Temperature Dependence and Mixing Studies

Table 1 shows that there was no difference in platelet count, percentage of neutrophils containing phagocytized platelets, or analyzer-generated WBC error flags when EDTA-anticoagulated samples were tested at 3 different temperatures (0 C-37 C). However, the presence of EDTA was essential for the phenomenon to occur, as no effects were seen in these variables when citrate, ACD, or heparin anticoagulants or finger-stick samples were used.

Table 2 shows the results of mixing studies using EDTA-anticoagulated PPP or washed platelets from citrated whole blood obtained from the affected subject combined with

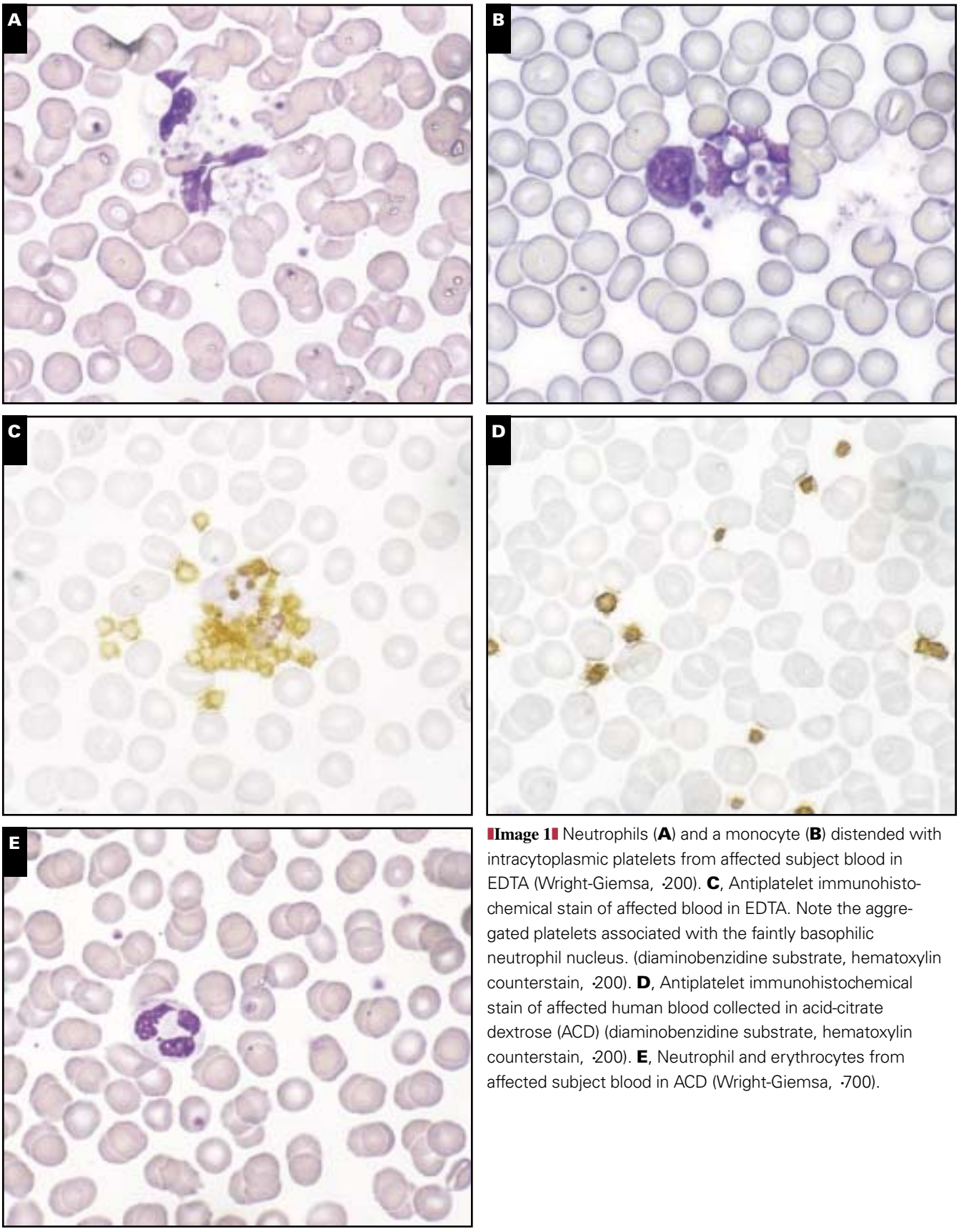

Image 1 Neutrophils (**A**) and a monocyte (**B**) distended with intracytoplasmic platelets from affected subject blood in EDTA (Wright-Giemsa, ·200). **C**, Antiplatelet immunohistochemical stain of affected blood in EDTA. Note the aggregated platelets associated with the faintly basophilic neutrophil nucleus. (diaminobenzidine substrate, hematoxylin counterstain, ·200). **D**, Antiplatelet immunohistochemical stain of affected human blood collected in acid-citrate dextrose (ACD) (diaminobenzidine substrate, hematoxylin counterstain, ·200). **E**, Neutrophil and erythrocytes from affected subject blood in ACD (Wright-Giemsa, ·700).

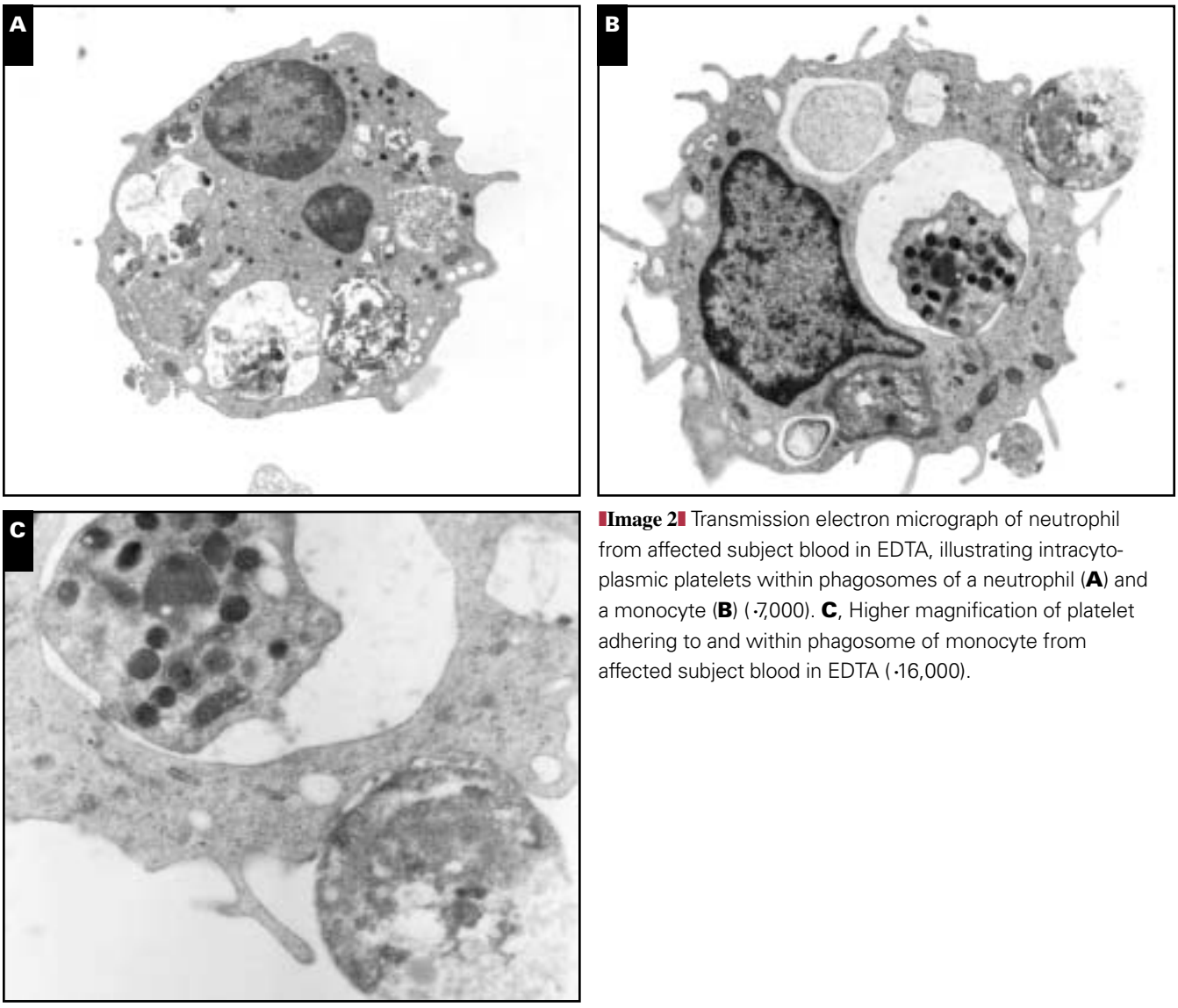

Image 2 Transmission electron micrograph of neutrophil from affected subject blood in EDTA, illustrating intracytoplasmic platelets within phagosomes of a neutrophil (**A**) and a monocyte (**B**) (·7,000). **C**, Higher magnification of platelet adhering to and within phagosome of monocyte from affected subject blood in EDTA (·16,000).

Table 1
Effect of Anticoagulant and Temperature on Platelet Phagocytosis Induction

Anticoagulant	Platelet Count, × 10^3/µL (× 10^9/L)*	Percentage of Neutrophils Containing Phagocytized Platelets†	WBC Error Flag Generated*
EDTA‡			
0 C-4 C	37.5 (37.5)§	98§	Yes
20 C-24 C	35.8 (35.8)§	97§	Yes
37 C	48.0 (48.0)§	92§	Yes
Citrate‡	265.4	0	No
Acid-citrate dextrose‡	267.1	0	No
Heparin‡	264.3	0	No
None (finger-stick)	ND	0	ND

ND, not done.
* Data generated on Cell-Dyn 3500 hematology analyzer, Abbott, Abbott Park, IL.
† Multiple smears prepared for each phase of testing. A total of 500 neutrophils per phase were evaluated.
‡ Data from sample prepared <2 minutes following venipuncture for 37°C sample and 15 minutes following venipuncture for 0°C-4°C and 20°C-24ºC EDTA samples and other anticoagulants. Data at 1, 3, and 5 hours after venipuncture were similar for EDTA samples and are not shown.
§ Significantly different from values obtained with citrated sample (Student *t* test, $P < .05$).

Table 2
Mixing Studies: Induction of Phagocytosis by Anticoagulant and Time Dependence

Experimental Condition	Platelet Count, × 10^3/μL (× 10^9/L)*	Percentage of Neutrophils Containing Phagocytized Platelets†	WBC Error Flag Generated*
EDTA-anticoagulated platelet-poor plasma from the affected subject + whole blood from the healthy donor			
Affected subject			
Immediate	241.9 (241.9)	0	No
15 min	242.4 (242.4)	0	No
1 h	152.2 (152.2)‡	19‡	Yes
3 h	106.7 (106.7)‡	42‡	Yes
Healthy donor			
Immediate	329.9 (329.9)	0	No
15 min	330.6 (330.6)	0	No
1 h	227.6 (227.6)‡	23‡	Yes
3 h	164.5 (164.5)‡	53‡	Yes
Washed platelets (citrated) from the affected subject + whole blood from the healthy donor			
Affected subject			
Immediate	287.9 (287.9)	0	No
15 min	288.7 (288.7)	0	No
1 h	276.9 (276.9)	0	No
3 h	202.4 (202.4)‡	9‡	No
Healthy donor			
Immediate	356.5 (356.5)	0	No
15 min	355.9 (355.9)	0	No
1 h	354.7 (354.7)	0	No
3 h	242.5 (242.5)‡	12‡	No

* Data generated on Cell-Dyn 3500 hematology analyzer, Abbott, Abbott Park, IL.
† Multiple smears were prepared for each phase of testing. A total of 500 neutrophils per phase were evaluated.
‡ Significantly different from values measured immediately (Student *t* test, $P < .05$).

whole blood from 2 healthy donors with blood group type O. PPP was more effective for transferring the phenomenon than washed platelets. The maximum percentage of donor neutrophils having phagocytized platelets after incubation with PPP was approximately 50%. Washed platelets retained the ability to initiate phagocytosis with donor EDTA-anticoagulated whole blood, although the percentage of affected cells was only 9% to 12%. Transfer of phagocytosis was also time dependent, with no effect immediately or at 15 minutes, but with a detectable difference at 1 hour and the maximal effect seen at 3 hours or 5 hours. Substantial decreases in the platelet count and increased generation of WBC error flags occurred only when control samples were combined with EDTA-anticoagulated PPP for 1, 3, or 5 hours. Similar decreases in the percentage of affected neutrophils were seen in smears prepared at 3 and 5 hours, suggesting the 3-hour values were a maximum effect (data not shown). Further mixing studies using PRP from healthy donors and the affected subject's washed platelets or citrated PRP from the affected subject plus whole blood from the healthy donor had no effect on platelet count, the presence of platelet phagocytosis, or WBC error flag generation. This confirmed that EDTA was essential for the phenomenon to occur (data not shown).

Direct Platelet Immunofluorescence Flow Cytometry Assay

The direct immunofluorescence test on the subject's platelets had a strongly positive result when EDTA was added to the sample. Fluorescence intensity exceeded that of the positive control. In contrast, when ACD-anticoagulated blood was tested alone, the direct immunofluorescence test result was negative.

Platelet Aggregation Studies

Consistent changes were detected in the 3 independent platelet function trials. Deviations from the control consisted of depressed primary aggregation with ADP, disaggregation of platelets following 3 to 4 minutes of exposure to ADP, failure to form the customary secondary wave of aggregation with ADP, and depressed ATP release in response to collagen, ADP, and arachidonic acid.

Figure 1 shows representative tracings of platelet aggregation in the subject compared with the control sample. As shown in Figure 1A, following collagen exposure, primary platelet aggregation of the subject sample was similar to that of the control sample. Collagen-induced ATP release was statistically less than that of the control sample but was still within the established reference range. Figure 1B shows the whole blood aggregation in response to ADP: disaggregation of platelets, depressed aggregation at 6 minutes, and depressed ATP release in the affected subject. In the PRP assay (Figure 1C), the control sample shows the typical smooth biphasic curve anticipated with ADP. In

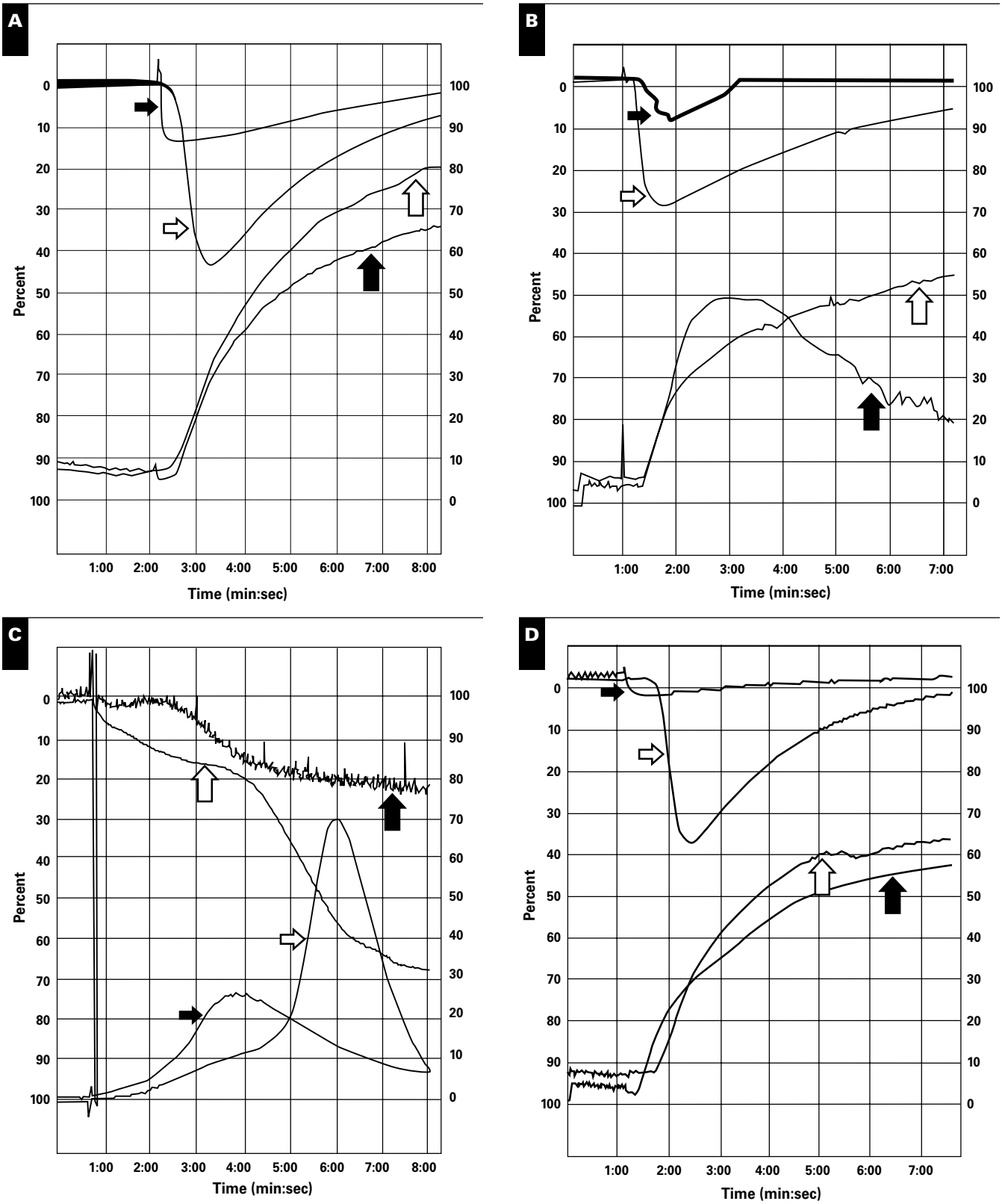

Figure 1 Platelet aggregation and adenosine triphosphate (ATP) release responses of healthy control and affected subjects under varied conditions. **A**, Citrated whole blood in 1 mg/mL collagen. **B**, Citrated whole blood in 1 mmol/L adenosine diphosphate (ADP). **C**, Platelet-rich plasma in 1 mmol/L ADP. **D**, Citrated whole blood in 50 mmol/L arachidonic acid. Closed arrows depict sample from the affected subject. Open arrows depict sample from the healthy control subject. Small arrows are associated with ATP release and large arrows with extent of platelet aggregation.

contrast, the subject sample clearly shows the lack of a secondary wave of aggregation, depressed overall aggregation, and an erratic tracing, probably also suggestive of an aggregation-disaggregation response (Figure 1C). ATP release was depressed in PRP and whole blood aggregation assays (Figure 1C). Figure 1D shows the whole blood platelet aggregation tracing associated with arachidonic acid. Overall aggregation was similar to that in the control sample, but arachidonic acid–stimulated ATP release was depressed. **Table 3** summarizes the effects of collagen, ADP, and arachidonic acid on whole blood and PRP aggregation and ATP release with values expressed as a percentage of control values. Approximately 2-fold decreases in maximal aggregation were seen with ADP, while responses to collagen and arachidonic acid were not statistically different from control. Depressed ATP release of 3 to 5 fold was seen in response to the addition of collagen, ADP, or arachidonic acid.

Discussion

The most common cause of pseudothrombocytopenia is platelet clumping or formation of platelet rosettes around neutrophils. This phenomenon is usually dependent on the presence of EDTA. Four large studies reported in 1984, 1988, and 1992 found a frequency of approximately 0.1% in the general population.[9-12] The subject in the present report differs from other reports because he demonstrated neutrophilic and monocytic phagocytosis of approximately 85% of all platelets. Although phagocytosis has been observed in some cases,[13,14] it always has been viewed as a rare event and has not been reported to encompass nearly all circulating platelets. The key commonality between the present case and more classic platelet satellitism is that platelet phagocytosis occurred only in the presence of EDTA and was strikingly absent with other anticoagulants such as citrate, ACD, and heparin and in finger-stick preparations.

The present case also demonstrated a strongly positive platelet immunofluorescent test result. This assay has been used widely to identify antibodies in the serum samples of persons demonstrating EDTA-induced pseudothrombocytopenia. In 1986, von dem Borne et al[6] introduced the idea of cryptantigens or hidden antigens and described the platelet immunofluorescent test. These antigens occur exclusively on platelets, most notably on the membrane glycoprotein IIb/IIIa complex. As indicated by their name, cryptantigens normally are not exposed on circulating platelets, but become exposed when the platelet membrane undergoes a conformational change as Ca^{2+} ions are removed by the chelator EDTA. The presence of these autoantibodies seems to be a hallmark in persons with EDTA-induced pseudothrombocytopenia, although the mechanism for this autoantibody production is unclear.

In most cases of EDTA-dependent phenomena, effects on platelets are greatest at room temperature or colder, and effects may even be eliminated if samples are kept at 37 C.[4,5,15,16] The present case varied from the norm by showing no difference in the degree of phagocytosis when tested from 0 C to 37 C. The ability to transfer platelet satellitism to normal donor platelets has been shown in other studies,[3,4] and transfer of the phagocytic properties also was demonstrated in the present study. The effect was not transferable if EDTA was not in the system, showing it to be an EDTA-dependent event. Surprisingly, a small number of washed platelets obtained from a citrated specimen of the affected subject also were phagocytized in EDTA-anticoagulated donor whole blood. This suggests that the predominant factor necessary for transfer resides within the plasma, although some component also may exist on the platelet surface and be reactive if EDTA is present.

Electron microscopy corroborated the light microscopic findings. Several studies have reported the formation of platelet dendrites and neutrophil pseudopods and, in general, a rather limited contact between the two cell types.[2,4,17] Still another study identified a greater number of glycogen particles in platelets participating in rosette formation than in free circulating platelets.[13] In the present study, approximately 85% of platelets were internalized and were in various stages of digestion. Some internalized platelets had completely normal intracellular morphologic features, while others were

Table 3
Effect on Platelet Aggregation*

	Whole Blood Aggregation		Platelet-Rich Plasma Aggregation	
Agonist	**Maximum Aggregation**	**ATP Release**	**Maximum Aggregation**	**ATP Release**
Collagen	82 ± 7	38 ± 4†	87 ± 6	45 ± 7†
ADP	46 ± 6†	24 ± 3†	52 ± 4†	36 ± 5†
Arachidonic acid	93 ± 4	17 ± 2†	91 ± 5	32 ± 4†

ADP, adenosine diphosphate; ATP, adenosine triphosphate.
* Data are expressed as mean ± SD percentage of control for 3 successive trials. Aggregation assessed 1, 2, and 5 months after identification of EDTA-dependent phagocytosis.
† Significantly different from results for healthy donor (Student *t* test, $P < .05$).

deficient in dense bodies and granules. This deficiency also was noted in platelets that had not been phagocytized. Although no general conclusion can be drawn owing to the overwhelming degree of phagocytosis evident, the lack of dense bodies and granules within platelets is interesting when coupled with the functional defects detected in platelet aggregation assays.

Although most studies report normal platelet aggregation in EDTA-dependent syndromes, 1 study[14] described a decreased primary aggregation with ADP, epinephrine, and collagen; failure to develop a secondary wave of aggregation in response to ADP; and persistent disaggregation with ADP. The response of the subject in the present study to ADP was remarkably similar. This similarity may be purely coincidental, and, unfortunately, ultrastructure was not assessed in the other study, so it is unclear whether the other subject also had a lack of dense body and granule formation. It is unlikely that the subject in the present study has a genetic deficiency of dense bodies and granules because there were many platelets observed with adequate numbers of these components. In addition, this person did not have a bleeding diathesis that would be anticipated with total deficiency of these structures. It may suggest, however, that the subject in the present study is more susceptible to platelet activation and degranulation. In support of this, disk to sphere formation, the first phase of platelet activation, was readily apparent in the ultrastructural platelet examination.

Although some reports suggest that EDTA-dependent platelet phenomena occur more frequently in ill or hospitalized patients, especially those with thrombotic complications,[7,13,18] others have shown that there is no correlation between the presence of disease and this phenomenon.[4,11,12,19] There was no evidence of disease or illness in the affected subject described in the present article, and although a quantitative functional platelet defect was identified in platelet aggregation assays, there was no evidence of bleeding diathesis. Interestingly, although there may be no increased health risks, the in vitro phenomenon may persist over long periods. The subject in the present study demonstrated qualitatively unchanged phagocytosis of platelets 1 year after his initial diagnosis. Similarly, a study that monitored long-term patient follow-up showed persistence of platelet satellitism 15 to 20 years later.[19] From the study of this case, it seems that marked phagocytosis of platelets can be an extension of the more classic platelet satellitism phenomenon, and it is also dependent on the presence of EDTA and patient autoantibody.

From Drug Safety and Evaluation, Pfizer Global Research & Development, Ann Arbor, MI.

Address reprint requests to Dr Criswell: Drug Safety and Evaluation, Pfizer Global Research & Development, 2800 Plymouth Rd, Ann Arbor, MI 48105.

Acknowledgments: We thank Kathy Toy for invaluable assistance with immunocytochemical staining and Greg Sobocinski and Matt Anstett for excellent technical assistance in preparation of ultrastructural samples and preparation of photographs.

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