



Neutrophil-Erythrocyte Rosettes in COVID-19

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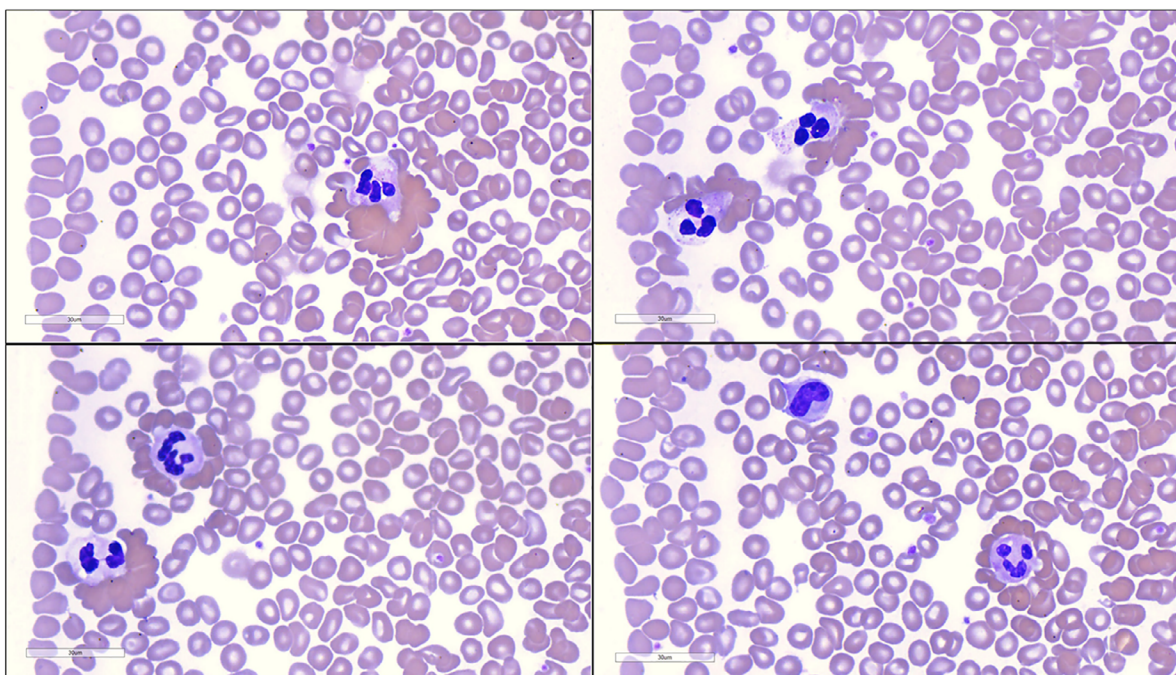


FIGURE 1 Neutrophil-erythrocyte rosettes in acute SARS-CoV-2 infection.

An 85-year-old Chinese female, known to have hypertension, hyperlipidemia, diabetes mellitus, atrial fibrillation, osteoporosis, and Alzheimer's dementia was admitted to the National Centre for Infectious Diseases, Singapore for clinical monitoring due to a mild COVID-19 infection. She had completed a primary series of COVID-19 vaccination with two doses of Pfizer BNT162b2 mRNA SARS-CoV-2 and received a booster dose of vaccination (BNT162b2 mRNA SARS-CoV-2), 1 month prior to her admission. Nasopharyngeal swab taken for COVID-19 real-time polymerase chain reaction was positive, with a cycle threshold value of 24.6, and she did not require supplemental oxygen throughout admission. The peripheral blood film (PBF) done on Day 2 of her symptoms showed numerous neutrophil-erythrocyte rosettes (FIGURE 1, Wright stain, 80 × objective, Motic EasyScan One), but no spherocytes, red cell agglutination, or reactive lymphocytes were observed. Monocyte-Erythroid rosetting was also not seen. Repeat PBF performed on a blood specimen taken with a lithium heparin tube (non-EDTA) as well as from finger prick showed persistence of neutrophil-erythrocyte rosettes, excluding pre-analytical causes. A previous PBF performed 3 years prior to her admission did not show any neutrophil-erythrocyte rosettes. Full blood count revealed a hemoglobin of 11.4 g/dl, white blood cell count of $5.8 \times 10^9/L$, and platelet count of $161 \times 10^9/L$. Hemolytic screen did not reveal any evidence of hemolysis, with normal lactate dehydrogenase levels of 461 U/L (reference range (RR): 270–550 U/L), total bilirubin of 18 $\mu\text{mol/L}$ (RR: 5–30 $\mu\text{mol/L}$), and a haptoglobin of 46 mg/dl (RR: 36–200 mg/dl). However, her absolute reticulocyte count was slightly elevated at $95.6 \times 10^9/L$ (RR: 25–85 $\times 10^9/L$). Autoimmune screen with anti-nuclear antibodies was negative and a pan computer-tomography scan of her neck, thorax, abdomen, and pelvis did not show any lung infiltrates or features of malignancy.

Further testing was performed to evaluate the cause of the neutrophil-erythrocyte rosettes. Specimens were sent to the Health Sciences Authority Blood Services Group (HSA BSG), Red Cell Reference Laboratory, where the direct Coombs test was weakly positive (1+) for anti-C3b and anti C3d. Testing of serum for red cell antibody identification showed an Anti-Mia, with the red cell eluate negative for common red cell antibodies. Serology testing (Elecsys Anti-SARS-CoV-2 S and Elecsys Anti SARS-CoV-2) on Day 3 of illness showed an elevated antibody titer against the SARS-CoV-2 spike (S) protein receptor binding domain of 250 U/ml and was negative for nucleocapsid-antigen (N-antigen). The red cell eluate was further tested with the Euroimmune anti-SARS-CoV-2 Spike S1 ELISA and was negative, suggesting that the neutrophil-erythrocyte rosettes seen were unlikely due to anti S1 IgG immunoglobulins. 2 months after recovery from COVID-19, the repeat PBF showed resolution of neutrophil-erythrocyte rosettes with a negative direct Coombs test and no biochemical features of hemolysis.

Neutrophil-erythrocyte rosettes are rare. They are associated with autoimmune hemolytic anemia (AIHA)^{1–4} and paroxysmal cold hemoglobinuria.⁵ The mechanism of neutrophil-erythrocyte rosettes in AIHA is still poorly defined. It is postulated to be due to a factor in the IgG fraction of immunoglobulin, primarily directed at some red blood cell surface antigen(s).⁶ Shulman et al² recently proposed the mechanism of the interaction of neutrophil surface Fc receptors with IgG1- or IgG3-coated erythrocytes. This usually requires a high titer of antibody-coated target cells to mediate neutrophil adhesion. In our patient, further testing with a

red cell flow cytometry for Immunoglobulin G, A, or complement could have identified the antibody; however, these studies are unavailable at our center and at the HSA BSG, and thus we have not been able to pursue confirmatory testing.

To our knowledge, this is the first reported case of COVID-19 associated neutrophil-erythrocyte rosettes, given the temporal association of neutrophil-erythrocyte rosettes with acute SARS-CoV-2 infection and subsequent resolution on convalescence, which is most likely secondary to an unidentified IgG antibody or complement. Clinicians should be aware of the rare possibility of neutrophil-erythrocyte rosettes in COVID-19 and exclude AIHA which is commonly associated with such cases.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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