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## Development and clinical application of nucleated red blood cell counting and staging on the automated haematology analyser XE-2100

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We initially developed a new flow cytometric (FCM) reference method for the enumeration and staging of nucleated red blood cells (NRBC) in 1997 [Wang et al., 1998 (XIth International Symposium on Technological Innovations in Laboratory Haematology, Banff, Canada, 1998); Tsuji et al., 1999 (Cytometry, 1999)]. The method used CD45 antibody and propidium iodide staining to separate NRBCs from other cells. Accuracy and precision were enhanced because larger numbers of cells were counted than was possible with the manual method. We also developed a method for automated NRBC counting on a haematology analyser, the XE-2100 (Wang, 1988). NRBC were separated from other cells using a special lysing buffer and a fluorescent dye. The XE-2100 was found to detect peripheral and cord blood NRBC accurately and precisely when compared with cell morphology or FCM control methods. The FCM NRBC staging method was established through the identification of different NRBC populations following the novel staining and lysing method. To evaluate the method further, we sorted samples containing NRBCs using a FACSort and investigated NRBC staging on the Sysmex XE-2100 based on the cell sorting results. Data were analysed using special software (ida). First, we used the data in various parameter combinations. We then established gates to classify the NRBC populations. Finally, we analysed blood specimens from patients with different types of diseases to explore possible clinical applications.

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