

# Red Blood Cell- Particle interaction for Enhanced Sedimentation

J.Liu, C.Vipulanandan, Ph.D., P.E.

Center for Innovative Grouting Materials and Technology (CIGMAT)

Department of Civil and Environmental Engineering

University of Houston, Houston, TX, 77204-4003

Tel: (713)743-4278; Email Address: jliu@mail.uh.edu, cvipulanandan@uh.edu

## Abstract

In order to enhance the separation by sedimentation, interaction of red blood cells with particle suspensions were investigated. In the particle suspension, nano to micro scale particles were used. Suspension to blood volume ratio used in this study was 1:1. Non-nucleated red blood cells (nn-RBC) were separated from nucleated red blood cells (n-RBC) and other blood cells in the whole blood, by the addition of selected solutions with nanoparticle or micro-scaled particles. Separation was observed within 30 min in the blood fluids. Observation through the optical microscopy showed the nucleated cells were largely enriched in the supernatant after the sedimentation separation.

## 1 Introduction

Red blood cells (RBC) are very important for body health, since they are used for oxygen transfer in the body by hemoglobin. The percentage of RBC in the whole blood should be maintained in a sustainable level. But for disease such as polycythemia, the concentration of RBC in the blood has to be reduced. For stem cell transplantation (Pahwa et al. 1994), RBC need to be depleted from the umbilical cord blood or bone marrow in order to eliminate interference of blood type difference, as antigens indicate blood types exist in mature red blood cells. So eliminating of mature red blood cells from bone marrow or peripheral blood is needed, for stem cell storage and transplantation, for curing disease like polycythemia, and for diagnosis of genetic disorders (Beutler 1975). Nano to micro scale particle suspensions were chosen in this study to increase the density of mature RBC by attaching on the cell surface with certain functional groups in the suspension used for separation, so that the nn-RBC can be depleted within short time by sedimentation, the n-RBC left includes the hematopoietic progenitor cells or the hematopoietic precursor cells (Telen 1990).

## 2 Objective

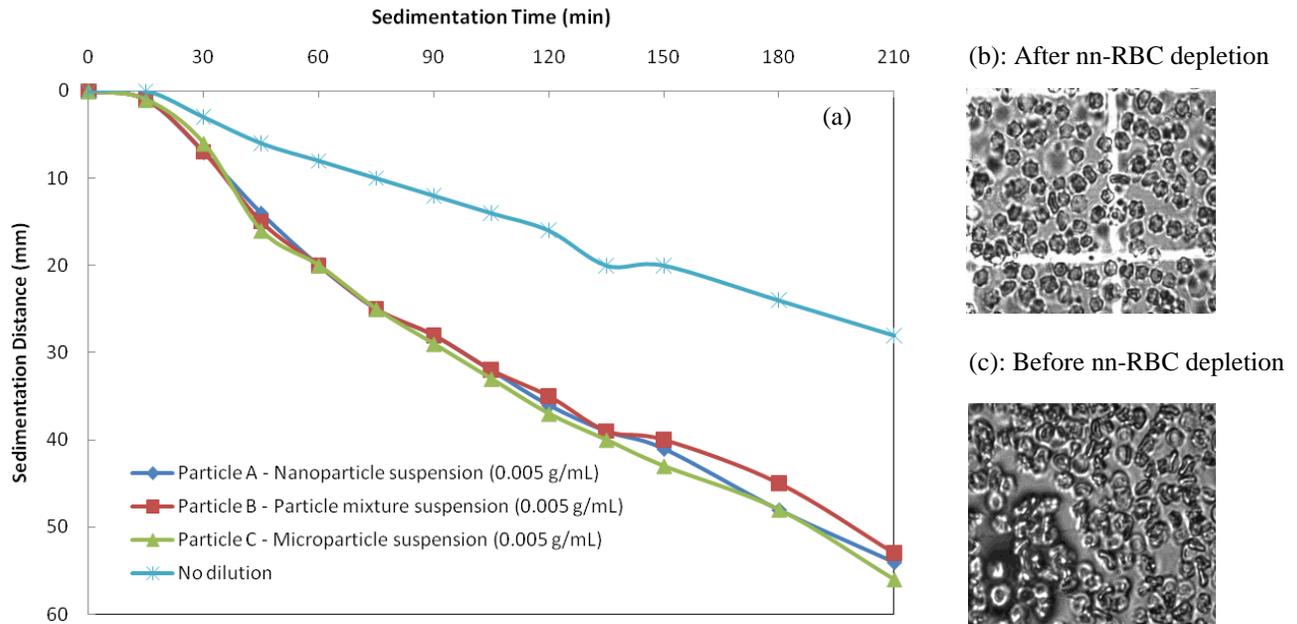
The objective of this study is to separate nn-RBC from n-RBC and other blood cells by sedimentation with nano or micro scale particle suspensions.

## 3 Materials and Methods

Blood sedimentation speed with different micro-scale and nano-scale particles in viscous liquid was tested. Dilution of 1:1 (volume of the solution : volume of the blood) was used. Used wintrobe tube for the sedimentation test. Inject 10 mL of the anti-coagulation whole blood or blood after dilution into the tube, set the wintrobe tube at an angle of 40° to horizontal, recorded the reading for blood sedimentation in 15 min interval. Particle A, B, and C were used here, where A was metal oxide nanoparticle produced in the laboratory, C was micro-scale clay particle, B was a combination of A and C. And the solution used here was a solution of gelatin. Natural sedimentation was used for depletion of the nn-RBC by only the media solution in the centrifugation tube, the supernatant was collected twice after 30 min sedimentation separately. Optical microscopy was used to count the number of the nucleated cells before and after sedimentation.

**4 Results and Discussion**

Sedimentation results are shown in Fig.1. All three particle suspensions used had almost the same effect, all showed tremendous enhancement for RBC sedimentation. In 30 min, compared to the control, the settlement distance doubled with all the suspensions. Though solution of particle B and C have slightly better RBC sedimentation effect, considering other possible separation method, particle B and particle A were chosen for further study. The collected nucleated RBC with the dilution medium and the original blood liquid with different dilution times was observed under optical microscopy by hemacytometer. It showed that mature RBC is bi-concave plated shaped cells, and other cells have irregular shape with nuclear inside. It can be told that 5 times dilution is good for optical microscopy observation, and after sedimentation by the suspension, a lot more of the nucleated cells were found in the collected supernatant, compared to the original whole blood (Fig.1(b) and (c)).



**Fig.1 Comparison of Red Blood Cell Sedimentation with Particle Suspensions**

**5 Conclusions**

1:1 time dilution was very effective for RBC sedimentation. The selected solution is a good medium for RBC sedimentation. The combination of the nanoparticle and the micro-scale clay particle, or the nanoparticle were effective for RBC sedimentation. Optical microscopy is a good tool to observe the nn-RBC depletion.

**6 Acknowledgements**

This study was supported by the CIGMAT in corporation with the Harvest Technologies Corporation, Boston, Massachusetts.

**7 References**

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