

Monitoring the properties of erythrocytes and leucocytes exposed to metal elements and compounds using flow cytometry

Název:

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Datum: **19.9.2014**

Reg.č.projektu: CZ.1.07/2.3.00/20.0148

Název projektu: Mezinárodní spolupráce v oblasti "in vivo" zobrazovacích technik

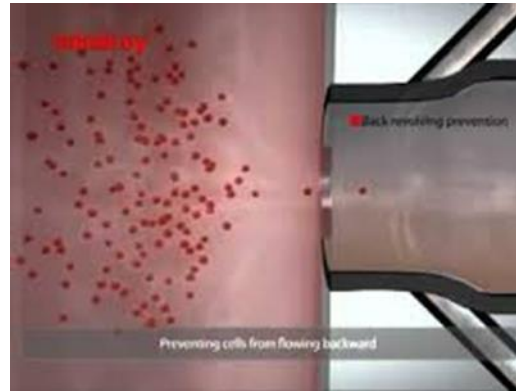


Hematology analyser BC 5800



- Number of blood cells can be measured on a hematology analyser.
- 24 parameters
- Leukocyte count and differential
- Erythrocyte count
- Platelet count
- Determination of the hemoglobin concentration in the blood

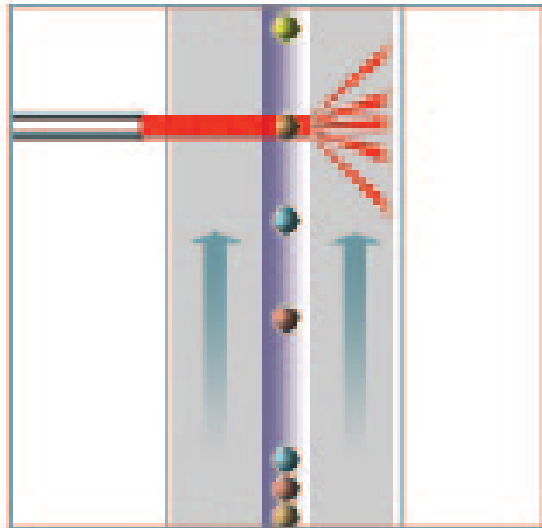
Hematology analyser



Principles

Impedance method for RBC and PLT counting

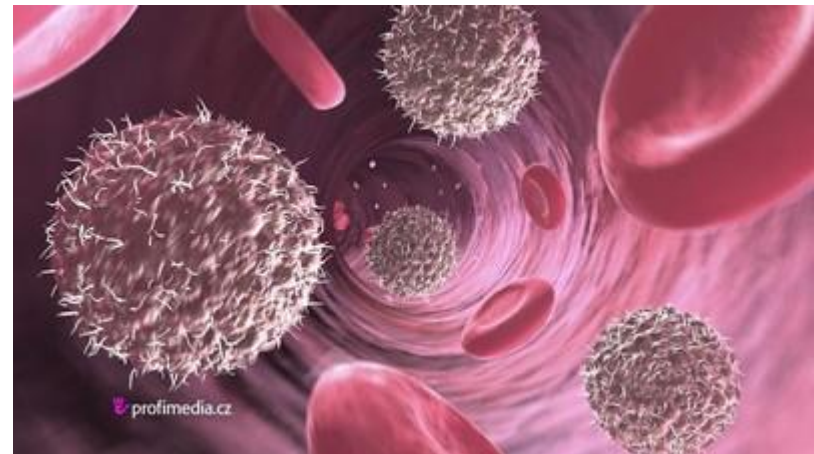
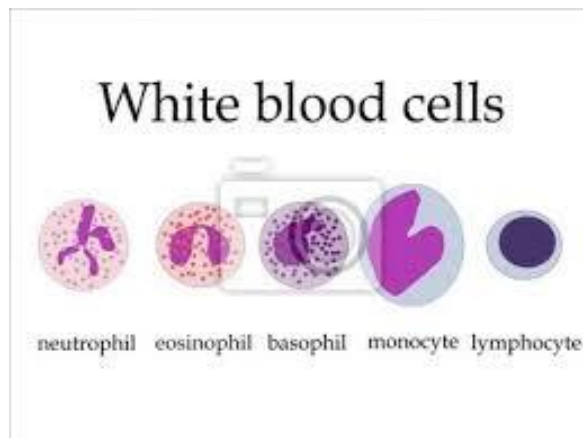
Cyanide free reagent for hemoglobin test



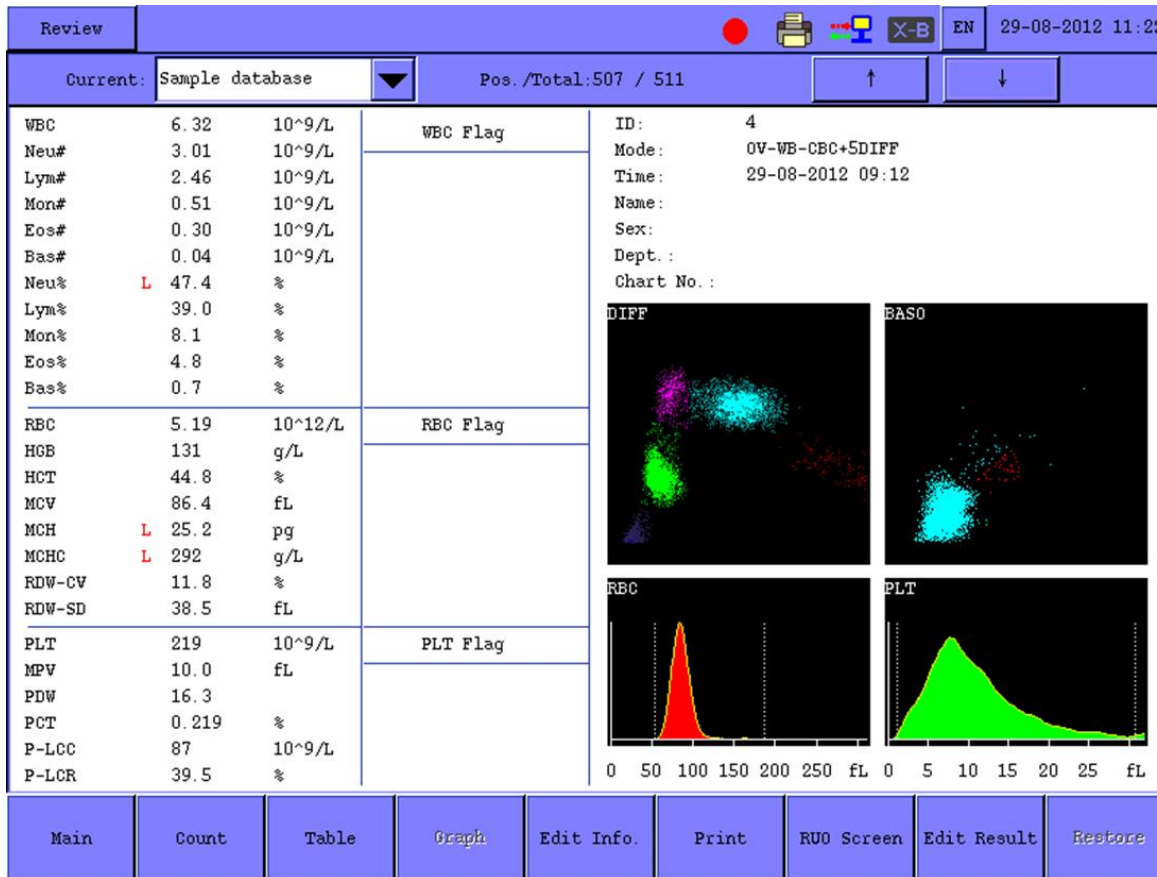
Flow cytometry + laser light scatter + chemical dye method for WBC differential analysis and WBC counting

Complete blood count

- Number of white blood cells (WBC) – leukocytes, differential count: lymphocytes (LYM), monocytes (MON), neutrophils (NEU), eosinophils (EOS), basophils (BAS) and their percentage content
- Number of red blood cells (RBC) – erythrocytes, hemoglobin concentration in the blood (HGB), hematocrit (HCT), RBC volume (MCV), mean amount of HGB in RBC (MCHC), average concentration of HGB in RBC (MCHC), distribution width of RBC (RDW)
- Number of platelets (PLT) – thrombocytes, distribution width of platelets (PDW), platelet volume (MPV)



Complete blood count



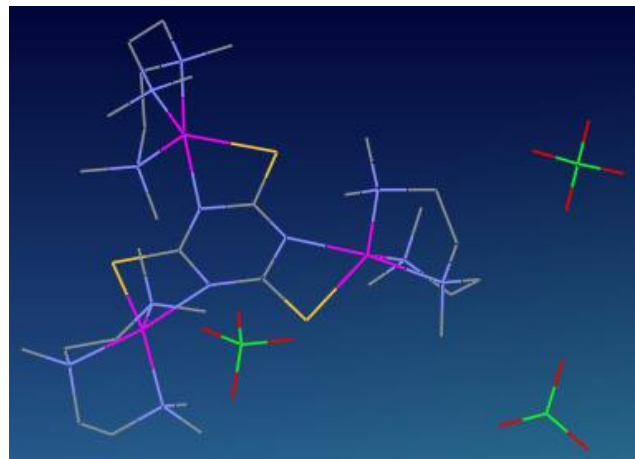
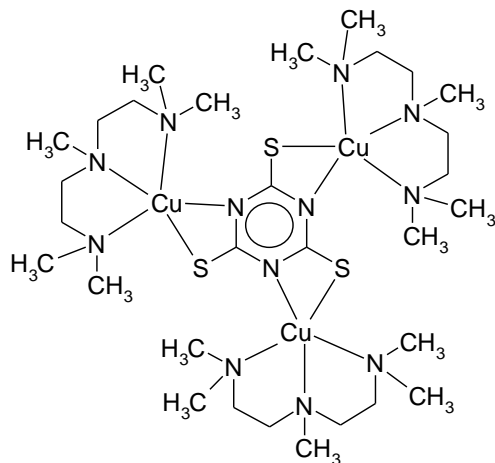
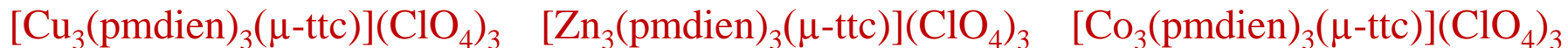
Result of hematological blood test – blood count:

WBC - leukocyte count ,
 RBC – erythrocyte count,
 PLT – platelet count, HGB-
 hemoglobin concentration in
 the blood

On the figures with black
 background: DIFF-
 differential except basophils
 (lymphocytes – green,
 monocytes – pink,
 neutrophils – blue,
 eosinophils-red, other cells -
 violet) , BASO-basophils
 (basophils-red, other cells -
 blue, RBC-distribution of
 erythrocytes, PLT-distribution
 of platelets

Aim of the work

- Investigate interactions of metal complexes (Cu, Zn and Co) and metal nanoparticles (PdNPs, RuNPs) with blood cells using hematology analyser
- Observe influence of metal compounds on blood cell count



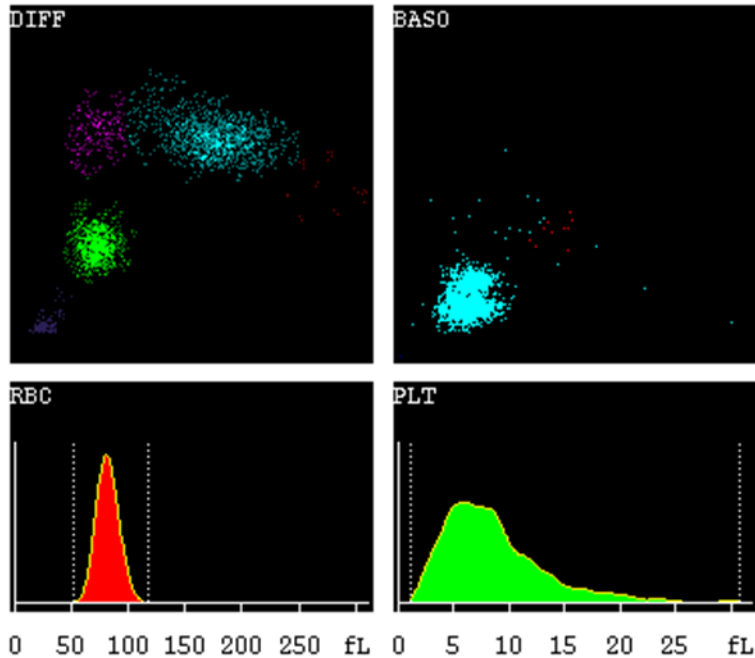
These metal complexes were cytotoxic to some tumor cell lines

PdNPs (PdCl_2)

RuNPs ($\text{RuCl}_3 \cdot 2.5 \text{H}_2\text{O}$)

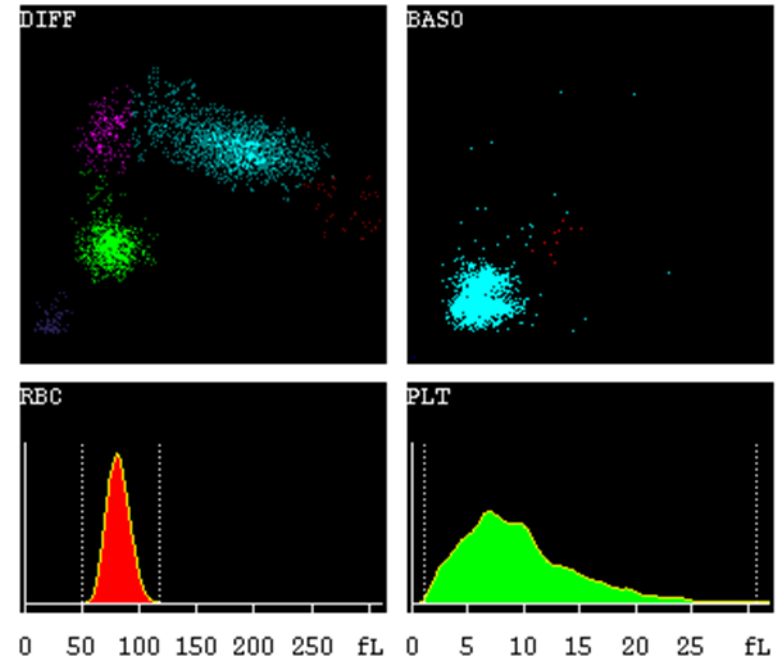
Control

Blood cells count diluted in phosphate buffer (1:2)



WBC 3.02×10^9 cells/mL
MON 0.23×10^9 cells/mL
EOS 0.11×10^9 cells/mL
RBC 1.84×10^{12} cells/mL
PLT 61×10^9 cells/mL

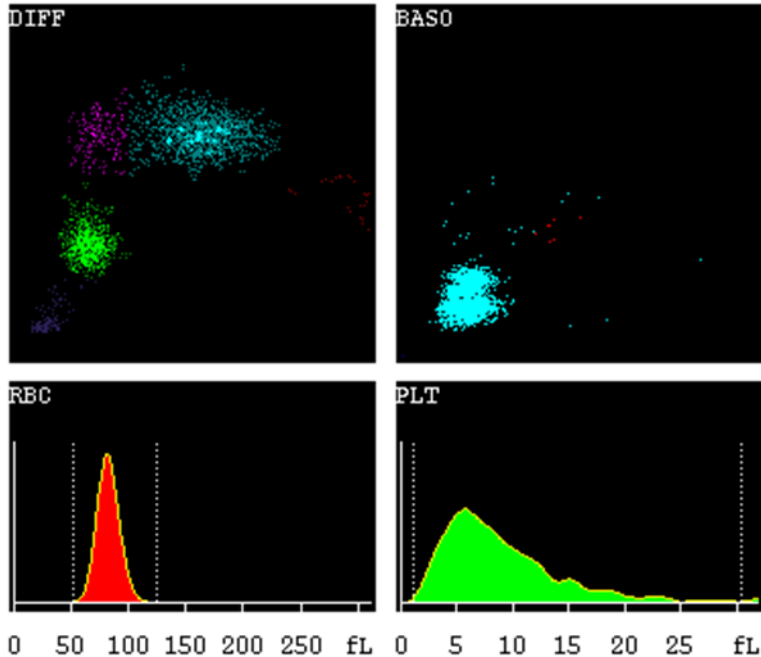
Blood cells count diluted in phosphate buffer (1:2) **after 1 hour**



WBC 3.32×10^9 cells/mL
MON 0.21×10^9 cells/mL
EOS 0.13×10^9 cells/mL
RBC 1.85×10^{12} cells/mL
PLT 62×10^9 cells/mL

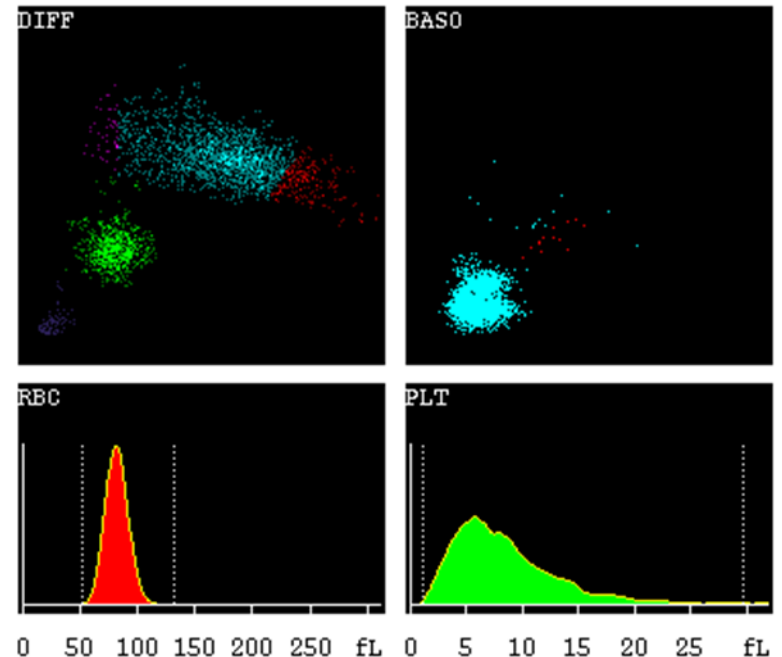
$[\text{Cu}_3(\text{pmdien})_3(\mu\text{-ttc})](\text{ClO}_4)_3$

Blood cells count diluted in phosphate buffer (1:2), Cu 345 $\mu\text{g/mL}$



WBC 2.98×10^9 cells/mL
MON 0.24×10^9 cells/mL
EOS 0.12×10^9 cells/mL
RBC 1.84×10^{12} cells/mL
PLT 56×10^9 cells/mL

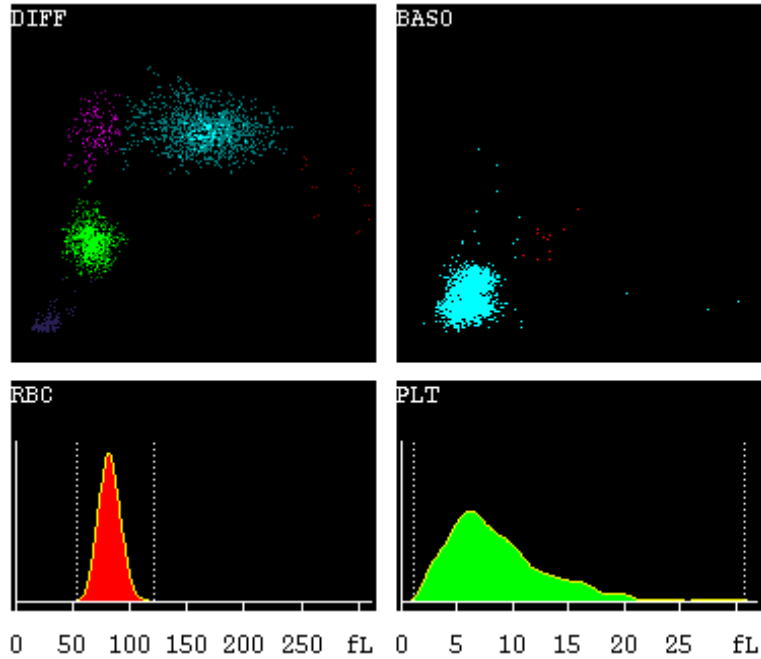
Blood cells count diluted in phosphate buffer (1:2), Cu 345 $\mu\text{g/mL}$ after 1 hour



WBC 3.64×10^9 cells/mL
MON 0.06×10^9 cells/mL
EOS 0.37×10^9 cells/mL
RBC 2.08×10^{12} cells/mL
PLT 51×10^9 cells/mL

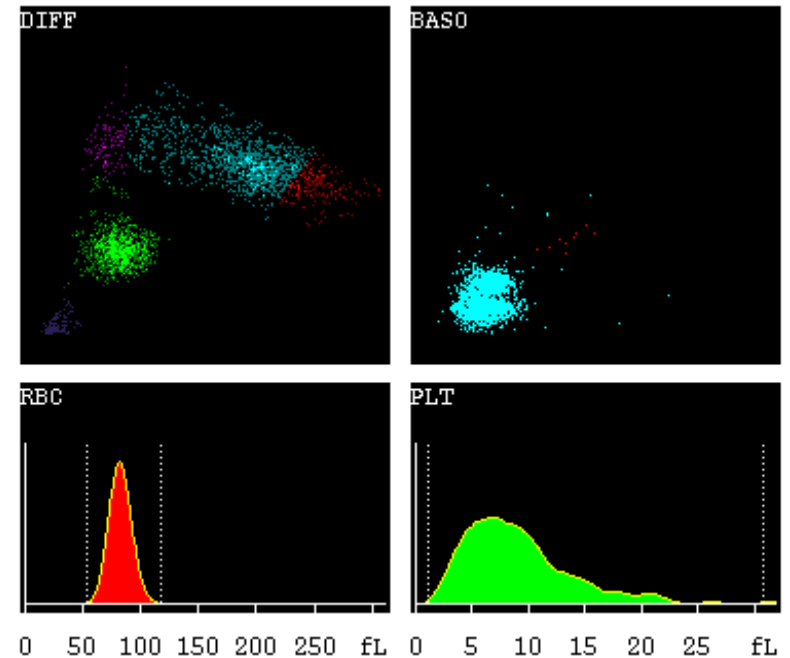
$[\text{Zn}_3(\text{pmdien})_3(\mu\text{-ttc})](\text{ClO}_4)_3$

Blood cells count diluted in phosphate buffer (1:2), Zn 345 $\mu\text{g/mL}$

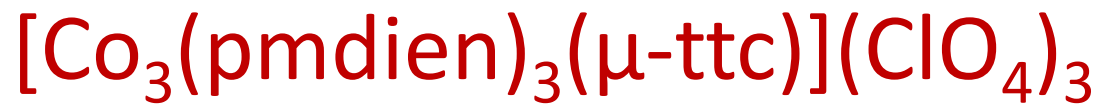


WBC 3.00×10^9 cells/mL
MON 0.17×10^9 cells/mL
EOS 0.13×10^9 cells/mL
RBC 1.85×10^{12} cells/mL
PLT 57×10^9 cells/mL

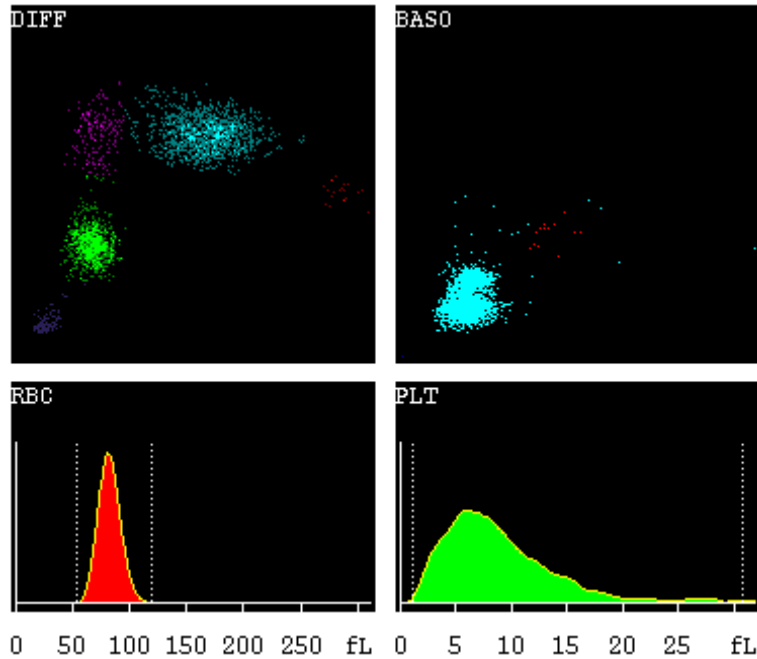
Blood cells count diluted in phosphate buffer (1:2), Zn 345 $\mu\text{g/mL}$ **after 1 hour**



WBC 2.99×10^9 cells/mL
MON 0.16×10^9 cells/mL
EOS 0.43×10^9 cells/mL
RBC 1.72×10^{12} cells/mL
PLT 64×10^9 cells/mL

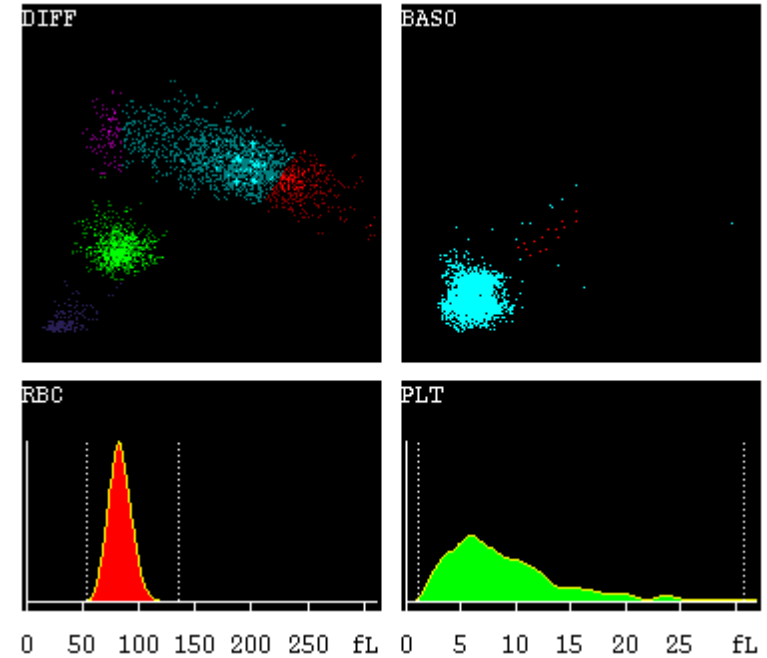


Blood cells count diluted in phosphate buffer (1:2), Co 345 μg/mL



WBC 2.97×10^9 cells/mL
 MON 0.18×10^9 cells/mL
 EOS 0.11×10^9 cells/mL
 RBC 1.86×10^{12} cells/mL
 PLT 59×10^9 cells/mL

Blood cells count diluted in phosphate buffer (1:2), Co 345 μg/mL **after 1 hour**

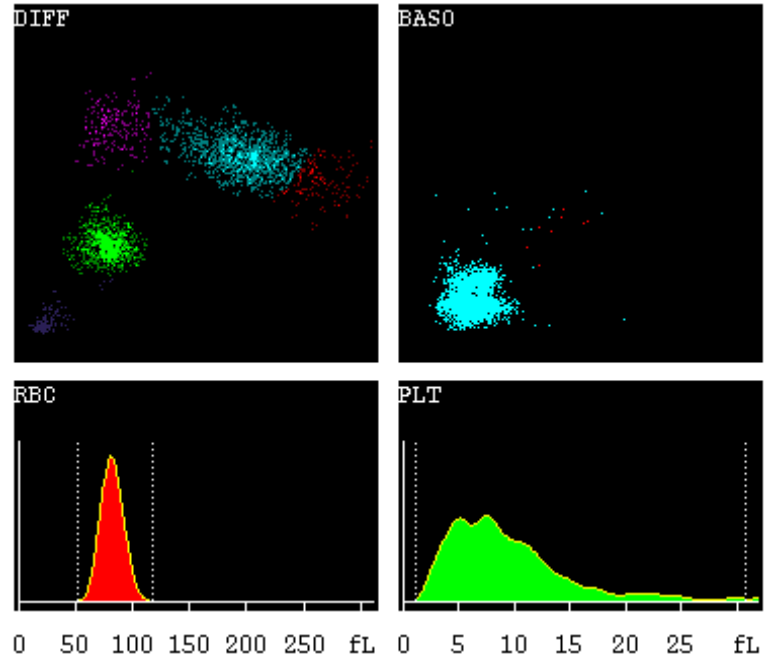
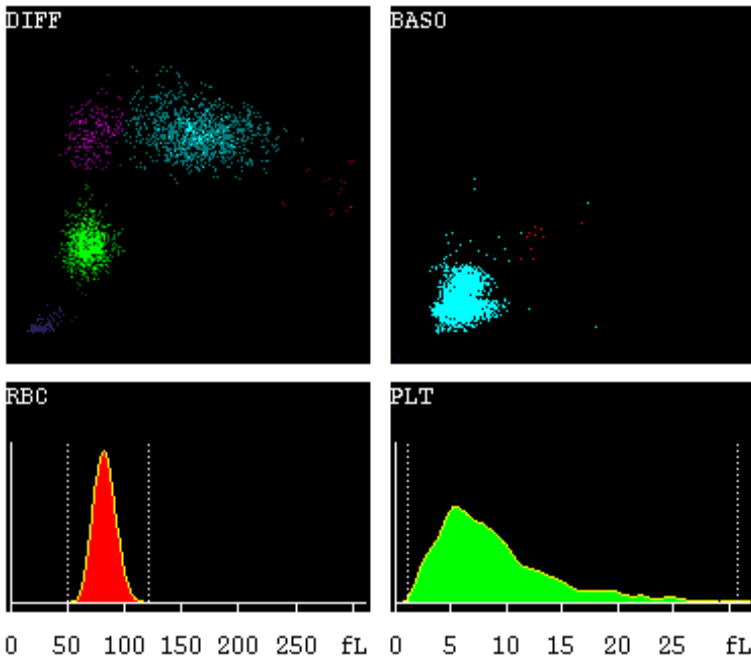


WBC 3.66×10^9 cells/mL
 MON 0.12×10^9 cells/mL
 EOS 0.54×10^9 cells/mL
 RBC 1.41×10^{12} cells/mL
 PLT 42×10^9 cells/mL

PdNPs (PdCl₂)

Blood cells count diluted in phosphate buffer (1:2), Pd 345 µg/mL

Blood cells count diluted in phosphate buffer (1:2), Pd 345 µg/mL **after 1 hour**



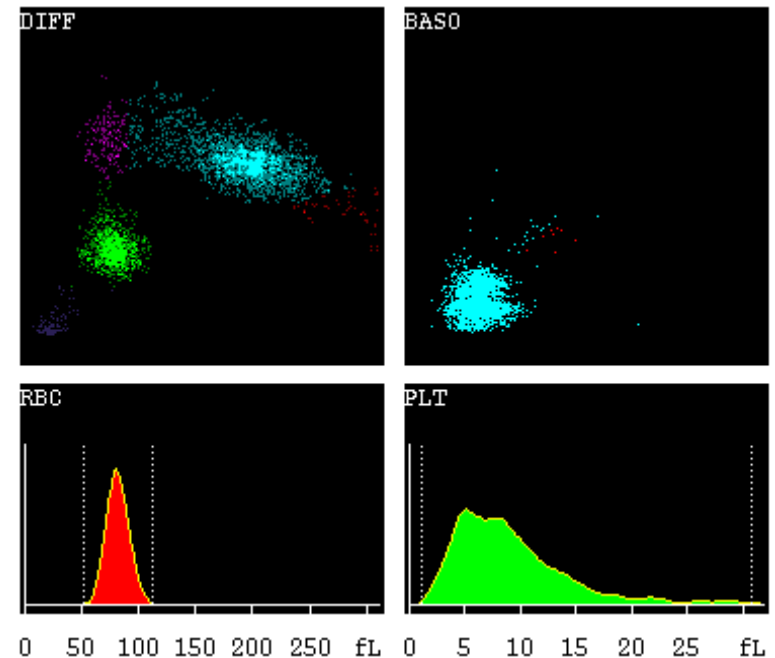
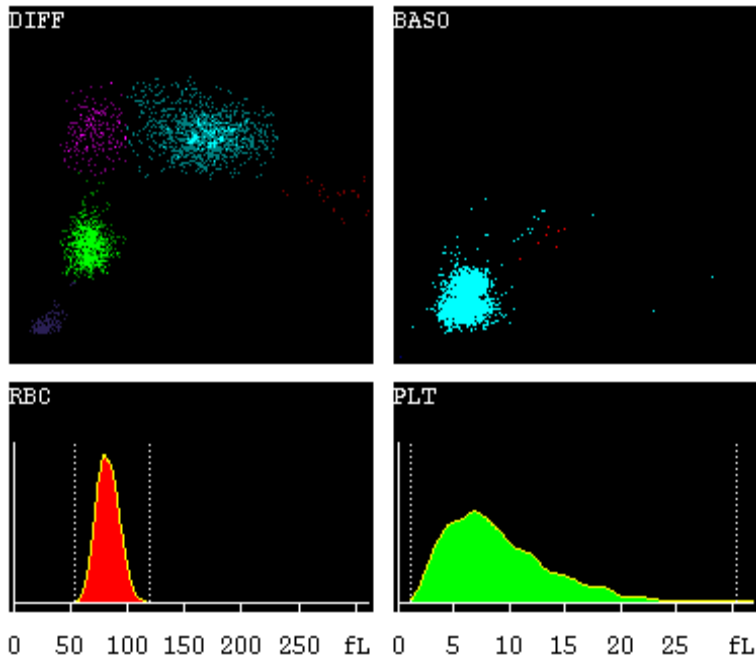
WBC 2.95×10^9 cells/mL
MON 0.21×10^9 cells/mL
EOS 0.11×10^9 cells/mL
RBC 1.87×10^{12} cells/mL
PLT 58×10^9 cells/mL

WBC 3.10×10^9 cells/mL
MON 0.24×10^9 cells/mL
EOS 0.26×10^9 cells/mL
RBC 1.81×10^{12} cells/mL
PLT 58×10^9 cells/mL

RuNPs ($\text{RuCl}_3 \cdot 2.5 \text{H}_2\text{O}$)

Blood cells count diluted in phosphate buffer (1:2), Ru 345 $\mu\text{g}/\text{mL}$

Blood cells count diluted in phosphate buffer (1:2), Ru 345 $\mu\text{g}/\text{mL}$ **after 1 hour**



WBC 3.08×10^9 cells/mL
MON 0.23×10^9 cells/mL
EOS 0.12×10^9 cells/mL
RBC 1.85×10^{12} cells/mL
PLT 60×10^9 cells/mL

WBC 3.35×10^9 cells/mL
MON 0.16×10^9 cells/mL
EOS 0.15×10^9 cells/mL
RBC 1.71×10^{12} cells/mL
PLT 60×10^9 cells/mL

Conclusions

- 3 metal complexes (Cu, Zn and Co) and 2 nanoparticles (Pd, Rd) were analysed for their effect on the blood cell count
- The biggest effect had metal complexes and nanoparticles on the number of eosinophils and monocytes
- The number of monocytes by metal complexes increased 3 times in comparison to control, the highest effect had Co complex

Acknowledgment

Grant agency

Financial support from the project

**NANOLABSYS CZ 1.07/2.3.00/20.0148 is highly
acknowledged**



Acknowledgment

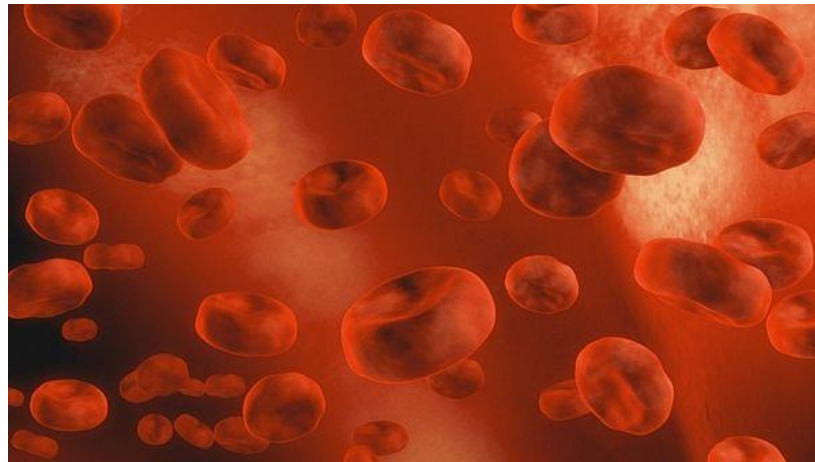
Prof. Ing. Rene Kizek, Ph.D.

Doc. RNDr. Pavel Kopel, Ph.D.

Ing. Jiri Kudr

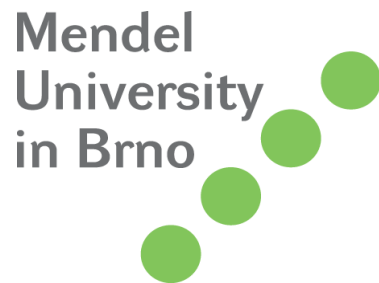
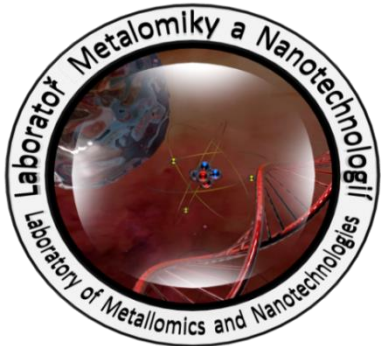
Ing. Lukas Nejd

Martina Stankova



Thank you for your attention





Thank you for your attention



evropský
sociální
fond v ČR



MINISTERSTVO ŠKOLSTVÍ,
MLÁDEŽE A TĚLOVÝCHOVY



OP Vzdělávání
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INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ