

# **Hyaluronic Acid-Based Nanocapsules as Efficient Delivery Systems of Garlic Oil Active Components with Anticancer Activity**

**Małgorzata Janik-Hazuka <sup>1</sup>, Kamil Kamiński <sup>1</sup>, Marta Kaczor-Kamińska <sup>2</sup>, Joanna Szafraniec-Szcęsny <sup>3</sup>, Aleksandra Kmak <sup>1</sup>, Hassan Kassassir <sup>4,5</sup>, Cezary Watała <sup>4</sup>, Maria Wróbel <sup>2</sup> and Szczepan Zapotoczny <sup>1,\*</sup>**

<sup>1</sup> Jagiellonian University, Faculty of Chemistry, Gronostajowa 2, 30-387 Krakow, Poland;

mal.janik@uj.edu.pl (M.J.-H.); kaminski@chemia.uj.edu.pl (K.K.); ola.kmak@student.uj.edu.pl (A.K.)

<sup>2</sup> Jagiellonian University Medical College, Faculty of Medicine, Chair of Medical Biochemistry, Kopernika 7, 31-034 Krakow, Poland; marta.b.kaczor@uj.edu.pl (M.K.-K.); mtk.wrobel@uj.edu.pl (M.W.)

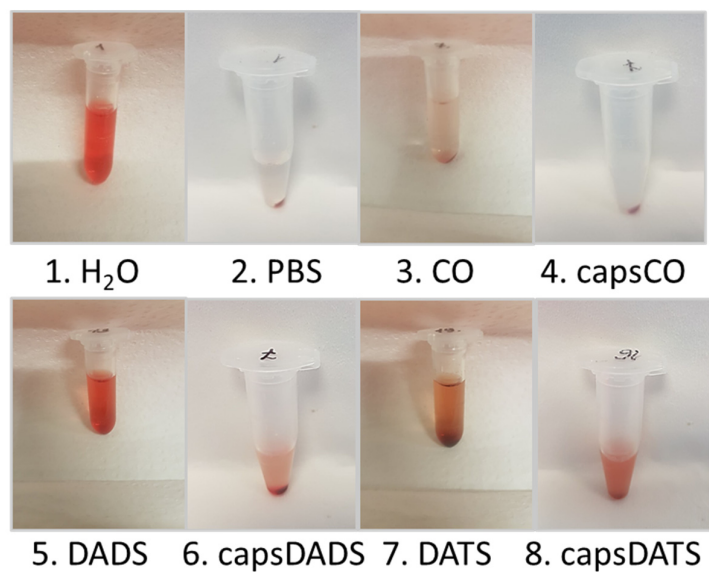
<sup>3</sup> Jagiellonian University Medical College, Faculty of Pharmacy, Department of Pharmaceutical Technology and Biopharmaceutics, Medyczna 9, 30-688 Krakow, Poland; joanna.szafraniec@uj.edu.pl (J.S.-S.)

<sup>4</sup> Medical University of Lodz, Department of Haemostasis and Haemostatic Disorders, Chair of Biomedical Sciences, Mazowiecka 6/8, 92-215 Lodz, Poland; hkassassir@cbm.pan.pl (H.K.); cezary.watala@umed.lodz.pl (C.W.)

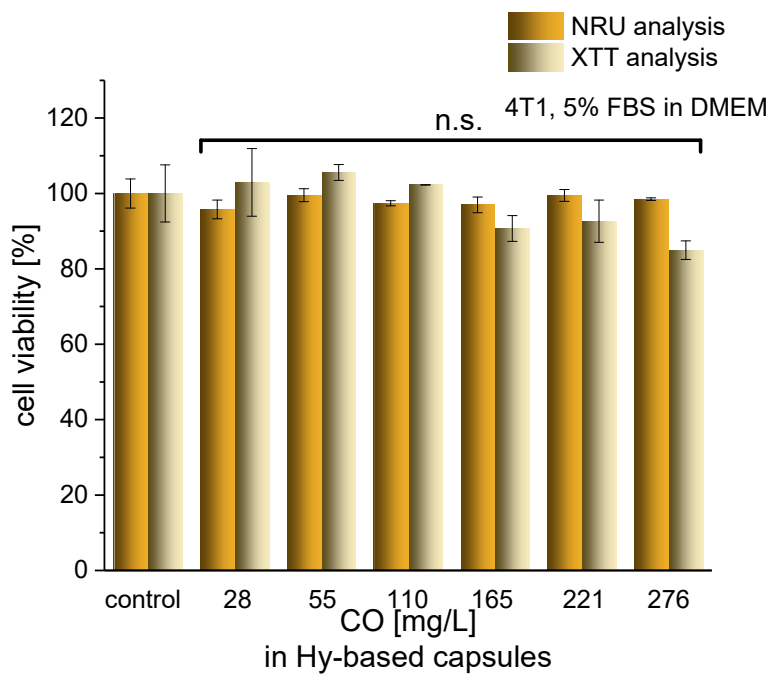
<sup>5</sup> Institute of Medical Biology, Polish Academy of Science, Lodowa 106, 93-232 Lodz, Poland

\* Correspondence: zapotocz@chemia.uj.edu.pl (S.Z.); Tel.: +48-12-686-2530

## **Results**



**Figure S1.** Hemolytic effects of tested emulsions: capsCO, capsDADS, capsDATS and free oils: the pictures of representative samples after the incubation with red blood cells (RBCs) for 5 min at 37 °C and the centrifugation at 5000 rpm for 5 min.



**Figure S2.** Cell viability (analyzed by NRU and XTT assays) after the treatment by Hy-based capsules with CO cores.

The data shown as mean  $\pm$  SD, (n=3) represent the amount of encapsulated and delivered compounds. The aqueous and oil phase were mixed in 1000:3 volume ratio. Significance estimated with the Mann-Whitney U-test; n.s., not significant.

**Table S1.** Averaged diameters of studied samples: capsules incubated in PBS and serum.

sample	size [nm]*	dispersity index (PdI)**
<b>capsCO-PBS</b>	520±25	0.243
<b>capsCO-serum</b>	48±11***	0.696***
<b>capsDADS-PBS</b>	468±19	0.358
<b>capsDADS-serum</b>	11±2***	0.582***
<b>capsDATS-PBS</b>	464±6	0.376
<b>capsDATS-serum</b>	12±2***	0.643***

Values represent the arithmetic mean  $\pm$  standard deviation for 9 donors, with each determination consisting of 8 (incubated with PBS) and 24 (incubated with serum) assays. The incubation time was 1 h.

\*Statistical significance, estimated with one-tailed Mann–Whitney U test or with unpaired Student's t-test (depending on the normality of data distribution), was: capsCO + PBS > capsCO + serum,  $P_{1,\alpha} < 0.0001$ ; capsDADS + PBS > capsDADS+ serum,  $P_{1,\alpha} < 0.0001$ ; capsDATS + PBS > capsDATS+ serum,  $P_{1,\alpha} < 0.0001$ ;

\*\* Statistical significance, estimated with one-tailed Mann–Whitney U test or with unpaired Student's t-test (depending on the normality of data distribution), was: capsCO + PBS < capsCO + serum,  $P_{1,\alpha} < 0.0001$ ; capsDADS + PBS < capsDADS+ serum,  $P_{1,\alpha} < 0.05$ ; capsDATS + PBS < capsDATS+ serum,  $P_{1,\alpha} < 0.01$ ;

\*\*\*change of average value due to multimodal distribution (see Figure 4 in the manuscript).