*Supplementary Materials and Methods*

All animals received humane care in accordance with the policies formulated by the institutional Animal Care Committee (University Health Network, Toronto: protocols AUP 2021 for rabbits, AUP 2150 for nude mice), the Animal for Research Act of the Province of Ontario and the Canadian Council on Animal Care. All animal studies were approved by the UHN Animal Care Committee.

*Accumulation of porphysomes in mouse orthotopic A549 lung tumor at 24 h post-injection*

After confirming the growth of A549 tumor in mice lung by micro-CT scanning, 20 mg/kg of porphysomes (total lipid) was administrated via the tail vain in mice. At 24 h post-injection, the animals were euthanized by sodium pentobarbital (Bimeda-MTC Animal Health Inc. Cambridge, ON, Canada) and organs of interest were dissected for *ex vivo* analysis: whole lung with tumors, esophagus, heart, kidney, spleen, small intestine, large intestine and hind limb muscle. Animals without porphysome injection were used as controls. Porphysome accumulation in lung and lung tumor was first visualized by *ex vivo* fluorescence imaging system (Maestro EX 2.10, Cambridge Research & Instrumentation, Inc., Waltham, MA, USA) with 575-605 nm excitation and >645 nm detection using 500 ms integration time. To further compare the porphysome accumulation in tumor and normal lung, tissues were cut to four sections as shown in Supplementary Figure S1A which were frozen in OCT gel using liquid nitrogen, sectioned (5 µm) and stained with nuclei-staining DAPI. The frozen slides were imaged by confocal microscopy using a 60X oil-immersion lens (633 nm excitation for porphyrin and 408 nm excitation for DAPI). In addition, for the biodistribution analysis, organs of interest were freshly weighed and homogenized in PBS, the porphyrin fluorescence in the supernatant was measured by spectrofluorimetry and the percent of injected dose per gram of tissue was calculated based on a standard porphyrin-lipid concentration curve. N=5 was used for the mouse study.

*Biodistribution of porphysomes in mice orthotopic tumor models at 48 h post-injection*

48 h after mice bearing orthotopic lung tumors (A549, H520 and H460) received iv injection of porphysomes, animals were euthanized by sodium pentobarbital (Bimeda-MTC Animal Health Inc. Cambridge, ON, Canada) and organ of interest were dissected (whole lung with tumors, esophagus, heart, kidney, spleen, small intestine, large intestine and hind limb muscle). Tissues were freshly weighed and homogenized in PBS, the porphyrin fluorescence in the supernatant was measured by spectrofluorimetry and the percent of injected dose per gram of tissue was calculated based on a standard porphrin-lipid concentration curve. N=5 was used for each animal group.

*Evaluation of intactness of porphysomes in phantom*

Since the fluorescence of intact porphysomes is highly quenched, we measured the fluorescence in phantoms containing porphysomes using a fluorescence imager (Maestro®: 575-605 nm excitation, >645 nm detection, 500 ms integration) to test whether the phantom formulation would affect the intactness of the porphysome nanostructure. Three porphysome concentrations in phantom were tested: 0 nM (control), 29 nM (=0.076% ID/g in the rabbit lung cancer model) and 58 nM (=0.16% ID/g in the rabbit lung cancer model); and two different phantoms were compared: with and without 1% Triton-100. Phantom with 1% Triton-100 was the control, in which porphysome nanostructure was completely disrupted and the fluorescence was fully unquenched and activated.

*Transpleural ablation of rabbit orthotopic VX2 tumor using straight-cut fiber (500mW)*

Since straight-cut fiber at 250 mW output ablated the tumor area with 3 mm diameter, the PTT efficiency of straight-cut fiber at higher output (500 mW) was evaluated using rabbit bearing orthotopic VX2 lung tumor. A transpleural approach after thoracotomy was used instead of the transbronchial approach because the optical fiber could be inserted into the tumor more accurately under direct palpation compared to brochoscopic laser fiber insertion (Note: that this is not possible in patients). At 48 h post-injection of porphysome, the rabbit lung containing the orthotopic VX2 tumor was exposed by surgery, and the lung was inflated and manually ventilated continuously. The tumor was stabilized by palpation and then an 18G needle was inserted through it to create a channel for the laser fiber. The straight-cut fiber (500 mW output) was placed into the center of the tumor and irradiated for 10 min. The lung was inflated and manually ventilated during the irradiation, mimicking the conditions of transbronchial ablation. Ablation of normal peripheral lung was performed in the same way. Rabbits were sacrificed by IV injection of sodium pentobarbital and the entire lung with the treated tumor was removed, fixed in Tissue-Tek OCT and stored at -80oC for frozen tissue sectioning to evaluate both morphological changes (H&E staining) and cell viability using NADH enzyme-activity staining. The size of ablated area was calculated based on the NADH staining images using the following formula:

 $V=\frac{4}{3}π\left(long radius\right)(short radius)^{2}$ (45)

The long and short radii were measured from the non NADH-stained area. When the ablation spread beyond the tumor boundary into adjacent lung, the radii were calculated by including only the tumor. Since the ablated volume by the diffuser type fiber should be proportional to the fiber length in the target tumor, the above formula is not exact but allows a straight-forward comparison of the results for the different conditions.