



سپهر بهار

سپهر بهار





In the name of God

Department of Bacteriology and Virology
School of Medicine.

Ph.D. Seminar

Title:

Bacterial-mediated cancer therapy (BMCT)

Presented by:

Abbasali Delarampour

Supervisor:

Dr. A. Bazargani



CONTENT

Introduction

Bacterial-mediated Cancer Therapy (BMCT)

Bacterial Based Anti-cancer Mechanisms

Review of The Studies

Advantage and Disadvantages

Conclusion

References



Introduction

Cancer

- **Cancer** is a deadly disease by the dysregulation of the cell cycle leading to uncontrolled cell division.
- **Cancer** one of the causes of mortality among the worldwide human population(**the second**).
- There are >200 different types of cancer, and each is classified by the type of the associated tissue.
- The World Health Organization (WHO), was estimated that at least 9 million patients are killed by cancer annually.

Most reported cases of cancer in men and women

Doi.org/10.3322/caac.21660

Men: All sites 854,790 (100%)

- Prostate (28%)
- Lung & bronchus (14%)
- Colon & rectum (9%)
- Urinary bladder (6%)
- Melanoma of the skin (5%)
- Kidney & renal pelvis (5%)
- Non-Hodgkin lymphoma (4%)
- Oral cavity & pharynx (3%)
- Leukemia (3%)
- Pancreas (3%)



Women: All sites 805,500 (100%)

- Female Breast (29%)
- Lung & bronchus (14%)
- Colon & rectum (9%)
- Uterine corpus (6%)
- Thyroid (6%)
- Non-Hodgkin lymphoma (4%)
- Melanoma of the skin (4%)
- Kidney & renal pelvis (3%)
- Pancreas (3%)
- Ovary (3%)



molecule



Conventional therapies for cancer

Doi. 10.1177/20503121211034366



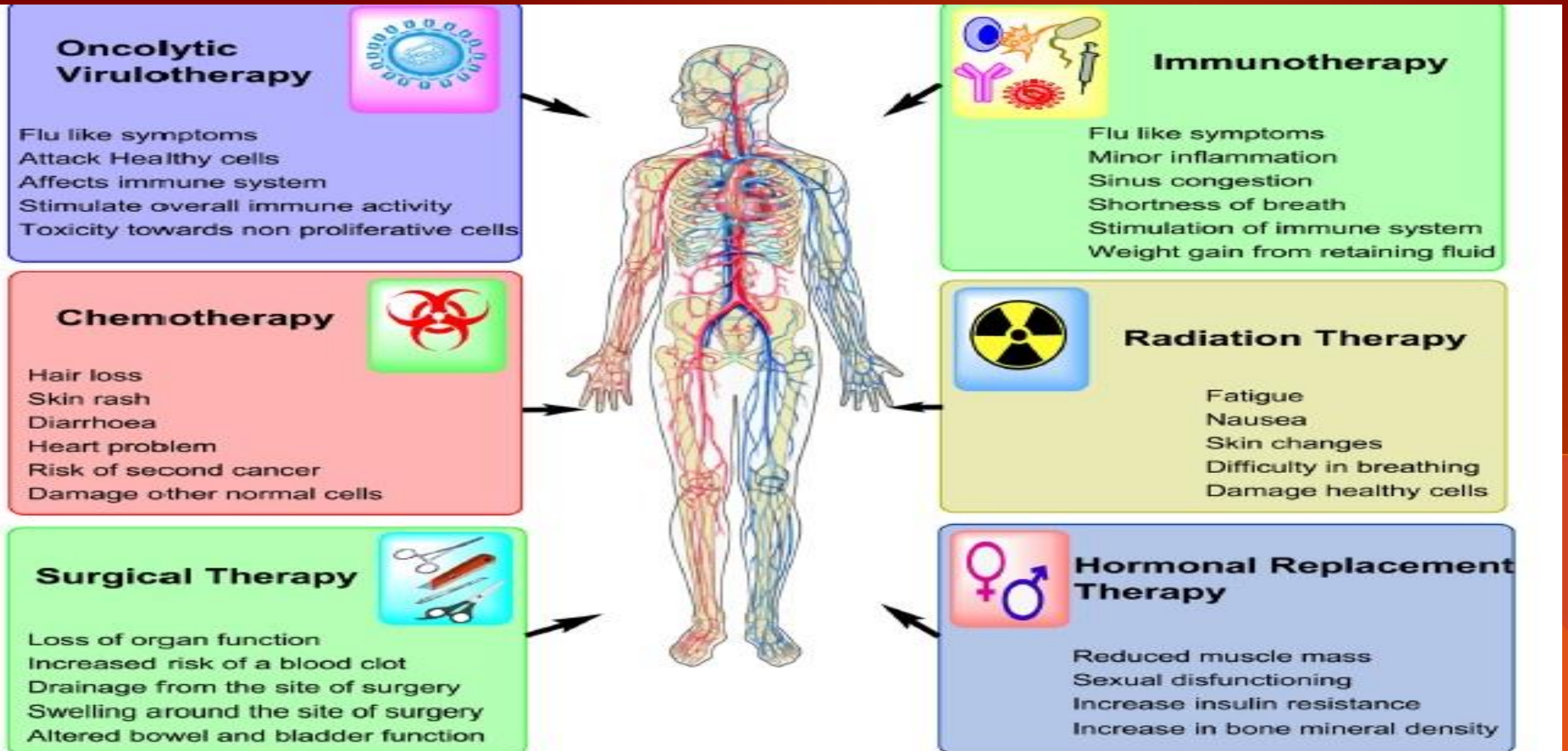
Treatment



- **Treatment** that a patient receives will depend on **the type of cancer** and how **advanced** it is.
- **Treatment** can be monotherapy or combination therapy and depends on **cancer origin, stage, location, and grade.**
- **Tumor surgery**  great way for tumor clearance and needs to be followed up with chemotherapy and radiotherapy.
- **Surgery and radiation therapy** are more effective against **local cancers.**
- **Chemotherapy**  is the only treatment option For **diffuse and malignant cancerous cells.**

Side effects associated with current cancer therapies

Doi.org/10.1016/j.semcancer.

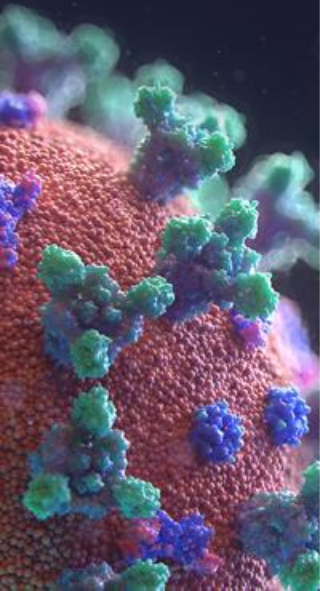




Bacterial-mediated cancer therapy (BMCT)

Bacterial-mediated cancer therapy (BMCT)

- Bacteria-mediated cancer therapy (BMCT) is a promising therapeutic strategy due to its unique properties.
- BMCT has unique properties including, **broad tumor targeting, delivery drug , facilitating the host's immune responses and tumor penetration capability.**
- *Streptococci* and *Clostridia* were the first bacteria used as live anticancer agents.
- *Salmonella, Mycobacterium, Bifidobacterium, Lactobacillus, Escherichia, Pseudomonas, Listeria, and Proteus.*



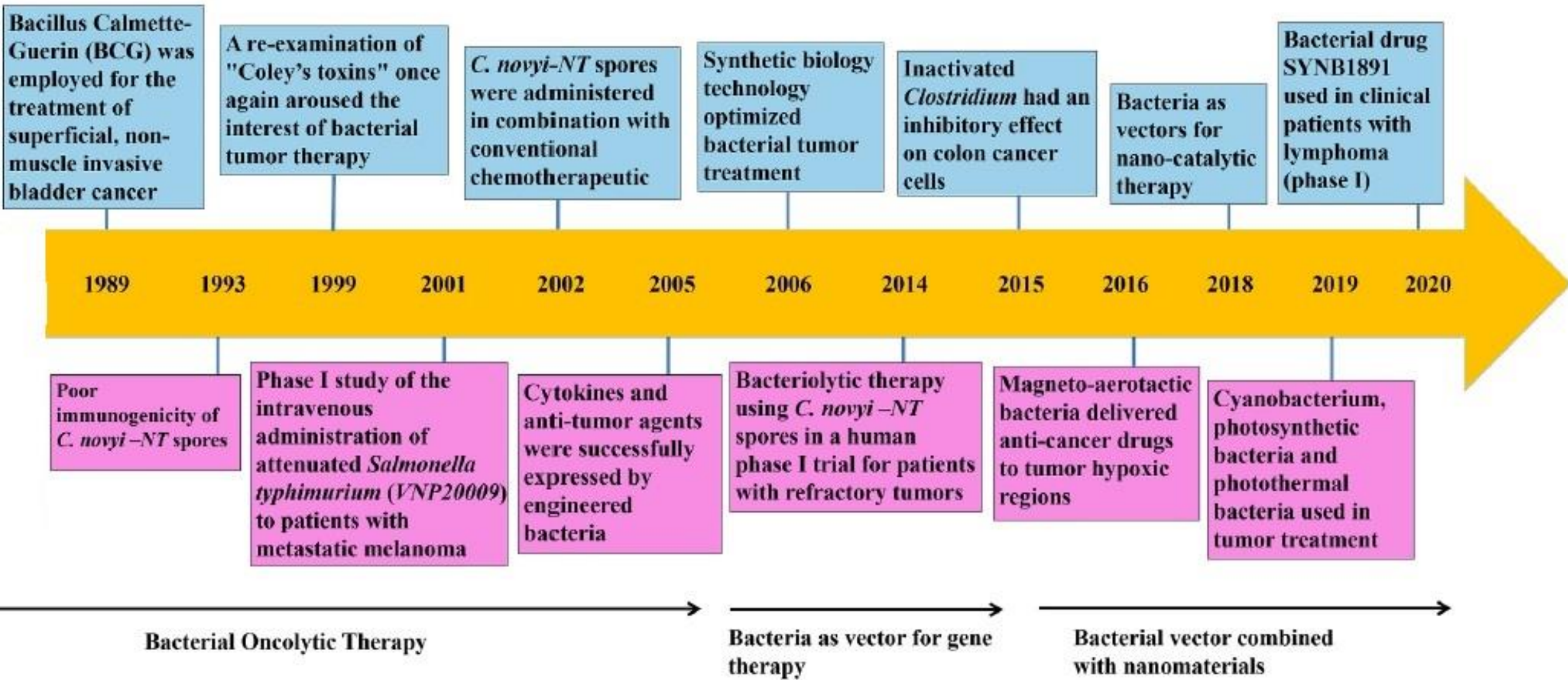


molecule

- Bacteria can be **engineered** to delivery of specific and diverse therapeutic drug into Tumor microenvironment (TME).
- Their **genetics can be modified** to produce and release specific compounds, which leads to tumor regression.

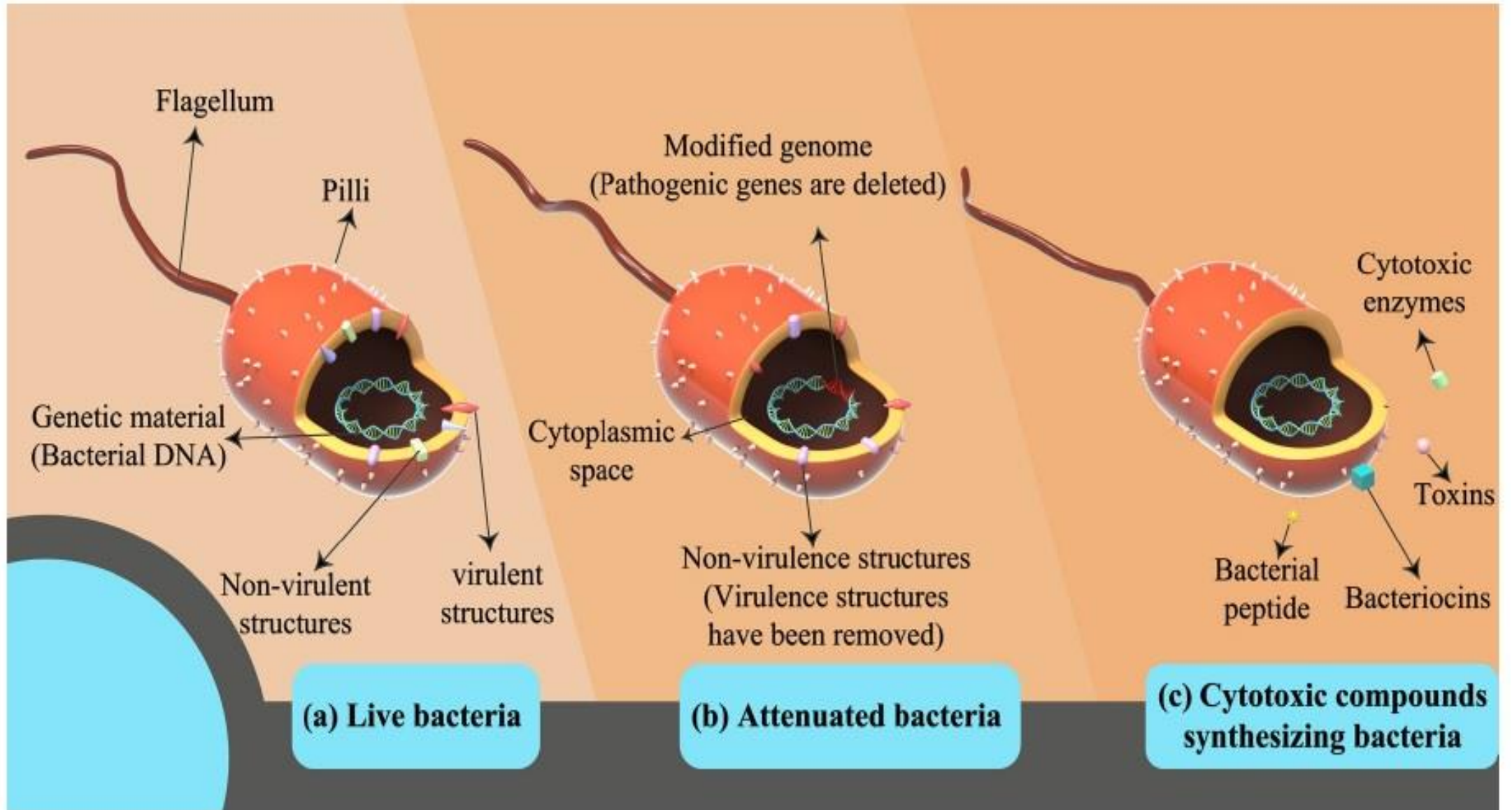


- **In 1813**, the observation of spontaneous tumor regression from concurrent *clostridial* infection.
- **In 1868**, The patients with cancer to be purposefully infected with bacteria and result cured.
- **In 1890**, inoperable tumors regression subsequent to being inoculated with inactivated *Serratia marcescens* and *Streptococcus pyogenes* (Coley's toxin).
- **In 1935**, tumor regression during therapy using filtrates from *Clostridium histolyticum*.



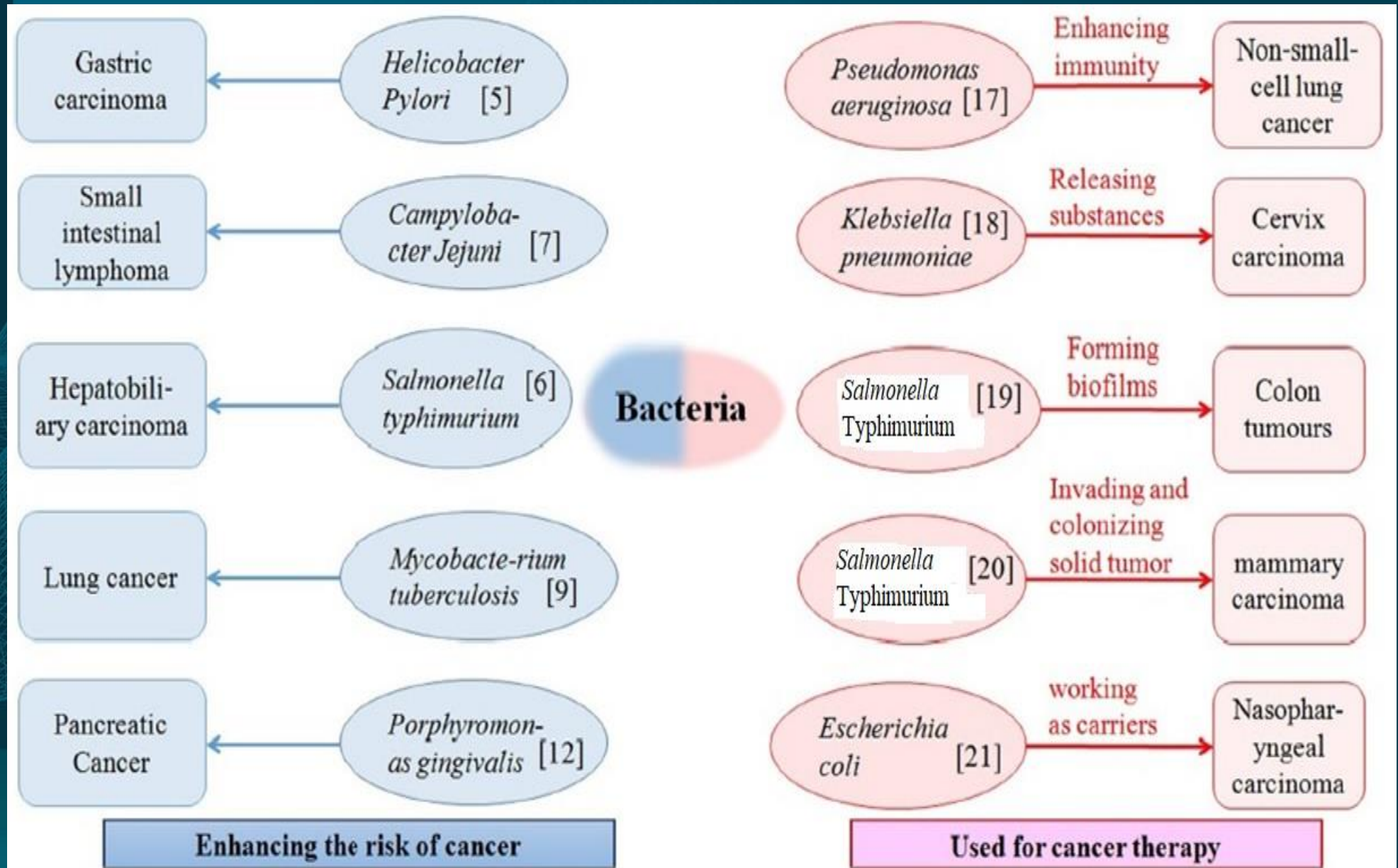
The used bacteria for cancer therapy

Doi.org/10.1007/s40487-021-00177-x



Bacteria strains and their dual roles

Doi.org/10.1186/s13027-018-0180-y



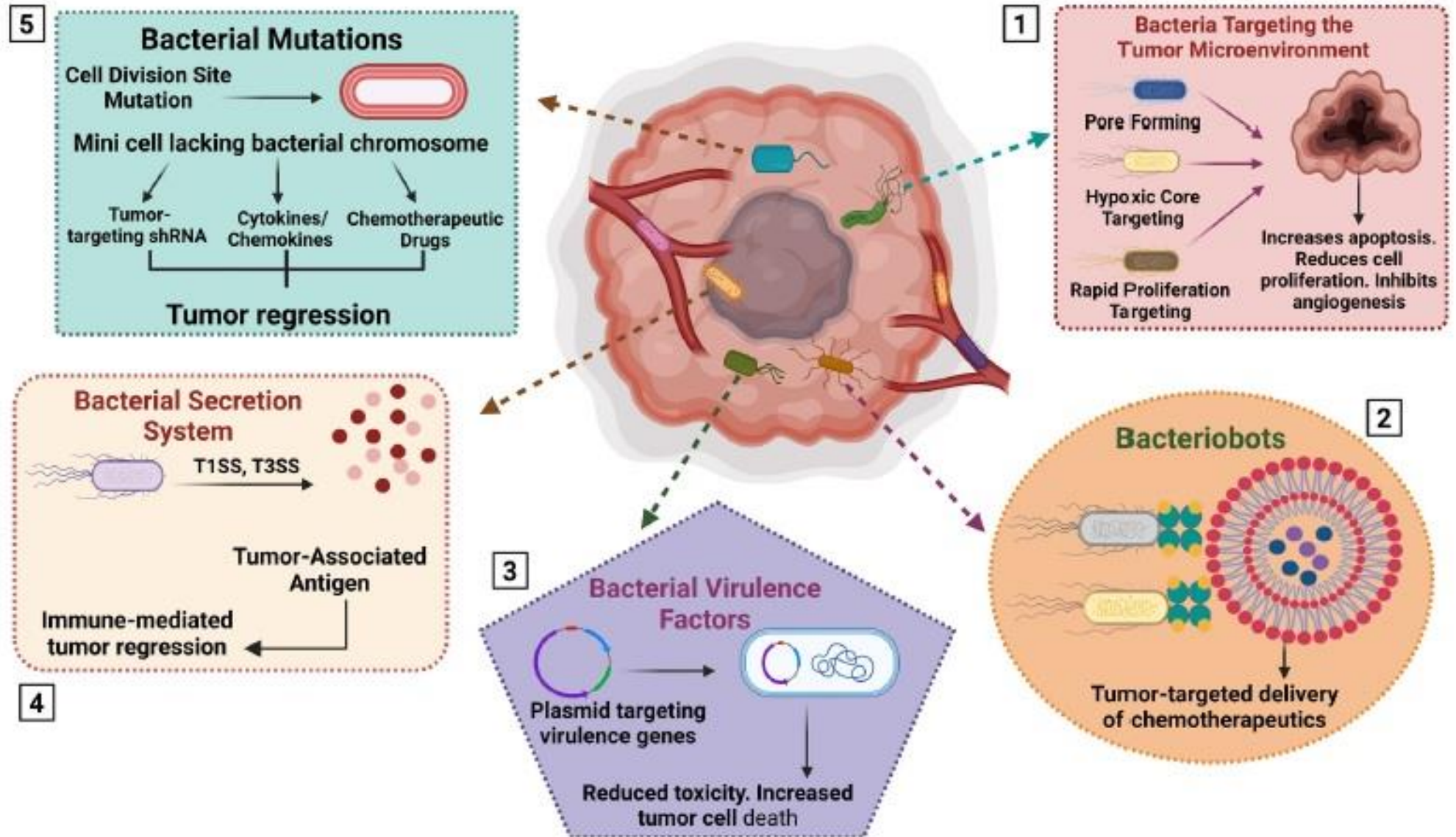


Bacterial Based Anti-cancer Mechanisms



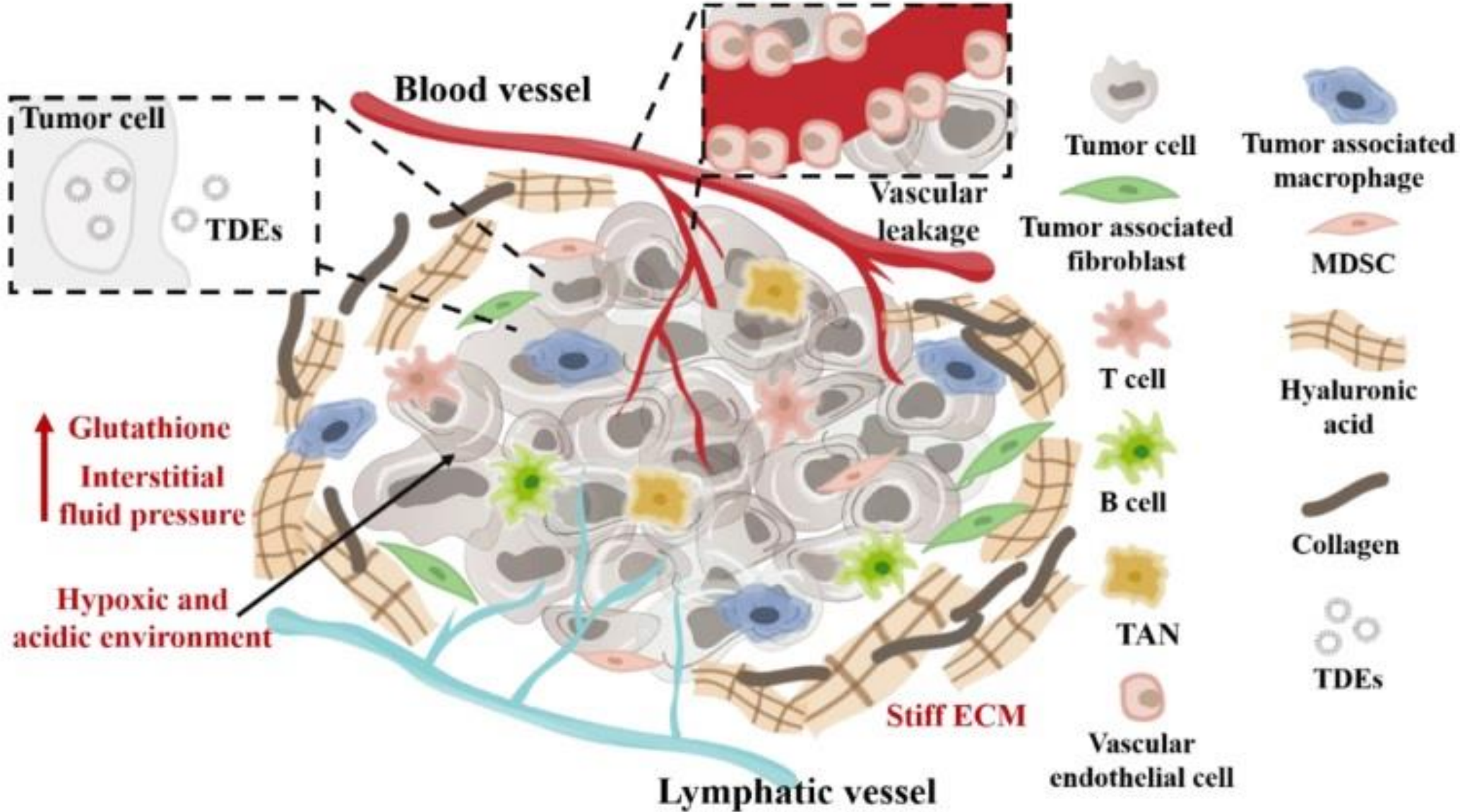
Bacterial Based Anti-cancer Mechanisms

Doi.org/10.1016/j.critrevonc.



Tumor-microenvironment(TME)

- The TME is a complex and diversely networked structure that affects the delivery of anticancer drugs.
- These characteristics include an acidic condition, overexpression of glutathione, hypoxic condition, immunosuppression and poor drug transportation.
- The TME include complex extracellular matrix (ECM), tumor-associated stromal cells and immune cells, endothelial cells, inflammatory cells, and lymphocytes.



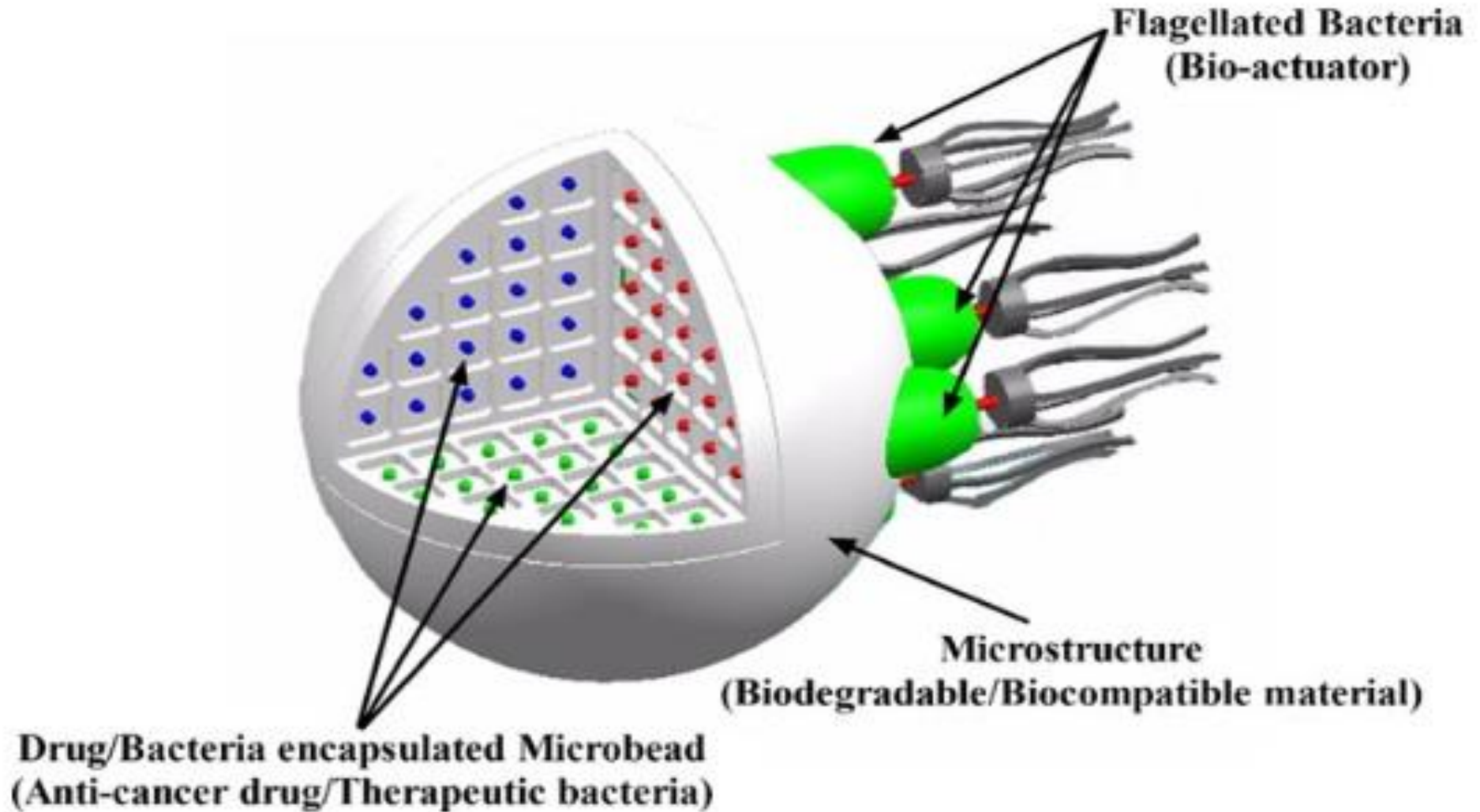
Bacterial Targeting of the TME

- **Anaerobic bacteria** specifically target the hypoxic environment of tumors initiating an inflammatory reaction resulting in tumor destruction.
- Oxygen concentrations in the TME are typically ≤ 10 mmHg, indicating a hypoxic condition.
- The metabolism of anaerobic bacteria in the hypoxic TME leads to lactic acid production, resulting in increased acidity.
- Hypoxia is a distinctive feature of rapidly growing solid tumors, leading to insufficient oxygen levels in the TME.

- Hypoxic conditions induce genetic changes in tumors result tumors withstand the effects of hypoxia-induced cell death and tissue necrosis.
- Bacteria adapt their growth and survival strategies within tumors, utilizing their **motility** and **diverse oxygen requirements** to thrive in this unique environment.
- *Salmonella spp* and *Clostridia spp* as carriers drug and gene delivery in cancer therapy used.

Bacteria-based microrobot(Bacteriobots)

- **Bacteriobots** (Bacteria-based microrobot), represent a novel class of devices that harness the capabilities of bacteria as micro-actuators and micro-sensors.
- Enabling the targeted delivery of diverse chemotherapeutics and therapeutic compounds to the deep regions of tumors.
- Adhere to cancer cells within the targeted tumor and to release potent anti-tumor agents, effectively eradicating the tumor.
- *S. marcescens*, *E. coli* and *Salmonella Typhimurium*, have been employed to develop bacteria-based microbots.





Bacterial virulence factors

- Virulence factors empower microbial pathogens to colonize and multiply within their host organisms.
- These factors are pivotal in facilitating immune evasion, immunosuppression, and nutrient acquisition.
- Certain virulence factors can contribute to the anti-tumor effects of bacteria.
- *Salmonella* Typhimurium strain VNP20009, which exhibits high specificity for tumor cells treatment.
- The mutations in genes like *rfaG* and *rfaD* can lead to the generation of truncated LPS within the host.

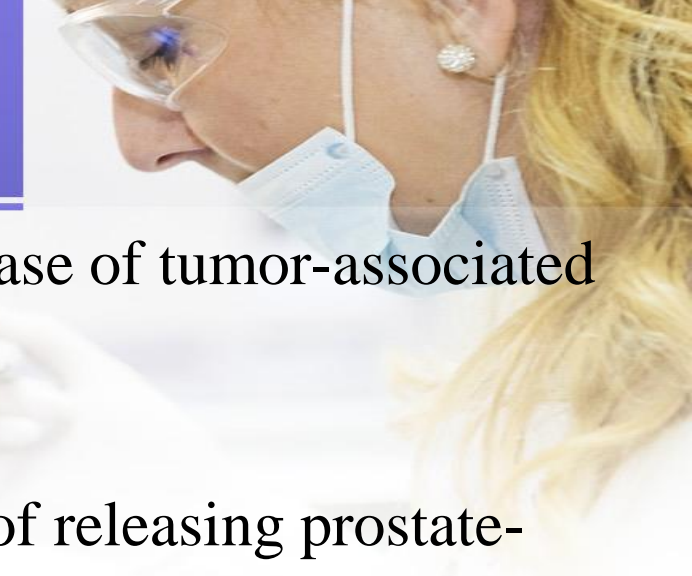


- These truncated LPS molecules exhibit reduced toxicity and contribute to a favorable anti-tumor response.
- Deleting the *Hyl* gene in *Listeria monocytogenes* can impair phagolysosome release and alter its cytotoxicity.
- The mutation of the *inlA* and *inlB* can impair invasion-related characteristics.

Bacterial secretion system



- Bacteria employ secretion systems to transport virulence proteins, which can be leveraged for innovative cancer therapies.
- Drugs can be more effectively and precisely delivered by with signaling molecules involved in bacterial secretion systems.
- The type III secretion system (T3SS) is a frequently exploited mechanism in cancer therapy, result the direct injection of bacterial proteins into the cytoplasm of host cells.
- This targeted delivery enables the manipulation of cellular processes and the therapeutic interventions against cancer.



- *S. Typhimurium* type 1 secretion systems for the expression and release of tumor-associated antigens and tumor-specific antigens in the field of cancer therapy.
- That immunizing mice with an engineered *S. Typhimurium* capable of releasing prostate-specific antigen (PSA) and induced the activation of $CD8^+$, leading to successful suppression of tumor growth.
- By employing genetic engineering techniques, researchers have successfully modified a live strain of *Pseudomonas aeruginosa* to deliver *Yersinia YopE* and *YopH* proteins into mammalian cells through the T3SS.

Bacteria mutations



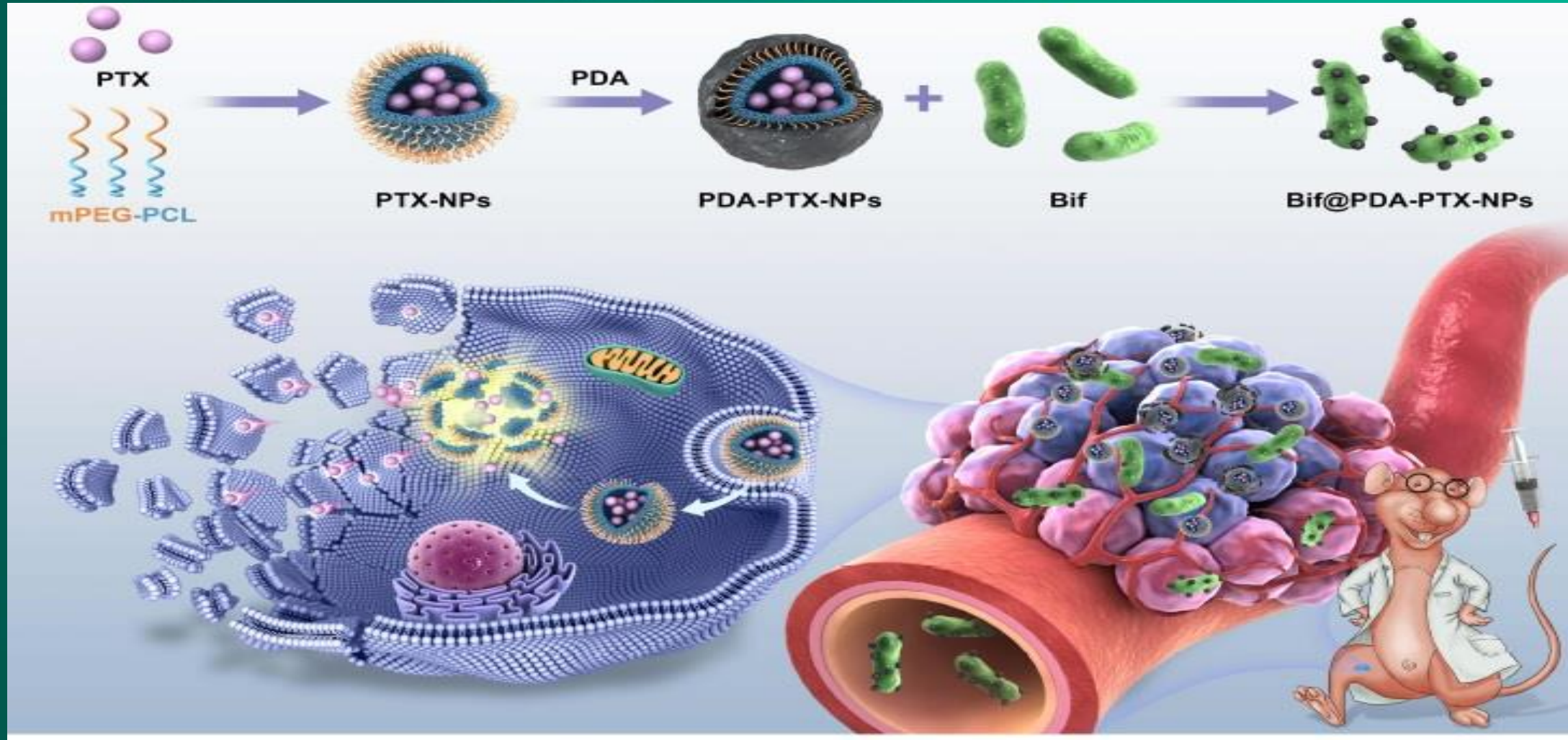
- **Aberrant cell division** in various rod-shaped bacteria, encompassing both Gram-positive and Gram-negative species, has been noted to produce **minicells**.
- **Minicells** that exhibit typical cellular structures such as membranes, ribosomes, RNA, and proteins but do not contain a bacterial chromosome.
- **Minicells** hold significant potential as a novel approach to drug delivery, as they possess essential virulence properties for tumor targeting.
- **Minicells** have demonstrated the ability to transfer genes to mammalian cells both in vitro and in vivo.
- *E.coli* or *S.enterica* can be genetically engineered to contain minicells loaded with chemotherapeutic medications.

A large, light blue pyramid with a slight 3D effect is centered on the slide. A white, rounded rectangular box is overlaid on the middle of the pyramid, containing the title text.

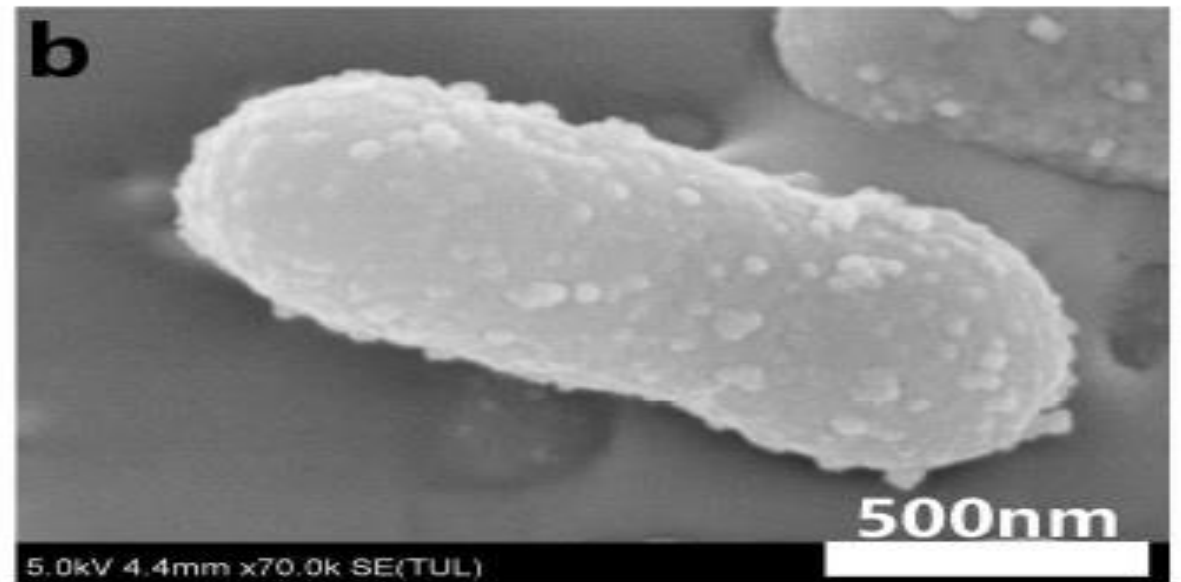
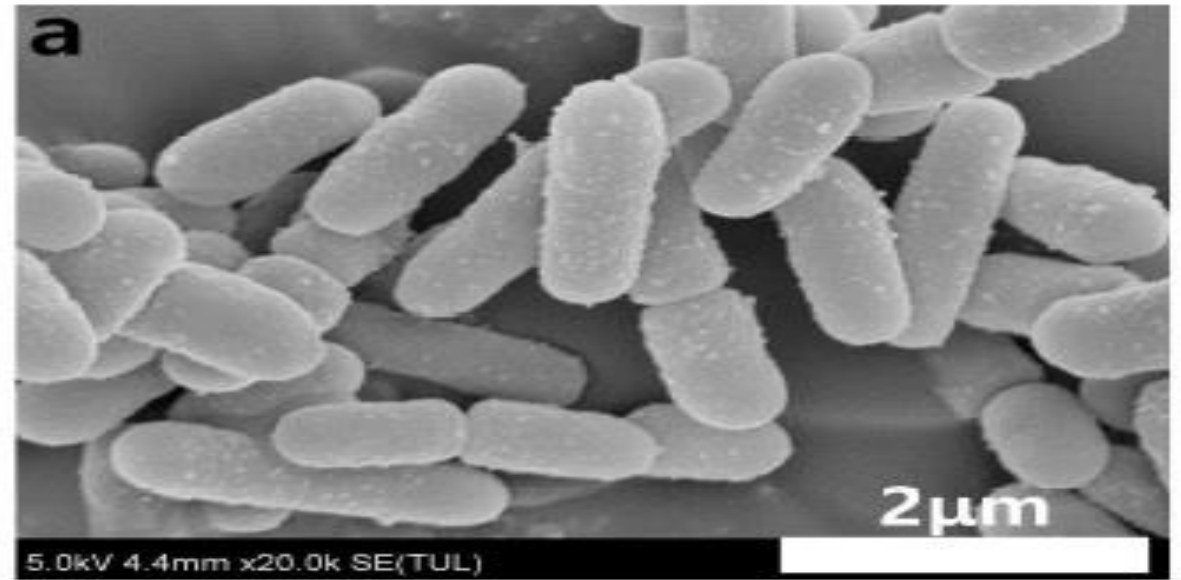
Review of The Studies

Bacteria-Driven Tumor Microenvironment

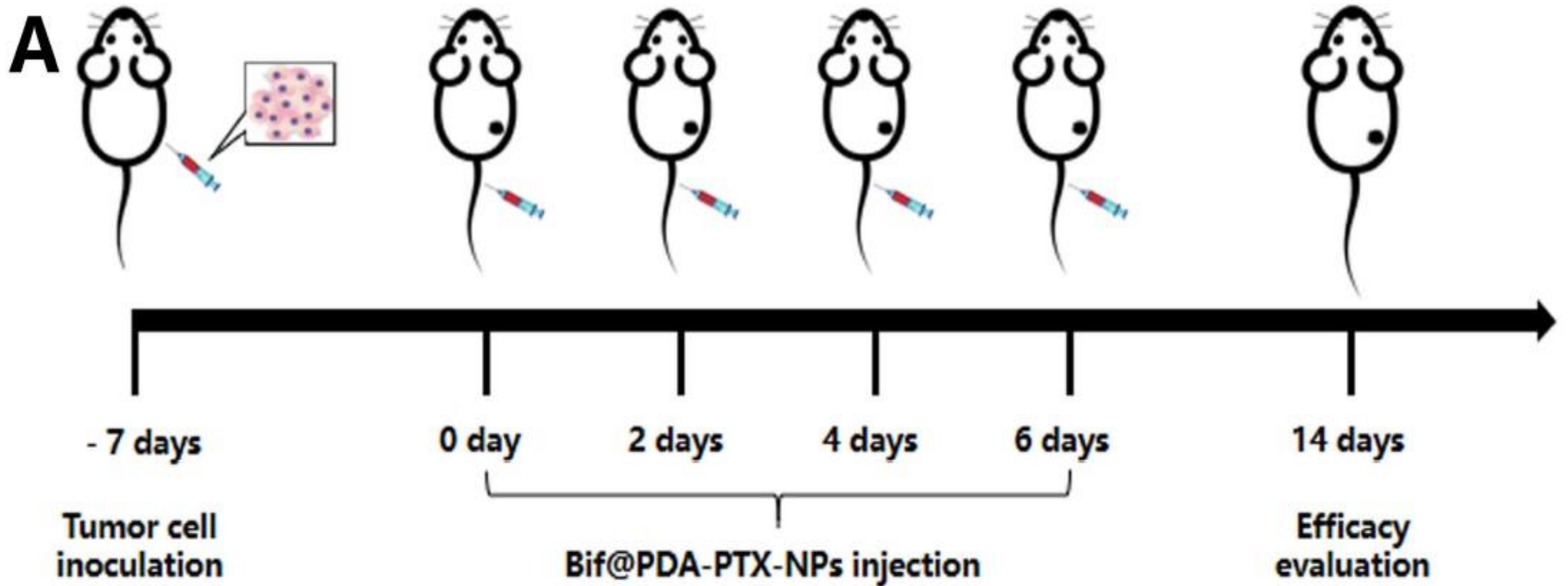
- *Bifidobacterium infantis* (Bif) is a probiotic, anaerobic bacteria that is harmless to humans and does not require any genetic modification.
- Synthesized paclitaxel loaded NPs (PTX-NPs) using Copolymer methoxy poly(ethylene glycol)-poly(ϵ -caprolactone) (mPEG-PCL) as the carrier, and coated the NPs with the biocompatible polydopamine (PDA).
- The PDA-PTX-NPs were incubated with *Bif* cells to generate the *Bif*@PDA-PTX-NPs biohybrids.
- Once these biohybrids reach the hypoxic zone of tumor tissues, the PTX-NPs are released in response to the reductive tumor environment and taken up by the tumor cells.



- SEM images of *Bif*@PDA-PTX-NPs.



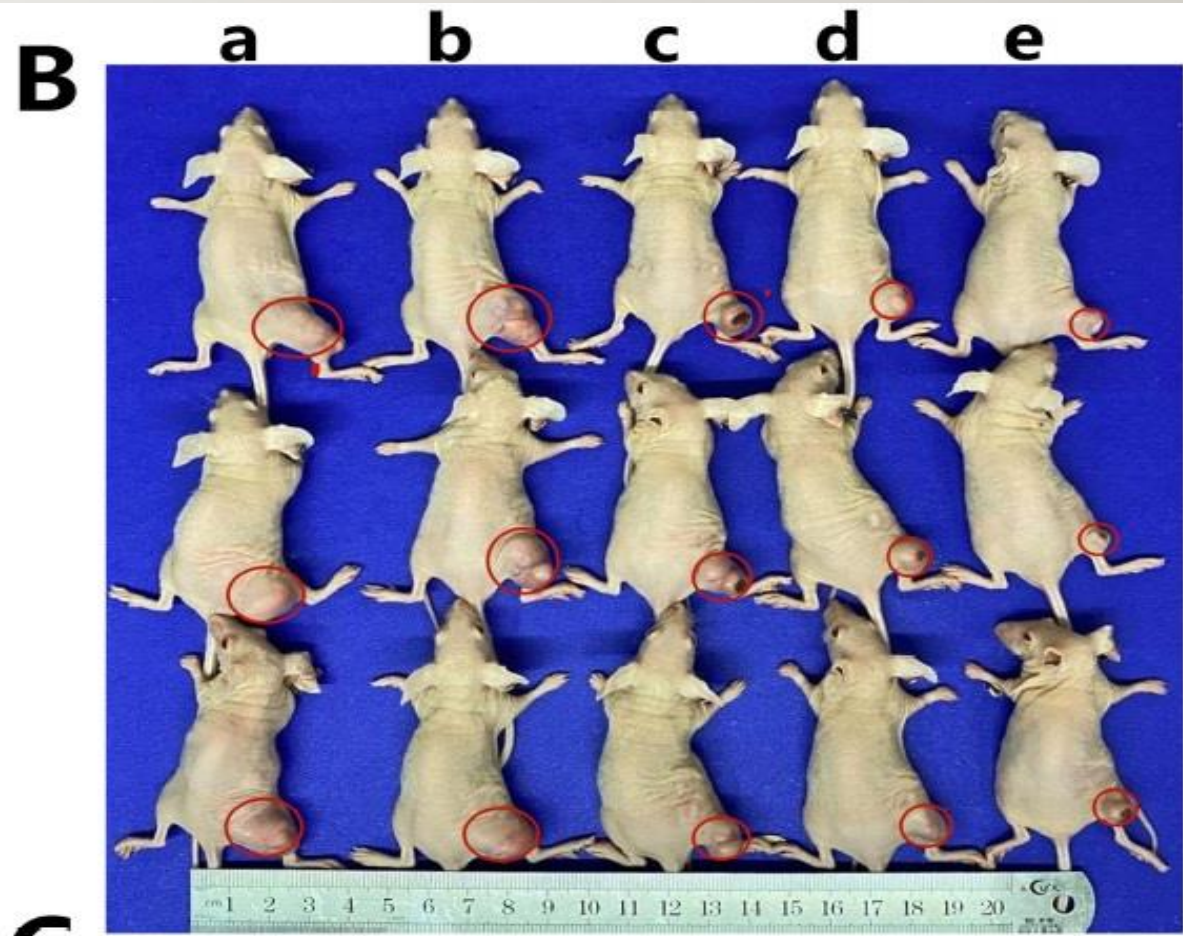
A549 tumor-bearing mice were used to evaluate in vivo anti-tumor effects.



a) NS(Control)-b) *Bif*@PDA-NPs-c) PTX-d) PDA-PTX-NPs-e) *Bif*@PDA-PTX-NPs.

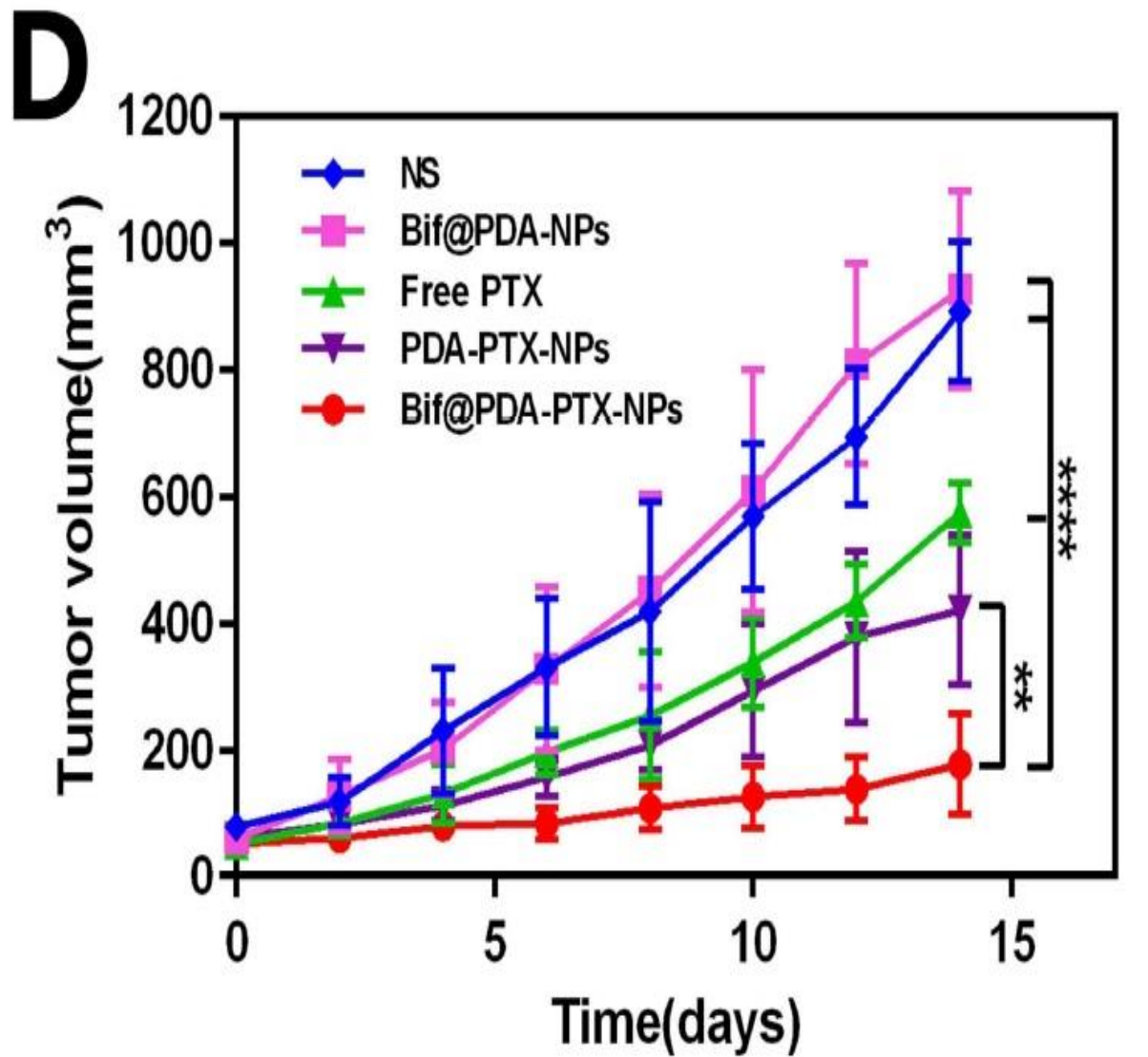
The photos of tumor-bearing mice on the 14th day after different treatments.

The photos of isolated tumors.

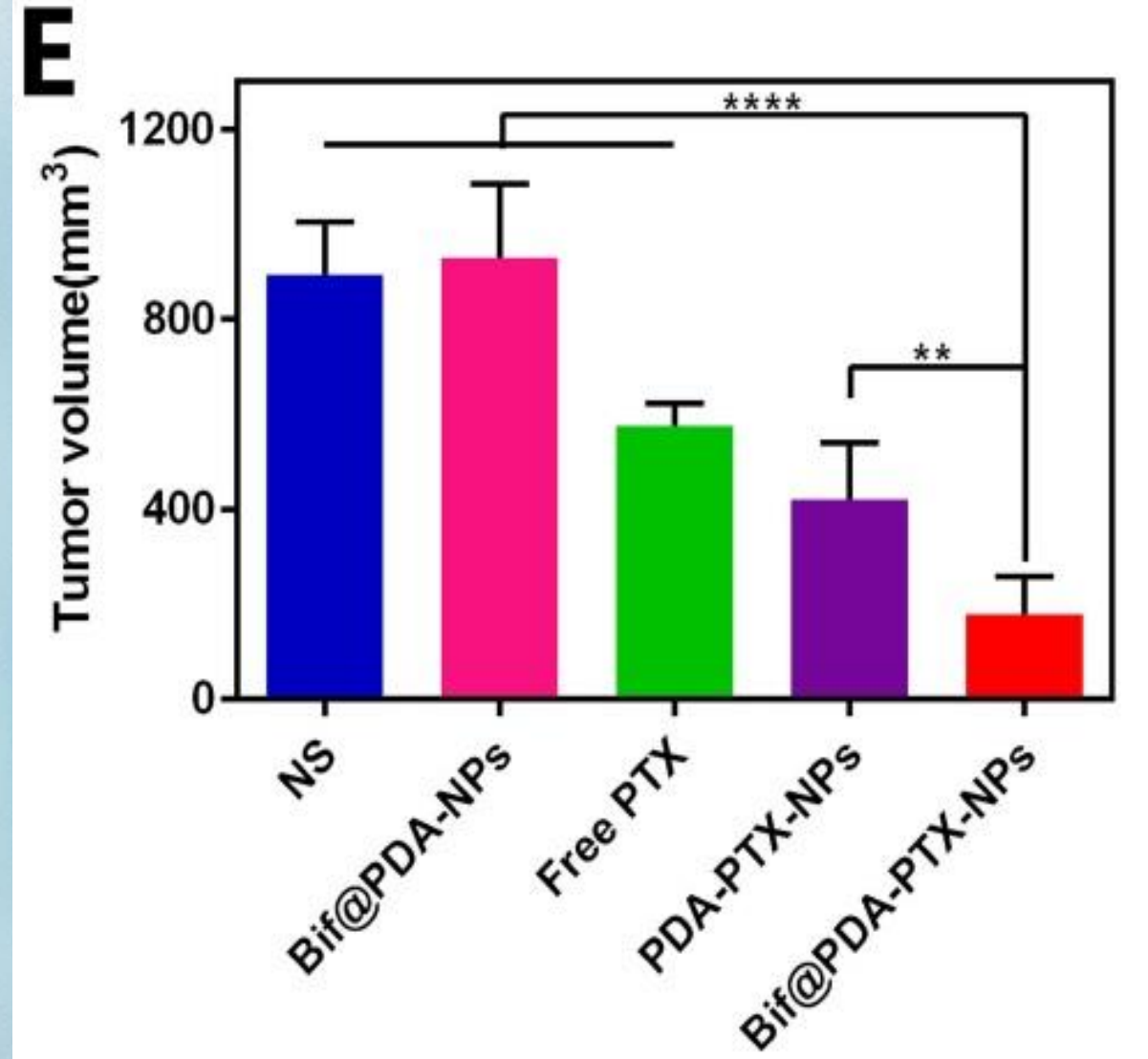


education

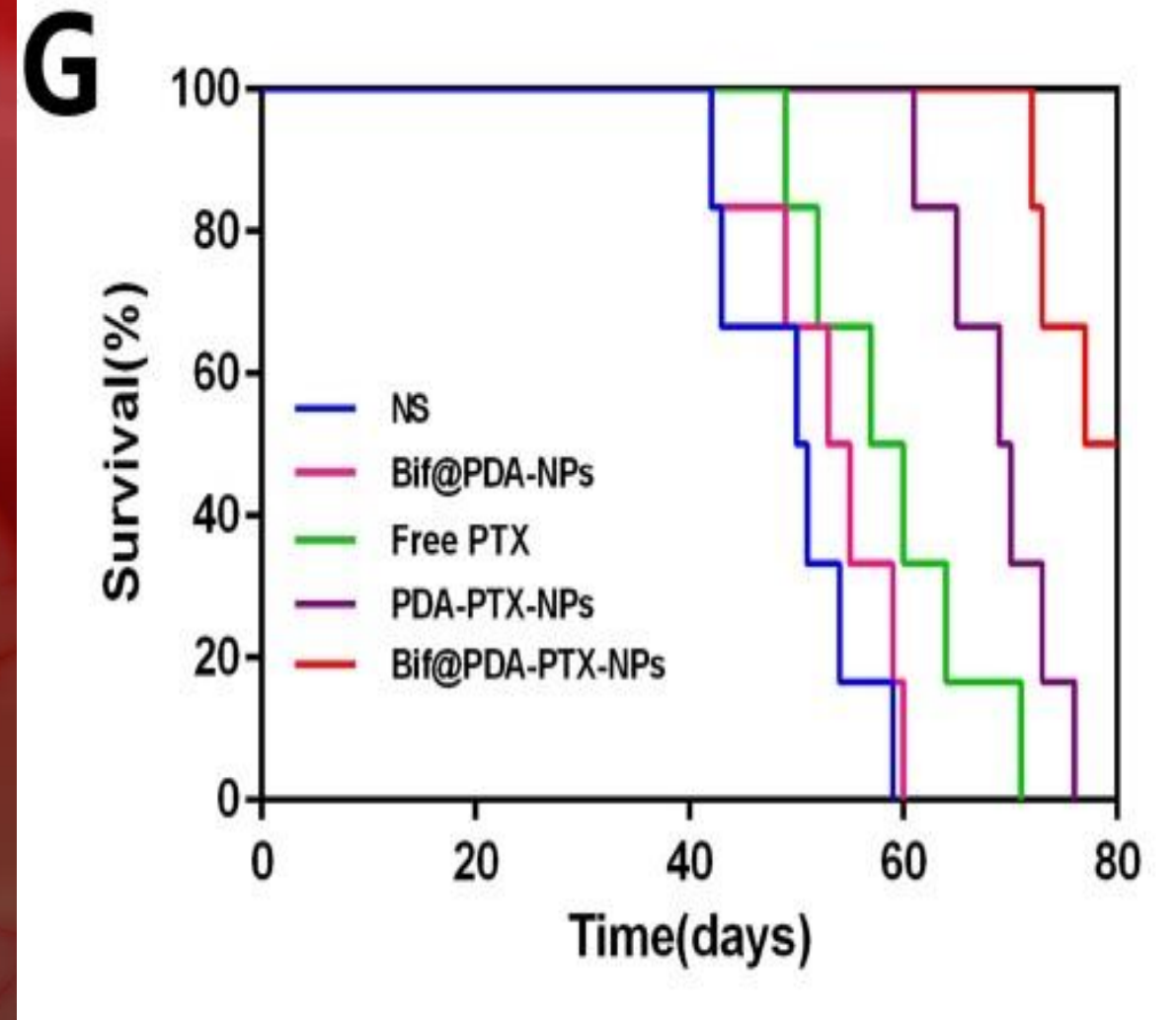
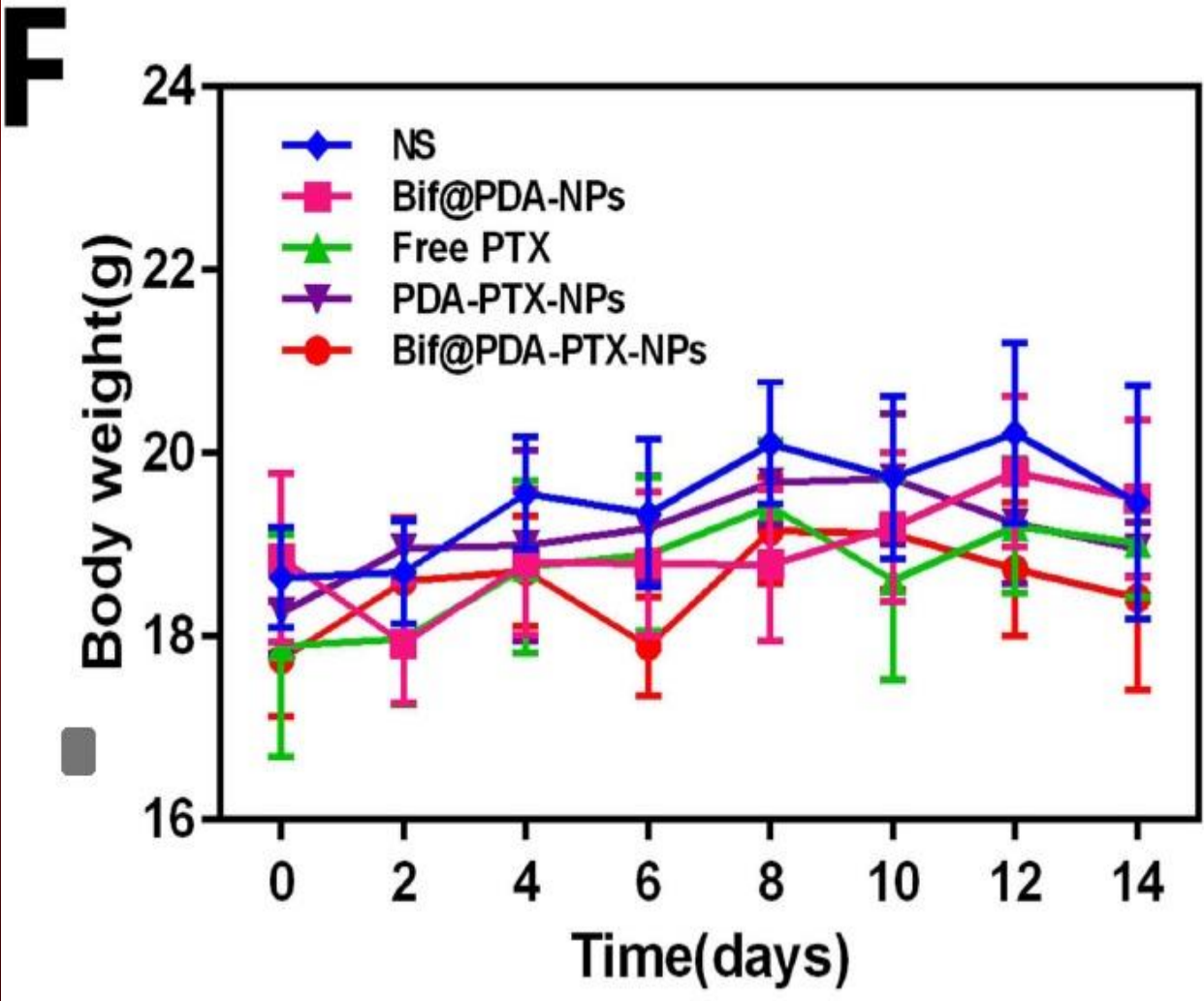
The tumor volume growth curves of mice in different groups.



The tumor volume of mice in each group on day 14 of treatment.

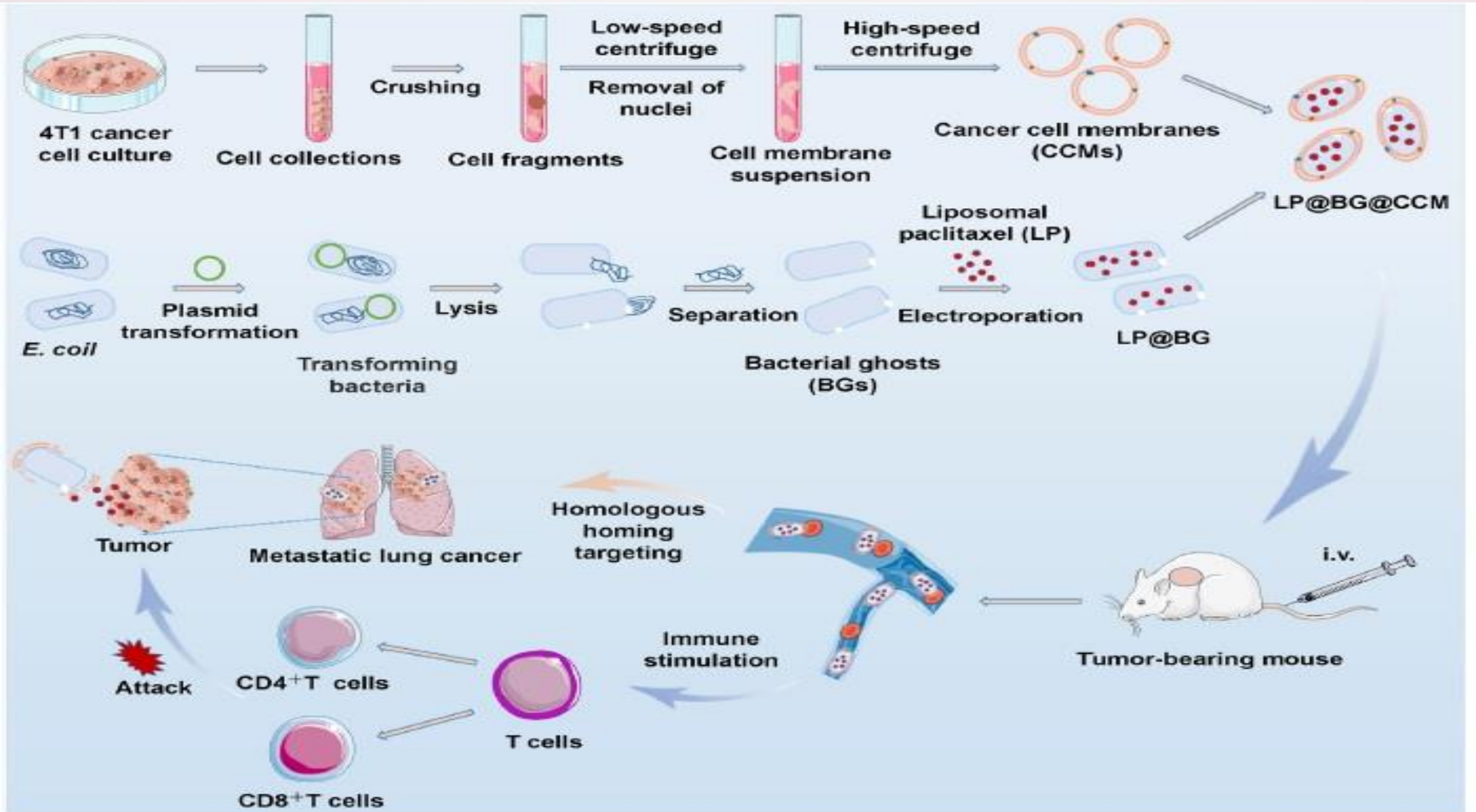


The body weight (F) and survival curves (G) of mice in each group.

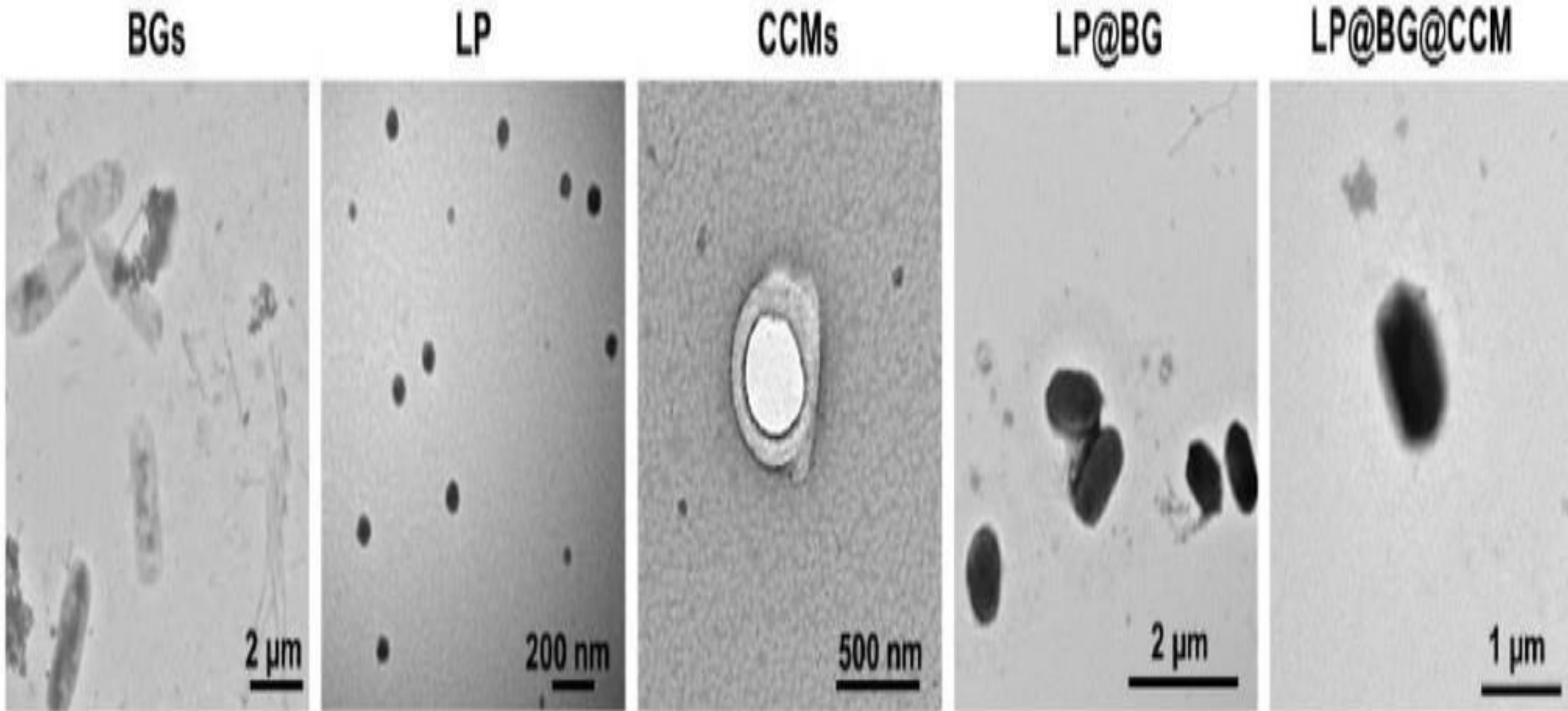


Bacterial ghost (BG) mediated Anti-cancer therapy

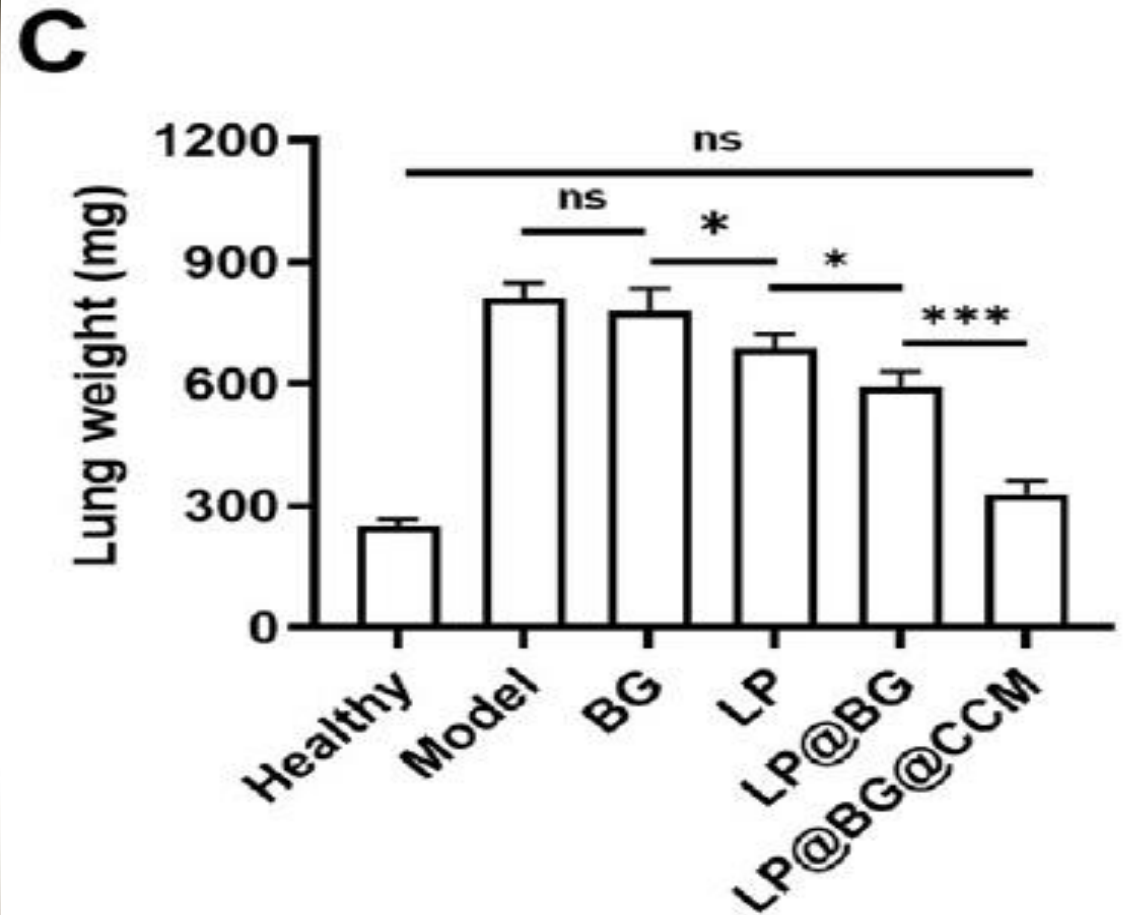
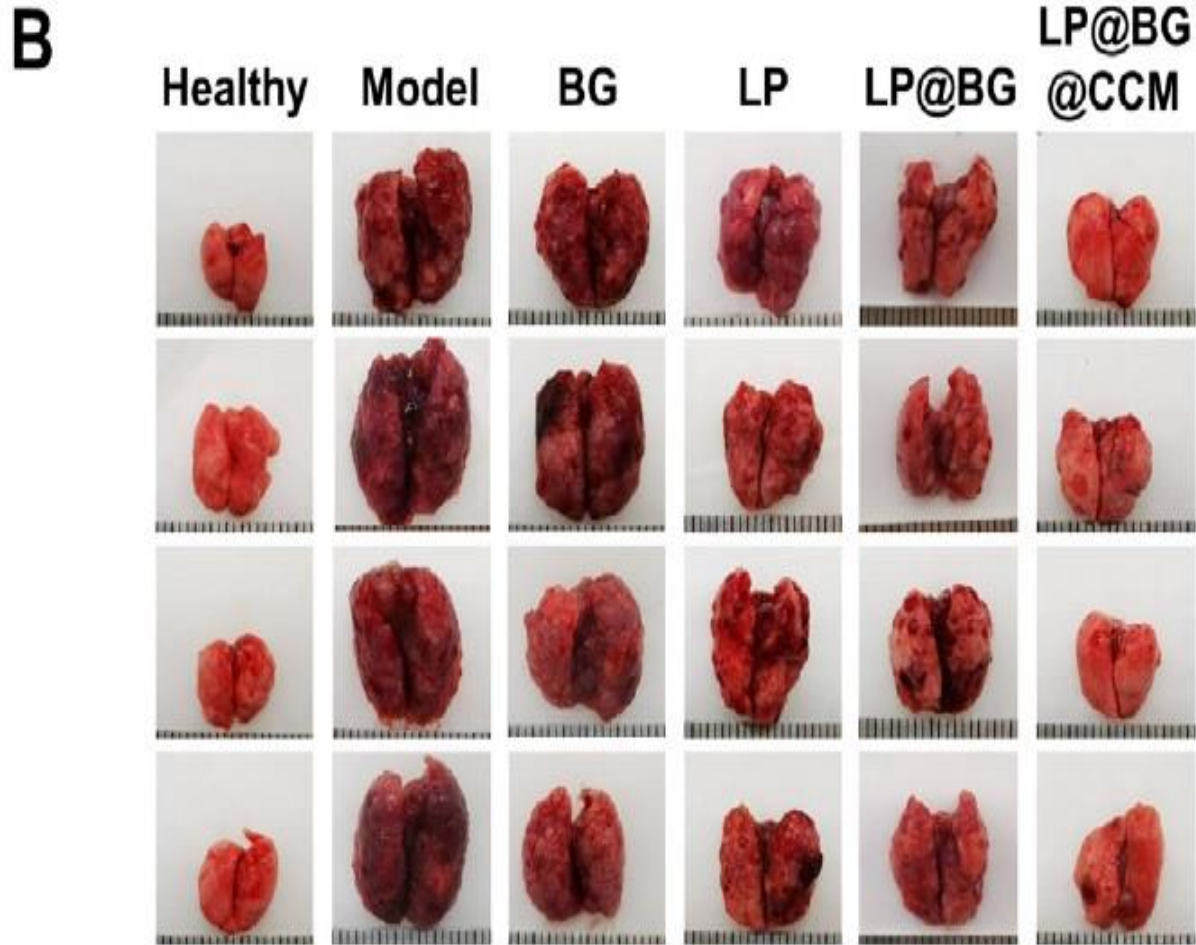
- BGs are empty cell envelopes derived from Gram-negative bacteria by bacteriophage ϕ X174 lysis.
- BGs own the complete cell morphology, including lipopolysaccharides, lipoproteins, peptidoglycans, and outer membrane proteins.
- Their strong immune stimulation effect is remained due to the presence of **lipopolysaccharide** and other cell wall components.
- **Cell membrane** coating on the surface of BGs is a good method to weaken the immune response of BGs.
- The cancer cell membranes (CCMs) is used, which are derived from cancer cell lysis.



Transmission Electron Microscopy (TEM) images of the formulations.



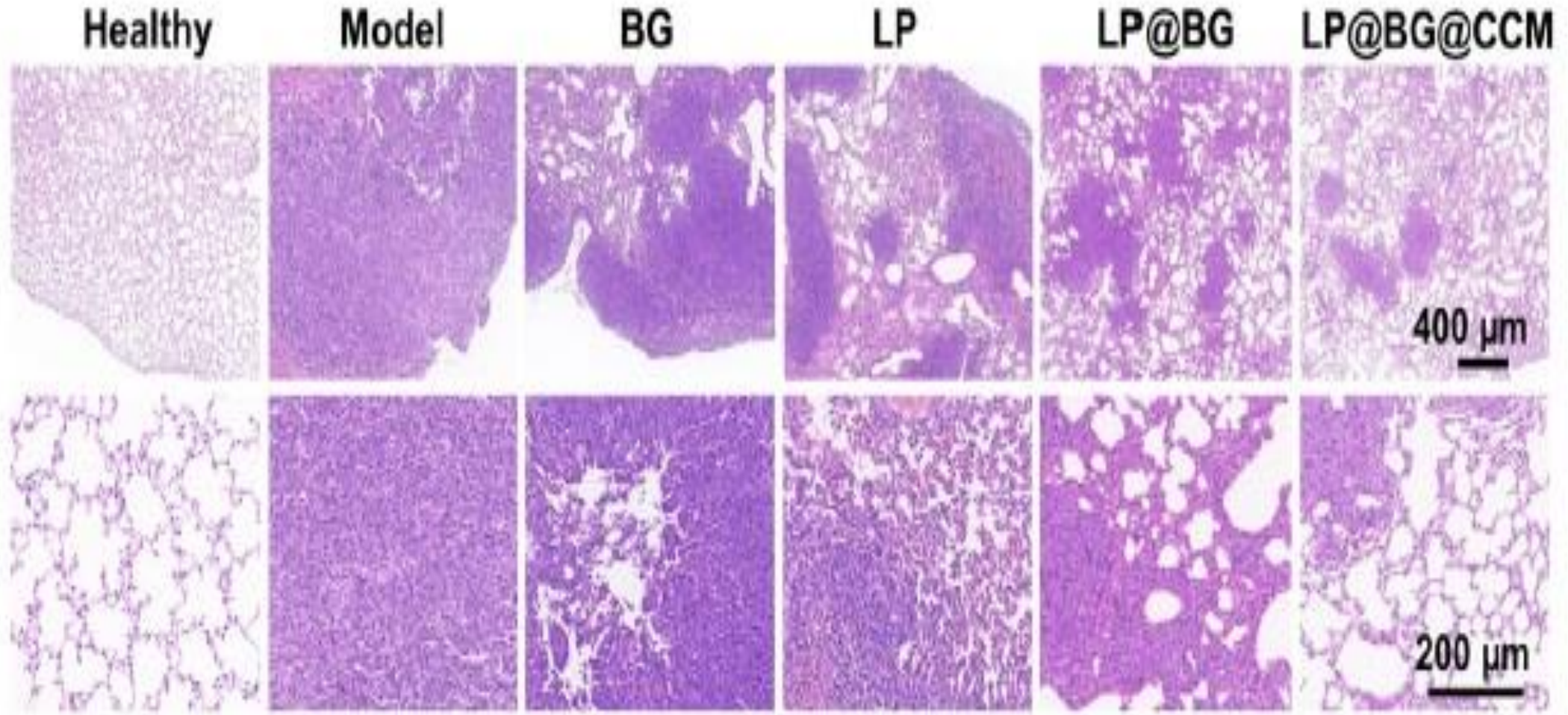
(B) Appearance and (C) the weight of the lungs of the various groups.



The H&E stained images

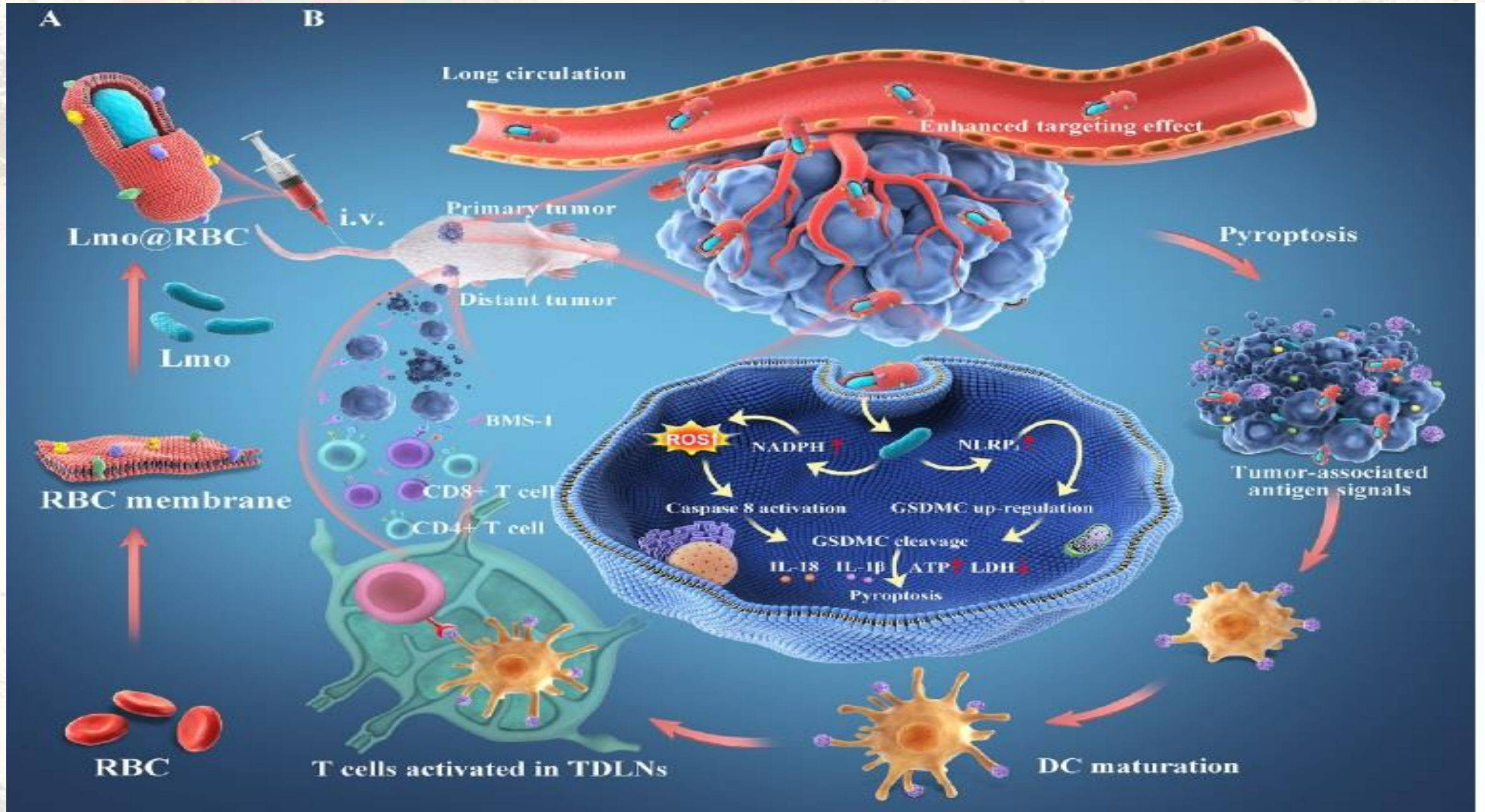
Doi.org/10.1016/j.apsb.2023.08.012

H&E



Bacterial-based immunotherapy

- *Listeria monocytogenes* (Lmo) has great potential for development as a cancer vaccine platform.
- Lmo in APC allows antigens to be processed in the MHC I and II molecules, resulting in robust CD₈⁺ and CD₄⁺ T cell-mediated immune responses.
- **Lmo@RBC** could directly kill tumor cells through reactive oxygen species (ROS) mediated by NADPH oxidase.
- **Lmo@RBC**, induce caspase-8 activation to up-regulate GSDMC expression to trigger the pyroptosis for tumor cells.

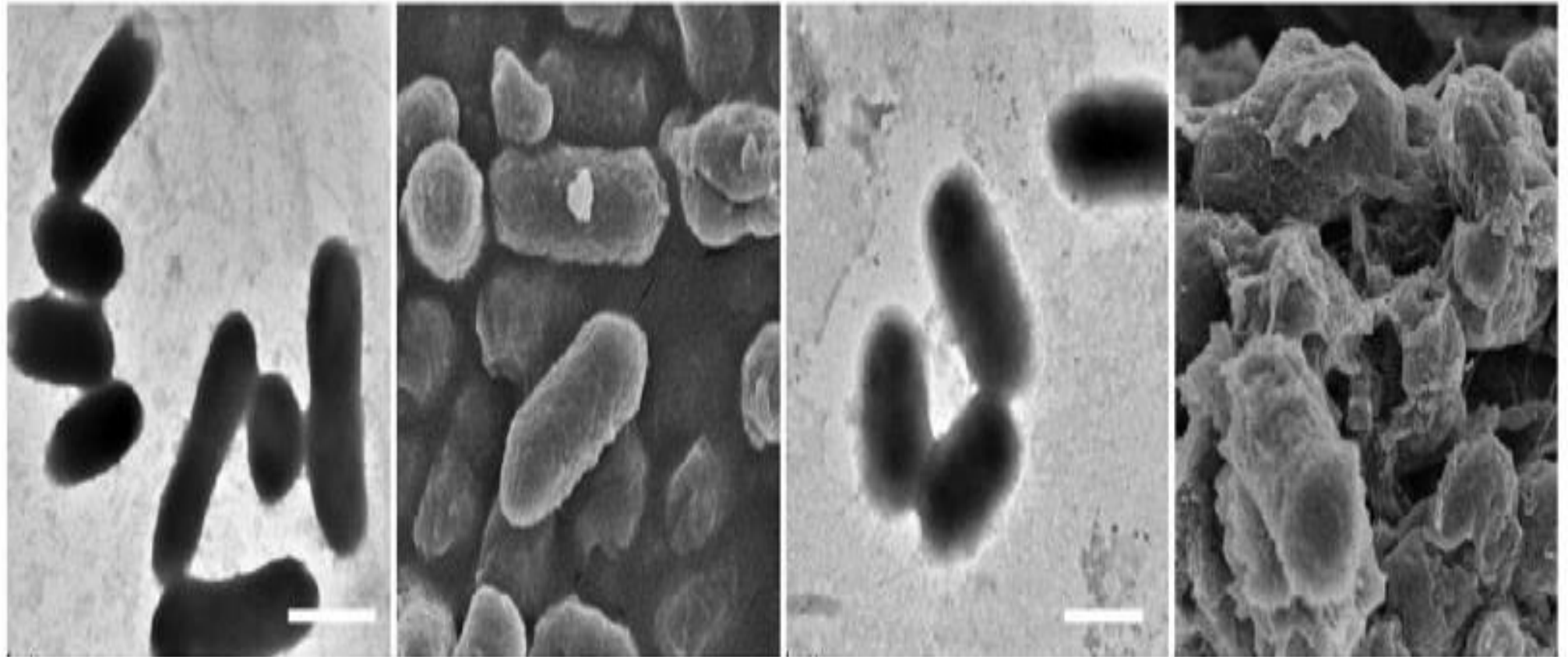


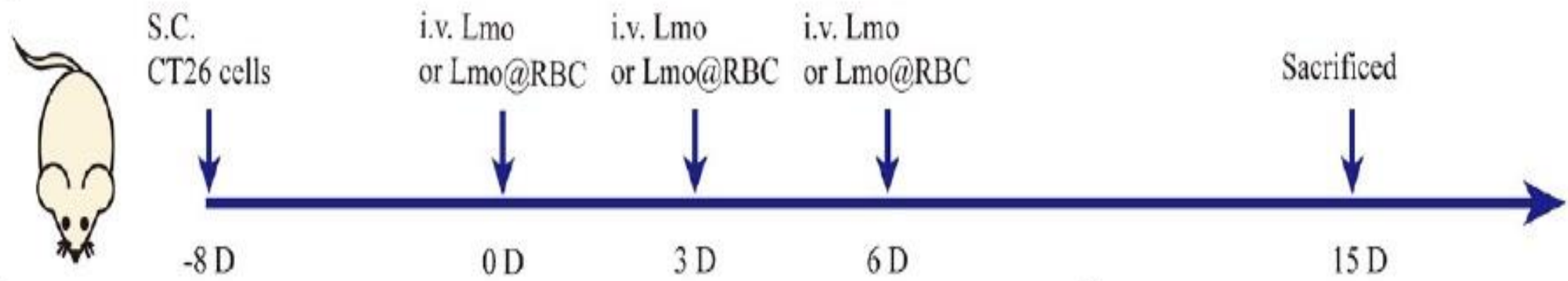
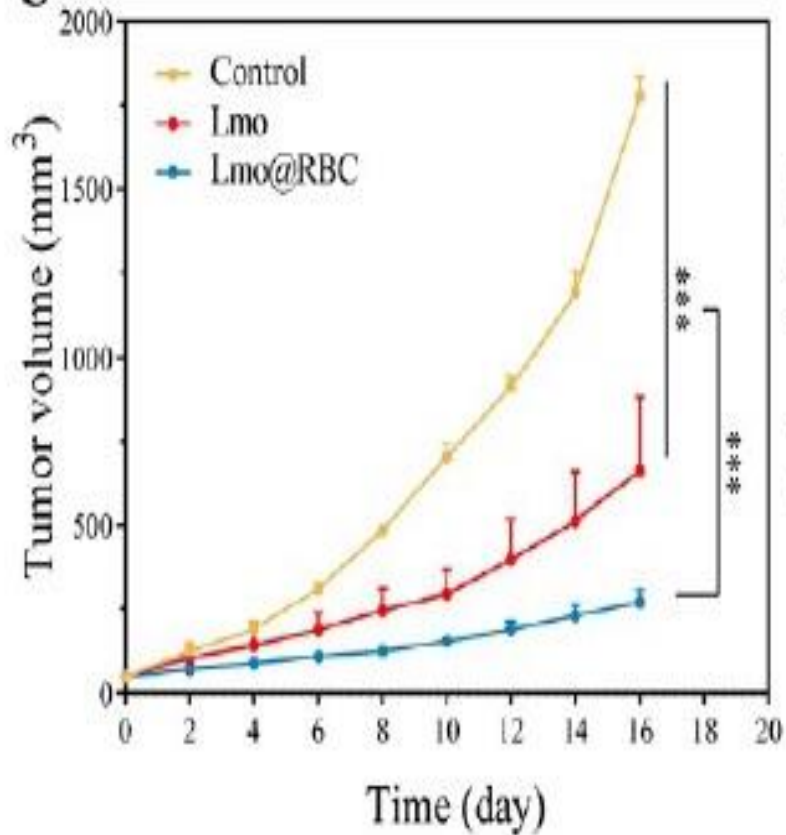
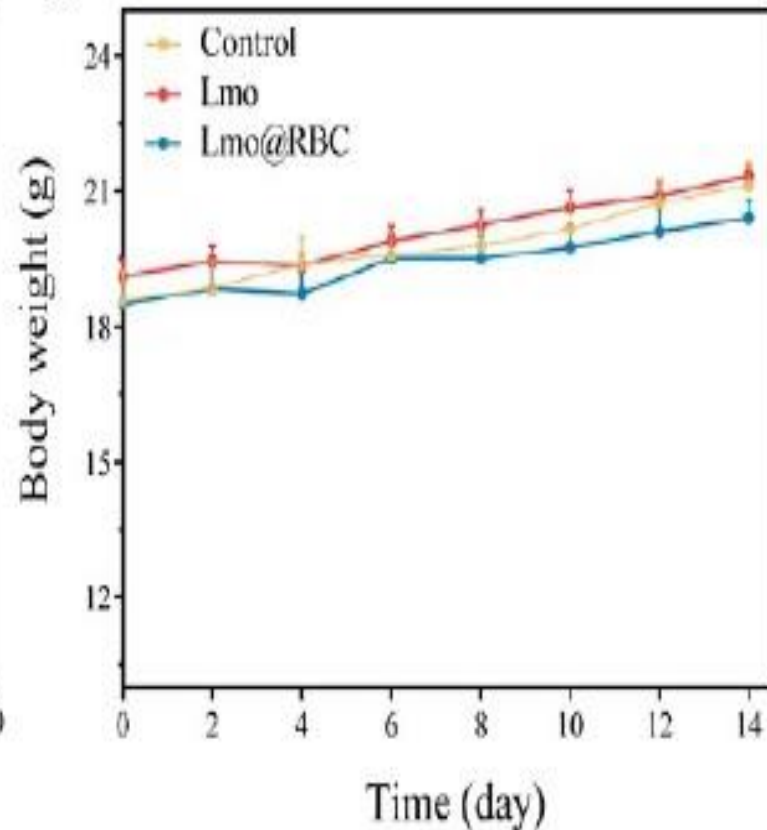
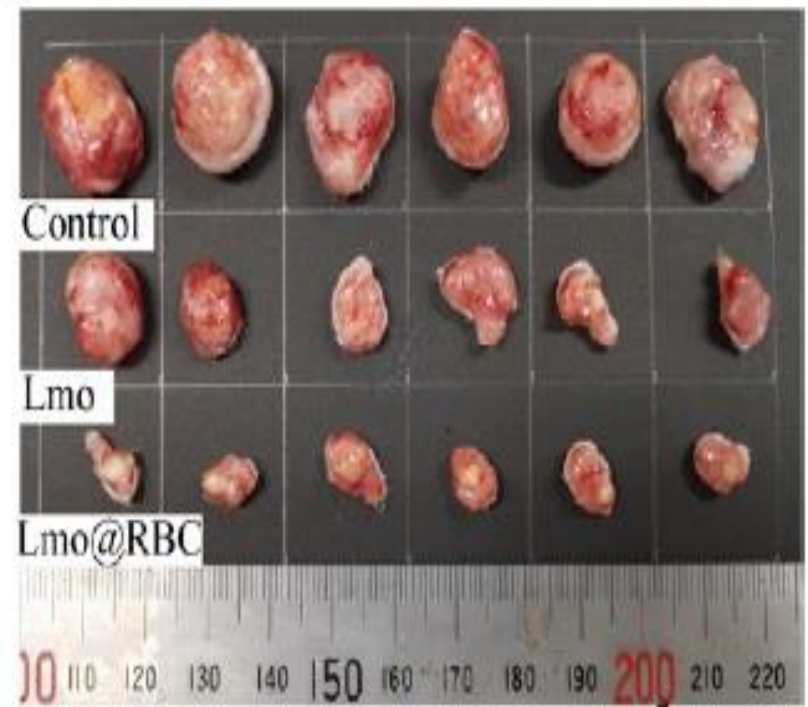


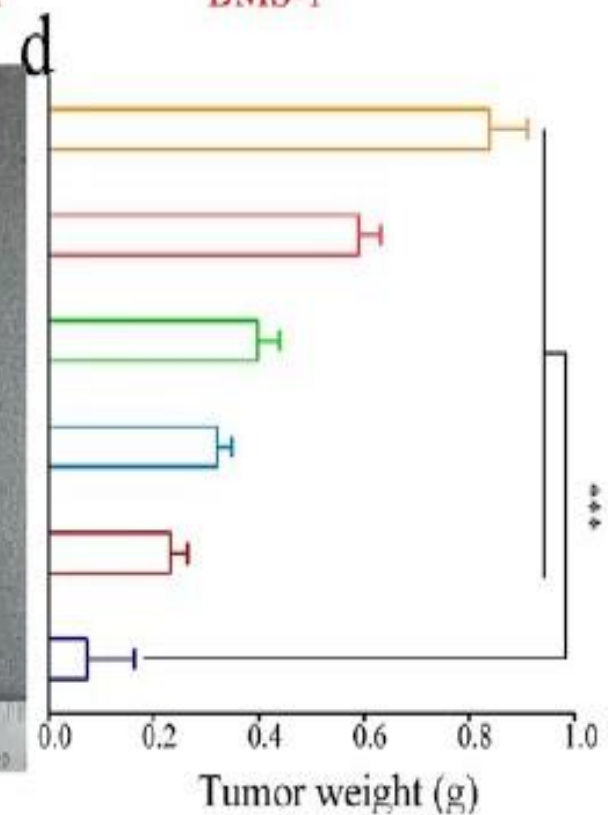
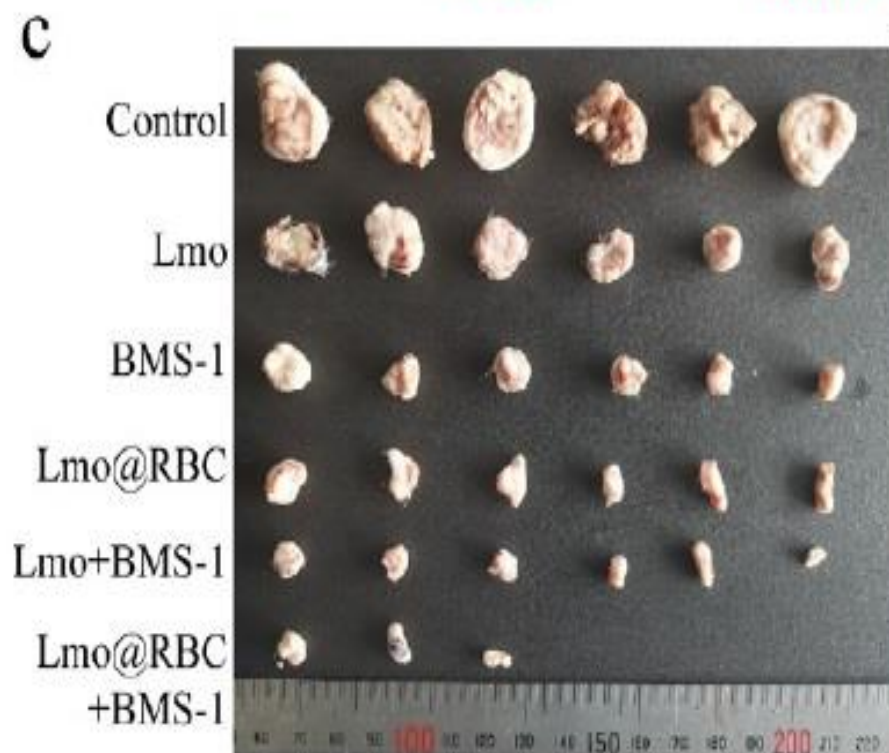
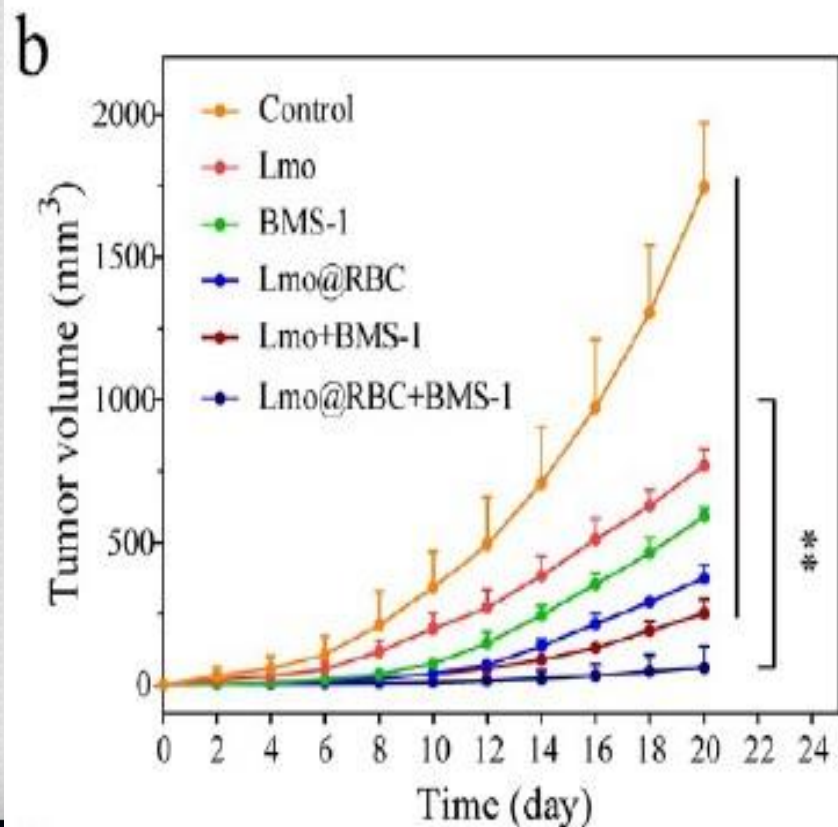
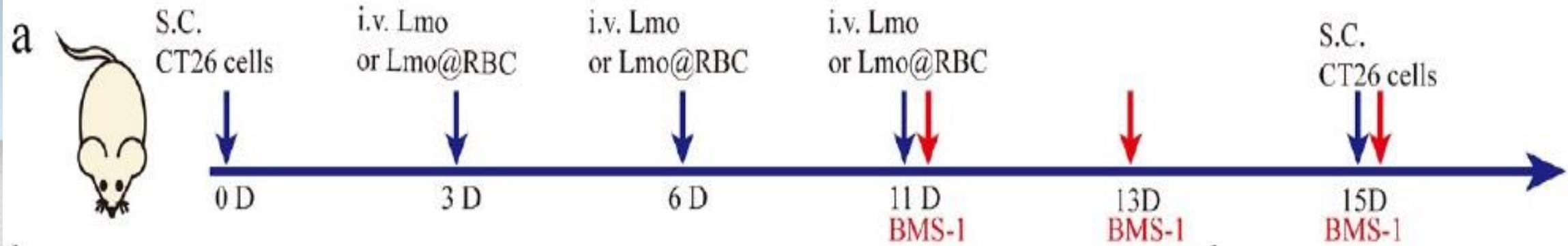
TEM and SEM image of uncoated Lmo and Lmo@RBC.

Lmo

Lmo@RBC



a**b****c****d**





Advantage and Disadvantages

Advantages

- BMCT has the advantage of high tumor-selectivity and mobility to overcome the limitation of conventional therapies.
- With bacterial therapy, other methods enhance their therapeutic potential.
- Bacterial therapy achieves sufficient tissue penetration.
- Bacteria may be easily manipulated and engineered.

Disadvantages

- Therapy is based on **attenuated strains**, the components of bacteria will **cause innate immunity**, and sequentially decreasing the number of bacteria colonized in solid tumors.
- The most of the administrated bacteria are unavoidably eliminated by **the reticuloendothelial system** before arrival at the tumor site.
- **The bacterial self-propagation** will lead to the therapeutic drugs or biomaterials detaching from the cell wall.
- Induction of **septic shock** due to high immunogenicity.
- A significant danger factor for living things can also be **systemic bacterial infections**.

Conclusion

- **Bacteria-mediated cancer therapy** is still in its infancy holding an immense potential to change the current forms of cancer therapy.
- **The main focus** at present is to effectively combine bacteria-based therapies and other relevant modalities, especially **immunotherapy** and **nanomedicine**.
- **The intelligent use** of bacterial can lead to the development of sustainable bacteria-mediated therapies for routine clinical applications.
- **Using engineered bacteria** alone or with conventional cancer treatment result to develop this treatment free of side effects.

REFERENCES

- **Zhao, Xiaoxuan Chen, Zhenghao Yang , Xiaodong Yang , Qiang Peng- Bacteria and tumor: Understanding the roles of bacteria in tumor genesis and immunology Mingming-Microbiological Research (2022).**
- **Md Kaiser Ali , Qing Liu , Kang Liang , Pei Li , Qingke Kong- Bacteria-derived minicells for cancer therapy-Cancer Letters (2020).**
- **Sandip Ghosh , Arghya Bhowmik, Kundlik Gadhave, Satabdi Datta- Bacterioboat—A novel tool to increase the half-life period of the orally administered drug Parmandeep Kaur-Science Advances(2022).**
- **Piyush Baidara, Santi M. Mandal-Bacteria and bacterial anticancer agents as a promising alternative for cancer therapeutics-Biochimie (2020).**

- **Shuaijie Ding, Zeming Liu, Chunyu Huang, Ning Zeng, Wei Jiang-Novel Engineered Bacterium/Black Phosphorus Quantum Dot Hybrid System for Hypoxic Tumor Targeting and Efficient Photodynamic Therapy-American Chemical Society(2021).**
- **Liang Yin, Zhan Meng, Yuxiao Zhang, Kaikai Hu, Wuya Chen-Bacillus spore-based oral carriers loading curcumin for the therapy of colon cancer-Journal of Controlled Release(2018).**
- **Dandan Ling, Xueli Ji, Ke Wang, Qiucheng Yan-Cancer cell membrane coated bacterial ghosts for highly efficient paclitaxel delivery against metastatic lung cancer-Acta Pharmaceutica Sinica(2024).**
- **Yao Liu, Yiping Lu, Bo Ning, Xiaomin Su, Binru Yang, Haiqing Dong-Intravenous Delivery of Living Listeria monocytogenes Elicits Gasdmermin- Dependent Tumor Pyroptosis and Motivates Anti-Tumor Immune Response-ACS Nano (2022).**

- **Yuhao Chen, Meng Du, Jinsui Yu, Lang Rao, Xiaoyuan Chen-Nanobiohybrids: A Synergistic Integration of Bacteria and Nanomaterials in Cancer Therapy-Bio Integration(2020).**
- **Jun Ying Fan, Yuan Huang, Yi Li, Tobias Achu Muluh-Bacteria in cancer therapy: A new generation of weapons-Cancer Medicine (2022).**
- **Huan Shi, Lan Chen, Yanlin Liu, Qinglian Wen-Bacteria-Driven Tumor Microenvironment-Sensitive Nanoparticles Targeting Hypoxic Regions Enhances the Chemotherapy Outcome of Lung Cancer-International Journal of Nanomedicine(2023).**

THANK
YOU

