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Cancer Traps: Implantable and On-Chip Solutions for Early Cancer Detection and Treatment

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Cancer continues to be a major global health issue causing millions of deaths annually. While traditional therapeutic methods may be effective in many cases, they may not be suitable for highly metastatic cancers. Moreover, the late detection of tumors, when they have already spread and are harder to treat, further exacerbates the challenge in managing this disease. As a result, there is a growing interest in developing complementary tissue-engineered approaches for early cancer diagnosis and treatment to enhance patient recovery. Bioengineered cancer traps have gained significant attention due to their efficacy and ease of use. These trapping systems employ (bio)chemical and mechanical strategies to selectively capture and limit the spread of cancer cells, leading to their eradication from the body. Furthermore, when integrated into microfluidic devices, these cancer traps-on-a-chip can be used for liquid biopsy and the early detection of circulating tumor cells and other tumor-derived material, allowing for precision medicine treatments. Herein, this innovative approach to cancer theranostics, including its mechanism of action, current stage of development, and potential advantages and limitations is discussed.

1. Introduction

Despite significant advances in research and treatment, cancer remains a leading cause of death globally. One contributing factor to this burden is the limitations of conventional treatments, such as chemotherapy and surgery, which are less effective for highly aggressive or recurrent cancers.[1] Additionally, traditional diagnosis methods, such as physical examination, imaging, biomarkers-based laboratory tests, and biopsies, are not very sensitive at early stages of tumorigenesis or take a significant amount of time to produce results, affecting patient prognosis.[2]

To address these challenges, there has been growing interest in developing complementary approaches to improve the efficacy of cancer diagnosis and therapy. In particular, bioengineered cancer traps have gained significant attention from the tissue engineering community as a potential solution to selectively capture cancer cells when implanted within the human body, avoiding their uncontrolled dissemination. This type of traps utilizes different engineered biomaterials (natural and synthetic) and mechanisms to selectively attract and capture cancer cells that have spread from the tumor, particularly after primary therapy, potentially leading to their eradication and reducing the need for more invasive treatments.[3] By being implantable, cancer traps have the advantage of being positioned in resected areas of a malignant tumor to target any residual invasive cells, or in specific organs or tissues where cells from the primary tumor typically metastasize, thus helping to prevent their uncontrolled dissemination.[4] Further, these traps can be used to provide continuous monitoring of cancer cell activity and therapy.[5] This can help to identify changes in the tumor that may indicate the need for additional treatment or a change in the treatment plan, or to detect the relapse of the tumor after therapy.[6] Indeed, the potential benefits of cancer traps for early cancer diagnosis and therapy have been explored in early-phase clinical trials, which have assessed their efficacy and safety for use in a clinical setting.[7] In addition, cancer traps can be incorporated within microfluidic devices to capture circulating tumor cells (CTCs) and other circulating tumor-related materials from bodily fluids, such as blood.[8,9] These devices use miniaturized channels and structures to manipulate and analyze small volumes of fluid, and can be designed to selectively capture CTCs based on their physical or (bio)chemical properties. The captured cells can be then analyzed for the identification of genetic mutations, protein expression, or other biomarkers to aid in diagnosis and treatment planning, leading to personalized medicine and theranostics.[10]

Cancer traps, whether in the form of implantable systems or integrated within microfluidic devices, present a wealth of opportunities for improving cancer diagnosis and treatment. Herein, we will shed light on the various ways in which cancer traps can be utilized in fighting cancer at various stages of progression, both in vivo and in vitro. We will focus on their current stage of development, their mechanisms of action, and potential

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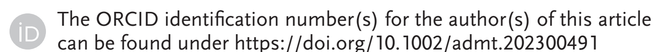
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advantages and limitations. Overall, this novel approach to cancer theranostics has the potential to reduce the spread of cancer cells, enable early cancer detection, boost our understanding of the disease, and improve the efficacy of treatments and overall patient prognosis.

2. Implantable Cancer Traps for In Vivo Capturing of Metastatic Cells

Implantable cancer traps use biocompatible materials, either naturally derived or synthetic, to attract and capture disseminating cancer cells. These traps can be broadly classified into two categories depending on their working mechanism: (bio)chemical- and mechanical-based (**Figure 1**). The former utilize (bio)chemical agents encapsulated into the material, such as specific antibodies, aptamers, chemoattractants, drugs, or other small molecules, to attract and capture cancer cells. Once trapped, these agents may block the migration and/or proliferation capabilities of cancer cells or induce their eradication from the body through the activation of the immune system.[11] Mechanical-based cancer traps, on the other hand, use physical mechanisms, such as microfilters, topological features, or physical entrapment, to capture cancer cells, which can then be removed from the body through similar (e.g., instruct the body/material to do it) or alternative methods, such as through a catheter or surgery. Independently of the selected capturing strategy, the traps can be either implanted in the tumor region after surgery or elsewhere as engineered biomimetic pre-metastatic niches exploiting the preference of certain types of tumors to colonize specific tissues, as described by the Paget´s *seed & soil* hypothesis.[12] In the following, these two types of cancer traps and some examples are discussed.

2.1. (Bio)chemical-Based Cancer Traps

Biochemical-based cancer traps are cutting-edge diagnostic and therapeutic tools that employ biocompatible and precisely functionalized biomaterials to attract and capture cancer cells when implanted into the body. In general, this type of cancer traps utilize hydrogels loaded or functionalized with one or several biological/chemical agents specific to cancer cells and not to healthy ones. For example, peptide-, protein-, or polysaccharide-based hydrogels decorated with cancer-specific ligands on their surface can be used to create personalized cancer traps that recapitulate the unique content of the tumor that supports metastasis.[13] Interestingly, the traps can be loaded with cancer-secreted exosomes to attract and capture cancer cells more efficiently,[14] or drugs to deliver therapeutic agents directly to the captured cancer cells, enhancing their therapeutic efficacy.

A typical example of a (bio)chemical cancer trap involves using polymeric scaffolds functionalized with specific agents, such as monoclonal antibodies, targeting receptors, proteins, etc., overexpressed on the surface of cancer cells.[15,16] For example, a silk fibroin scaffold mimicking the native bone marrow was developed for capturing breast and prostate cancer cells, which frequently spread to the bones. By infusing BMP-2 and inserting the scaffold

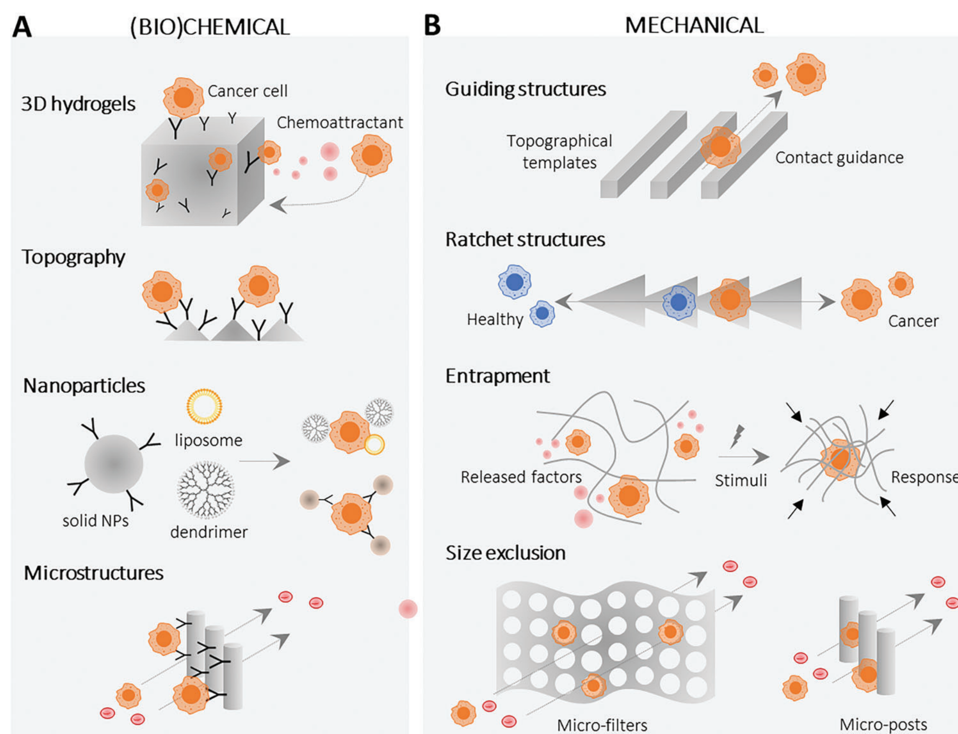

Figure 1. Cancer traps types and their mechanisms of action. Main types of A) (bio)chemical- and B) mechanical-based cancer traps. The formers include 3D hydrogels and scaffolds, topographic structures, nanoparticles, and 3D microstructures functionalized with biorecognition elements for the detection of cancer cells or other tumor-derived material. The latter include (bio)chemical-free guiding and ratchet-like structures, physical entrapment materials, or size exclusion methods.

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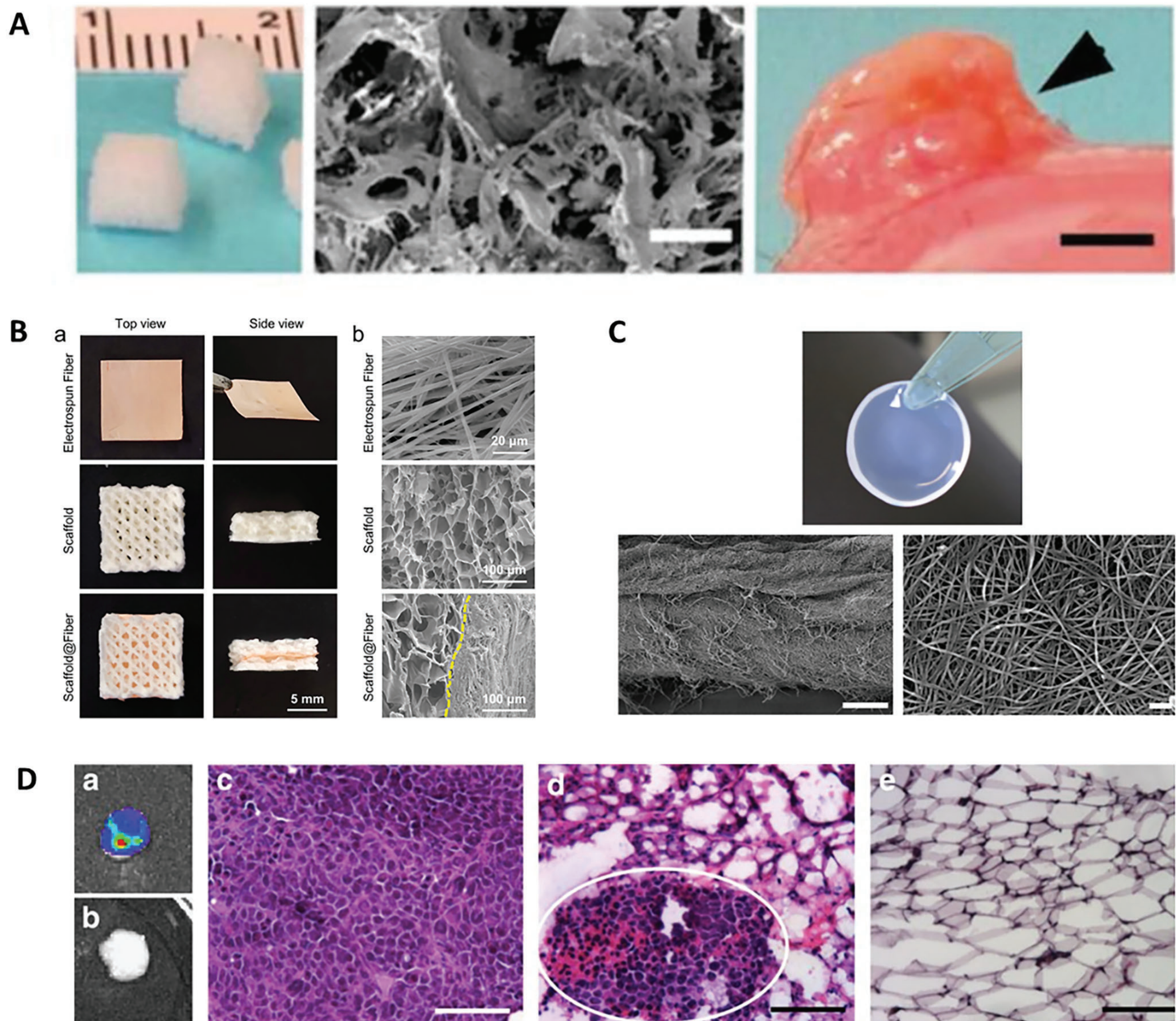

Figure 2. (Bio)chemical-based cancer traps. A) Macroscopic (left) and scanning electron microscopy—SEM (mid) images of BMP-2-loaded silk fibroin scaffolds mimicking the native bone marrow and their in vivo implantation (right) for capturing disseminating cancer cells. Reproduced with permission.[17] Copyright 2015, Elsevier. B) Left: Macroscopic top and side images of the 3D-printed composite scaffold made of gelatin, chitosan, and sodium alginate loaded with drugs. Right: Higher magnification SEM view of the scaffold and fibers. Reproduced with permission.[20] Copyright 2022, The American Chemical Society. C) Top: Image of the chemoattractant-loaded bacterial cellulose nanofibrous membrane developed to attract and treat glioblastoma cells in vivo. Bottom: SEM images of the membranes showing their random fibrillary network. Reproduced with permission.[23] Copyright 2019, The American Chemical Society. D) a,b) Microporous sponge-like and disk-shaped scaffolds made from polylactic-*co*-glycolide used for the in vivo capture of highly metastatic breast cancer cells. c–e) Bioluminescence images of the scaffold implanted in fat pads showing the presence of tumor cells or mock surgeries. Reproduced with permission.[29] Copyright 2015, Springer Nature.

under the skin of a mouse, bone growth was accelerated and red bone marrow was created. This resulted in the accumulation of metastatic breast cancer cells in the functional scaffold, serving as an in vivo trap (**Figure 2**A).[17] The implanted scaffolds can also release chemoattractants, such as SDF-1, TGF-α, TGF-β, FGF, PDGF, and various CCL and CXCL proteins,[18] among others, and chemotherapeutic drugs to attract and directly eradicate disseminating cancer cells. Once captured, the cells can be directly eliminated by the encapsulated drugs or surgically removing the trap. The release of the compounds can be done simultaneously or in sequence, which could be advantageous. In this case, the compounds can be organized in multiple layers with well-defined degradability and release kinetics.[19]

Undoubtedly, tumor relapse is a major challenge in cancer treatment, especially in postoperative therapy where bleeding and dispersed tumor cells can lead to recurrence. Cancer traps can help reduce this risk by capturing the remaining cells with the potential of spreading and deliver controlled doses of

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chemotherapy locally. This strategy was recently reported, where a composite material made of a 3D-printed scaffold of gelatin, chitosan, and sodium alginate loaded with combretastatin A4 and an electrospun fiber of polyvinyl alcohol-polylactic-*co*-glycolide (PLGA) loaded with doxorubicin was utilized to prevent bleeding and metastasis (Figure 2B).[20] This multifunctional scaffold was tested in a mouse model to investigate whether it could prevent prostate tumor recurrence after surgery. The release of combretastatin A4 and doxorubicin disrupted blood vessels and inhibited the growth of peripheral RM-1 prostate tumor cells, respectively. This synergistic strategy offered several benefits over more traditional approaches as demonstrated by the number of works recently published describing similar combinatory strategies.[21] However, some concerns with trap retrieval after the therapeutic cargo is exhausted may limit the in vivo applicability of this trapping strategy. This issue can be addressed by using biodegradable materials, such as polycaprolactone, which degrade over time after trapping and killing the cancer cells, reducing the need for surgical removal and minimizing the risk of long-term complications.[22] This is especially relevant for those regions with difficult access, such as the brain, as demonstrated recently through a cancer trap for treating glioblastoma in vivo (Figure 2C).[23] In this case, the trap was based on nanofibrous mesh loaded with a chemoattractant to attract residual tumor cells and successfully tested in a rat glioma model.

In manufacturing cancer traps, different biomaterials have been employed containing specific physical and (bio)chemical properties that allow them to attract and capture cancer cells.[24] Some examples include polymeric materials,[3,25] hydrogels such as silk fibroin,[26] and even metallic materials, such as gold and silver.[27] These biomaterials can be used alone or in combination with other biological elements, such as proteins or antibodies, to create cancer traps that are effective at capturing and removing cancer cells from the body, and can be designed to bind to specific markers or receptors that are expressed on the surface of cancer cells.[16,28] This allows cancer traps to be more selective in their targeting, increasing their effectiveness in removing cancer cells while minimizing the risk of damaging healthy ones. One type of U.S. Food and Drug Administration (FDA)-approved biomaterial that has been developed for trapping cancer cells inside the human body is a sponge-like material made from PLGA, a biocompatible polymer, loaded with the chemokine CCL22 to attract immune cells to induce a local immune response (Figure 2D).[29] This material was designed to be placed in a specific location in the body, such as the abdomen or the chest cavity. The sponge-like structure of the material allowed to collect cancer cells when implanted under the skin near the lymph nodes by creating environments similar to where cells naturally accumulate. Once the cancer cells were captured, the cancer cell-laden sponges could be removed and analyzed to determine the type and stage of cancer. This information could be used to guide treatment decisions and monitor the effectiveness of therapy. Recently, this trapping strategy was exploited not for capturing cancer cells but for absorbing the excess of chemotherapy drugs avoiding systemic toxic effects, thus showcasing the versatility of this type of approach.[30]

Besides hydrogels and scaffolds, nanoparticles, such as metals (e.g., gold and silver), dendrimers, lipids, or self-assembled nanomaterials, among others, can also be used for trapping cancer cells.[31] These approaches are well-suited for capturing those cells that transit along the bloodstream, namely, CTCs, which are considered responsible for the hematological dissemination of the tumor to distant tissues.[32] Gold and magnetic nanoparticles coated with cancer-specific antibodies have been widely used to create cancer traps targeting cancer cells in the bloodstream.[33] These traps can be combined with hyperthermia to heat up cancer cells to increase the effectiveness of chemotherapy.[34] Next, dendrimers, highly branched nanoscale polymers, have also been used to create traps by attaching cancer-specific ligands to their surface and deliver drugs directly to the cancer.[35] Finally, the self-assembly of molecules or synthetic nanostructures provides precise targeting capabilities in cancer therapy.[36] For instance, the use of self-assembled peptide-Au nanohybrids capable of forming higher-order structures has shown promising anticancer effects and holds potential for the development of targeted peptide-based nanomedicine tailored to the biochemical properties of the tumor microenvironment.[37] Moreover, when combined with stimuli-responsive hydrogels, self-assembled materials derived from peptides can generate a synergistic effect for capturing and effectively eliminating cancer cells, opening up exciting opportunities not only in cancer therapeutics but also in other therapeutic areas.[38]

Overall, biochemical-based cancer traps offer a valuable solution for the early diagnosis and treatment of cancer, reducing the risk of relapse and enhancing therapeutic efficacy. The versatility of these traps allows for the integration of various biological and chemical agents making them a useful tool for improving cancer outcomes. Nonetheless, there are some challenges, such as the possibility of the therapeutic cargo affecting healthy cells, which raises concerns about their practical use. Thus, further efforts or strategies are required to advance the clinical potential of cancer traps.

2.2. Mechanical-Based Cancer Traps

Mechanical-based cancer traps utilize pure physical mechanisms to capture and eliminate cancer cells, avoiding the potential harmful effects of (bio)chemical-based traps on healthy cells. To date, a myriad of mechanical-based cancer traps has been described, displaying different working mechanisms but with the common feature of being biomarker-independent platforms. Examples include the use of size-sensitive materials (e.g., microposts, porous membranes, and others) selective to large cancer cells, particularly CTCs; topological surfaces, such as guiding templates; or distinct types of physical forces, such as optical, acoustic, electrical, inertial, or magnetic forces (see Section 3). An example of a size-sensitive cancer trap are microfilters, a device similar to a dialysis membranes, which utilize a physical barrier to selectively trap cancer cells while allowing normal cells and other smaller molecules to pass through.[39] Many different filter designs with a diversity of pore sizes have been reported being most of them tested and validated in micro- or millifluidic devices in a sort of liquid biopsy-on-a-chip approach (see Section 3).[40] This type of size-sensitive microfiltration devices has been designed with the final aim to be implanted, typically through a minimally invasive procedure, in a blood vessel selectively capturing CTCs as they flow through the bloodstream and preventing them from reaching distant parts in the body. They

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can additionally be located next to the tumor, e.g., after surgery, to attract and trap disseminating cancer cells that were left behind as previously mentioned, thus reducing the risk of cancer cells forming new tumors. One potential limitation of microfilters is clogging when saturated with cancer cells or even debris, potentially leading to a decrease in the efficiency of the trapping process. Clogging can also lead to a reduction in the fluid flow being filtered and to an increase in blood pressure, which could lead to severe consequences. Additionally, clogging can make it more difficult to properly monitor and track the progression of the tumor, as it can interfere with the ability to accurately assess the number of cancer cells present in the body. Furthermore, it can also increase the risk of complications and side effects associated with the treatment, as it can make it more difficult to properly control the conditions within the body. Overall, albeit very efficient, size exclusion approaches may have serious limitations, particularly when utilized in vivo.

Physical entrapment has been proposed as an effective, label-free method of mechanically trapping cancer cells within a biomaterial that alters its shape in response to changes in the tumor's environment. This trap is usually placed near the tumor, where the body's temperature or acidic environment serves as a trigger for capturing the cells on the spot, as demonstrated using silk fibroin hydrogel.[41] The change in the shape of the silk hydrogel (from a random coil to a crystalline β-sheet form) transformed the cancer-friendly microenvironment into a harsh one, trapping and eliminating glioblastoma cancer cells likely due to limited oxygen and nutrient access in the hydrogel (**Figure 3**A). One of the advantages of this approach is its potential clinical applications since the gel's viscoelastic properties allow it to be injected near or on the tumor, where the high concentration employed (16%) causes it to solidify quickly. These hydrogels could be further optimized to deliver drugs or other therapeutic agents to the trapped cancer cells. However, the main limitation of this method is its incapability to differentiate between cancerous and noncancerous cells. Hence, a more sophisticated approach, or adaptation, may be necessary, such as customizing the hydrogel with a molecule that attaches to receptors only found in cancer cells, allowing for their specific capture. This concept was recently tested using a heat-sensitive hydrogel with imprinted sialic acid and powerful binding receptors.[42] The trap showed significant success in trapping HepG-2 cancer cells when tested in a lab environment that mimicked the human body temperature. Interestingly, when the temperature was decreased to 25 °C, the still-living cancer cells could be quickly recovered for further examination. The trap's accuracy was proven through a blood sample study that acted as a liquid biopsy model, showcasing a remarkable capturing ability. A similar approach using simple carbohydrate derivatives and an external trigger was employed to successfully trapping and killing cancer cells.[43] This approach exploited the elevated production of alkaline phosphatase (ALP) by cancer cells to trigger the self-assembly into nanofibers and subsequent gelation of the material around the cells, reducing their metabolic activity and leading to cell death. This approach is remarkable for its selectivity toward cancer cells that produce higher levels of ALP than healthy ones.

In recent years, the biophysics community has been closely studying the potential of topological approaches to control cell movement. Among all the works published during the last years, one particularly unique approach showed how an array of periodic asymmetrical cues, such as triangular adhesive patches or polymeric channels similar to a ratchet mechanism, can be used for guiding cells (or other out-of-equilibrium entities),[44] and therefore, with the potential for being employed as chemical-free cancer traps.[45] Indeed, this topological trapping method—ratchetaxis—has already been applied to differentiate between different cell types by encouraging their movement toward opposite directions, making it possible to categorize cells based on size, type, or invasiveness.[46] This innovative technique was suggested as a means of utilizing nontoxic materials to create implantable cancer traps, where tumor cells would be directed toward the trap keeping other cells away from being trapped. Finally, other more conventional methods based on the contact guidance of cells using microfabricated structures have been described for guiding the motion of cells, promoting their capture. These structures, such as parallel grooves or lines, are much simpler than ratchetaxis-based structures, but their efficiency in capturing cancer cells can be threatened because cell directionality is stochastic. In some cases, this is not a major problem, such as in the case where aligned polymeric nanofibers were used as guiding templates to attract glioblastoma cells toward an extracortical cytotoxic collagen-based hydrogel trap in vitro and importantly, validated using a mouse model (Figure 3B).[47] Other topographical structures have been reported for capturing disseminating cancer cells, including artificial biomimetic nanotopographies.[48] This strategy pioneered the development of a novel concept referred to as NanoVelcro CTC assays, where different nanosubstrates recapitulating the working mechanism of Velcro were designed for efficient CTC capture (Figure 3C).[49] Similarly, 3D-like structures, such as electrospun fibers or polymeric brushes, have been utilized to trap CTCs with higher yield due to the larger surface-to-volume ratio.[50]

Finally, other mechanical-based mechanisms have been described to capture cancer cells selectively, being some of them very sophisticated (and promising), such as using magnetic levitation forces.[51] Each cell type has a unique levitation profile and differences in levitation and density between cancer and noncancerous cells can be identified. As such, this makes density a robust "biomarker" for the label-free identification, and capture, of cancer cells. Indeed, implantable magnetic scaffolds have demonstrated a remarkable ability for the in vivo capture of CTCs. Specifically, a vascular-like system containing adhesive sites and the wireless magnetothermal response was recently reported for the continuous capture and removal of CD44-positive CTCs in vivo (Figure 3D).[52] The device was made of poly(L-lactic acid) fibers encapsulated with FeCo nanocrystals and modified with hyaluronic acid and gelatin to form artificial adhesion sites for capturing the cells. Cell death was accomplished through magnetothermal ablation under an alternating magnetic field. Notably, the efficacy of this approach was validated in a rat metastasis model with successful removal of captured CTCs and normal function of adjacent blood vessels. Despite the demonstrated effectiveness, this trap suffers from limitations, including the challenging integration into the vessel, even though it has been designed to prevent vascular blockage and induce potential vascular regeneration. In any case, this type of approach could be tested using reductionistic microfluidic models of tumor blood

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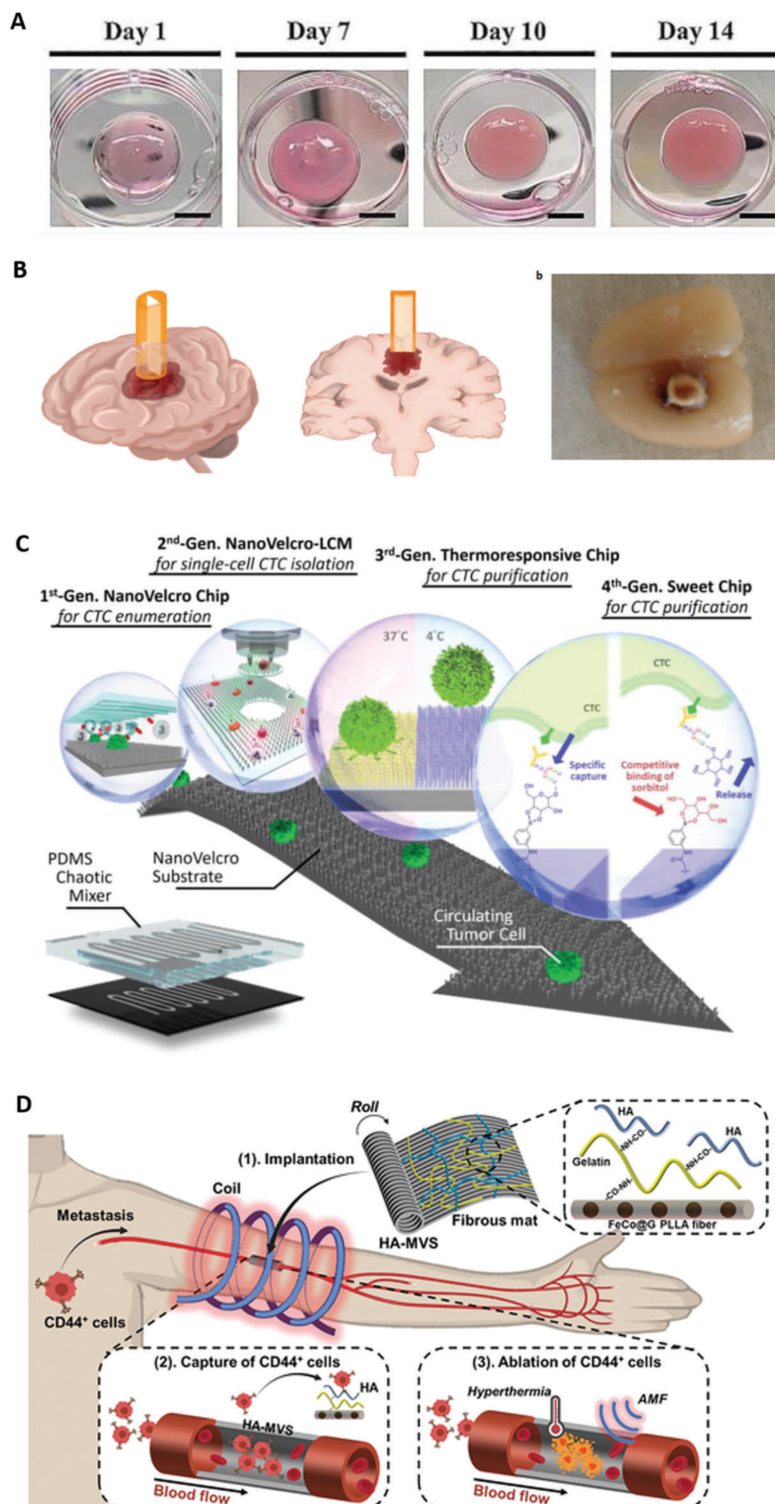

Figure 3. Mechanical-based cancer traps. A) Time sequence images over 14 days of human glioblastoma cell-laden silk fibroin hydrogels turning β-sheet structural transition for the capture and killing of cancer cells. Reproduced under the terms of the Creative Commons – Attribution 4.0 International CC BY 4.0.[41] Copyright 2018, The Authors, published by Public Library of Science. B) Trapping of glioblastoma cells through a tubular conduit working as a guiding template. Reproduced with permission.[47] Copyright 2014, Springer Nature. C) Graphic illustration of the NanoVelcro assays for CTC enumeration, isolation, and purification among four different generations. The embedded nanostructures increase the affinity between the CTCs and the surface. The polydimethylsiloxane chaotic mixers enhance the contact frequency between the cells and the capturing structures. Reproduced with permission.[49] Copyright 2018, Elsevier. D) Scheme describing the step-by-step capture and removal of CD44+ CTCs by the vascular-like integrated trapped device. Reproduced with permission.[52] Copyright 2022, Wiley-VCH GmbH.

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Table 1. Cancer trap-on-a-chip summary.

Mechanism	Isolation type	Capturing method	Application	Ref.
(Bio)chemical	Active	EpCAM-coated microposts	CTCs isolation – lung cancer	[61]
		EpCAM-coated microposts	CTCs isolation – lung cancer	[62]
		CD9 and EpCAM-coated herringbone	Exosomes isolation – ovarian cancer	[74]
		CRISPR-RNA	Nucleic acids isolation	[78]
Mechanical	Active	Optical forces	CTCs isolation – colorectal cancer	[63]
		Acoustic forces	CTCs isolation – prostate cancer	[64]
		Acoustic forces	CTCs isolation – multiple cancers	[65]
		Electrical forces	CTCs isolation – prostate cancer	[67]
		Inertial forces	CTCs isolation – breast/bladder cancer	[68]
		Magnetic forces	CTCs isolation – breast cancer	[72]
	Passive	Size exclusion	Exosomes isolation – breast cancer	[75]
		Size exclusion; deformability	CTCs isolation – breast cancer	[59]
		Size exclusion	CTC clusters isolation – multiple cancers	[60]

vessels to optimize the trapping design and avoid unexpected complications.[53]

3. Microfluidic Systems for Trapping Cancer-Derived Material: Cancer Trap-on-a-Chip

Early cancer detection can lead to a significant boost in survival rates as the tumor can be treated more efficiently, whether through surgery or less intense medication treatments. The relevance and effectiveness of some of the screening tests available for certain types of cancers tests has generated some controversy.[54] Some other cancers still lack relevant biomarkers for their early detection.[55] Microfluidics technology has become a highly effective tool for precisely identifying, capturing, and analyzing cancer-derived material, such as CTCs and clusters in peripheral blood.[9,56] Increased CTCs levels on peripheral blood are directly correlated with a low patient prognosis; therefore, their capture and analysis are of utmost clinical importance. Similarly, other biomarkers present in blood plasma (or other bodily fluids like urine or saliva),[57] such as circulating tumor DNA (ctDNA), tumor-derived extracellular vesicles (EVs), or other proteins (e.g., antibodies), can be employed for liquid biopsy applications.[58] Despite the potential for early detection through the capture of these tumor-derived materials, several hurdles make this a challenging task, mainly their low number present in plasma at the initial stages of tumorigenesis or their small size. Microfluidic devices exploit the unique physical (size, density, deformability, and others) and biological properties of CTCs (and other tumor-derived material) to separate them in high purity from other blood components in specific regions of a microfluidic chip where they can be captured and isolated. As such, this type of liquid biopsy device can be referred to as *cancer trap-on-a-chip* platforms. By doing so, CTCs can be accurately analyzed in a manner that provides valuable information about the molecular profile of the tumor and potential drug sensitivity. This information can be used to guide personalized treatment strategies and monitor disease progression.

Two main methods have been used for the microfluidic isolation of tumor-derived biomarkers, mainly CTCs: *active* and *passive* (**Table 1**). Active methods utilize external forces, such as the application of magnetic fields or electric fields for the manipulation and separation of CTCs from other blood cells, or cell-targeting strategies, such as antibodies or aptamers, to actively capture these rare cells. In contrast, passive methods rely on inherent physical properties of CTCs, such as size, deformability, and density, allowing their selective trapping or migration within the microfluidic device without external forces.[59,60] Indeed, integrating filters, such as microposts, within microfluidic channels is a simple but effective approach to capture CTCs. These posts can be decorated with specific antibodies for improving cell trapping yield (Figure 1). The most popular approach is based on higher EpCAM expression on CTC surfaces. However, this biomarker is not universally expressed by CTCs leading to potential false negative results and limiting the overall efficacy of the trapping method. Despite these limitations, the use of EpCAM as a CTC biomarker remains widespread due to its simplicity and ease of use, and several microfluidic platforms containing anti-EpCAM antibody-coated microposts have been described to concentrate CTCs in reduced sample sizes (Table 1).[61,62]

The nonspecificity of biomarker recognition through antibodies has stimulated the interest in exploiting the physical characteristics of CTCs for their selective isolation, including optical, acoustic, electrical, inertial, or magnetic forces. Optical forces have attracted much attention for the remote separation of CTCs in a gentle and efficient manner, even though the similar optical properties between the tumor and other blood cells may limit their efficacy. Several strategies have been recently proposed to improve the yield of CTCs isolation, including molecular binding technology to form red blood cells—tumor cell aggregates to significantly change the refractive index and enable their discrimination through optical tweezers (**Figure 4**A and Table 1).[63] Similarly, acoustic forces exploit sound waves to remotely sort CTCs (Table 1).[64,65] These forces are generated by sending high-frequency sound waves through a liquid medium, creating pressure gradients and velocity fields that can be used to manipulate the cells within the microfluidic channel (Figure 4B). Next, electrical forces have also been employed to capture CTCs on-chip, mainly through dielectrophoretic forces that exploit the

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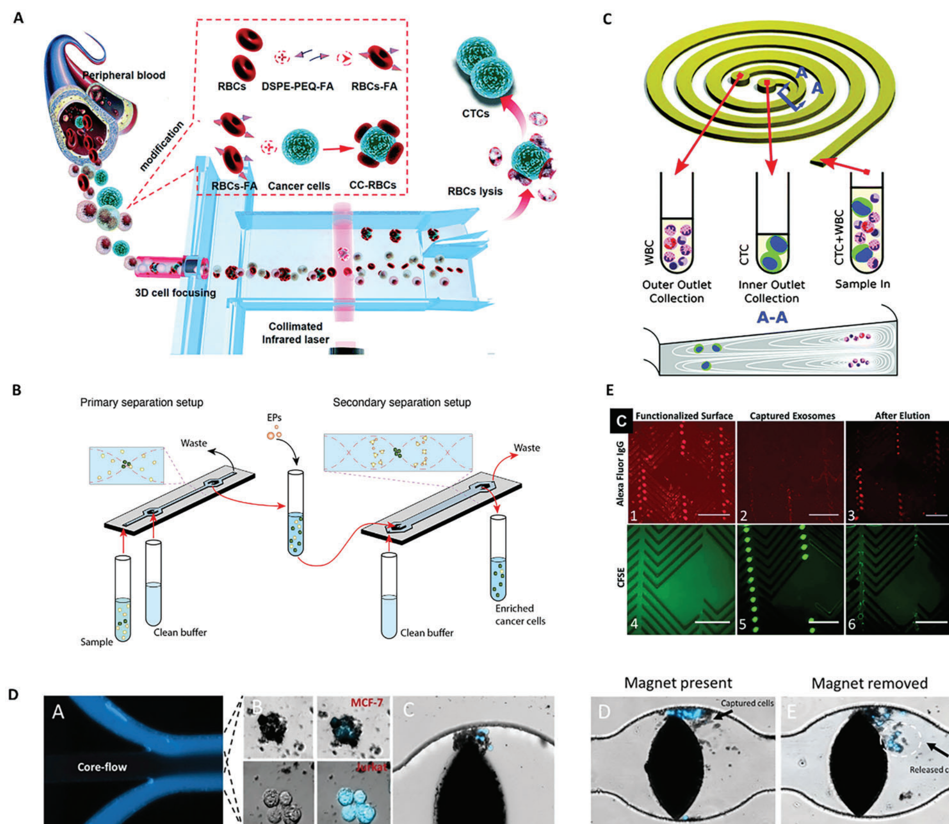

Figure 4. Microfluidic-based cancer traps. A) Optical-based methodology for CTCs isolated. The strategy involves binding tumor cells to homologous red blood cells resulting in noticeable differences in size and refractive index, which can then be separated from other blood cells using an optofluidic system under laser illumination. Reproduced with permission.[63] Copyright 2019, The Royal Society of Chemistry. B) Two-step acoustophoretic method for CTCs isolation from red blood cell lysed whole blood. The method uses an initial acoustofluidic pre-separation followed by a purging step to remove contaminating white blood cells. Reproduced under the terms of Creative Commons – Attribution 4.0 International CC BY 4.0.[64] Copyright 2021, The Authors, published by American Chemical Society. C) Spiral microfluidic device for the ultra-fast, label-free enrichment of CTCs from clinically relevant blood volumes. The technique utilizes Dean vortex flows and inertial lift forces to separate CTCs from smaller hematologic components, resulting in high purity and detection of CTCs from cancer patients. Reproduced under the terms of Creative Commons – Attribution-NonCommercial 3.0 Unported CC BY-NC 3.0.[68] Copyright 2014, The Authors, published by Royal Society of Chemistry. D) Magnetic separation of CTCs. Optical microscopes images of a) the microfluidic co-flow; b) hybrid microgel-decorated MCF7 breast cancer cells, and Jurkat cells; c) chamber with the captured cells by the micromagnets; and d,e) accumulation of MCF7 breast cancer cells after capturing and their magnetic release. Reproduced under the terms of Creative Commons – Attribution 4.0 International CC BY 4.0.[72] Copyright 2023, Wiley–VCH GmbH. E) Microfluidic chip containing a herringbone pattern coated with specific antibodies to capture ovarian cancer-derived exosomes. Reproduced under the terms of Creative Commons CC BY.[74] Copyright 2018, published by Royal Society of Chemistry.

differences in the dielectric properties of CTCs in response to an inhomogenous electric field.[66] For example, a new microfluidic device was recently reported for capturing CTCs using dielectrophoresis, which utilizes the pores of a porous membrane as traps for CTCs. The working mechanism was based on the dielectrophoretic force to efficiently capture and hold the CTCs for further analysis. The chip's performance was simulated and then physically tested, showing the ability to detect rare cells in a large cell population (Table 1).[67] Similarly, the physical differences with other blood components are in the foundation of CTCs discrimination through hydrodynamic inertial forces. In this case, the distinct size and density of CTCs enable their selective capture (Figure 4C and Table 1).[68] Typically, microfluidic designs include a spiral design whose curvature and number of turns are essential for selective CTCs isolation.[69] Finally, magnetic forces have also been described for capturing cancer cells. For this, magnetic nanoparticles can be designed to specifically target and bind to CTCs, mainly by functionalizing them with specific antibodies against well-known receptors (e.g., EpCAM, Her-2, EGFR, CD146, CD44, etc.) expressed in the outer membrane of cancer cells, allowing them to be captured and removed from circulation. This type of immunomagnetic separation is indeed a highly explored concept[70] and has been applied for the positive enrichment of CTCs. Of special mention is the well-known CellSearch commercial apparatus approved by the FDA that utilizes antibody-coated magnetic nanoparticles for the detection and capture of CTCs.[71] However, this system has relatively low sensitivity, is time-intensive, and requires a large sample volume. Due to the small number of CTCs found in 1 mL of blood (ranging from 1 to 100 CTCs), microfluidics is therefore better suited for the manipulation of low fluid volumes and biological tissues, particularly cells. Additionally, the aforementioned biomarkers

may not be present in all the CTCs as they may turn into a mesenchymal phenotype, leading to false negative findings. To minimize this though, a combination of several antibodies has been proposed to enhance the number of entrapped CTCs. In any case, several microfluidic-based platforms have been reported using magnetic nanoparticles decorated with specific antibodies to trap and isolate CTCs from blood samples rapidly. However, immunomagnetic separation in whole blood typically suffers from low capturing efficiency. Recently, soft micromagnet patterns with optimized geometry and magnetic material were integrated into a bilayer microfluidic chip to enhance the capture efficiency of CTCs labeled with magnetic nano/hybrid microgels (Figure 4D and Table 1).[72] The chip was specifically designed to optimize the capturing efficiency, allowing for high purity of target cells and real-time monitoring of their behavior. This method offers a simple, low-cost, and robust strategy for early-stage diagnosis and tracking of cancer-associated biomarkers. However, some limitations of this approach are the introduction of foreign beads, which may affect downstream analysis of the captured cells, and the antibody conjugation of the beads may be time-consuming and expensive.

The portfolio of microfluidic-based techniques for isolating and trapping CTCs is extensive, encompassing a variety of approaches that exploit the different mechanical and biological properties of CTCs. As discussed in Section 2, these techniques can vary in their specificity, sensitivity, and efficiency, making it essential to carefully consider the desired outcomes and limitations when selecting the best approach for a particular application. In addition, the cancer traps-on-a-chip used for isolating CTCs could also be adapted to capture other tumor-derived material, such as EVs, secreted by cancer cells.[73] These vesicles have been identified as pivotal mediators in cancer progression. They contain important information about the primary tumor, making their trapping and analysis a crucial aspect of liquid biopsies. These biopsies can provide noninvasive early diagnosis and lead to the development of personalized therapeutic strategies and better patient follow-up. An example of this strategy is the use of a microfluidic platform containing a herringbone pattern coated with antibodies targeting tumor-derived exosomes associated with ovarian cancer (Figure 4E).[74] The aim was to isolate exosomes from small amounts of serum from patients, and interestingly, the results showed that the concentration of tumor-derived exosomes increased as the disease progressed. This example highlights the potential of exosome analysis as a noninvasive tool for monitoring cancer progression. However, applying microfluidic chips in capturing EVs demands careful consideration of various factors to ensure efficient isolation and trapping. First, the size exclusion criteria must be adjusted to accommodate the smaller size of EVs compared to CTCs, for instance, by including smaller pores or by implementing filtration techniques that specifically target the size range of EVs (Table 1).[75] Similarly, the sensitivity of the detection methods must be enhanced. This can be achieved by incorporating on-chip highly sensitive detection methods, such as electrochemical (e.g., amperometry, impedance spectroscopy, etc.) or optical (e.g., Raman, colorimetric, fluorescence, etc.) biosensors, as recently reviewed.[73] More sophisticated methods include the combination of microfluidics and CRISPR technology to develop CRISPR-based biosensors for nucleic acid detection that may aid in the early detection of cancer.[76] This type of biosensors can target and cleave specific ctDNA fragments secreted by tumors containing, e.g., gene mutations, single-nucleotide polymorphism, or DNA methylation, among others, and microfluidics can process and analyze the samples quickly and accurately.[77] This combination of disruptive technologies has already enabled the accurate detection of ctDNA fragments, providing valuable information on the presence of cancer and genetic diseases (Table 1).[78,79] Though still in its early stages, this approach has the potential to transform cancer diagnosis and treatment, leading to better patient outcomes and a deeper understanding of the human genome.

Additional advantages of using microfluidic platforms for capturing tumor-derived material include their ability to process small sample volumes with a high level of precision and control, resulting in rapid processing times and facilitating high-throughput analysis, particularly beneficial for limited sample volumes such as liquid biopsies. Furthermore, microfluidic devices offer improved sensitivity, selectivity, and specificity by incorporating specific capture agents, enhancing the detection and isolation of CTCs, EVs, and ctDNA from complex biological samples. Integration with downstream analysis techniques, as discussed earlier, allows for seamless processing and analysis within the microfluidic device or transfer to other analytical platforms like polymerase chain reaction or sequencing systems, reducing the risk of sample loss or contamination.

Despite these advantages, there are limitations to consider when working with cancer traps-on-a-chip. Potential issues include clogging and the possibility of false-positive results due to nonspecific binding. Additionally, scalability and manufacturing consistency pose challenges in microfluidics, requiring attention to ensure reproducibility and scalability of devices across different laboratories or manufacturing processes. Addressing these limitations will further enhance the utility of microfluidics in tumor-derived material capture and analysis.

Finally, it is essential to contemplate the specificity of the isolation method of small tumor-derived materials, particularly EVs and ctDNA, since they can come from various tissues. To target these "biomaterials" derived from cancer cells, additional trapping methods may be necessary. One such approach is immunoaffinity-based isolation methods to selectively capture the targeted EVs and ctDNA. For the former, antibodies or aptamers targeting specific surface markers can be immobilized on microstructures within the microfluidic device. For the latter, specific probes or primers targeting known genetic mutations or alterations present in ctDNA can be exploited for their selective capture and retention. In conclusion, the use of microfluidic chips in EV and ctDNA isolation and characterization requires a thoughtful approach that considers various technical factors, such as size exclusion, detection sensitivity, and specificity, to achieve successful results.

4. Discussion

The potential of cancer traps to improve the efficacy of cancer diagnosis and treatment has motivated a growing body of research and development in this field, with the final goal of transforming the way we detect, treat, and monitor cancer. The relevance of using biomaterials-based cancer traps in early cancer detection and treatment is demonstrated by extensive in vivo research,

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including clinical trials,[80] tested in different tumor types (e.g., breast,[22] prostate,[81] melanoma,[82] ovarian,[14] and others). The cancer cells captured by these traps can offer crucial insights into the origin of the tumor. Importantly, these traps can be integrated into disruptive technologies, such as microfluidic platforms, to capture and examine CTCs and other tumor-derived material, such as EVs, ctDNA, or proteins. The use of these liquid biopsy methods is already widespread in academia and their adoption gaining more acceptance by the industry and the clinical community.

Similarly, implantable biomaterial-based traps are becoming a promising strategy to capture metastatic cells in vivo, particularly after surgery. For this, several challenges associated with the material employed to manufacture the traps may need to be addressed. One challenge is developing materials that are biocompatible and do not cause adverse reactions in the body, such as inflammation or other adverse reactions. Another challenge is developing materials that are able to effectively capture cancer cells without interfering with the normal functioning of the body. For example, a microfilter or sponge used to capture cancer cells in the bloodstream or lymphatic vessels should not block the flow of fluid or cause damage to vessels. In addition to these challenges, there are also logistical challenges associated with the use of implantable biomaterials. For example, these materials may need to be removed from the body after a certain period of time, which could require additional surgical procedures, particularly, if the captured cells need to be analyzed. Alternatively, the use of biodegradable biomaterials is preferred. Despite these issues, large efforts have been invested in the development of implantable cancer traps. In the future, this technology may play a role in the treatment of metastatic cancer and may help to improve the prognosis for patients. While these materials show promise, further research is needed to fully understand their safety and effectiveness and to determine their optimal use in clinical practice.[7]

5. Conclusion

Biomaterials-based cancer traps offer a promising complementary approach to traditional cancer therapies. These innovative medical devices utilize various mechanisms to selectively capture and remove cancer cells from the body, potentially reducing the need for more invasive treatments and improving patient outcomes. (Bio)chemical-based cancer traps utilize specific chemical agents to target and bind to cancer cells, while mechanical-based cancer traps utilize physical mechanisms to capture and remove cancer cells. Both types of cancer traps display advantages and limitations but have already shown promising results in capturing disseminating cancer cells. Furthermore, the use of microfluidics in combination with cancer traps has already demonstrated the potential to revolutionize liquid biopsy approaches to improve early cancer diagnosis by enabling the rapid and efficient capture, analysis and characterization of cancer cells, and other tumor-derived biomaterials, providing a wealth of information about the tumor at an early stage. Importantly, this information can guide the selection of the most effective treatment options and potentially improve patient outcomes.

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Conflict of Interest

The authors declare no conflict of interest.

Keywords

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