Organic semiconducting nanomaterials-assisted phototheranostics in near-infrared-II biological window

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Abstract
Phototheranostics conducted in the near-infrared-II (NIR-II) biological window exhibits high superiorities relative to that conducted in the first near-infrared (NIR-I) window due to higher penetration depth, higher signal-to-noise ratio of imaging, and improved maximum permissible exposure. Currently, most existing agents developed for NIR-II phototheranostics are limited to inorganic nanoparticles and small-molecule dyes, which suffer from the issues of long-term biotoxicity and poor photostability, respectively. Organic semiconducting nanomaterials (OSNs) constructed from π-conjugated organic semiconducting polymers or oligomers have unique advantages including high photostability, flexible optical features, and good biocompatibility due to the totally organic and biologically inert constituents. These advantages make OSNs more promising for in vivo phototheranostics. We here make a brief review about recent progress of OSNs-assisted phototheranostics conducted in the NIR-II biological window. NIR-II fluorescence imaging and photoacoustic imaging are mainly reviewed as diagnostic modalities to guide various treatments. Besides, multimodal phototheranostics using OSNs in NIR-II window is also described. At last, the challenges and outlook for OSNs-assisted NIR-II phototheranostics are discussed.

KEYWORDS
cancer therapy, fluorescence imaging, organic semiconducting nanoparticle, photoacoustic imaging, second near-infrared window
Phototheranostics refers to the diagnosis-guided treatment toward diseases based on light, which is becoming an emerging interdisciplinary technique with the combination of advanced diagnostic and therapeutic approaches. In phototheranostics, the light-based bioimaging, such as fluorescence imaging (FI), chemiluminescence imaging (CI), photoacoustic imaging (PAI), and photothermal imaging, plays the most effective role for disease detection and monitoring. Meanwhile, phototherapy, especially near-infrared (NIR) light-triggered phototherapy, has unique superiorities compared with the traditional therapeutic methods (eg, chemotherapy, radiotherapy, and surgery). For example, photothermal therapy (PTT), which is based on the nonradiative decay-induced photothermal conversion mechanism under light excitation, has significant advantages including minimal invasiveness, high selectivity to lesion sites, flexible maneuverability, and excellent therapeutic effect. Besides, photodynamic therapy (PDT) also acts as a decent therapeutic approach toward cancer or other diseases based on the generation of reactive oxygen species (ROS) induced by the photosensitizer (PS)-assisted light energy transfer to surrounding oxygen. Thus, rational combination of advanced imaging and therapeutic techniques not only enables accurate and effective treatment toward diseases but also provides real-time monitoring of therapeutic efficacy during and after the therapeutic process, which has attracted tremendous attention in biomedical field.

In phototheranostics, light source is an indispensable element. Compared with ultraviolet and visible light, NIR light shows relatively weak interactions with biological tissues, which is more suitable for in vivo theranostics. Nevertheless, current phototheranostics are mainly performed using the NIR light with a short wavelength ranging from 650 to 950 nm (NIR-I biological window), which undergoes a limited tissue penetration depth (around 1 cm) principally due to the light scattering in living subjects, thereby restricting the translational study of phototheranostics to some extent. By contrast, light scattering can be significantly suppressed at NIR-II biological window (1000-1700 nm) in various biological tissues, such as skin, brain, and muscle (Figure 1A), which is beneficial to increase the tissue penetration depth of phototheranostics. Besides, some major endogenous light absorbers including blood, skin, and fat exhibit a relatively weak absorption in NIR-II window (Figure 1B).

This is also favorable for in vivo phototheranostics. For instance, the weak extinction capability of endogenous light absorbers in the NIR-II window will lead to a remission of background fluorescence, photoacoustic, or photothermal signals, thereby improving the signal-to-noise ratio (SNR) for imaging and minimizing the side effects for PTT. Furthermore, biological tissues are usually more tolerant to NIR-II light, thus permitting much higher maximum permissible exposure (MPE) compared with that for NIR-I light. Such higher MPE makes it possible to utilize a laser with increased power density to conduct phototheranostics in vivo, which will further improve the imaging SNR and therapeutic outcomes.

Not all of the diseases can express endogenous molecules with NIR-II absorption or emission; the development of exogenous agents with effective responsiveness to NIR-II light is of great importance for NIR-II phototheranostics. Currently developed agents for bioimaging and therapy in NIR-II window are mainly focused on inorganic nanomaterials (eg, quantum dots, rare-earth nanoparticles, and metallic nanoparticles), small molecules, and nanoparticles.

**FIGURE 1** Interactions between biological tissues and light with different wavelengths. (A) Scattering coefficients of different biological tissues toward light with different wavelengths ranging from visible to NIR-II window. Reprinted with permission from Springer Nature. (B) Effective attenuation coefficients of some biological components including oxygenated whole blood, deoxygenated whole blood, skin, and fat as a function of different wavelengths of light. Reprinted with permission from Springer Nature.
molecule dyes (SMDs),\textsuperscript{27-29} and organic semiconducting nanomaterials (OSNs).\textsuperscript{30-32} For such kind of inorganic nanomaterials, the potential biotoxicity resulting from heavy metal ions and metabolic issues should be considered further. SMDs such as cyanine derivatives and porphyrin are widely exploited for NIR-II phototheranostics with good biocompatibility. However, these organic dyes often encounter the issues of poor photostability and unsatisfactory tumor accumulation. In contrast, OSNs prepared from $\pi$-conjugated organic semiconducting polymers (OSPs) or oligomers (OSOs) have unique advantages for in vivo phototheranostics. On the one hand, OSNs consist of entirely organic and biologically inert constituents, thereby intrinsically circumventing the issues associated with inorganic nanomaterials including heavy metal ion-induced toxicity to living organisms.\textsuperscript{33} On the other hand, OSNs exhibit higher photostability relative to some SMDs,\textsuperscript{34} more excellent light-harvesting ability,\textsuperscript{35} more tunable absorption and emission profiles,\textsuperscript{36} and more controllable size\textsuperscript{27} compared with other inorganic optical nanomaterials. All these merits make OSNs promising for diverse biomedical applications in NIR-II window, especially in vitro or in vivo bioimaging (eg, cell imaging and tracking,\textsuperscript{38} tumor imaging,\textsuperscript{39} lymph node mapping,\textsuperscript{40} cardiovascular imaging,\textsuperscript{41} and real-time imaging of drug-induced hepatotoxicity\textsuperscript{42}) and imaging-guided therapy (eg, PTT\textsuperscript{43} and surgery\textsuperscript{40}).

In this contribution, we review the recent progress of OSNs-assisted phototheranostics conducted in the NIR-II biological window (Scheme 1). We first introduce the features and advantages of NIR-II FI and review the NIR-II FI-guided therapy including surgery, chemotherapy, and PTT. Then NIR-II PAI-based phototheranostics using OSNs is summarized. Subsequently, multimodal phototheranostics in NIR-II window such as NIR-II FI/PAI-guided PTT/PDT is described. At last, a brief summary is given and the challenges and outlook of OSNs used for NIR-II phototheranostics are discussed.

2 | **NIR-II FI-GUIDED THERAPY**

In vivo NIR-II FI has become a research hotspot recently due to its salient superiorities over the FI conducted in visible light or NIR-I light region. Irradiation of NIR-II light or detecting NIR-II photons provides reduced light scattering in biological tissues together with weaker autofluorescence, which is beneficial to improve the penetration depth and SNR of imaging.\textsuperscript{44} The NIR-II window can be further separated into NIR-IIa and NIR-IIb subwindows, which are identified as the spectra ranging from 1300 to 1400 nm and 1500 to 1700 nm, respectively.\textsuperscript{45} The inverse wavelength dependence of photon scattering\textsuperscript{46} reflects that the longer wavelength in NIR-IIb window lead to weaker light scattering in the whole NIR-II region. Furthermore, the autofluorescence of biological tissues can be minimized beyond 1500 nm,\textsuperscript{47} further improving the imaging SNR. These indicate the high potential of NIR-IIb window for clinical imaging and imaging-guided therapy.

2.1 | **Surgery**

Surgery is an old and widely employed technique for disease therapy especially tumor elimination. For the surgeon, a big challenge is to visually delineate the profile of lesions and identify their margins for accurate treatment. NIR-II FI is competent for this task because it possesses high sensitivity as well as excellent spatial and temporal resolutions. To date, NIR-II FI-guided surgery mainly focuses on tumor resection, sentinel lymph nodes resection, and vascular embolization. Hong et al reported the synthesis of benzobisthiadiazole (BBT)-based semiconducting oligomer nanoparticles (H1 NPs) and the application for sentinel lymph nodes surgery.\textsuperscript{48} Oligomer H1 (Figure 2A) was modified by PEGylated amphiphile (DSPE-mPEG\textsubscript{5000}) to prepare biologically used H1 NPs, which showed an average diameter of 70.0 nm (Figure 2B). The emission peak of H1 NPs was located at about 1100 nm, making it possible for NIR-II FI in vivo. After verifying the biocompatibility, H1 NPs were injected into C57BL/6J mice to visually determine the sentinel lymph node via NIR-II
FI. The sentinel lymph node was then completely resected without unwanted damage to surrounding normal tissues (Figure 2C), benefiting from the high imaging sensitivity and resolution of NIR-II FI assisted by H1 NPs.

Later, Cheng et al developed a diketopyrrolopyrrole-based semiconducting polymer nanoparticle (PDFT1032) as a NIR-II fluorescent probe for in vivo NIR-II FI and imaging-guided tumor surgery. PDFT1032 was constructed from furan-containing diketopyrrolopyrrole polymers and amphiphilic DSPE-mPEG via nanoprecipitation approach (Figure 2D), which exhibited ideal monodispersity and homogeneity with a particle size of 68 nm in PBS. Optical investigations revealed a strong absorption peak at 809 nm with a main emission peak at 1032 nm of PDFT1032 dispersed in PBS solution (Figure 2E). Toxicity studies showed that PDFT1032 had favorable biocompatibility both in vitro and in vivo, indicating high promise for clinical application. Then an orthotopic osteosarcoma model was established to demonstrate the tumor surgery under the guidance of NIR-II FI. After intravenous injection of PDFT1032, the tumor profile can be clearly visualized with high contrast, providing accurate resection of orthotopic lesions (Figure 2F). Even more powerful, the residual lesions after surgery and micrometastasis, as well as lymph node, can be sensitively identified using PDFT1032-assisted NIR-II FI (Figure 2G), thus making
it possible for further resection to prevent recurrence of cancer to the greatest extent. In addition, embolotherapy, which may be a more suitable therapeutic approach for patients who show a poor response to chemotherapy or have recurrent or unresectable tumors, was also successfully conducted with the help of NIR-II FI. Upon the intravenous injection of PDFT1032 into mice with a tumor located at the proximal femur for 5 min, the branched femoral artery that supports the tumor and its vascular network were vividly depicted (Figure 2H [i and ii]). Mediated by NIR-II FI, the temporary thrombus was established by using a vessel clamp to obstruct the blood flow. Subsequently, the major artery was incised and the tumor was resected with the successful retention of femoral artery (Figure 2H [iii and iv]), and this reflected high practical feasibility of NIR-II FI for vascular surgery.

### 2.2 Chemotherapy

FI is an extensively used tool to indicate drug delivery and release, as well as to monitor the efficacy of chemotherapy mainly due to its high imaging sensitivity and temporal resolution. However, most existing FI techniques for the guidance of chemotherapy are limited to visible light and NIR-I window, thus compromising the further clinical translation toward deep tissues. To date, NIR-II FI-guided chemotherapy assisted by OSNs is rarely reported. For example, based on their previous work on developing a strategy to improve the NIR-II fluorescence quantum yield by mimicking organic environment in aqueous media, Dai et al further reported OSNs composed of organic semiconducting oligomer (FE), anticancer drug (paclitaxel), and amphiphilic polymer (PS-g-PEG) for NIR-II FI-guided chemotherapy. FE was designed as a “S-D-A-D-S” structure with BBT core (Figure 3A), which was proved as an optimal architecture for relatively high NIR-II fluorescence efficiency. Then, PS-g-PEG was employed to encapsulate PA and paclitaxel to prepare water-dispersed nanotheranostics, which were further modified with folic acid (FA) for cancer targeting (Figure 3B). In vitro imaging was first investigated by using 4T1 cells and HEK-293T cells, which are FA-receptor positively and negatively expressed cells, respectively. NIR-II FI reflected that 4T1 cancer cells treated by FA-modified OSNs showed nearly 5.6-fold signal enhancement compared with HEK-293T cells that received the same treatment (Figure 3C), thus verifying the effective targeting ability. In vivo experiments showed a gradual signal (NIR-II signal above 1300 nm) increase at tumor site after intravenous injection of FA-modified OSNs, which exhibited the maximum tumor to normal tissue signal ratio (T/NT) of (20.0 ± 2.3) at 24 h postinjection (Figure 3D). By contrast, OSNs without FA modification indicated a lower accumulation at tumor area (maximum T/NT = 12.9 ± 1.4). The specific targeting makes it possible to deliver paclitaxel to tumor region to the greatest extent, thereby achieving the optimal therapeutic effect. Subsequently, the chemotherapy process of FA-modified OSNs was monitored by recording the NIR-II fluorescence signals overtime from tumor region, which indicated an obvious tumor inhibition effect and complete elimination of tumor after 20 days (Figure 3E).

Compared with some conventional drugs for chemotherapy, such as doxorubicin (DOX), paclitaxel, and camptothecin, Pt(II) metallacycles-based supramolecular coordination complexes (SCCs) service as not only efficient anticancer drugs but also biosensors for life-related analytes. Nevertheless, such SCCs often have limitations such as poor photostability and insufficient tumor uptake. Particularly, there is lacking in effective imaging techniques with deep penetration depth to guide the chemotherapy using Pt(II) metallacycles-based SCCs. To overcome this, Stang et al designed a multifunctional nanotheranostics that consists of Pt(II) metallacycles-based SCCs, “D-A-D”-structured semiconducting oligomers, and DSPE-mPEG5000 (Figure 4A). This nanoformula combined therapeutic drugs and imaging agents together to realize NIR-II FI-guided chemotherapy toward cancer (Figure 4A). In vivo diagnosis was conducted in HepG2 tumor models. After injection of the nanoformula, NIR-II fluorescence signals from the tumor site increased gradually over time and reached a maximum at 24 h postinjection, and the tumor could be clearly depicted at this time point (Figure 4B). It should be noted that the NIR-II fluorescence can be detected from the tumor region even after 14 days, thus making it possible for long-term monitoring of the therapeutic response. In vivo therapy exhibited that the nanoformula showed the optimal antitumor efficacy in living mice compared with control groups (mice treated with PBS or cisplatin) (Figure 4C), which can be attributed to the effective delivery of Pt(II)-based drug to tumor sites. Besides, changes of mice body weight and survival curves indicated a much better biocompatibility of the nanoformula relative to the free Pt drug (Figure 4D), which provided a biologically available nanotheranostic platform for NIR-II FI-guided chemotherapy using Pt(II) metallacycles-based SCCs.

### 2.3 Photothermal therapy

OSNs are an excellent class of fluorescent nanomaterials due to their high extinction coefficient, high fluorescence brightness, large Stokes shift, and high photostability. Besides, NIR-absorbing OSNs generally show ideal pho-
to thermal capability due to nonradiative decay process under light excitation. This difunctional fluorescence and photothermal signal output of OSNs lays a theoretical foundation for FI-guided PTT. For NIR-II FI-guided PTT, two main problems should be concerned. The first one is how to simultaneously improve the NIR-II fluorescence and photothermal signals because they are competitive processes. By constructing J-aggregates from the preparation process of semiconducting bispyrrole-sq-bispyrrole (SQP) nanoparticles, Fan et al introduced a novel strategy to enhance the NIR-II fluorescence brightness and PTT effect in a simultaneous manner. SQP was first designed with electron donor-acceptor architecture (Figure 5A) and successfully synthesized with 13% yield. Then water-dispersed nanoparticles were obtained by encapsulating SQP into an amphiphilic copolymer (F-127). In this process, nanoparticles (SQP-NPs(\text{J})) with both H- and J-aggregate features can be formed when SQP was initially dissolved in tetrahydrofuran, whereas only H-aggregates (SQP-NPs(\text{H})) were observed when the initially used solvent was dichloromethane. The redshifted absorption peak at approximately 901 nm of SQP-NPs(\text{J}) allowed enhanced NIR-II brightness compared with SQP-NPs(\text{H}) by 4.8-fold improvement under the same concentration (Figure 5B). Furthermore, the sharp absorption peak resulting from J-aggregation made it possible for effective PTT conducted in longer wavelength. In vivo imaging of mice brain and body showed remarkable superiority of SQP-NPs(\text{J}) relative to SQP-NPs(\text{H}) with much stronger fluorescence brightness. With the NIR-II fluorescence visualization of tumor, LED light lamp (810 nm, 0.8 W/cm²) was employed to irradiate the tumor-bearing mice after injection of SQP-NPs(\text{J}), which exhibited excellent tumor inhibition effect.
The second problem is the limited excitation laser with relatively short wavelength in NIR-I window for most reported NIR-II FI. In such, light scattering still cannot be ignored, although NIR-II fluorescence signals are recorded. To improve the deep tissue imaging quality and therapeutic efficiency, Fan et al demonstrated the application of 1064-nm activatable NIR-II FI-guided NIR-II PTT by using squaraine-based semiconducting polymer nanoparticles (PSQPNs-DBCO). As mentioned above, Fan’s group developed squaraine-based nanoparticles excited by a wavelength of 808 nm to achieve bright NIR-II FI and PTT. Based on this, squaraine-based polymer (PSQP) with redshifted excitation and emission wavelength was further obtained by extending the conjugated length of semiconducting backbone. Water-dispersed nanoparticles (PSQPNs) were prepared by self-assembly process using amphiphilic DSPE-PEG$_{5000}$-NH$_2$, which were further modified with alkynyl groups on the nanoparticle surface to obtain PSQPNs-DBCO (Figure 5C). The PSQPNs-DBCO showed maximum emission peak at 1290 nm under 1064 nm laser excitation, making it possible for NIR-II-triggered NIR-II FI and therapy. As expected, PSQPNs-DBCO exhibited enhanced SNR by fourfold compared with the reported TT-3T CPs, which is a 808-nm light-responsive NIR-II fluorescence dye. This proved the high superiority of NIR-II FI triggered by NIR-II light relative to that triggered by NIR-I light, mainly resulting from the suppressed photon scattering of NIR-II light in biological tissues. To further improve the imaging quality and PTT efficacy, biorthogonal labeling strategy (Figure 5D) was employed both in vitro and in vivo, which indicated extra improvement of imaging SNR by 2.5-fold in the tumor region compared with the no labeling group. All these merits of PSQPNs-DBCO permitted 1064-nm laser-triggered NIR-II FI and PTT, realizing specifically targeted diagnosis and efficient treatment toward colorectal tumor in living mice.

### 3.1 NIR-II PAI

PAI is a noninvasive diagnostic technique that relies on photoacoustic effect, which is developing to a widely used imaging modality for biomedical applications. Compared with conventional FI that relies on a “light in/light out” process, PAI is a “light in/sound out” approach, which is based on the thermal expansion of target tissue or light-absorbing agents under nanosecond laser irradiation. Therefore, PAI perfectly inherited the advantages of FI and ultrasonic imaging, providing high sensitivity, high spatial resolution, and deep penetration depth. Current PAI principally adopts NIR-I laser as the excitation light source. However, PAI conducted in NIR-II window has rarely been developed.
As NIR-II light has intrinsic superiorities of reduced photon scattering, deeper penetration depth, and higher MPE, PAI performed in NIR-II window should be more advantageous for in vivo applications especially for deep tissues. In addition, the acoustic signal attenuates far less than light in biological tissues, thus enabling NIR-II PAI superior over NIR-II FI to some extent. For example, Bian et al developed a semiconducting polymer-based nanoprobe (OSPNs⁺) for PAI and tracking of stem cells in the NIR-II window. Comparison study showed that NIR-II PAI had much higher SNR relative to NIR-I PAI at the same OSPNs⁺ concentration and imaging depth. Specifically, when the sample concentration was fixed at 0.286 mg/mL, the NIR-II photoacoustic signals from...
3.2 Photothermal therapy

The optical properties of OSPs or OSOs can be easily tuned by constructing the semiconducting backbone with different electron donor-acceptor pairs, thereby realizing red-shift of the absorption band to NIR-II window. In addition, OSPs- or OSOs-based OSNs have excellent light-capturing capability, strong photostability, good biocompatibility, and design flexibility, which make them good candidates for NIR-II PAI-guided PTT. To date, OSPs that have strong NIR-II absorption profile are mainly focused on BBT-, thiadiazolobenzotriazole-, thiadiazoloquinoxaline-, and thiophene-fused benzodifurandione-based oligo (p-phenylenevinylene)-based skeleton structures, and their in vivo phototheranostics were widely investigated.

Fan et al designed a series of semiconducting polymer nanoparticles (SPNs) with absorption beyond 1000 nm for efficient PAI-guided PTT in the NIR-II window. In this work, a new class of OSPs (SP1, SP2, and SP3) with D-A1-D-A2 structure were developed via adopting two-acceptor strategy, in which the donor (D, thiophene) and the first acceptor (A1, diketopyrrolopyrrole [DPP]) were totally the same, whereas the second acceptor (A2) can be finely tuned via employing acceptors with different electron-withdrawing properties (Figure 6A). Different acceptors resulted in different absorption and photothermal conversion abilities. Among these three SPNs, SPN3 with the strongest electron-withdrawing moiety showed not only a far-reaching absorption peak of 1300 nm, but also high photothermal conversion efficiency (PCE) of 60% at 1064 nm. Strong PA signals from SPN3 at 1300 nm were detected and good PTT effect was achieved under the guidance of PAI in living mice. This study demonstrated the methodology for exploring NIR-II SPNs with maximum performance through a molecular engineering approach.

Among various malignant tumors, gliomas are the most aggressive and wildly uncontrolled. There still remains a huge challenge for precise theranostics of gliomas mainly due to the strongly infiltrative feature, the existence of blood-brain barrier (BBB), and the relatively deep sites of niduses inside the brain. For this, Liu et al developed NIR-II absorptive OSNs for targeted PAI and imaging-guided PTT toward brain tumor through scalp and skull (Figure 6B). BBT-based OSP was designed and synthesized, which was encapsulated by DSPE-PEG-Mal to obtain water-dispersed PI NPs. Then c-RGD was further decorated to PI NPs surface, achieving PIRGD NPs. The PIRGD NPs showed strong absorption in NIR-II region, and the extinction coefficient in 1064 nm was measured as high as 22.6 L/g/cm in aqueous solution. In vitro studies showed that cancer cells treated with PIRGD NPs exhibited higher photoacoustic brightness than that treated with PI NPs by 3.5-fold, indicating the positive contribution of c-RGD to enhance tumor cell uptake. In the presence of 1064-nm laser irradiation, both P1 NPs- and PIRGD NPs-treated tumor cells showed severe apoptosis compared with the control group. However, “PIRGD NPs + laser” led to higher cell killing effect than “PI NPs + laser,” indicating enhanced PTT efficacy of PIRGD NPs in vitro. Upon intravenous injection of nanoparticles, the brain tumor was gradually visualized over time via PAI, which reflected that the prepared nanoparticles can cross the BBB to effectively accumulate into brain tumor. As expected, the photoacoustic signals of brain tumor from mice treated with PIRGD NPs were always higher than that treated with PI NPs at different postinjection time points, revealing the effective targeting ability of PIRGD NPs in living body. Under the guidance of PAI, PTT toward orthotopic brain tumor was performed at 24 h postinjection. During 1064-nm laser irradiation, the maximum temperature of brain from PIRGD NPs-treated mice reached 48.4°C, higher than that from PI NPs-treated mice (43.6°C). Impressively, the size of brain tumor in “PI RGD NPs + laser” group showed the smallest compared with the “PI NPs + laser” and control groups as indicated by hematoxylin and eosin staining of brain slices after PTT. This work first demonstrated NIR-II PAI-guided PTT toward orthotopic brain tumor through scalp and skull by using OSNs, providing a vanguarding exploration on deep tissue phototheranostics.

However, abovementioned water-dispersible OSNs were mainly fabricated by using an amphiphilic encapsulating matrix (eg, Pluronic F-127 or DSPE-PEG). The introduction of encapsulating matrix may suffer the problems of nanoparticles instability and dissociation during circulation in living body. With these concerns, Bian et al designed an organic semiconducting polymer amphiphile (OSPA) for PAI-guided PTT under 1064-nm laser illumination (Figure 6C). This OSPA was equipped with a D-A-conjugated backbone as the photoacoustic/photothermal generator and covalently modified with poly (ethylene glycol) (PEG) responsible for water solubility and biocompatibility. Different from reported NIR-II PAI/PTT agents prepared using nanoprecipitation method, the OSPA itself is an amphiphilic polymer, and thus it can self-assemble into nanoparticles independent on extra encapsulating matrix, enhancing structural stability. Except for structural stability, the OSPA showed relatively high PCE and good biocompatibility. Comparison studies revealed higher penetration depth of NIR-II PAI over NIR-I PAI using OSPA,
making it a good candidate for deep tissue imaging and therapy. In vitro experiments showed excellent PAI and PTT efficacy of OSPA toward 4T1 cancer cells. Good therapeutic effect of the OSPA was also confirmed in in vivo experiments under the guidance of PAI using 1064-nm laser irradiation. This study provided a good example with respect to NIR-II light-absorbing OSNs with single-component architecture for PAI-guided PTT.

It is well known that semiconducting polymer backbones are usually big and rigid, generally leading to the problem of hard metabolism, which may cause long-term toxicity concern. To address this issue, Yuan’s group reported a metabolizable semiconducting polymer dots (Pdots) for NIR-II phototheranostic. In this study, ultrasmall Pdots (S-Pdots) with average diameter of 4 nm were fabricated via a modified nanoprecipitation method. The
as-prepared Pdots showed excellent photothermal performance with a high PCE of 53.1%, guaranteeing their anticancer capability. On the other hand, after exerting the photothermal effect and ablating tumors, the S-Pdots can be more quickly metabolized from body by comparison with the Pdots with larger size, which eliminate the potential long-term toxicity concern. NIR-II PAI experiments visualized the Pdots accumulation trend at tumor site with prolonged time and determined the optimal therapeutic time points for next NIR-II PTT. A good tumor eradication efficacy was achieved in vivo under 1064-nm laser irradiation. This work provided a new thought for developing metabolizable OSNs for NIR-II PAI-guided PTT, which may accelerate the clinical translation of OSNs in biomedical field.

4 | NIR-II MULTIMODAL PHOTOTHERANOSTICS

4.1 | Multimodality versus single modality

Multimodal phototheranostics conducted in NIR-II biological window has unique advantages compared with single-modal theranostics. For diagnosis, the combination of various imaging modalities into one entity can achieve complementarity of functions to provide more accurate diagnosis. For example, FI is a rapid diagnostic technique and featured with high imaging sensitivity. However, the imaging resolution is relatively poor mainly due to the light scattering. Magnetic resonance imaging (MRI) is a noninvasive approach for tumor detection that provides high physiological and anatomical resolution, and should be a good partner for FI.71 Similarly, multimodal therapy is also more powerful to obtain a satisfying therapeutic outcome than single-modal treatment. For instance, although PTT shows perfect tumor ablation capability, the residual tumor tissues would be able to recrudesce resulting from the acquired thermal-resistance ability of residual cancer cells mainly due to the existence of heat-shock protein.72 PDT is also a noninvasive phototherapeutic technique, which is able to cause cell apoptosis or death by toxic ROS resulting from PS-assisted light energy transfer to surrounding oxygen. Nevertheless, the oxygen-dependent property of PDT largely restricts its practical use in tumor treatment due to the hypoxic microenvironment for most of the malignant tumors.73 Therefore, the combination of different therapeutic modalities will make up for each other’s shortcomings and thus improve the efficiency of tumor killing. Taking together, multimodal theranostics has significant superiorities to realize accurate and high-efficiency therapy to the greatest extent.

4.2 | Multicomponent architecture-based OSNs

The most convenient and facile approach to endow OSNs with multimodal theranostic capability is to integrate diverse functional components into one entity. Fan et al developed an “All-in-One” phototheranostics based on organic semiconducting small molecule (DPP-BT) for NIR-II fluorescence/PAI-guided photothermal/photodynamic/chemo combination therapy (Figure 7).74 The DPP-based semiconducting small molecule was first synthesized, which served as the photoacoustic/NIR-II fluorescence signal generator and the PDT/PTT therapeutic agent. Then the multifunctional nanoparticles, (DPP-BT/DOX) NPs, were prepared via self-assembled process using DPP-BT, anticancer drug (DOX), organic phase-change material (PCM), amphiphilic lecithin, and DSPE-PEG-FA. In the presence of a single-NIR laser (730 nm) irradiation, (DPP-BT/DOX) NPs can not only emit strong NIR-II fluorescence and photoacoustic signals, but also exhibit excellent photothermal and photodynamic performances. In addition, the PCM matrix melted in the heat from photothermal effect of DPP-BT, accelerating the release of loaded DOX to further improve the overall therapeutic efficiency. In vitro studies showed a gradual signal increase of both PAI and NIR-II FI from Hela cells treated with (DPP-BT/DOX) NPs under increasing concentrations, which verified the effective cellular uptake. In vivo studies showed that the NIR-II fluorescence and photoacoustic signals from tumor region gradually enhanced over time after injection of (DPP-BT/DOX) NPs, and the signal maximum reached at 24 h postinjection, indicating high-efficiency accumulation of (DPP-BT/DOX) NPs into tumor sites. At the optimal postinjection time point of 24 h, a 730-nm laser (1.0 W/cm2) was employed to irradiate the tumor region for 10 min. The combinational PTT/PDT/chemo therapy in “P(DPP-BT/DOX) NPs + laser” group showed much more efficient tumor suppression compared with “P(DPP-BT) NPs + laser” group and those without laser irradiation, thus proving the enhanced therapeutic outcome of (DPP-BT/DOX) NPs in a synergistic manner under single-laser excitation.

Apart from the combination of fluorescence and PAI as the diagnostic approach, some other clinical imaging techniques, such as positron emission tomography (PET) and MRI, can also be integrated into a hybrid nanosystem for multimodal imaging-guided therapy. For example, Cheng et al reported the integration of three different components (mesoporous silica nanoparticle, melanin dot, and supported lipid bilayer) for constructing the multifunctional hybrid platform.75 In this platform, mesoporous silica nanoparticle served as a uniform nanocarrier to adsorb melanin dot, which acted as a chelator to 64Cu2+...
Multicomponent architecture-based OSNs for multimodal phototheranostics. Schematic illustration of the preparation process of P(DPP-BT/DOX) NPs and NIR-II FI/PAI-guided PDT/PTT/chemotherapy against tumor. Reprinted with permission from Wiley.

The supported lipid bilayer was then employed to envelop the nanoparticles for colloidal stability and reduction of unwanted release of loaded melanin dot. Subsequently, the semiconducting small molecule, CH-4T, was embedded into the lipid bilayer to endow the nanoplatform with NIR-II FI capability. Impressively, the fluorescent brightness of CH-4T improved significantly (~4.27 times enhanced) after accommodation by lipid bilayer, which is beneficial for effective NIR-II FI. The developed bimodal nanoplatform was then applied in NIR-II FI/PET imaging-guided tumor surgery. Because PET has high imaging sensitivity and deep penetration depth, it is an ideal tool for tumor diagnosis and delineation. Meanwhile, thanks to the amplified NIR-II fluorescence, the nanoplatform was successfully utilized to delineate the tumor profile and vasculature, thereby providing a visualized window to precisely guide the tumor resection.

4.3 Single-component architecture-based OSNs

“One component, multiple functionalities” is more advantageous in practical use because nanoprobes with multicomponent architecture are generally constructed via a complicated procedure, which makes it difficult to reproduce. Besides, the prepared nanoprobes often
suffer from the issue of poor structural stability due to the possible leakage of multitudinous components inside the nanoprobes. Thus, the development of single-component architecture-based nanoprobe will avoid these problems to a great extent. Fan et al developed a single component-based OSNs constructed from organic semiconducting oligomer (DPP-BDT) and DSPE-mPEG5000 for NIR-II FI/PAI-guided PDT/PTT (Figure 8). The DPP-BDT was first designed and synthesized, which showed strong absorption in NIR-I region and emission in NIR-II window. Through nanoprecipitation approach, multifunctional OSNs with good water solubility, ideal biocompatibility, and high photostability were achieved. In the presence of NIR light irradiation, the prepared OSNs could simultaneously generate hyperthermia and toxic ROS. In vitro studies showed excellent cancer cells killing efficacy by using the OSNs together with light irradiation. In vivo NIR-II FI/PAI indicated an effective accumulation of the OSNs at tumor sites, which made it possible for therapy. Under the guidance of dual-modal imaging, synergistic treatment toward tumor was conducted, showing excellent tumor inhibition effect. By the similar way, Wang et al developed a DPP-based semiconducting polymer (DPP-TT) and further modified it into a versatile phototheranostic nanoagent for NIR-II fluorescence/photoacoustic/thermal imaging-guided PTT. In spite of the advantages of single-component architecture-based OSNs, development of OSPs or OSOs with desired versatility is still a challenge, which should be further investigated.

5  |  SUMMARY AND OUTLOOK

In summary, recent developments of OSNs-assisted phototheranostics in the NIR-II window were briefly reviewed. NIR-II phototheranostics have remarkable superiorities compared with NIR-I phototheranostics in practical use, such as deep penetration depth and high imaging SNR. This is mainly attributed to the reduced photon scattering of NIR-II light in biological tissues, weak absorption of NIR-II light by tissues, and weak autofluorescence in NIR-II region. Although numerous nanoagents have been developed for NIR-II phototheranostics, most existing fluorophores or light absorbers for NIR-II FI or PAI are mainly focused on inorganic nanomaterials and small...
molecule dyes, which may suffer from the issues of long-term biotoxicity and poor photostability, respectively. Development of a new class of nanomaterials with excellent optical properties and good biocompatibility is highly desired to advance NIR-II phototheranostics.

OSNs are becoming a promising generation of nanomaterials for NIR-II phototheranostics due to their flexible absorption and emission band, structural versatility, and biologically inert feature. Nevertheless, several concerns remain to be addressed for OSNs-assisted NIR-II phototheranostics. First, the NIR-II fluorescence of OSNs is prone to quench in aqueous environment due to the “aggregation-caused quenching” effect of NIR-II fluorophores in water. Although aggregation can lead to the enhancement of photoacoustic signal to some extent, the attenuated fluorescence will cause poor temporal resolution for in vivo imaging. Besides, most of currently developed NIR-II OSNs can only be excited by NIR-I light to emit NIR-II fluorescence. OSNs with NIR-II excitation and NIR-II emission features are scarcely explored. Second, biosafety concern. Although reported OSNs showed excellent biocompatibility and no obvious toxicity to small animals, hepatobiliary metabolism is the major pathway for the clearance of OSNs. Therefore, the long-term toxicity of OSNs to major organs remains to be further evaluated. To solve this problem, development of OSNs with good biodegradability or ultrasmall size (below 5 nm) to realize fast renal clearance is of great significance and in high demand.78 Third, the structural diversity of OSPs or OSOs should be further broadened because NIR-II light-absorbing OSPs or OSOs are still rarely explored. This relies on the development of fundamental research and basic theory to guide the construction of new OSPs or OSOs with NIR-II responsiveness. With the rapid development of materials science, nanotechnology, and advanced theranostic instruments, these problems will eventually be solved in the near future, furthering the clinical translation of OSNs-assisted NIR-II phototheranostics to make contributions to improve human health.

CONFLICT OF INTEREST
The authors declare no conflict of interest.

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