Pharmacological Research 117 (2017) 261–266

Contents lists available at ScienceDirect

Pharmacological Research

journal homepage: www.elsevier.com/locate/yphrs

Original Article

Polymer-coated nanoparticles: Carrier platforms for hydrophobic water- and air-sensitive metallo-organic compounds

Daniel Valdeperez[a,1], Tianqiang Wang[a,b,1], Jens P. Eußner[c], Bastian Weinert[c], Jianyuan Hao[b], Wolfgang J. Parak[a,d], Stefanie Dehnen[c,*], Beatriz Pelaz[a,*]

[a] Fachbereich Physik, Philipps-University Marburg, Renthof 7, 35037 Marburg, Germany
[b] School of Microelectronics and Solid State Electronics, University of Electronic Science and Technology of China, Chengdu 610054, China
[c] Fachbereich Chemie and Wissenschaftliches Zentrum für Materialwissenschaften, Philipps-University Marburg, Hans-Meerwein Straße, 35043 Marburg, Germany
[d] CIC biomaGUNE, San Sebastián, Spain

ARTICLE INFO

Article history:
Received 27 June 2016
Received in revised form
19 December 2016
Accepted 22 December 2016
Available online 23 December 2016

Keywords:
Nanoparticles
Metallodrugs
Drug carriers
Catalytic activity
Clusters

ABSTRACT

Many of the relevant compounds for anticancer therapy are metal-based compounds (metallodrugs), being platinum-based drugs such as cisplatin, carboplatin (Paraplatin®), and oxaliplatin (Eloxatin®) the most widely used. Despite this, their application is limited by issues such as cell-acquired platinum resistance and manifold side effects following systemic delivery. Thus, the development of new metal-based compounds is highly needed. The catalytic properties of a variety of metal-based compounds are nowadays very well known, which opens new opportunities to take advantage of them inside living cells or organisms. However, many of these compounds are hydrophobic and thus not soluble in aqueous solution, as they lack stability against water or oxygen presence. Thus, versatile platforms capable of enhancing the features of these compounds in aqueous solutions are of importance in the development of new drugs. Surface engineered nanoparticles may render metallodrugs with good colloidal stability in water and in complex media containing high salt concentration and/or proteins. Herein, polymer coated nanoparticles are proposed as a platform to link insoluble and water/oxygen sensitive drugs. The linkage of insoluble and oxygen sensitive tin clusters to nanoparticles is presented, aiming to enhance both, the solubility and the stability of these compounds in water, which may be an alternative approach in the development of metal-based drugs. The formation of the cluster-nanoparticle system was confirmed via inductively coupled plasma mass spectrometry experiments. The catalytic activity and the stability of the cluster in water were studied through the reduction of methylene blue. Results demonstrate that in fact the tin clusters could be transferred into aqueous solution and retained their catalytic activity.

1. Introduction

Nanomaterials have been extensively used as drug carriers through different approaches such as polymeric nanoparticles (NPs) [1,2], dendrimers [3,4], polymersomes [5], liposomes [6,7], or capsosomes [8]. The preferred approaches in the design of these nanocarriers are based on organic NPs. Nevertheless, the use of inorganic NPs as drug carriers has been also widely explored [9]. The synthesis of inorganic NPs has greatly evolved in the last years due to their great number of applications. Nowadays it is possible to produce NPs in a reproducible manner [10], having well-defined physicochemical properties and a good colloidal stability in physiological media. Among all the materials used to produce inorganic NPs, gold has arisen as one of the currently most suitable for bioapplications, due to its good properties (*e.g.,* optical properties) and biocompatibility. Gold NPs (Au NPs) have been applied in nanomedicine with different purposes such as imaging (*e.g.,* optoacoustic agents, computed tomography, *etc.*), therapeutic agents (*e.g.,* photothermal therapy), sensors [11] and as drug carriers [12,13]. When designing drug carriers based on inorganic NPs, the surface nature is crucial to determine their features *in vitro* and *in vivo*. As example, Kim et al. demonstrated that the dominant mechanism for NP uptake in particle delivery in three-dimensional tumour tissue was defined by the surface charge [14]. According to their results, positive NPs will be more effective for drug deliv-

* Corresponding authors.
E-mail addresses: dehnen@chemie.uni-marburg.de (S. Dehnen), beatriz.pelazgarcia@physik.uni-marburg.de, beatriz.pelaz@gmail.com (B. Pelaz).
[1] These authors contributed equally.

http://dx.doi.org/10.1016/j.phrs.2016.12.034
1043-6618/

ery due to their greater uptake, while negative NPs will perform better to deliver drugs in deep tissues due to their faster diffusion [14]. The use of amphiphilic polymers to coat inorganic NPs has been extensively reported as a powerful method to generate a controlled surface on NPs of different materials, sizes and shapes [15,16]. Other polymers, such as block copolymers have been also used [17,18].

One of the major drawbacks of many drugs is their poor water solubility, which by using nanocarriers can be overcome, leading to increased solubility of several orders of magnitude (*e.g.,* placlitaxel [19]). Several insoluble drugs (*e.g.,* paclitaxel, doxorubicin [14], or oxaliplatin [20]) have been linked to Au NPs by direct linkage on the NP surface or by using hydrophobic interactions to load the drug on the NPs [21]. The use of hollow Au NPs as drug containers has been also reported [22]. Min et al. proved the higher efficiency of cisplatin, when it was combined to gold nanorods as compared to the free drug [23].

Metal-based drugs are very relevant in both, therapy and diagnosis, the most notorious example being platinum-based drugs for cancer treatment. Remarkably, in 2014, 10%–20% of all patients suffering from cancer were treated with cisplatin or other platinum-based drugs (data from http://www.cancer.gov/research/progress/discovery/cisplatin). Cisplatin is considered to be the best chemotherapeutic to treat testicular cancer (http://www.cancer.gov/research/progress/discovery/cisplatin) and it is broadly applied also to treat other cancers, including bladder, cervical, ovarian, lung, or head and neck cancer. There are several Pt-based drugs that have been approved for their use in the clinic worldwide, such as carboplatin, oxaliplatin or nedaplatin. All of these drugs present *cis*-conformation. Nevertheless, satraplatin has broken the limiting condition of using the *cis*-configuration for the Pt (II) complexes, proving that other configurations are suitable [24].

Other approved anticancer (semi-)metallodrugs based on other metals such as ruthenium or arsenic are NAMI-1, RAPTA, KP1019, and Darinaparsin [24]. To fight cancer, (semi-)metallodrugs are applied also in therapy as radiopharmaceuticals (*e.g.,* technetium, indium or gadolinium-based drugs) [24], or as photochemotherapeutic drugs (*e.g.,* arsenic, antimony or bismuth-based drugs) [25]. However, these drugs have found mainly application as antiparasitics and antimicrobials. For example, melarsoprol ((2-(4-amino)-(4,6-diamino-1,3,5-triazin-2-yl)-phenyl-1,2,3-dithiarsolan-4-methanol)) is used as antiparasitics and rantidine bismuth citrate (RBC, Pylorid, Tritec) is applied to treat ulcers in the gastrointestinal tract. These compounds have arisen as the "perfect" complement or substitutes to treat resistant bacteria [26,27]. Other uses in medicine are their application as antiarthritics, antidiabetes, antivirals, to treat gastric or cardiovascular disorders [24]. Remarkably, most of these drugs contain only a single metal atom. Only few examples like BBR3464 (triplatin tetranitrate) contains three Pt atoms, opening the gate to new drug designs [25].

The usage of organotin(IV) compounds in industry and agriculture is global. Moreover, their medical application as anticancer, antiviral, antimicrobial, antihypertensive agents among others has been evaluated [28]. Although their activity is good, they present unwanted toxicity issues and side effects. Generally, the composition of these compounds can be described as R_nSnL_{4-n}. R represents an organic ligand, and L is an anionic specie [28]. The anti-cancer activity of organotin(IV) compounds has been related with their capability to inhibit ATP synthase inducing apoptosis. However, the exact mechanism of apoptosis induction of organotin(IV) compounds is still not completely clear [29]. Examples of this behaviour are tributyltin (TBT) chloride and dibutyltin (DBT) dichloride. One of these compounds, the triphenyltin 2-phenyl-1,2-triazole-4-caboxylate, exhibits higher anticancer activity against HeLa cells than cisplatin [30]. The activity of these tin compounds is intrinsically related with their chemical structure and due to various toxicity issues, widely described in the literature, the search for new organotin(IV) compounds for bioapplications is still open. Several examples based on dinuclear and polynuclear species have been described, as well as the application of polymers based on these compounds [31]. Polymers and polynuclear compounds exhibit better anti-cancer activity than cisplatin or the monomeric compounds. Recent papers discuss that there is still a long way to go before the fully understanding of the behaviour of metallodrugs in biology is achieved [25,31]. Some common rules have been described in the literature to help in the design of these kind of drugs [25]. Nevertheless there are many examples of drugs exhibiting a great anticancer behaviour breaking those rules, including di- and tri- nuclear cationic Pt(II) compounds [25].

Recently, the potential use of metal complexes as catalysts for chemical transformations in living organisms has attracted much attention [32,33]. These characteristics combined with the acquired resistance of many cancers to cisplatin treatment invites to explore the behaviour of new compounds. Well-defined metal chalcogenide clusters can be synthesized in a controlled manner, ranging from few atoms to several nanometre big architectures [34,35]. Organo-functionalized tin sulfide $[(R^FSn)_4S_6]$ clusters ($R^F = CMe_2CH_2C(O)Me$), are interesting compounds. In the presence of the R^F ligand used here, they adopt a so-called "double-decker" like architecture, in which two Sn_2S_2 rings are parallel arranged and bridged by two further bridging S ligands. The R^F ligands can be further extended by condensation reaction with other molecules, like for instance ferrocene units [36]. Remarkably, ferrocenes and ferrocenyl derivatives present antiproliferation properties [27]. The size of the inorganic Sn/S core, as well as the substituents can be easily tuned by according reaction conditions and the choice of R^F or its extension (hydrazone groups in this case), which also affects further physical and chemical properties. Thus, considering the above described review regarding drug design, these compounds may be a new alternative to the current organotin compounds. However, one of their major limitations is their poor stability in the presence of oxygen and water [36]. The presence of oxygen typically drives these compounds to their degradation to elemental sulfur, tin oxides, organic tin hydroxides (*e.g.*, $RSn(OH)_3$) or mesityl oxide [37,38].

In this work, we propose the use of polymer coated Au NPs as platform for the stabilization of organotin sulfide clusters in aqueous solutions. Thus, Au NPs were coated using an amphiphilic polymer previously modified with the clusters. The presence of the clusters on the NP surface was confirmed *via* inductively coupled plasma mass spectrometry (ICP-MS). The functionality of the clusters linked to the NPs was proven using a catalytic colorimetric reaction based on the reduction of methylene blue, and the stability over time of the clusters was investigated.

2. Results and discussion

Au NPs of *ca.* 7 nm core diameter d_c were prepared using the Brust-Schiffrin method (*cf.* the supporting information). These NPs are capped with dodecanethiol and are insoluble in water. Thus, an amphiphilic polymer was used to transfer them to aqueous solutions. Poly(maleic-alt-anhydride) modified with dodecylamine (PMA) [15] or its modifications [39] are commonly used amphiphilic polymers for this purpose [16,40]. The synthesis of this polymer is typically carried out anhydrous tetrahydrofuran (THF). The organotin clusters are stable in this solvent, and thus can be linked to the polymer in THF.

Prior to the transfer of the NPs to water, the PMA was modified with the previously synthesized organotin clusters (cf. the supporting information). This reaction was performed in a controlled