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Polymeric micelles as a new drug carrier system and their required considerations for clinical trials

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Importance of the field: A polymeric micelle is a macromolecular assembly composed of an inner core and an outer shell, and most typically is formed from block copolymers. In the last two decades, polymeric micelles have been actively studied as a new type of drug carrier system, in particular for drug targeting of anticancer drugs to solid tumors.

Areas covered in this review: In this review, polymeric micelle drug carrier systems are discussed with a focus on toxicities of the polymeric micelle carrier systems and on pharmacological activities of the block copolymers. In the first section, the importance of the above-mentioned evaluation of these properties is explained, as this importance does not seem to be well recognized compared with the importance of targeting and enhanced pharmacological activity of drugs, particularly in the basic studies. Then, designs, types and classifications of the polymeric micelle system are briefly summarized and explained, followed by a detailed discussion regarding several examples of polymeric micelle carrier systems.

What the reader will gain: Readers will gain a strategy of drug delivery with polymeric carriers as well as recent progress of the polymeric micelle carrier systems in their basic studies and clinical trials.

Take home message: The purpose of this review is to achieve tight connections between the basic studies and clinical trials.

Keywords: anticancer drug, block copolymer, EPR effect, polymeric micelle, targeting, toxicity

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1. Introduction

Polymeric micelles have lately appeared as a new type of drug carrier; the preceding drug carriers were micro(nano)particles, liposomes and non-micelle-forming polymeric carriers. The study of polymeric micelle drug carriers started in the 1980s [1-4], and these carrier systems were recognized as one of the most potent drug carrier types in the 1990s [5-13]. Then, in the 2000s, several significant related clinical trials [14-23] got underway, while more and more R&D projects were conducted.

A polymeric micelle is a macromolecular assembly composed of an inner core and an outer shell. The polymeric micelles can have a spherical or a cylindrical shape, depending on the chemical structure and chain length of the macromolecules. For the purpose of drug targeting, most polymeric micelle studies have dealt with the spherical shape, whereas a very limited number of the filamentous shape systems have been studied [24,25]. Therefore, this review deals only with the spherical polymeric micelles. A spherical polymeric micelle structure forms from block copolymers or graft copolymers [26]. **Figure 1** illustrates the formation of a polymeric micelle structure resting on an AB-type block copolymer in which two polymer chains are of a tandem-connected form. Drug molecules are incorporated into the inner core of the micelle through both chemical conjugation and physical

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Article highlights.

- In this review, I discuss toxicities of the polymeric micelle carrier systems as well as pharmacological activities of the block copolymers insofar as these subjects have not been well discussed irrespective of their importance in clinical applications.
- There are two types of drug incorporation into the inner core of the polymeric micelles, namely chemical conjucation and physical entrapment.
- Other than targeting, three functions can be obtained with the polymeric micelles.
- In the system in Kabanov's report, the block copolymer worked as a biologically active agent rather than as a carrier to specific sites.
- In the limited situation, the research team reported on three important observations, which merit an explanation and a discussion here.
- There are six widely known clinical trials regarding polymeric micells drug carrier systems.

This box summarises key points contained in the article.

entrapment. Such a micellar structure forms if one segment of the block copolymer can provide enough interchain cohesive interactions in a selective solvent. For the cohesive interactions, hydrophobic interactions have been used most because most drug molecules possess a hydrophobic character. Typically, the polymeric studies on drug carriers have involved AB- or ABA-type block copolymers because the close relationship between micelle-forming behavior and the structure of polymers can be evaluated more easily with these types of block copolymer than with the other types.

In this review, toxicities of the polymeric micelle carrier systems are discussed as well as pharmacological activities of the block copolymers insofar as these subjects have not been well discussed irrespective of their importance in clinical applications. (Please see other excellent reviews [27-32] for more general information on the polymeric micelle drug carrier systems.) In this explanation of these toxicity and activity subjects, anticancer targeting systems are referenced because most studies on polymeric micelle drug carrier systems have concerned anticancer targeting to solid tumors. **Figure 2** summarizes the developmental process of drug targeting systems, which has comprised the basic study stage, the preclinical study stage and the clinical trial stage. The basic study includes syntheses of polymers, preparation of polymeric micelles containing drugs, *in vitro* cytotoxicity tests, *in vivo* anticancer activity tests and *in vivo* pharmacokinetic analyses. The basic study includes *in vivo* toxicity tests, but these tests span a narrower examination range than do preclinical tests. In the preclinical tests, anticancer activity is examined with reference to a greater variety of cancer models, and pharmacokinetic analyses are carried out in greater detail than is the case of the basic study. Furthermore, in the preclinical stage, toxicities undergo detailed examination relative to various kinds of animals. Clinical trials are carried out in three phases (from Phase I to Phase III). In Phase I trials, the two chief objectives concern toxicity evaluation and determination of a recommended dose for the Phase II trials.

I would argue that, among these three stages, there are slight but significant differences in the importance of evaluations. As illustrated in **Figure 2**, the importance of anticancer activity tests is greater in the basic study than in the preclinical test. On the other hand, the importance of toxicity tests is lower in the basic study than in the preclinical and clinical tests. These differences reflect the purposes and functions of each stage, but the importance of the toxicity evaluations in the basic studies should be stressed, in particular for researchers in the basic sciences. The two overall reasons for emphasizing this point are as follows.

(1) The critical-factor difference between the basic study and the clinical trial. In basic studies, *in vivo* toxicity is assayed at least in anticancer activity tests because high anticancer activity is obtained at the maximum tolerated dose that does not provide lethal toxicity to experimental animals. In this circumstance, one or only a few types of toxicity are critical owing to homogeneity of the experimental animals. Inhibition of the critical toxicity(ies) can be a good strategy for the anticancer activity enhancement. Therefore, basic study researchers tend to pay attention only to one or a very small number of toxicities that are correlated to the maximum tolerated dose. By contrast, many more types of toxicity must be seriously considered and observed in the clinical trials because of the heterogeneity of human patients. A certain type of toxicity that is very mild in the basic study can be critically toxic to some patients owing to genetic characteristics or to a slight disorder in some functions of the patients as well as to physiological differences between the experimental animals and the human beings. If a proportion of the patients receiving the critical (lethal) toxicity is high (e.g., 10%), a drug is dropped from the clinical test, even for anticancer drug cases.

(2) A drug carrier system shows unexpected toxicity in the clinical tests. The unexpected toxicity of an approved drug carrier system is a well-known phenomenon. A liposomal anticancer drug targeting system containing doxorubicin [33,34] was approved in 1995 against Kaposi's sarcoma, followed by FDA approval for treating ovarian cancer. This liposomal system can be targeted to solid tumors owing to its successful poly(ethylene glycol) surface modification, which allows this liposome to use the enhanced permeability and retention effect (EPR effect) [35-37]. The EPR effect is a passive targeting mechanism based on hyperpermeability of the tumor vasculature. One of the most serious adverse effects of this liposome in human clinics is hand-foot syndrome, in which there are observable developed palmar-plantar skin eruptions on the hands and the feet [38,39]. This hand-foot syndrome is never observed with doxorubicin alone or with this liposome carrier alone. This correlation means that the

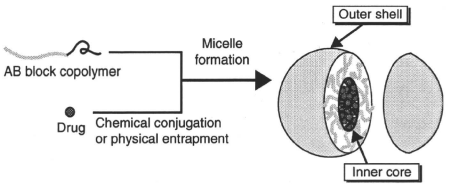

Figure 1. Design of polymeric micelle drug carrier system.

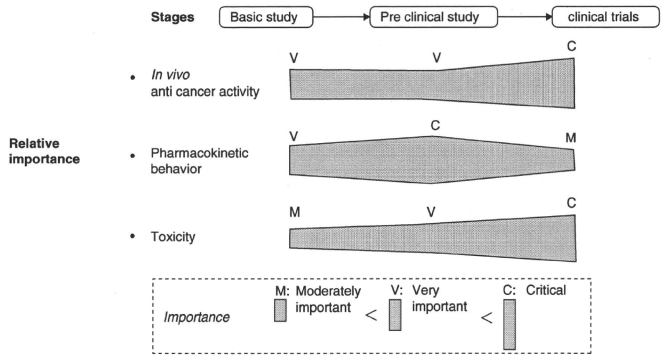

Figure 2. Importance of evaluations on each developmental stage.

adverse effect is observed only in this liposomal doxorubicin carrier system. Furthermore, it is generally believed that delivery to skin tissue is well inhibited for the carriers showing the EPR effect. This hand-foot syndrome was not reported in basic studies of this liposomal doxorubicin; at least, the hand-foot syndrome was not a dose-limiting toxic side effect in mouse anticancer activity assays. Therefore, the appearance of the hand-foot syndrome as a severe side effect in clinical trials was totally unexpected. If skin toxicity had undergone a detailed examination, the hand-foot syndrome could have been reported even in basic studies using mouse models. However, such detailed examinations cannot be conducted in either the basic studies or the preclinical studies for less important side effects surfacing in the mouse models unless some special information related to this toxic effect is present.

This story about the liposomal doxorubicin system does not lead to a necessary conclusion that there should be detailed examinations for all types of toxicity, including unexpected ones. A powerful lesson from the liposomal doxorubicin story is that the system constitutes a good strategy by which basic researchers can pay attention to mild but characteristic side effects in the basic studies.

Drawing on the above-mentioned facts and viewpoints, in this review toxicities of the polymeric micelle targeting systems in animal experiments are summarized and discussed, particularly non-dose-limiting toxicities (i.e., mild toxicities) both of the carrier systems and of the carrier block copolymers.

2. Characterizations and classifications of polymeric micelle drug carrier systems

Before proceeding to the topics in Section 3, which presents some examples of polymeric micelle drug carrier systems, the polymeric micelle systems will be classified depending on their purposes and functions relative to drug delivery system drug delivery system (DDS). This classification should facilitate readers' grasp of the materials in later chapters. Furthermore, the advantages and characteristics of the polymeric micelle are briefly summarized.

2.1 Types of drug incorporation

There are two types of drug incorporation into the inner core of the polymeric micelles, namely chemical conjugation and physical entrapment. Physical entrapment using hydrophobic interactions can be applied to many drugs [40-42], as most drug molecules possess a hydrophobic moiety(ies) (even in the case of water-soluble drugs), and because functional groups that are required for chemical conjugation are not necessary for physical entrapment. In this type of polymeric micelle system, toxicities of carrier block copolymers need not be considered seriously because block copolymers without chemically conjugated drug molecules are expected to be biologically inactive or much less active than the incorporated drug. This type of system can be applied well, particularly to anticancer drug carrier systems because toxicities or pharmacological activities of non-drug-binding block copolymers are believed to be much less intense than those of highly cytotoxic anticancer drugs.

The second type of drug incorporation, chemical conjugation, occurs through chemical bond formation between a drug molecule and the inner-core-forming block of the block copolymer [43-53]. In this type, drug release by cleavage of a chemical bond is an important issue in most systems. One common reaction for this cleavage is hydrolysis. Owing to the phase separation of the inner core from both the outer shell and an outer environment, the drug's access to water molecules, hydrogen ions, hydroxyl ions and hydrolytic enzymes is considerably inhibited. Therefore, the cleavage rate is expected to be much lower than occurs with conventional polymer–drug conjugates. In turn, micelle structures can facilitate the quick release of a drug if the hydrophobicity of the bound drug contributes to inner core association for micelle formation. As the drug molecules are released, the drug release can be accelerated owing to a decrease in inner core hydrophobicity. From this point of view, Li and Kwon [50] have designed methotrexate-conjugated block copolymer micelles, in which the drug release rate can be controlled with conjugated drug amounts. The other possible strategy for chemical conjugation is an alternative action mechanism for the drugs. In a physically entrapped system, only released drugs are expected to express pharmacological activity, even though the delivery systems change both the whole body's distribution and the intracellular distribution. In the case of chemical conjugation, a drug may show activity in both a released form and in a conjugated form. If the conjugated form can express activity, the chemical conjugation systems may overcome the multi-drug resistance induced by P-glycoprotein, as the P-glycoprotein is not expected to result in an efflux of the polymer–drug conjugates to the cells' exteriors. Regarding toxicity concerns, the chemically conjugated drug may pose a risk of a new type of toxicity that was not observed in the corresponding free drug, as the former drug's pharmacokinetic and pharmacodynamic behaviors can be very different from those of the free drug.

2.2 Functions of drug carriers

One important function of the polymeric micelle drug carriers is targeting. As described in Section 2.3, the polymeric micelles possess an inherent ability to target solid tumors owing to the micelles' appropriate size for the EPR effect, which is a passive targeting mechanism to solid tumors.

Other than targeting, three functions can be obtained with the polymeric micelles. The first function is controlled release of a drug. Timing and duration of drug actions can be controlled by controlling drug-release rates from the micelles [51,52,54,55]. This function is not distinctly recognized because both targeting and controlled release functions are achieved in one polymeric micelle system in many cases. In these cases, it is more difficult to prove the contribution of the controlled release for better therapeutic effects than to prove the targeting contribution.

The second function is pharmacological activities of the carrier polymers. In most polymeric micelle systems, block copolymers work only as carriers without showing pharmacological activities. Alternatively, some block copolymers show pharmacological activities. In the former case, toxicity concerns of the carrier block copolymers are not as serious as in the latter case.

There are two types of pharmacologically active copolymer. The first type is the case where block copolymers have a biological activity. Kabanov *et al.* reported that poly(propylene oxide)-*b*-poly(ethylene oxide) block copolymers had an inhibitory activity on P-glycoproteins that play an important role in multi-drug resistance [8,9,19,68,79,70]. Section 3.1 introduces this topic in detail. The second type is a case where drug molecules are attached to the block copolymers. This type of system may have an action mechanism completely different from that of the corresponding free drug (e.g., a cytotoxic action in the conjugate form without a need for the drug release). Such an unusual action mechanism could help result in better pharmacological effects in such matters as circumvention of P-glycoprotein-driven multi-drug resistance. In turn, the new action mechanism has another potential: to cause new toxic side effects that are not found in the corresponding free drug.

The third function of the polymeric micelle systems is solubilization of water-insoluble drugs. Water-insoluble or hardly soluble drugs cannot be safely injected into the bloodstream. One method to overcome this problem is the use of solubilizers that transform these insoluble drugs into a pseudo-soluble status. Water-miscible organic solvents and low-molecular-mass surfactants are representative solubilizers for this purpose. However, these solubilizers are frequently observed showing very high toxicity. Therefore, a DDS carrier that simply solubilizes a water-insoluble drug without any other function is of great value in chemotherapy. Several studies reported that polymeric micelle carriers showed high functions for the solubilization [13,56-62]. This desired function is discussed in Section 2.3, and examples of the solubilization are explained in Sections 3.2 and 3.3.

2.3 Advantages and disadvantages of polymeric micelle carrier systems

Advantages and disadvantages of the polymeric micelle systems as drug carriers are summarized and discussed briefly in this section. Table 1 summarizes the advantages.

The first advantage is the very small size of polymeric micelles. Polymeric micelles are formed typically in a diameter range from 10 to 100 nm, with a substantial narrow distribution. This size range of particles is considered preferable for the attainment of stable, long-term circulation of the carrier system in the bloodstream because the larger particles are actively captured in the reticuloendothelial system and because the smaller particles are rapidly excreted from the kidneys. Alternatively, the small size of polymeric micelles is a great benefit in the sterilization processes associated with pharmaceutical production. Polymeric micelles are easily and inexpensively sterilized by filtration, itself resting on the use of typical sterilization filters with 0.45 or 0.22 μm pores.

The second advantage is the high structural stability of polymeric micelles as compared with the micelles forming from low-molecular-mass surfactants. The high structural stability of polymeric micelles stated above is an important key to *in vivo* delivery in micellar forms (not in single polymer chain form).

The third advantage is the high water solubility of the polymeric micelle drug carrier system incorporating a large amount of hydrophobic drugs [63]. Generally, in conventional polymer–drug conjugate systems, a loss of the water solubility of the polymeric carrier resulting from the introduction of a hydrophobic drug creates a serious problem. Several research groups reported this problem of the drug–polymer conjugates in syntheses [64-66] and in their intravenous injections [67]. Polymeric micelles can incorporate a large number of hydrophobic drug molecules in the large volume of the micelles' inner core, and simultaneously the micelles can maintain their water solubility by inhibiting the intermicellar aggregation of the hydrophobic cores and by using a hydrophilic outer shell layer that works as a barrier against the intermicellar aggregation.

The beneficial character of low toxicity may be described as the fourth advantage. Generally, polymeric surfactants are known to be less toxic than low-molecular-mass surfactants such as sodium dodecyl sulfate. Furthermore, in theory polymeric micelles are considered to be very safe in relation to chronic toxicity. Possessing a much larger size than critical filtration values in the kidney, polymeric micelles can evade renal filtration, even if the molecular mass of the constituting block copolymer is lower than the critical molecular mass for renal filtration. Of course, potential toxicities of polymers and degradation products of polymers (for biodegradable polymers, for example, inflammation induction due to acidic products from polyesters) must be considered. On the other hand, all polymer chains can be released (as single polymer chains) from the micelles during a long time period. This phenomenon results in the complete excretion of the block copolymers from the renal route if the polymer chains are designed with a lower molecular mass than the critical value for renal filtration. Such a result constitutes an advantage of polymeric micelles over the conventional (non-micelle forming) and non-biodegradable polymeric drug carrier systems.

The fifth advantage is separated functionality. Polymeric micelles are composed of two phases: inner core and outer shell. Various functions required for drug delivery systems can be shared by these structurally separated phases. Each phase can play different roles in drug delivery. The outer shell is responsible for interactions with biocomponents such as proteins and cells. These interactions determine pharmacokinetic behavior and the biodistribution of drugs; therefore, the *in vivo* delivery of drugs may be controlled by the outer shell segment independently of the inner core, which is responsible for pharmacological activities through drug loading and release. This heterogeneous structure is more favorable in the construction of highly functionalized carrier systems than in the conventional (non-micelle-forming) polymeric carrier systems, as properties of both phases are freely and independently controlled through a selection of the polymer chains that are appropriate for each segment of block copolymers.

Here, the disadvantages of the polymeric micelle systems will be explained. One disadvantage that cannot easily be recognized in scientific papers is a fact that relatively high levels of polymer chemistry are needed in the polymeric micelle studies. As illustrated in **Figure 1**, an AB type of block copolymer is one of the most favorable structures for the formation of polymeric micelles possessing well-defined shape, size and diameter distribution. The architecture of the AB block copolymer is very simple (two polymer blocks are connected in a tandem manner), but in general its synthesis is more difficult than that of random polymers, where different units are aligned on a polymer chain in a random manner. (As a complete explanation of the difficulties and the limitations of block copolymer syntheses would require more space than is available for this paper, please see other references or textbooks on polymer chemistry.) As a result of the synthesis-related difficulty, choices of monomer units and their combinations are considerably limited. In some cases where laboratories can synthesize a desired block copolymer, it is difficult to synthesize the block copolymer on a large industrial scale in a highly reproducible manner. (In contrast to this general situation, a limited number of block copolymers for drug carriers are produced on an industrial scale and are commercially available. The typical example is Pluronics, poly(ethylene oxide)-*b*-poly(propylene oxide)-*b*-poly(ethylene oxide) block copolymers.) In addition to the above-mentioned problem, some studies have reported cases in which substantial optimization relative to the chemical structures and the chain lengths of the block copolymers was essential for successful drug targeting [61,62,63,68]. This finding indicates that high levels of polymer chemistry are critical for the syntheses of the controlled chemical structures and chain lengths.

Table 1. Advantages of polymeric micelle drug carrier systems.

1. Small diameter with narrow distribution (10 – 100 nm)
2. High structural stability
3. High water solubility
4. Low toxicity
5. Separated functionality

The other disadvantage of the polymeric micelle systems is the immature technology for drug incorporation in a physical manner. Yokoyama *et al.* reported that physical incorporation efficiencies were dependent on drug incorporation methods [61]. At present, there seems to be no universal incorporation method applicable to any polymer. Therefore, researchers must find an appropriate incorporation method for each drug through trial and error. Furthermore, in some methods the drug incorporation may be difficult on a large industrial scale, but easy and efficient on a small laboratory scale. The scale problem is more serious than the polymer synthesis matter because physical factors (e.g., diffusion, solvent exchange rate) are influenced very strongly by the scales in the drug incorporation processes, such as solvent exchange through dialysis membrane. Therefore, more scientific and engineering studies are necessary for significant development on the incorporation technology.

Also, it is worthwhile mentioning that the control of micelle dissociation and drug release rate is essential for drug targeting, and that this control of these matters is sometimes technically difficult to optimize for the targeting, although this is not a disadvantage of the polymeric micelle systems.

3. Reports concerning toxicities of the polymeric micelle systems and biological activities of carrier block copolymers

This section introduces reports concerning toxicities of the polymeric micelle systems and biological (pharmacological) activities of carrier block copolymers.

3.1 Kabanov's report

Kabanov and his colleagues used a poly(ethylene oxide)-*b*-poly(propylene oxide)-*b*-poly(ethylene oxide) block copolymer (ABA type, commercial name Pluronic®) for their polymeric micelle drug delivery system [8,9,19,69-71]. In this system, the block copolymer worked as a biologically active agent rather than as a carrier to specific sites. They reported that Pluronic polymers specifically inhibited the ATP production in mitochondria. P-glycoprotein, which plays an important role in multi-drug resistant cancer cells, shows the efflux action of anticancer drug in an ATP-dependent manner. Consequently, the drug efflux action is inhibited by a Pluronic polymer through the inhibition of ATP production. By using this block copolymer, they reported successful circumvention of the multi-drug resistance both *in vitro* and *in vivo*. This is an innovative application of a synthetic polymer to cancer chemotherapy.

This activity of the block copolymer is very interesting and rather unexpected, as Pluronic polymers lack the functional groups (e.g., charged groups such as carboxyl and amine groups and bulky hydrophobic groups such as long acyl groups) that are expected to have strong interactions with proteins. Kabanov *et al.* showed that occurrences of this activity featured Pluronic polymers possessing an appropriate hydrophilic/hydrophobic balance and appropriate chain lengths. Furthermore, research has not shown any other synthetic polymer that shows P-glycoprotein inhibitory activity. Consequently, it seems that this very interesting and useful activity for cancer chemotherapy is obtained with a very limited number of polymers. However, researchers must pay attention to biological activities of carrier polymers because the mitochondrion and P-glycoprotein inhibitory activities run the risk of raising the levels of toxic side effects in the normal organs and tissues irrespective of the success that Kabanov *et al.*'s animal cancer model had in obtaining enhanced *in vivo* anticancer activity. The presence of this activity can be found relatively easily in the *in vitro* cytotoxic assays (e.g., through determination of drug influx and efflux amounts into/from cells in the presence/absence of polymers). If researchers observe that an anticancer drug's *in vitro* activity increases in strength owing to the drug's combinatory use with a polymer, an advisable subsequent step is to examine the P-glycoprotein in greater detail.

3.2 Yokoyama's report

Yokoyama *et al.* have reported several polymeric micelle anticancer drug targeting systems since the late 1980s, and most of this research team's reports involved the basic study stage. Therefore, the main focus of these reports was on enhanced anticancer activity, improved biodistribution and pharmacokinetic behavior, and related physicochemical characterizations of the carrier systems, whereas descriptions of the carrier systems' toxicities and of the carrier polymers' biological activities were relatively limited. In this limited situation, the research team reported on three important observations, which merit an explanation and a discussion here.

In a series of papers, Yokoyama and co-workers reported enhanced *in vivo* anticancer activity and tumor-specific drug delivery of an anticancer drug, doxorubicin, with drug incorporation into a polymeric micelle carrier [72-78]. These reports focus mainly on *in vivo* anticancer activity and on tumor targeting; however, the reports treat some important information about toxicity. At this point, some details should be noted concerning the chemical structure of the research team's doxorubicin–micelle system because the polymer structure is correlated with toxicity concerns. **Figure 3B** shows the block copolymer structure. Doxorubicin (DOX) was chemically conjugated to aspartic acid residues of the poly(ethylene

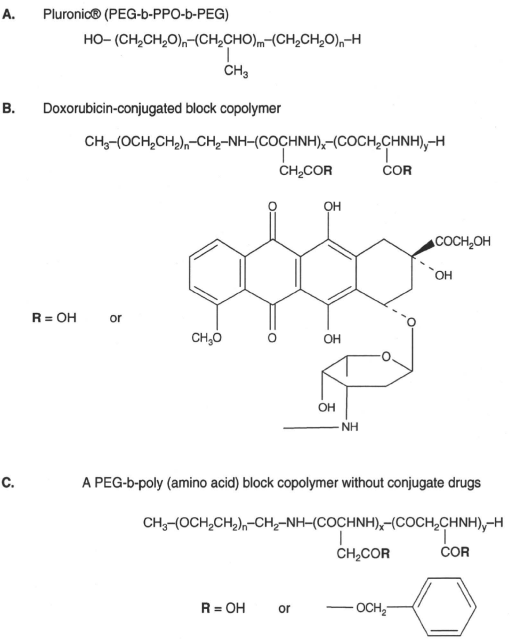

Figure 3. Chemical structures of block copolymer used as polymeric micelle carriers.

glycol)-*b*-poly(aspartic acid) block copolymer through amide bond formation. The poly(ethylene glycol) polymer block was hydrophilic, whereas the DOX-substituted poly(aspartic acid) chain was hydrophobic. Therefore, the obtained drug–block copolymer conjugate (PEG–P(Asp(DOX))) formed micellar structures owing to its amphiphilic character. Doxorubicin was further incorporated into the inner core by means of physical entrapment using hydrophobic and π–π interactions with the chemically conjugated DOX molecules. As a result, polymeric micelles containing both the chemically conjugated and the physically entrapped DOX in the inner core were obtained with the PEG outer shell. Yokoyama *et al.* reported that the carrier polymer PEG–P(Asp(DOX)) did not show any *in vitro* or *in vivo* activities, and that the cytotoxic and anticancer activities were obtained with physically entrapped DOX [78]. The inactivity of the chemically conjugated DOX resulted from the fact that DOX molecules were directly conjugated, without any spacer, to the aspartic acid residues of the block copolymer through amide bonds. These 'direct' amide bonds were considered to be too stable for its cleavage, which provides pharmacologically active free DOX. Therefore, for the purpose of anticancer activity assays, the block polymer can be considered only a carrier, not a biologically active species. On the other hand, attention must be paid to the *in vivo* toxicities attributable to the polymers conjugating drug molecules, particularly in chronic toxic side effects that are not examined in simple toxicity assays done in the basic studies focusing on anticancer activity evaluations.

In another report [76], Yokoyama *et al.* analyzed pharmacokinetic behaviors of the DOX polymeric micelle system as well as enhanced *in vivo* antitumor activity. In this analysis, they showed tumor-selective delivery; the accumulated DOX physically entrapped in the micelle was an amount approximately ninefold larger than the amount of free DOX, whereas the accumulation of the micellar DOX (physically entrapped) in normal organs and tissues was smaller than or the same as the accumulation of free DOX. This report identifies an interesting pharmacokinetic behavior of the micellar DOX. Within 1 h of intravenous (i.v.) injection, accumulated amounts of the micellar DOX in liver were smaller than those

of free DOX. This inequality indicates that the targeting strategy based on the EPR effect was effective in this targeting system even for liver that is known to possess pores large enough for micelles' extravasation in the liver's vasculature. By contrast, 4 h and later, after i.v. injection, this situation was reversed. The micellar DOX showed larger accumulation amounts in liver than free DOX. This inequality resulted from rapid clearance of free DOX in liver through the liver's metabolic activity for drugs, whereas the micellar DOX concentration in liver did not undergo a significant drop, probably because the physically entrapped DOX in the micelle core was largely protected from the metabolic activity. Consequently, the concentration of the micellar DOX was several-fold larger than that of the free DOX. This fact implies that the nonspecific distribution (e.g., at liver) may be an important concern even for successful targeting systems. In another report [74], Yokoyama *et al.* examined toxic side effects of the DOX polymeric micelle system in a composition similar to that of the former one. For liver toxicity, this research team examined pathological observations as well as alanine aminotransferase (ALT)- and aspartate transaminase (AST)-level measurements in blood on day 10 after the first drug injection. The conclusion of this toxicity evaluation was that the toxic side effects of the DOX polymeric micelle were shown in the same pattern as that of free DOX. This means that the liver toxicity was not especially higher than the other side effects for the DOX polymeric micelle. However, considerable attention must be paid to chronic liver toxicity that could not be evaluated in that report up to day 10.

In this DOX-containing polymeric micelle system, the liver toxicity does not seem to be a serious problem because neither unusual nor unexpected toxic side effects were observed in its Phase I clinical trial [14,79,80]. The appearance of the side effects showed the same pattern as that of free DOX in this human clinical trial, where patients underwent longer observations (e.g., several weeks to several months) than was the case in the report's basic study stage.

As explained above, chronic liver toxicity was not serious for the DOX-containing polymeric micelle system. Most scientists in this field posit that the considerable slowness of the metabolism of the incorporated drug compared with the free drug is a universal phenomenon of polymeric micelle drug carrier systems owing to the isolated drug-containing inner core from the outer aqueous environment, where a liver's metabolic enzymes work. Therefore, chronic liver toxicity merits careful examination not only for the polymeric micelle systems, but also for all nano-sized drug carrier systems such as PEG-coated liposomes.

The second important study is Yokoyama and co-worker's report concerning toxicity evaluation of a polymeric micelle carrier lacking drug incorporation [81]. A micelle-forming block copolymer whose chemical structure is shown in **Figure 3C** had been used as a carrier for an anticancer compound camptothecin [61,62,68,82-85] and synthetic retinoids [86-90] such as all-*trans* retinoic acid. The research team analyzed toxicities by conducting pathological examinations that used rats. No pathological abnormality was found for a considerably high dose (200 mg/kg × 5). The team, however, observed significant activation of the mononuclear phagocytic system (MPS) in several organs such as the spleen and liver. Selective accumulation of the polymeric micelle at the MPS was confirmed in an immunohistochemical analysis. These results were obtained with a drug-free polymeric micelle. By contrast, the team concluded that the MPS suffered considerable damage from incorporated cytotoxic drugs in anticancer drug-carrying polymeric micelles. That is why the MPS activation phenomenon seems less important than the other side effects for cytotoxic anticancer drug-carrying systems. However, this MPS-related phenomenon may be important if the polymeric micelle carrier systems are applied to delivering drugs that are much less toxic than typical anticancer drugs.

The third relevant study for biological activity of polymeric micelle carriers is one reported recently concerning the accelerated blood clearance (ABC) phenomenon [91]. ABC is a phenomenon where clearance rates of carrier systems from the bloodstream are raised substantially at repeated injections [92-95]. This ABC phenomenon has been well studied with PEG-coated liposomes that have long-circulating characters at the first injection. A PEG-coated liposome is injected (the first dose) intravenously at an appropriate dose, and then the same liposome is injected (the second dose) according to an appropriate interval (5 – 7 days). This phenomenon occurs owing to immunological activity induced at the first dose, and the change in the clearance is considerable; for example, a case could arise where 90% of the injected dose is circulating 2 h after the first injection and where < 5% of the injected dose is circulating in blood 2 h after the second injection. As polymeric micelles with PEG outer shells have the same profile as that of the PEG-coated liposomes in terms of the PEG outer layer, it is of great interest to know whether these polymeric micelles induce the ABC phenomenon or not. Yokoyama and Oku *et al.* reported the first observation of polymeric micelles' display of the ABC phenomenon [91]. These researchers injected three kinds of polymeric micelle at the first dose, followed by an injection of a PEG-coated liposome at the second injection. The ABC phenomenon was observed only for one polymeric micelle, it was not observed for the other two polymeric micelles. This indicates that some polymeric micelles can induce the ABC phenomenon. At present, the essential properties of the micelle for ABC induction are unknown and will be elucidated in future studies.

The ABC phenomenon is related to the above-mentioned uptake of carrier systems at the MPS and activation of the MPS in the following two aspects.

(1) The existing research suggests that ABC induction develops at the liver [92,93] or the spleen [94,95], both of which are typical organs relative to the MPS. Therefore, the control of uptake extent at the MPS can be a key factor in ABC induction.

(2) It was reported that the ABC phenomenon ceased when the PEG-coated liposomes contained a cytotoxic drug at animal experimental and human clinical levels. Therefore, the importance of the ABC phenomenon becomes greater when drug carriers are applied to pertinent drugs that are much less cytotoxic than typical anticancer drugs. The application to the less cytotoxic drug is a new and important direction of DDS.

3.3 Clinical trials

There are six widely known clinical trials regarding polymeric micelle drug carrier systems. **Table 2** summarizes these trials. The objectives for the use of the polymeric micelles are different among these six examples: tumor targeting is the objective for four examples (a – d) [96,97], solubilization of water-insoluble drugs is the objective for one other example (e), and circumvention of multi-drug resistance (MDR) is the objective for the remaining example (f).

Kabanov *et al.* designed and formulated SP-1049C for MDR circumvention on the basis of this research team's basic studies ([8,9,19,69-71]; see Section 3.1). In this system, an anticancer drug, doxorubicin, was physically encapsulated in the hydrophobic inner core of the Pluronic micelles. After being intravenously injected, however, doxorubicin is very rapidly released from the micelle. Owing to this rapid release, concentrations in blood and pharmacokinetic behaviors of the SP-1049C's doxorubicin were almost the same 5 min post-injection as those of the free doxorubicin injection case. I consider that this rapid release stemmed from two factors: the Pluronic micelle inner cores were of relatively low hydrophobicity and of the fluidic state. In the SP-1049C system, the anticancer drug and the biologically active block copolymer are delivered separately to solid tumor cells, and then the MDR circumvention effect will take place there.

Genexol-PM incorporates paclitaxel in a micelle formed from the poly(ethylene glycol)-*b*-poly(DL-lactide) block copolymer [20-23,42]. This micelle possesses a glassy inner core composed of the poly(DL-lactide) block, but drug release in blood occurred very rapidly after intravenous injection, in almost the same manner as that of SP-1049C. Consequently, the pharmacokinetic behavior of the incorporated drug paclitaxel is almost the same as that of the conventional formulation (Taxol). However, Genexol-PM can be superior to Taxol in terms of safe solubilization of water-insoluble drugs. The conventional formulation, Taxol, contains a large amount of surfactant called Cremophor EL. This surfactant has the function of making paclitaxel soluble in water, but accompanying this function is a set of substantial toxic side effects resulting from this surfactant's toxicities. The poly(ethylene glycol)-*b*-poly(DL-lactide) block copolymer is practically a non-toxic substance and successfully plays a role in the solubilization. Therefore, the solubilization is of great value to cancer chemotherapy. As a large incorporation capacity for hydrophobic drugs is a strong advantage of polymeric micelle carriers, more and more drugs may be applied to the polymeric micelle systems for the drug solubilization purposes.

In clinical trials, Japanese, US and British teams examined the remaining four polymeric micelles in relation to solid-tumor targeting. Chemical structures of the inner-core-forming polymer blocks vary depending on the incorporated drug, whereas the poly(ethylene glycol) chain is used for the outer shell in all cases. Tumor targeting is the primary objective of these carrier systems. However, these systems possess another objective: the system has a function to solubilize a water-insoluble drug in the same manner as does Genexol-PM. Matsumura and co-workers reported that NK-105 incorporating paclitaxel showed highly tumor-selective delivery in murine tumor models [14,15,18,40,41]. In clinical stages, NK-105 can have two solubilization-related advantages over the conventional paclitaxel formulation, Taxol. The first advantage attributable to NK-105 is its relatively low toxic side effects, which reflect the fact that the block copolymer is much less toxic than Cremophor EL used in Taxol. The second advantage attributable to NK-105 is that it does not need the premedication that Taxol requires for reducing its own side effects.

We must wait for the final results of Phase II and III trials to answer the question 'Are the polymeric micelle systems valuable in cancer chemotherapy?' While waiting for the answer, we can take comfort in the fact that Phase I clinical studies have already yielded important information concerning toxic side effects. Even for targeted drugs, serious side effects arise because doses are escalated until dose-limiting toxicities become observable. The important obtained information revealed the toxicity profiles of the polymeric micelle drugs, which turned out to be the same as those of the corresponding free drugs. Most of the toxic side effects of the polymeric micelle drugs appear to result from the carriers' release of the drug in the bloodstream. The absence of uncommon and unexpected types of toxicity is a very meaningful fact that can contribute to the safety of clinical use. Not enough clinical results have been obtained to draw a general conclusion that synthetic block copolymers can be safely used in clinical stages, at least for cancer chemotherapy. However, basic study researchers and clinicians must develop their studies while keeping in mind this potential clinical advantage of a drug carrier.

4. Conclusion

In drug targeting with polymeric micelle drug carrier systems, research has reported less information concerning the toxicities of this carrier system than concerning either the systems' therapeutic activities or the systems' pharmacokinetic behaviors. However, even in this limited information, valuable characteristics of the polymeric micelle (no incidence of unexpected or serious toxicities such as hand-and-foot syndrome and toxicities of Cremophor EL) can be known, and these characteristics must be used for further development of

Table 2. Polymeric micelle anticancer drug carrier systems in clinical trials.

Code	Trade name	Primary objective	Incorporated drug	Progress	Company	Refs
a	NK-911	Targeting	Doxorubicin	Phase II	Nippon Kayaku Co., Japan	[14,15,17]
b	NK-105	Targeting	Paclitaxel	Phase II	Nippon Kayaku Co., Japan	[14,15,18,40,41]
c	NC-6004	Targeting	Cisplatin	Phase I	Nanocarrier Co., Japan	[14,15,96,97]
d	NK-012	Targeting	SN-38	Phase I	Nippon Kayaku Co., Japan	[14,15,44-49]
e	Genexol-PM	Solublilization	Paclitaxel	Phase II	Samyang Corp., Korea	[20-23,42]
f	SP-1049C	Anti-MDR effect	Doxorubicin	Phase II	Supratek Pharma, Inc., Canada	[8,9,19,68-70]

the carrier system. Further, some micelle-forming block copolymers show unique pharmacological activities, which perhaps can serve as a basis for new and innovative chemotherapeutic strategies.

5. Expert opinion

In this section, important points are summarized.

(1) Purpose of carrier use. Most pertinent studies state that the use of polymeric micelle carriers serves a drug-targeting purpose, although a considerable proportion of clinical trials are aimed at the other purposes describe below. The drug targeting with polymeric micelle carriers is of high potential owing to the carriers' several physicochemical advantages, such as very small diameters. Two more purposes, solubilization of water-insoluble drugs and circumvention of multi-drug resistance, are unique and valuable regardless of whether or not they accompany the targeting purpose.

(2) Pharmacological activities of block copolymers. Researchers must check whether or not micelle-forming block copolymers alone show any pharmacological activity in the basic study stage, although this type of activity-positive case is not so common according to previous reports. This activity can be evaluated by means of *in vitro* cell culture examinations because drug incorporation generally lowers *in vitro* drug activities for the activity-negative cases. If enhanced or considerably high activities are found, researchers are encouraged to conduct not only further *in vitro* studies to elucidate mechanisms of the activity, but also *in vivo* examinations.

(3) *In vivo* toxicities of the polymeric micelle drug carrier systems. *In vivo* toxicities and their profiles of the polymeric micelle drug carrier systems must be carefully observed in the basic study stage even if some toxicities are so mild that they do not qualify as dose-limiting toxicities. The information concerning the toxicities is valuable in developments for the clinical trial stage even though the information is such that the toxicity profile is the same as the profile of the corresponding free drug.

(4) Some features of the polymeric micelle drug carrier systems in clinical trials. At present, clinical results of the polymeric micelle drug carrier systems are obtained in a very limited manner as compared with liposomal drug carrier systems. Even in this situation, the fact that neither unexpected nor very different profiles of toxicities have been observed is a very valuable one. This is a potentially great advantage of the polymeric micelle system, but many more clinical and basic examinations are required for satisfactory proof of this advantage.

Declaration of interest

M Yokoyama may receive financial benefits from patents NK-911, NK-105 and NC-6004 (summarized in Table 2) when these pharmaceutics go to market after their approval.

Bibliography

Papers of special note have been highlighted as either of interest (•) or of considerable interest (••) to readers.

1. Bader H, Ringsdorf H, Schmidt B. Watersoluble polymers in medicine. Angew Chem Int Ed Engl 1984;123/124:457-85

•• **This was the first paper concerning polymeric micelle drug carrier systems.**

2. Pratten MK, Lloyd JB, Ringsdorf H. Micelle-forming block copolymers: pinocytosis by macrophages and interaction with model membranes. Makromol Chem 1985;186:725-33

3. Yokoyama M, Inoue S, Sakurai Y. Molecular design for missile drug: synthesis of adriamycin conjugated with IgG using poly(ethylene glycol)-poly (aspartic acid) block copolymer as intermediate carrier. Makromol Chem 1989;190:2041-54

4. Kabanov AV, Alakhov VY, Kabanov VA, et al. The neuroleptic activity of haloperidol increases after its solubilization in surfactant micelles: micelles as microcontainers for drug targeting. FEBS Lett 1989;258:343-5

• **This reported the first enhancement in the in vivo pharmacological activity of drug by the use of polymeric micelles.**

5. Yokoyama M, Okano T, Inoue S, et al. Toxicity and antitumor activity against solid tumors of micelle-forming polymeric drug and its extremely long circulation in blood. Cancer Res 1991;51:3229-36

•• **This reported the first case of in vivo antitumor effect enhancement by the use of polymeric micelle carriers.**

6. Kwon GS, Yokoyama M, Kataoka K, et al. Enhanced tumor accumulation and prolonged circulation times of micelle-forming poly(ethylene oxide-aspartate) block copolymer-adriamycin conjugates. J Control Release 1994;29:17-23

• **This provided the first proof of tumor targeting with polymeric micelles.**

7. Yokoyama M, Okano T, Kataoka K, et al. Selective delivery of adriamycin to a solid tumor using a polymeric micelle carrier system. J Drug Targeting 1999;7:171-86

•• **This paper clearly proved that significant antitumor effect enhancement resulted from tumor targeting effects of a polymeric micelle carrier.**

8. Batrakova EV, Alakhov VY, Kabanov AV, et al. Anthracycline antibiotics non-covalently incorporated into the block copolymer micelles: in vivo evaluation of anti-cancer activity. Br J Cancer 1996;74:1545-52

9. Kabanov AV, Alakhov VY. Micelles of amphiphilic block copolymers as vehicles for drug delivery. In: Alexandridis P, Lindman B, editors, Amphiphilic block copolymers: self assembly and applications. Elsevier, Netherlands; 1997. p. 1-31

• **A good review for biological activities of block copolymers.**

10. Yu BG, Okano T, Kwon GS, et al. Polymeric micelles for drug delivery: solubilization and haemolytic activity of amphotericin B. J Control Release 1998;53:131-6

11. Rolland A, O'Mullane J, Petrak K, et al. New macromolecular carriers for drugs. I. Preparation and characterization of poly (oxyethylene-b-isoprene-b-oxyethylene) block copolymer aggregates. J Appl Polym Sci 1992;44:1195-203

12. Inoue T, Nakamae K, Hoffman AS, et al. An AB block copolymer of oligo(methyl methacrylate) and poly(acrylic acid) for micellar delivery of hydrophobic drugs. J Control Release 1998;51:221-9

13. Zhang X, Burt HM, Hunter WL, et al. An investigation of the antitumor activity and biodistribution of polymeric micellar paclitaxel. Cancer Chemother Pharmacol 1997;40:81-6

14. Matsumura Y, Kataoka K. Preclinical and clinical studies of anticancer agent-incorporating polymer micelles. Cancer Sci 2009;100:572-9

15. Matsumura Y. Polymeric micellar delivery systems in oncology. Jpn J Clin Oncol 2008;38:793-802

16. Matsumura Y. Poly (amino acid) micelle nanocarriers in preclinical and clinical studies. Adv Drug Deliv Rev 2008;60:899-914

• **A good review of clinical studies of polymeric micelle systems.**

17. Matsumura Y, Hamaguchi T, Watanabe N, et al. Phase I clinical trial and pharmacokinetic evaluation of NK911, a micelle-encapsulated doxorubicin. Br J Cancer 2004;91:1775-81

18. Hamaguchi T, Kato K, Matsumura Y, et al. A phase I and pharmacokinetic study of NK105, a paclitaxel-incorporating micellar nanoparticle formulation. Br J Cancer 2007;97:170-6

19. Danson S, Alakhov V, Ranson M, et al. Phase I dose escalation and pharmacokinetic study of pluronic polymer-bound doxorubicin (SP1049C) in patients with advanced cancer. Br J Cancer 2004;90:2085-91

20. Kim DW, Kim SY, Heo DS, et al. Multicenter phase II trial of Genexol-PM, a novel Cremophor-free, polymeric micelle formulation of paclitaxel, with cisplatin in patients with advanced non-small-cell lung cancer. Ann Oncol 2007;18:2009-14

21. Lee KS, Chung HC, Ro J, et al. Multicenter phase II trial of Genexol-PM, a Cremophor-free, polymeric micelle formulation of paclitaxel, in patients with metastatic breast cancer. Breast Cancer Res Treat 2008;108:241-50

22. Park SR, Oh DY, Kim HK, et al. A multi-center, late phase II clinical trial of Genexol (paclitaxel) and cisplatin for patients with advanced gastric cancer. Oncol Rep 2004;12:1059-64

23. Kim TY, Kim DW, Bang YJ, et al. Phase I and pharmacokinetic study of Genexol-PM, a cremophor-free, polymeric micelle-formulated paclitaxel, in patients with advanced malignancies. Clin Cancer Res 2004;10:3708-16

24. Christian DA, Minko T, Discher DE, et al. Flexible filaments for in vivo imaging and delivery: persistent circulation of filomicelles opens the dosage window for sustained tumor shrinkage. Mol Pharm 2009;6:1343-52

25. Geng Y, Minko T, Discher DE, et al. Shape effects of filaments versus spherical particles in flow and drug delivery. Nat Nanotechnol 2007;2:249-55

26. Tuzar Z, Kratochvil P. Block and graft copolymer micelles in solution. Adv Coll Interface Sci 1976;6:201-32

27. Aliabadi M, Lavasanifar A. Polymeric micelles for drug delivery. Expert Opinion Drug Deliv 2006;3:139-62

•• **An excellent review of polymeric micelle drug carrier systems.**

28. Kwon GS. Polymeric micelles for delivery of poorly water-soluble compounds. Crit Rev Ther Drug Carrier Syst 2003;20:357-403

29. Adams ML, Lavasanifar A, Kwon GS. Amphiphilic block copolymers for drug delivery. J Pharm Sci 2003;92:1343-55
- **A good review of polymeric micelle drug carrier systems.**

30. Yokoyama M. Block copolymers as drug carriers. Crit Rev Ther Drug Carrier Syst 1992;9:213-48
- **A good review of polymeric micelle drug carrier systems focusing on polymer chemistries.**

31. Yokoyama M. Polymeric micelles for the targeting of hydrophobic drugs. In: Kwon GS, editor, Drug and pharmaceutical sciences polymeric drug delivery systems, Taylor & Francis vol. 148; 2005. p. 533-75
- **A good review of polymeric micelle drug carrier systems.**

32. Yokoyama M. Polymeric micelles as nano-sized drug carrier systems. In: Y Tabata, editor, Nanoparticles for pharmaceutical applications. American Scientific Publishers; Boca Raton 2007. p. 63-72

33. Tardi PG, Boman NL, Cullis PR. Liposomal doxorubicin. J Drug Target 1996;4:129-40

34. Gabizon AA, Shmeeda H, Zalipsky S. Pros and cons of the liposome platform in cancer drug targeting. J Liposome Res 2006;16:175-83

35. Matsumura Y, Maeda H. A new concept for macromolecular therapeutics in cancer chemotherapy: mechanism of tumoritropic accumulation of proteins and the antitumor agent smancs. Cancer Res 1986;46:6387-92
- •• **This is the original paper for the EPR effect.**

36. Maeda H, Sawa T, Konno T. Mechanism of tumor-targeted delivery of macromolecular drugs, including the EPR effect in solid tumor and clinical overview of the prototype polymeric drug SMANCS. J Control Release 2001;74:47-61

37. Maeda H, Bharate GY, Daruwalla J. Polymeric drugs for efficient tumor-targeted drug delivery based on EPR-effect. Eur J Pharm Biopharm 2009;71:409-19

38. Lorusso D, Di Stefano A, Scambia G, et al. Pegylated liposomal doxorubicin-related palmar-plantar erythrodysesthesia ('hand-foot' syndrome). Ann Oncol 2007;18:1159-64

39. Gordon KB, Tajuddin A, VonRoenn J, et al. Hand-foot syndrome associated with liposome-encapsulated doxorubicin therapy. Cancer 1995;75:2169-73

40. Negishi T, Koizumi F, Matsumura Y, et al. NK105, a paclitaxel-incorporating micellar nanoparticle, is a more potent radiosensitising agent compared to free paclitaxel. Br J Cancer 2006;95:601-6

41. Hamaguchi T, Matsumura Y, Kakizoe T, et al. NK105, a paclitaxel-incorporating micellar nanoparticle formulation, can extend in vivo antitumour activity and reduce the neurotoxicity of paclitaxel. Br J Cancer 2005;92:1240-6

42. Kim SC, Kim DW, Seo MH, et al. In vivo evaluation of polymeric micellar paclitaxel formulation: toxicity and efficacy. J Control Release 2001;72:191-202

43. Nagano T, Yasunaga M, Matsumura Y, et al. Antitumor activity of NK012 combined with cisplatin against small cell lung cancer and intestinal mucosal changes in tumor-bearing mouse after treatment. Clin Cancer Res 2009;15:4348-55

44. Kuroda J, Kuratsu J, Matsumura Y, et al. Potent antitumor effect of SN-38-incorporating polymeric micelle, NK012, against malignant glioma. Int J Cancer 2009;124:2505-11

45. Nakajima TE, Yanagihara K, Matsumura Y, et al. Antitumor effect of SN-38-releasing polymeric micelles, NK012, on spontaneous peritoneal metastases from orthotopic gastric cancer in mice compared with irinotecan. Cancer Res 2008;68:9318-22

46. Saito Y, Yasunaga M, Matsumura Y, et al. Enhanced distribution of NK012, a polymeric micelle-encapsulated SN-38, and sustained release of SN-38 within tumors can beat a hypovascular tumor. Cancer Sci 2008;99:1258-64

47. Sumitomo M, Koizumi F, Matsumura Y, et al. Novel SN-38-incorporated polymeric micelle, NK012, strongly suppresses renal cancer progression. Cancer Res 2008;68:1631-5

48. Nakajima TE, Yasunaga M, Matsumura Y, et al. Synergistic antitumor activity of the novel SN-38-incorporating polymeric micelles, NK012, combined with 5-fluorouracil in a mouse model of colorectal cancer, as compared with that of irinotecan plus 5-fluorouracil. Int J Cancer 2008;122:2148-53

49. Koizumi F, Hamaguchi T, Matsumura Y, et al. Novel SN-38-incorporating polymeric micelles, NK012, eradicate vascular endothelial growth factor-secreting bulky tumors. Cancer Res 2006;66:10048-56

50. Li Y, Kwon GS. Methotrexate ester of poly (ethylene oxide)-block-poly (2-hydroxyethyl-L-aspartamide). Part 1: effects of the level of methotrexate conjugation on the stabliity of micelles and on drug release. Pharm Res 2000;157:607-11
- **An important paper for a strategic design of polymeric micelle systems; drug release control through the control of chemically conjugated drug amount.**

51. Bae Y, Fukushima S, Kataoka K. Design of environment-sensitive supramolecular assemblies for intracellular drug delivery: polymeric micelles that are responsive to intracellular pH change. Angew Chem Int Ed Engl 2003;42:4640-3

52. Bae Y, Fukushima S, Kataoka K. Preparation and biological characterization of polymeric micelle drug carriers with intracellular pH-triggered drug release property: tumor permeability, controlled subcellular drug distribution, and enhanced in vivo antitumor efficacy. Bioconjug Chem 2005;16:122-30

53. Bae Y, Nishiyama N, Kataoka K. In vivo antitumor activity of the folate-conjugated pH-sensitive polymeric micelle selectively releasing adriamycin in the intracellular acidic compartments. Bioconjug Chem 2007;18:1131-9

54. Lee ES, Gao Z, Bae YH. Recent progress in tumor pH targeting nanotechnology. J Control Release 2008;132:164-70

55. Na K, Sethuraman VT, Bae YH. Stimuli-sensitive polymeric micelles as anticancer drug carriers. Anticancer Agents Med Chem 2006;6:525-35

56. Yokoyama M, Satoh A, Matsumura Y, et al. Incorporation of water-insoluble anticancer drug into polymeric micelles and control of their particle size. J Control Release 1998;55:219-29

57. Matsumura Y, Yokoyama M, Kakizoe T. Reduction of the adverse effects of an antitumor agent, KRN 5500 by incorporation of the drug into polymeric micelles. Jpn J Cancer Res 1999;90:122-8
- **This describes the first report concerning the *in vivo* advantages of the solubilization of a water-insoluble drug.**

58. Mizumura Y, Matsumura Y, Kakizoe T. Incorporation of the anticancer agent KRN 5500 into polymeric micelles diminishes the pulmonary toxicity. Jpn J Cancer Res 2002;93:1237-43

59. Lavasanifar A, Samuel J, Kwon GS. Block copolymer micelles for the encapsulation and delivery of amphotericin B. Pharm Res 2002;19:418-22

60. Lavasanifar A, Samuel J, Kwon GS. Micelles self-assembled from poly(ethylene oxide)-block-poly(N-hexyl stearate L-aspartamide) by a solvent evaporation method; effect on the solubilization and hemolytic activity of amphotericin B. J Control Release 2001;77:155-60

61. Yokoyama M, Opanasopit P, Okano T. Polymer design and incorporation method for polymeric micelle carrier system containing water-insoluble anti-cancer agent camptothecin. J Drug Target 2004;12:373-84

62. Opanasopit P, Yokoyama M, Okano T. Block copolymer design for camptothecin incorporation into polymeric micelles for passive tumor targeting. Pharm Res 2004;21:2003-10

63. Yokoyama M, Kwon GS, Kataoka K. Preparation of micelle-forming polymer-drug conjugates. Bioconjugate Chem 1992;3:295-301

64. Hoes CJT, Potman W, Feijen J. Optimization of macromolecular prodrugs of the antitumor antibiotic adriamycin. J Control Release 1985;2:205-13

65. Duncan R, Kopeckova-Rejmanova P, Kopecek J. Anticancer agents coupled to N-(2-hydroxypropyl)methacrylamide copolymers I. Evaluation of daunomycin and puromycin conjugates in vitro. Br J Cancer 1987;55:165-74

66. Endo N, Umemoto N, Hara T. A novel covalent modification of antibodies at their amino groups with retention of antigen-binding activity. J Immunol Methods 1987;104:253-8

67. Zunino F, Pratesi G, Micheloni A. Poly (carboxylic acid) polymers as carriers for anthracyclines. J Control Release 1989;10:65-73

68. Yamamoto T, Yokoyama M, Maitani Y, et al. What are determining factors for stable drug incorporation into polymeric micelle carriers? Consideration on physical and chemical characters of the micelle inner core. J Control Release 2007;123:11-8

69. Kabanov AV, Batrakova EV, Alakhov VY. Pluronic block copolymers for overcoming drug resistance in cancer. Adv Drug Deliv Rev 2002;54:759-79

70. Kabanov AV, Batrakova EV, Alakhov VY. An essential relationship between ATP depletion and chemosensitizing activity of Pluronic block copolymers. J Control Release 2003;91:75-83

71. Sharma AK, Zhang L, Li S, et al. Prevention of MDR development in leukemia cells by micelle-forming polymeric surfactant. J Control Release 2008;131:220-7

72. Yokoyama M, Inoue S, Sakurai Y, et al. Preparation of adriamycin-conjugated poly (ethylene glycol)-poly(aspartic acid) block copolymer. A new type of polymeric anticancer agent. Makromolekulare Chem Rapid Commun 1987;8:431-5

73. Yokoyama M, Miyauchi M, Inoue S, et al. Characterization and anti-cancer activity of micelle-forming polymeric anti-cancer drug, adriamycin-conjugated poly (ethylene glycol)-poly(aspartic acid) block copolymer. Cancer Res 1990;50:1693-700

74. Yokoyama M, Okano T, Inoue S, et al. Toxicity and antitumor activity against solid tumors of micelle-forming polymeric drug and its extremely long circulation in blood. Cancer Res 1991;51:3229-36

75. Kwon GS, Yokoyama M, Kataoka K, et al. Enhanced tumor accumulation and prolonged circulation times of micelle-forming poly(ethylene oxide-aspartate) block copolymer-adriamycin conjugates. J Control Release 1994;29:17-23

76. Yokoyama M, Okano T, Fukushima S, et al. Selective delivery of adriamycin to a solid tumor using a polymeric micelle carrier system. J Drug Targeting 1999;7:171-86

77. Yokoyama M, Okano T, Kataoka K, et al. Improved synthesis of adriamycin-conjugated poly(ethylene oxide)-poly(aspartic acid) block copolymer and formation of unimodal micellar structure with controlled amount of physically entrapped adriamycin. J Control Release 1994;32:269-77

78. Yokoyama M, Fukushima S, Okano T, et al. Characterization of physical entrapment and chemical conjugation of adriamycin in polymeric micelles and their design for in vivo delivery to a solid tumor. J Control Release 1998;50:79-92

79. Nakanishi T, Matsumura Y, Yokoyama M, et al. Development of the polymer micelle carrier system for doxorubicin. J Control Release 2001;74:295-302

80. Tsukioka Y, Matsumura Y, Kakizoe T, et al. Pharmaceutical and biomedical differences between micellar doxorubicin (NK911) and liposomal doxorubicin (Doxil). Jpn J Cancer Res 2002;93:1145-53

81. Kawaguchi T, Yamamoto T, Yokoyama M, et al. Histological study on side effects and tumor targeting of a block copolymer micelle on rats. J Control Release 2009;136:240-6

• **Histological study concerning toxicities of a block copolymer.**

82. Opanasopit P, Y Maitani, T Okano, et al. Influence of serum and albumins from different species on stability of camptothecin-loaded micelles. J Control Release 2005;104:313-21

83. Watanabe M, Yokoyama M, Maitani Y, et al. Preparation of camptothecin-loaded polymeric micelles and evaluation of their incorporation and circulation stability. Int J Pharm 2006;308:183-9

84. Kawano K, Yokoyama M, Maitani Y, et al. Enhanced antitumor effect of camptothecin loaded in long-circulating polymeric micelles. J Control Release 2006;112:329-32

85. Hayama A, Kawano K, Maitani Y, et al. Polymeric micelles modified by folate-PEG-lipid for targeted drug delivery to cancer cells in vitro. J Nanosci Nanotechnol 2008;8:3085-90

86. Kawakami S, Opanasopit P, Hashida M, et al. Biodistribution characteristics of all-trans retinoic acid incorporated in liposomes and polymeric micelles following intravenous administration. J Pharm Sci 2005;94:2606-15

87. Chansri N, Kawakami S, Hashida M, et al. Anti-tumor effect of all-trans retinoic acid loaded polymeric micelles in solid tumor bearing mice. Pharm Res 2008;25:428-34

88. Okuda T, Kawakami S, Hashida M, et al. Block copolymer design for stable encapsulation of N-(4-hydroxyphenyl) retinamide into polymeric micelles in mice. Int J Pharm 2008;357:318-22

89. Okuda T, Kawakami S, Hashida M, et al. Enhanced in vivo antitumor efficacy of fenretinide encapsulated in polymeric micelles. Int J Pharm 2006;373:100-6

90. Satoh T, Kagechika H, Yokoyama M, et al. Encapsulation of the synthetic retinoids Am80 and LE540 into polymeric micelles and the retinoids' release control. J Control Release 2009;136:187-95

91. Koide H, Yokoyama M, Oku N, et al. Particle size-dependent triggering of accelerated blood clearance phenomenon. Int J Pharm 2008;362:197-200

92. Dams ET, Storm G, Boerman OC, et al. Accelerated blood clearance and altered biodistribution of repeated injections of sterically stabilized liposomes. J Pharmacol Exp Ther 2000;292:1071-9

• **The first report of the ABC phenomenon.**

93. Laverman P, Carstens MG, Storm G, et al. Factors affecting the accelerated blood clearance of polyethylene glycol-liposomes upon repeated injection. J Pharmacol Exp Ther 2001;298:607-12

94. Ishida T, Ichihara M, Kiwada H, et al. Spleen plays an important role in the induction of accelerated blood clearance of PEGylated liposomes. J Control Release 2006;115:243-50

95. Ishida T, Kiwada H. Accelerated blood clearance (ABC) phenomenon upon repeated injection of PEGylated liposomes. Int J Pharm 2008;354:56-62

96. Nishiyama N, Matsumura Y, Kataoka K, et al. Novel cisplatin-incorporated polymeric micelles can eradicate solid tumors in mice. Cancer Res 2003;63:8977-83

97. Uchino H, Matsumura Y, Kakizoe T, et al. Cisplatin-incorporating polymeric micelles (NC-6004) can reduce nephrotoxicity and neurotoxicity of cisplatin in rats. Br J Cancer 2005;93:678-87

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高分子ミセルの薬剤学分野への応用

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1. はじめに

高分子ミセルの薬物キャリヤーとしての研究は1980年代に始まったもので[1~3]，薬物キャリヤーとしては新しい部類に入るであろう．よって，読者にとって馴染みがなかったり，薬物キャリヤーとしての検討が余りなされていない部分があるのが現状である．一方で，抗がん剤を内包したDDSシステムの臨床試験が2000年代から始まり[4]，現在，世界で6件の臨床試験が行われている．現時点では，その研究・開発はまだ熱しているとは言えないが，臨床試験の結果などによっては今後大きく発展する余地があるのが，高分子ミセル薬物キャリヤーと言えよう．

以上の状況下で，高分子ミセル薬物キャリヤーの特徴を製剤学的な観点からまとめておくことは意義があると思う．特に，余り書かれることの少ない(短所については原著論文の種になりにくいため，記述されることが少ないので)高分子ミセルキャリヤーの短所についてまとめることは重要と思われる．

2. 高分子ミセルとは[5~8]

「高分子ミセル」とはその名の通り，高分子から成るミセル構造のことである．ある高分子の中に溶媒に溶けやすい部分（ここではAとする）と，その溶媒中で会合する高分子鎖（Bとする）が共存した場合に，Bの部分が会合して形成する構造がミセルである．この会合力として一般的なのは疎水性相互作用である．封入する薬物が疎水性である場合が多いためで，典型的な高分子ミセルキャリヤーではB鎖は疎水性鎖になる．その他には静電相互作用によってミセル形成及び薬物封入がなされるが，封入対象が核酸やタンパク質のように高分子で荷電密度が高いものの場合だけである．

図1にAB型ブロックコポリマーからなる高分子ミセル型薬物キャリヤーシステムの例を示す．これは直鎖状の高分子鎖であるA鎖とB鎖が直列につながった形のブロックコポリマーであり，高分子ミセル型薬物キャリヤーシステムとして最も典型的なものである．水溶性のA鎖としては，ポリエチレングリコール（PEG）が用いられることが圧倒的に多い．このAB型ブロックコポリマーが，数十～数百個会合してB鎖が疎水性の内核を，A鎖は親水性の外殻を構成した球状のミセル構造を形成する．ミセルに導入する薬物は，B鎖に化学的に結合するか，B鎖が形成するミセル内核に物理的に主に疎水性相互作用で封入される．

低分子両親媒性脂質から成るミセル構造の場合に知られているように，ミセルには大別すると球状ミセルと棒状ミセルがある．高分子ミセルの場合にも，棒状のミセルが薬物キャリヤーとしての検討が始められたが[9]，本総説は球状ミセルのみを扱う．

高分子科学での高分子ミセルに関する基礎研究の歴史は古く，1970年代にはすでにかなり広範に研究がなされていたが[10]，薬物キャリヤーとして応用する研究は，1984年[1]になって開始されたものである．その歴史については，参考文献[6,7]を参照されたい．

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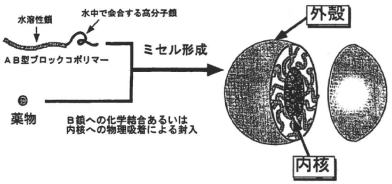

図1　高分子ミセル薬物キャリヤーシステムの構築

表1　高分子ミセル薬物キャリヤーとしての特徴

特長	
	1. ナノサイズの小さな粒径（10～100 nm）
	2. ミセル構造の高い安定性
	3. 様々なタイプの薬物封入が可能
	4. 低い毒性
	5. 機能的な材料設計が容易
短所	
	1. 比較的高度な高分子合成が必要
	2. 薬物封入法が未発達

3. 高分子ミセルの特長

高分子ミセル薬物キャリヤーシステムの特長と短所を表1にまとめた．本項では特長を説明する．

3.1　ナノサイズの小さな粒径

球状の高分子ミセルでは，その直径が10～100 nmの範囲内で得られる．高分子ミセルとしてはこの範囲の粒径が得られることは通常のことであるが，一般的にこのナノサイズの範囲の微粒子を得ることは技術的に困難が伴う．よって，小さな粒径の粒子を得る方法として，高分子ミセルが優れている側面がある．また，その粒径分布も比較的狭いものとなる．例えば動的光散乱で測定した重量換算平均粒径±標準偏差が50±10 nmといった値である（標準偏差は用いるブロックコポリマーの分子量分布等によって増減する）．

また，粒径に関する大きな特長として，これらの粒径はミセル形成操作のみで得られ，粒径をそろえる後操作の必要がないことである．逆に，リポソームで行われているようにエクストルージョンによって粒径を小さくできないとの報告がある[11]．これは薬物キャリヤーとして用いる高分子ミセルの内核が固体状態の硬いものであるためと考えられる．

上記のような小さな粒径であることは，フィルターろ過による容易な滅菌操作を可能とし，製剤上の大きな利点となっている（もちろんこれはミクロンサイズあるいはそれに近い粒径の凝集が形成しないように，高分子組成と封入の量が適切に選択されていることが前提である）．

ドラッグターゲティングに関してこの範囲の粒径は，固形がん組織へのターゲティングなどで有用な大きさである．その理由は，この粒径であると腎臓からの排出を逃れ，かつ肝臓・脾臓の細網内皮系に捕捉されにくいために，血中循環性を高く保つことが可能である．また，固形がん組織へのパッシブターゲティングの1つの方法であるEPR効果が利用できることである．がん組織血管の透過性亢進と，リンパ系による排出抑制によって，ナノサイズのキャリヤーは本質的に固形がん部位に選択的に蓄積しやすい性質がある．この性質はEPR効果（Enhanced Permeability and Retention effect）と呼ばれ，1986年に前田，松村によって提唱された[12,13]．EPR効果は，高分子ミセルに限らずに，リポソームや合成高分子の固形がんへのターゲティングにも適用されているが，これらのキャリヤーの中で高分子ミセルは，このEPR効果を利用した疎水性の強い抗がん剤ターゲティングキャリヤーとして大変優れていると考えられる．なぜなら，EPR効果を示すためにはその大きさが5 nm～200 nmであることが求められるとともに，その表面は親水的で荷電においては中性か弱く負に帯電していることが必要である[14]．高分子ミセルでは，疎水性や正荷電の薬物を多量に封入しても疎水性内核を親水性の外殻がとり囲んでいるので，表面物性は親水的であり，EPR効果発現のための条件を満たしているからである．

3.2　ミセル構造の高い安定性

高分子ミセルは高分子の会合によって形成しているために，ターゲティングのキャリヤーとして有用であるためには，血液中でターゲティングに必要な期間ミセル構造が安定に保たれる必要がある．一般的に低分子界面活性剤から成るミセルに比べ，高分子ミセル構造は安定である．その高い安定性はミセル内核が高分子鎖間の複雑な絡み合い，相互作用により構築されていることに起因し，静的（低い臨界ミセル濃度）及び動的（小さなミセル解離速度定数）な両側面で高い安定性を有することが高分子ミセルシステムの特長である．静的な側面では，高分子ミ

セルの臨界ミセル濃度の典型的な値は1～10 μg/mLと小さく，血液中でも高分子濃度を臨界ミセル濃度より高い濃度で投与することが可能となる．

一方，動的な安定性を記述する解離速度定数はミセル構造が1本の高分子鎖に解離する速度であり，生体内では臨界ミセル濃度よりも重要な因子であるように考えられる．その理由は，生体には血液中の高分子濃度を下げる要因が多数有り（尿排泄や，各種マクロファージによる貪食など），ミセル構造の濃度を高く保つには，ミセル内核を硬い性質の高分子で構成して解離速度を小さくすることが有効と考えられるからである．但し，これまでの研究で静的な安定性と動的な安定性のどちらがどのくらい重要であるかを定量的に扱った報告はない．

3.3 様々なタイプの薬物封入が可能

前項目までは，疎水性のミセル内核に疎水性の低分子薬物を封入するタイプを中心に説明を行ってきた．薬物には疎水性部分を含むものが多いので，疎水性相互作用を利用することが最も適用範囲が広いためである．しかし，「はじめに」に記述したように高分子ミセルを形成する性質（ミセル内核の会合を生み出す相互作用）は疎水性相互作用に限定されない．水系の溶媒中，内核でB鎖が会合を起こすような相互作用を起こせばミセル形成し得るということである．疎水性相互作用の他に，静電相互作用，水素結合などこの相互作用に該当する．静電相互作用，水素結合を利用すれば，DNAやタンパク質などの高分子をミセルに封入することに応用される．また，金属錯体の抗がん剤であるシスプラチンを結合させた高分子ミセルシステム[15,16]がある．結合させたシスプラチン自身，ある程度の疎水性を有するが，ミセル形成の中心的な役割を果たしているのが高分子鎖間の架橋構造であることが他の場合と異なっている．シスプラチン分子中心の白金イオンに複数の高分子側鎖が配位結合して，高分子間に架橋構造が生ずることでミセル内核が形成している．

3.4 低い毒性

まず，高分子ミセルキャリヤー自体の生体内毒性について詳細に解明されたわけではく，表2に示す6つの臨床試験[4,17～19]が行われている現時点までで，大きな問題となっている副作用がないという現状であるということをまずお断りしておく．その上で，概念的な高分子ミセルの毒性の低さから説明させていただく．

EPR効果を利用した固形がんターゲティングでは，血液中での循環性を高めるためには腎臓のろ過作用によって排出されない充分な大きさのキャリヤーサイズである必要がある．この要件を満たしたキャリヤーは一方において，生体外へ排出されないことによる長期に蓄積毒性の懸念がある．もちろん，肝臓からの胆汁排泄の経路もあるのであるが，腎臓からの排泄が確保されていることは，現時点では重要なことと考えられている．高分子ミセルのように会合性でない材料では，高い血液循環性と長期的な腎臓からの排出性を両立するには，材料を生体内分解性とする必要がある．一方，高分子ミセルは，高分子の会合によってミセル構造が構築されているので，血液循環性が必要な期間にはミセル構造によって腎臓からの排出を免れ，長期的には高分子ミセルは1本1本の高分子鎖に解離する用に設計することが可能である．この設計では，蓄積毒性を及ぼす心配が概念上ないことになる．このためには，ブロックコポリマーの分子量を約3万以下にすればよい．

次に，筆者が研究・開発に関係したシステムについての毒性について述べる．まず，マウスがんモデルを用いた評価では，最大耐量を基準に評価を行うため，非選択的に正常組織・臓器に分布した抗がん剤による副作用が顕著に起きている状態での評価であり，高分子ミセルキャリヤー自体の毒性を評価することは困難な場合が多い．このような限られた範

表2 抗がん剤含有高分子ミセルの臨床試験

薬剤名	封入抗がん剤	主目的	段階	開発会社
NK-911	Doxorubicin	固形がんターゲティング	Phase II	日本化薬（株）
NK-105	Paclitaxel	固形がんターゲティング	Phase II	日本化薬（株）
NC-6004	Cisplatin	固形がんターゲティング	Phase I	ナノキャリア（株）
NK-012	SN-38	固形がんターゲティング	Phase I	日本化薬（株）
Genexol-PM	Paclitaxel	薬剤の可溶化	Phase II	Samyang., 韓国
SP-1049C	Doxorubicin	多剤薬剤耐性克服	Phase II	Supratek Pharma, Inc., カナダ

囲の評価であっても，内包する抗がん剤が元々有していた副作用と異なるタイプの副作用が見られるかは大切な事項である．これまでの検討ではそのような副作用は観察されることはなかった[20,21]．但し，最近の検討[22]では病的な変化はないものの，肝臓，脾臓のマクロファージなどのMPSの活性化が観察された．抗がん剤を内包する高分子ミセルでは，その殺細胞活性によってこれらの活性化は観察されないが，高分子ミセルを殺細胞性の抗がん剤よりマイルドな薬物に適用する場合には，このような高分子ミセルキャリヤーの生体への作用は十分検討する意義のあることと考えられる．

薬物ターゲティングの臨床では，キャリヤー自体，あるいはキャリヤーによるターゲティングの特性によって元来の抗がん剤にはない特有の副作用が報告されている．リポソーム製剤におけるHand and foot syndromeであり[23,24]，抗体製剤におけるinfusion-related reactionがよく知られた例である．これまでに明らかになった高分子ミセル抗がん剤の臨床報告[18,19]では，キャリヤーシステム特有の副作用は観察されていない．

以上の結果をもって，「高分子ミセルキャリヤーには毒性はない」とか「抗がん剤を対象とした場合には全く問題はない」と言うことはできず，更なる基礎的・臨床的な検討の積み上げが必要である．しかし，少なからず行われた動物実験・臨床試験の中でキャリヤー特有の毒性が認められないという事実は，毒性面から見た高分子ミセル（あるいは広義に合成高分子キャリヤー）の高いポテンシャルを示していると考えられる．

3.5 機能的な材料設計が容易

図1に示したように，ブロックコポリマーのA鎖がミセル内核を形成し，B鎖が内核を形成する．ミセル外殻はタンパク質や細胞などの生体成分と相互作用を通して体内動態・分布を決定する．一方，内核は薬物を封入しそれを放出することで，薬効発現の役割を担う．すなわち，ブロックコポリマーのそれぞれの鎖に合目的な機能を与え，それを最適化するように化学構造を選択することが可能となる．このように，機能分離性はシステム設計の観点からみると大きな利点となり，内核を構成する高分子鎖と外殻を構成する高分子鎖とに薬物キャリヤーシステムとして必要とされる機能を分割して付与することで，より高性能のシステムが達成可能となる．

4. 高分子ミセルの短所

次に高分子ミセルキャリヤーの短所と考えられる2つの事項を述べる．

4.1 比較的高度な高分子合成が必要

典型的な高分子ミセルを得るためには，合成高分子としては一般的ではない部類のブロックコポリマーが必要であり，ブロックコポリマーの合成には比較的高度な高分子合成技術が必要とされる場合が多い．この必要性に関する説明は他の総説[25]に譲る．

また，単に高分子ミセルを形成すれば必ず薬物キャリヤーとして有用というわけではなく，ブロックコポリマーの化学構造にかなり左右されることも[26]，高分子合成化学が重要であることを示している．逆に見ると，高分子合成化学の技術がない環境での高分子ミセルキャリヤー研究・開発が遂行しにくいということであり，大きな進歩を得るためには短所であると考えられる．

4.2 薬物封入法が未発達

マイクロスフィアやリポソームなど研究と開発の歴史が長いキャリヤーシステムに比べて，新しい高分子ミセルでは，薬物を安定にかつ高効率でミセル内核に封入する方法[8,27~29]についての検討が未発達である．また，実験室でのスケールではうまく封入できても，それを臨床に向けたスケールに大きくする方法論も今後進歩させる必要を感じる．

5. おわりに

本総説では，高分子ミセル薬物キャリヤーの製剤的な特徴に述べた．本キャリヤーシステムは，水溶性高分子，リポソームなど他のキャリヤーシステムにはないいくつかの特徴があることを述べた．また，比較的に高度な高分子合成が必要なことなど，急速な発展を阻害する要因についても述べた．それにもかかわらず，DDSで果たし得る大きな可能性を考慮すれば，技術的な困難を克服して研究・開発する価値のあるキャリヤーシステムであると信じる．

引用文献

1) H. Bader, H. Ringsdorf , B. Schmidt, Watersoluble polymers in medicine, *Angew Chem.*, **123/124**, 457–485 (1984).
2) M. Yokoyama, S. Inoue, Y. Sakurai, *et al.*, Prepa-

ration of adriamycin-conjugated poly(ethylene glycol)-poly(aspartic acid) block copolymer, A new type of polymeric anticancer agent, *Die Makromolekulare Chemie Rapid Communications*, **8**, 431–435 (1987).
3) A.V. Kabanov, V.Y. Alakhov, V.A. Kabanov, *et al.*, The neuroleptic activity of haloperidol increases after its solubilization in surfactant micelles: Micelles as microcontainers for drug targeting, *FEBS Lett.*, **258**, 343–345 (1989).
4) Y. Matsumura, K. Kataoka, Preclinical and clinical studies of anticancer agent-incorporating polymer micelles, *Cancer Sci.*, **100**, 572–579 (2009).
5) M. Yokoyama, Block copolymers as drug carriers, *Crit. Rev. Ther. Drug Carrier Systems*, **9**, 213–248 (1992).
6) M. Yokoyama, Polymeric micelles for the targeting of hydrophobic drugs, In G.S. Kwon, ed., Drug and Pharmaceutical Sciences, Vol. 148, Polymeric Drug Delivery Systems, Taylor & Francis, pp. 533–575, 2005.
7) M. Yokoyama, Polymeric micelles as nano-sized drug carrier systems, In Y. Tabata, ed., Nanoparticles for Pharmaceutical Applications, American Scientific Publishers, 63–72, 2007.
8) M. Aliabadi, A. Lavasanifar, Polymeric micelles for drug delivery, *Expert Opinion Drug Delivery*, **3**, 139–162 (2006).
9) Y. Geng, T. Minko, D.E. Discher, *et al.*, Shape effects of filaments versus spherical particles in flow and drug delivery, *Nat. Nanotechnol.*, **2**, 249–255 (2007).
10) Z. Tuzar, P. Kratochvil, Block and graft copolymer micelles in solution, *Adv. Coll. Interface Sci.*, **6**, 201–232 (1976).
11) M. Yokoyama, A. Satoh, Y. Matsumura, *et al.*, Incorporation of water-insoluble anticancer drug into polymeric micelles and control of their particle size, *J. Control. Release*, **55**, 219–229 (1998).
12) Y. Matsumura, H. Maeda, A new concept for macromolecular therapeutics in cancer chemotherapy: Mechanism of tumoritropic accumulation of proteins and the antitumor agent smancs, *Cancer Res.*, **46**, 6387–6392 (1986).
13) H. Maeda, T. Sawa, T. Konno, Mechanism of tumor-targeted delivery of macromolecular drugs, including the EPR effect in solid tumor and clinical overview of the prototype polymeric drug SMANCS, *J. Control. Release*, **74**, 47–61 (2001).
14) 横山昌幸, ナノ粒子とドラッグターゲティング, 機能材料, **22**, 22–27 (2002).
15) M. Yokoyama, Y. Sakurai, K. Kataoka, *et al.*, Introduction of cisplatin into polymeric micelle, *J. Control. Release*, **39**, 351–356 (1996).
16) N. Nishiyama, Y. Matsumura, K. Kataoka, *et al.*, Novel cisplatin-incorporated polymeric micelles can eradicate solid tumors in mice, *Cancer Res.*, **63**, 8977–8983 (2003).
17) Y. Matsumura, Poly (amino acid) micelle nanocarriers in preclinical and clinical studies, *Adv. Drug Deliv. Rev.*, **60**, 899–914 (2008).
18) Y. Matsumura, T. Hamaguchi, N. Watanabe, *et al.*, Phase I clinical trial and pharmacokinetic evaluation of NK911, a micelle-encapsulated doxorubicin, *Br. J. Cancer*, **91**, 1775–1781 (2004).
19) T. Hamaguchi, K. Kato, Y. Matsumura, *et al.*, A phase I and pharmacokinetic study of NK105, a paclitaxel-incorporating micellar nanoparticle formulation, *Br. J. Cancer*, **97**, 170–176 (2007).
20) M. Yokoyama, T. Okano, S. Inoue, *et al.*, Toxicity and antitumor activity against solid tumors of micelle-forming polymeric drug and its extremely long circulation in blood, *Cancer Res.*, **51**, 3229–3236 (1991).
21) M. Yokoyama, T. Okano, S. Fukushima, *et al.*, Selective delivery of adriamycin to a solid tumor using a polymeric micelle carrier system, *J. Drug Targeting*, **7**, 171–186 (1999).
22) T. Kawaguchi, T. Yamamoto, M. Yokoyama, *et al.*, Histological study on side effects and tumor targeting of a block copolymer micelle on rats, *J. Control. Release*, **136**, 240–246 (2009).
23) D. Lorusso, A. Di Stefano, G. Scambia, *et al.*, Pegylated liposomal doxorubicin-related palmar-plantar erythrodysesthesia ('hand-foot' syndrome), *Ann. Oncol.*, **18**, 1159–1164 (2007).
24) K.B. Gordon, A. Tajuddin, J. VonRoenn, *et al.*, Hand-foot syndrome associated with liposome-encapsulated doxorubicin therapy, *Cancer*, **75**, 2169–2173 (1995).
25) 横山昌幸, 薬物キャリアに用いられる高分子材料, 特集 DDS に利用される高分子化学, *Drug Delivery System*, **23**, 610–617 (2008).
26) T. Yamamoto, M. Yokoyama, Y. Maitani, *et al.*, What are determining factors for stable drug incorporation into polymeric micelle carriers? Consideration on physical and chemical characters of the micelle inner core, *J. Control. Release*, **123**, 11–18 (2007).
27) G.S. Kwon, M. Yokoyama, T. Okano, *et al.*, Block copolymer micelles as vehicles for hydrophobic drugs, *Colloids and Surfaces, B: Biointerfaces*, **2**, 429–434 (1994).
28) M. Yokoyama, T. Okano, K. Kataoka, *et al.*, Improved synthesis of adriamycin-conjugated poly (ethylene oxide)-poly(aspartic acid) block copolymer and formation of unimodal micellar structure with controlled amount of physically entrapped adriamycin, *J. Control. Release*, **32**, 269–277 (1994).
29) M. Yokoyama, P. Opanasopit, T. Okano, *et al.*, Polymer design and incorporation method for polymeric micelle carrier system containing water-insoluble anti-cancer agent camptothecin, *J. Drug Targeting*, **12**, 373–384 (2004).

High Resolution Brain Imaging with Combined Parallel-hole and Pinhole Collimation

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Abstract– A brain single photon emission computed tomography (SPECT) imager is simulated for obtaining high resolution brain scans for diagnosing brain ischemia. In this simulation the camera consists of one large field of view detector with parallel-hole collimation and a smaller field of view high resolution detector with pinhole collimation. The parallel-hole collimated detector images the whole brain and acquires data without truncation. It localizes areas of particular diagnostic interest, and also provides support information for the reconstruction of data acquired by the pinhole collimated detectors. The pinhole collimated detector images small regions of the brain. It provides high resolution truncated projections, from which a high resolution region of interest (ROI) is obtained. The reconstruction is performed using a maximum a posteriori (MAP) estimate with total variation regularization within the ROI. The low resolution image from the parallel-hole collimated detector is used as prior information. This improves the quantitation for the interior problem. The combination of a large field of view parallel-hole collimated detector and a smaller field of view high resolution pinhole detector improves the quantitation in the simulated brain imaging study. It makes use of the high sensitivity of the pinhole collimator while compensates for the degradation in the reconstructed image due to interior problem caused by the small field of view of the pinhole collimator. Our simulations show potential for clinical application, where the quantitation of cerebral blood flow (CBF) and cerebral vascular reactivity (CVR) are valuable in diagnosis of ischemia, and the quantitation of benzodiazepine receptor density is important in evaluating neuronal damage due to ischemic effects.

I. INTRODUCTION

The quantitation of cerebral blood flow (CBF) and cerebral vascular reactivity (CVR) are valuable in diagnosis of ischemia, and the quantitation of benzodiazepine receptor density is important in evaluating neuronal damage due to ischemic effects. To better evaluate cerebral autoregulation, the Department of Investigative Radiology at the National Cardiovascular Center Research Institute in Osaka, Japan is designing a high resolution single photon emission computed tomography (SPECT) imager for obtaining high resolution brain. The camera consists of one large field of view detector imaging the whole brain and multiple smaller field of view high resolution detectors imaging small regions of the brain (see Fig. 1). The large field of view detector provides images without truncation that localize areas of particular diagnostic interest and provide support information for the reconstruction of high resolution regions of interest (ROIs) from high resolution truncated projections obtained with the small field of view detectors. The work presented in this paper provides simulations which show that the camera improves the quantitation for the interior reconstruction problem.

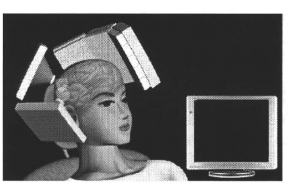

Figure 1. Large field of view detector for imaging whole brain and smaller field of view detectors for imaging ROIs.

As has been noted, with a small object-to-detector distance, both sensitivity and resolution of pinholes are better than that of the parallel-hole collimator [1]. However, the advantage is guaranteed at the expense of a small field of view. Imaging with a high resolution small field of view camera provides truncated projections. The reconstruction of these projections involves determining the solution to the interior problem in local tomography. The interior problem in medical imaging refers to the situation where the region-of-interest (ROI) is totally contained within the object. For instance, in SPECT, the interior problem happens when the projections passing through the region outside the ROI are truncated due to a small field-of-view detector or a short detector-to-object distance in the case of converging collimation. The interior problem has been

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