Special Section on Pharmacokinetics and ADME of Biological Therapeutics—Minireview

The In Vivo Pharmacokinetics of Block Copolymers Containing Polyethylene Glycol Used in Nanocarrier Drug Delivery Systems

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ABSTRACT

Polyethylene glycol (PEG) is one of the most commonly used synthetic macromolecular polymers for modifying small molecule drugs, peptides, proteins, or nanodrug delivery systems to improve their water solubility, biocompatibility, and stability. Block copolymers containing PEG have been widely used in nanodrug delivery systems such as solid lipid nanoparticles, polymeric nanoparticles, polymeric micelles, and liposomes. To date, although numerous PEGylated nanodrug delivery systems have been developed, only a few have been approved for clinical application. Poor safety and effectiveness are important reasons for the high failure rate of nanodrug delivery system clinical trials. These factors are not only related to the loaded drugs and released drugs but are also related to the nanocarriers. Therefore, investigating the in vivo spatiotemporal fate of block copolymers containing PEG used in nanodrug delivery systems is necessary and important for evaluating their safety, efficacy, and toxicity. In this article, we will review the information that has been reported about the absorption, distribution, metabolism, and excretion of block copolymers containing PEG. We believe this review is helpful to understand the biologic fate of block copolymers containing PEG.

SIGNIFICANCE STATEMENT

This review describes pharmacokinetic study of block copolymers containing polyethylene glycol. The main focus of this paper is the in vivo fate of these polyethylene glycol-related copolymers after their release from nanocarriers. This review is helpful for understanding the in vivo fate of block copolymers containing polyethylene glycol used in nanocarrier drug delivery systems.

Introduction

During these years, great progress has been made in the development of nanodrug delivery systems (NDDSs) (Fan et al., 2021; Rai et al., 2021). NDDSs are specially designed as drug delivery vehicles to deliver active pharmaceutical ingredients to their target locations, which have numerous advantages such as improved targeting, decreased toxicity, and prolonged circulation time (Aggarwal et al., 2022). Polyethylene glycol (PEG) is composed of repeating oxyethylene subunits. It is one of the most commonly used synthetic macromolecular polymers for modifying drugs with various molecular mass or NDDSs to improve their water solubility, stability, and biocompatibility (Fu et al., 2021). Furthermore, block copolymers containing polyethylene glycol such as polyethylene glycol-poly(lactic acid) (PEG-PLA), polyethylene glycol-poly(caprolactone) (PEG-PCL), poloxamers, polyethylene glycol-poly(lactic-co-glycolic acid) (PEG-PLGA), and polyethylene glycol-diestearoyl-sn-glycero-3-phosphoethanolamine (PEG-DSPE) have been commonly used as pharmaceutical excipients in the preparation of solid lipid nanoparticles, polymeric micelles, polymeric nanoparticles, and liposomes (Owens and Peppas, 2006; Markovsky et al., 2012). These macromolecular polymers containing PEG could offer sterical barriers to other NDDSs and blood components such as opsonin proteins, preventing their macrophage phagocytosis and prolonging their circulation time in blood, consequently as ‘invisible features’ in vivo. In particular, the end groups of these synthetic macromolecular polymers can be connected to different targeting ligands, which could enhance the bioavailability, decrease the side effects, and especially target specific locations. Figure 1 is the overview of PEGylated carrier systems for drug delivery. However, to date,
although numerous PEGylated nanodrug delivery systems have been developed, only a few of them (e.g., Genexol, Doxil, Krystatexxa, and several marketed LNP formulations) are clinically approved (Kim et al., 2001; Gabizon et al., 2003; Li et al., 2020). Moreover, many problems of these approved PEGylated nanodrug delivery systems have not been fully explained, especially the in vivo fate of loaded drugs and polymer excipients, which are very important for the safety and effectiveness of these PEGylated nanodrug delivery systems. Furthermore, numerous reasons contribute to the high failure rate of clinical trials of the

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**Fig. 1.** Overview of PEGylated carrier systems for drug delivery.

**Fig. 2.** Chemical structures of PEG (A), PEG-PLA (B), PEG-PCL (C), Poloxamer (D), PEG-PLGA (E), and PEG-DSPE (F).
NDDSs, such as potential toxicity and unsatisfactory pharmacokinetic behavior. These defects are associated with both NDDSs and the nanocarriers that are released in vivo. Therefore, investigating the in vivo spatiotemporal fate of block copolymers containing PEG used in NDDSs (i.e., their absorption, distribution, metabolism, and excretion) is necessary and important for evaluating their safety, effectiveness, and toxicity.

In this article, we have concentrated on the following PEG-related block copolymers: PEG, PEG-PLA, PEG-PCL, poloxamers, PEG-PLGA, and PEG-DSPE. The chemical structures of these synthetic macromolecular polymers are illustrated in Fig. 2. The existing information related to the absorption, distribution, metabolism, and excretion (ADME) of these block copolymers containing polyethylene glycol in vivo are summarized in Fig. 3. PEG and PEG copolymers can be absorbed into blood across the gastrointestinal, nasal epithelia, lung epithelial membranes, and skin. PEG-related block copolymers are relatively easy to be accumulated in the liver, spleen, lung, and kidney. These polymers can be eliminated in urine, bile, and feces. The summary of ADME information for these block copolymers containing PEG is shown in Table 1. Distribution and excretion of these PEG-related copolymers are shown in Fig. 4. We believe that this review is helpful for understanding the pharmacokinetics of block copolymers containing PEG.

**Pharmacokinetics of PEG.** Polyethylene glycol (PEG) polymers are composed of repeating ethylene oxide units, and their general formula is HO-(CH₂CH₂O)nH. Hydrophilic and neutral polymers of this kind have been authorized by the US Food and Drug Administration for human oral, dermal, and intravenous applications. PEG-related polymers are widely used in pharmaceutical industry as solubilizers, stabilizers, and control releasing modifiers or conjugated with small molecular compounds or proteins (PEGylation) and drug delivery systems, which can increase the hydrophilicity, prolong residence time in circulation, and reduce the immunogenicity and antigenicity of these chemicals. The in vivo pharmacokinetics of PEGylated drug delivery systems are usually well characterized during their development phases. However, information about the in vivo pharmacokinetics of PEG polymers is relatively sparse. During these years, it has been reported that PEG can cause vacuolation in animal tissues (Irizarry Rovira et al., 2018; Fletcher et al., 2019). Some studies also indicated that adverse reactions of PEG polymers usually occur through complement (C) activation, resulting in hypersensitivity reactions that can cause anaphylactic shock (Chanan-Khan et al., 2003; Fruijtier-Polloth, 2005; Szébeni, 2005). Moreover, Wang et al. (2020) reported that PEG can act as P-glycoprotein inhibitors, which may influence the absorption and efflux of the medical compounds taken at the same time. The uptake of PEG polymers with small molecular mass (<2 kDa) occurs through passive diffusion, whereas the uptake of PEG polymers with large molecular mass (>5 kDa) occurs through both passive diffusion and caveolae-mediated endocytosis. Fasinu and coworkers (2013) found that PEG10K and PEG20K were able to act as CYP3A4 inhibitors and inhibit the metabolism of felodipine. Therefore, PEG may not be as nontoxic and bioinert as we usually think. Detailed information about the pharmacokinetics of PEG polymers is necessary and important for evaluating the safety, effectiveness, and toxicity of PEGylated drug delivery systems in their clinical applications. It is helpful for establishing a relationship to the functional effects of the PEGylated drug delivery systems, particularly when administered chronically.

Absorption, distribution, metabolism, and excretion of different sized PEG polymers in various species with different administration routes have previously been reported (Soderholm et al., 1993; 1996; 1997; He et al., 1998; Webster et al., 2007; Pelham et al., 2008; Baumann et al., 2019; Su et al., 2019; Wang et al., 2020). PEG polymers can be absorbed across the gastrointestinal, nasal epithelia, lung epithelial membranes, and skin. Absorption of PEG polymers is size dependent, decreasing with increasing molecular mass. For example, He and colleagues (1998) studied the oral bioavailability of PEG400 and PEG900 in rats and dogs after oral and intravenous administrations. In their study, the mean oral bioavailability of PEG400 in rats and dogs is 23.8% and 112.2%, respectively. The mean oral bioavailability of
<table>
<thead>
<tr>
<th>Administration Route</th>
<th>Animal Models</th>
<th>Absorption</th>
<th>Distribution</th>
<th>Metabolism</th>
<th>Excretion</th>
</tr>
</thead>
<tbody>
<tr>
<td>PEG</td>
<td>Intratracheal, Intravenous, Intranasal, &amp; Gastrointestinal</td>
<td>Mice, Rats, Dogs, Guinea Pigs, Cats, Rabbits, &amp; Humans</td>
<td>PEG can be absorbed across gastrointestinal, nasal epithelia, lung epithelial membranes, &amp; skin. Size dependent, decreasing with increasing molecular mass. Mean oral bioavailability of PEG400 is 23.8% in rats &amp; 112.2% in dogs; mean oral bioavailability of PEG9000 is 3.24% in rats &amp; 16.4% in dogs. Extent of absorption after intranasal &amp; gastrointestinal administrations to rats is &lt;60% for PEG6000; ~14% for PEG10000, respectively; ~4% &amp; ~2% for PEG20000, respectively.</td>
<td>Mainly distribute to heart, liver, spleen, lung, &amp; kidney (PEG10K, PEG20K, PEG40K, &amp; PEG polymers with molecular mass from 6 kDa to 190 kDa).</td>
<td>PEG polymers can be metabolized to carboxylic acids by alcohol dehydrogenase &amp; CYP3A5s. Alcohol dehydrogenase plays a major role in the metabolism of PEG. PEG polymers can also be metabolized by sulfotransferases to form sulfated metabolites. PEG polymers can inhibit P-glycoprotein &amp; CYP3A4 (PEG400 &amp; PEG1K).</td>
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<tr>
<td>PEG-PLA</td>
<td>Intravenous</td>
<td>Rats</td>
<td>N/A</td>
<td>PEG-PLA may be metabolized into PEG &amp; lactic acid under the action of esterase.</td>
<td>Mainly distribute to spleen, liver, &amp; kidney (mPEG2000-PLA2500 &amp; mPEG2000-PLA2000).</td>
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<tr>
<td>PEG-PCL</td>
<td>Intravenous</td>
<td>Rats</td>
<td>N/A</td>
<td>PEG-PCL may be metabolized into PEG &amp; PCL under the action of esterase.</td>
<td>Mainly distribute to liver, kidney, &amp; spleen (mPEG5000-b-PCL5000).</td>
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<tr>
<td>PEG-PLGA</td>
<td>Intravenous</td>
<td>Rats</td>
<td>N/A</td>
<td>PEG-PLGA may be metabolized into PEG, lactic acid, &amp; glycolic acid under the action of esterase.</td>
<td>Mainly distribute in liver &amp; spleen.</td>
</tr>
<tr>
<td>PEG-DSPE</td>
<td>Intravenous</td>
<td>Rats</td>
<td>N/A</td>
<td>PEG-DSPE may be metabolized into PEG &amp; DSPE under the action of esterase. DSPE-PEG2000 &amp; DSPE-PEG5000 do not inhibit activities of CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6 &amp; 3A4 significantly.</td>
<td>Mainly distribute in lung, fat, liver, &amp; spleen (DSPE-PEG2000 &amp; DSPE-PEG5000).</td>
</tr>
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PEG900 in rats and dogs is 3.24% and 16.4%, respectively. Oral bioavailability decreased with increasing molecular mass in the same species. Amidon and colleagues (Donovan et al., 1990) examined the absorption of PEG600, PEG1000, and PEG2000 across the gastrointestinal and nasal epithelia in rats. For PEG600, the extent of absorption is less than 60% after intranasal and gastrointestinal administrations; for PEG1000, the extent of absorption is about 14% and 9% after intranasal and gastrointestinal administrations; for PEG2000, the extent of absorption is about 4% and 2% after intranasal and gastrointestinal administrations. The permeabilities of both the gastrointestinal and the nasal mucosa showed similar molecular mass dependencies for PEG polymers. Furthermore, Gursahani and coworkers (2009) studied the absorption behavior of fluorescently labeled PEG polymers with different molecular mass across epithelial membranes in rat lung. Their results indicated that PEG synthetic polymers could be absorbed across epithelial membranes with first-order kinetics after intratracheal administration to rats. They also found that PEG polymers with small molecular mass (<2 kDa) were effectively cleared within 48 hours, whereas large PEG polymers (>5 kDa) remained in lung cells and tissues for 7 days. The size of PEG polymers plays an important role in controlling their absorption rate and absorption mechanism. The same absorption results were found in humans. For example, Shaffer and coworkers' (1950) study showed that the oral absolute bioavailability of PEG500 and PEG1000 in human volunteers is about 57% and 9.8%, respectively, whereas the oral bioavailability of PEG6000 in human volunteers is basically zero. Moreover, Pelham et al. (2008) found that oral absolute bioavailability of PEG3350 in healthy individuals is only 0.17%; most of the PEG3350 was not absorbed into the blood but excreted in the feces. As for the transdermal administration route, PEG polymers with small molecular mass can only be absorbed minimally through the intact skin, and PEG polymers with large molecular mass (>4 kDa) cannot be absorbed through the intact skin at all (Fruijtier-Polloth, 2005). However, PEG polymers can penetrate through injured skin regardless of their molecular mass (Herold et al., 1982).

The tissue distribution of PEG polymers depends on many factors, such as size and route of administration. Extravascularly, PEG polymers with small molecular mass may permeate tissues more quickly than PEG polymers with higher molecular mass. In particular, large PEG synthetic polymers have relatively slower renal clearance and greater potential to accumulate within tissues and cells. It was observed by standard histologic examination that the intracellular PEG synthetic polymers can be accumulated within the lysosome, eventually leading to vacuolation and distension (Baumann et al., 2014). Several studies about the tissue distribution of PEG polymers have been reported. For instance, (Longley and coworkers (2013) found that PEG40K polymers administered by i.v. injection with a dose of 1250 mg/kg to mice could be monitored in most tissues like liver, lung, kidney, heart, gastrointestinal tract, spleen, fat, bladder, testes, eyes, muscle, and brain. The concentrations of PEG40K polymers were relatively high in liver, kidney, lung, and heart, and the maximum concentration of PEG40K was detected in the kidney. Rudmann and colleagues (2013) used immuno-histochemical assay to study the cellular distribution of PEG10k, PEG20K, and PEG40K after i.v. injection of 100 mg/kg in rats. In their study, 3 months after i.v. administration, they found that PEG10K, PEG20K, and PEG40K were still detected in liver, lung, heart, spleen, kidney, and choroid plexus. Moreover, the concentrations of both PEG10K and PEG20K polymers were higher in hepatic Kupffer cells, alveolar macrophages, and renal tubule epithelium than in the other tissue cells. In contrast, the concentrations of PEG40K polymers were higher in choroid plexus epithelial cells, renal interstitial macrophages, and splenic subcapsular red pulp macrophages. Yamaoka and coworkers (1994) examined the tissue distribution of PEG polymers after i.v. administration to mice. Their results indicated that large PEG polymers...
were retained longer in blood circulation compared with small PEG polymers. With the increase of the molecular mass of PEG polymers from 6 kDa to 190 kDa, the half-life of the synthetic PEG polymers in blood circulation was extended from 18 minutes to 24 hours. The concentrations of these related PEG polymers in the tissues such as liver, muscle, skin, and bone are higher than those in other tissues. Vascular permeability is an important factor for the time dependence of the accumulation of PEG polymers in tissues. Their results also showed that small PEG polymers tended to translocate freely from blood to various tissues and return to blood again via diffusion, whereas large PEG polymers transferred more slowly from blood circulation to different tissues. Their results indicated that chronic administration of high molecular mass PEG polymers may lead to serious accumulation and cause potential toxicity in related tissues. Generally speaking, PEG polymers are relatively easy to be accumulated in the following tissues: highly perfused tissues; tissues containing mononuclear phagocyte system, and neovascular tissues with low lymphatic drainage (Markovsky et al., 2012).

So far, there have been some reports on metabolism of PEG polymers (Hunt et al., 1982; Webster et al., 2007; Su et al., 2019). Shaffer and coworkers’ (1950) study with PEG400 synthetic polymers showed that the subunit ethylene glycol is not generated as the metabolite of PEG synthetic polymers in humans. The main metabolic pathway of PEG polymers is the oxidation of the terminal alcohol group to carboxylic acid group. For instance, Hunt et al. (1982) developed an analytical method based on triple-quadrupole mass spectrometry technique and detected both the diacid and hydroxyl acid metabolites of PEG polymers in urine and serum samples of a burn patient who was treated with an antibacterial cream containing PEG. Friman et al. (1990; 1993) detected carboxylic acid metabolites of PEG900 in cat bile after intravenous injection. They also calculated the percentages of both PEG polymers and their carboxylic acid metabolites in cat bile. Their results indicated that 74% of PEG molecules were excreted in cat bile unchanged and 26% of PEG molecules were oxidized to carboxylic acids. Moreover, PEG polymers can be metabolized by sulfotransferases. Roy and colleagues (1987; 1988) observed the sulfated metabolites of PEG400 and PEG1000 in rat and guinea pig in vitro liver preparations. It was also reported that a small amount of oxalic acid can be released after PEG metabolism (Fruitijter-Polloth, 2005). In general, the phase I oxidation of PEG polymers is mainly mediated by alcohol dehydrogenase in mammalian systems. It was also reported that the oxidation of PEG synthetic polymers can be mediated by CYP450s (Herold et al., 1989; Veronese and Pasut, 2005). Compared with alcohol dehydrogenase, CYP450s play a lesser role for the oxidation of PEG polymers. Furthermore, it was reported that the acid metabolites of PEG polymers are related to their adverse effects such as acidosis and hypercalcemia (Bruns et al., 1982).

PEG polymers can be retained in urine, bile, and feces. The excretion of PEG polymers is associated with many factors such as size, shape, and administration routes. For intravenous administration, urinary excretion is the main excretion pathway for PEG polymers. He et al. (1998) studied the excretion of PEG400 and PEG900 in rats and dogs after i.v. administration. In their study, PEG400 and PEG900 were mainly excreted unchanged in urine. Mean 24-hour urinary recoveries of total radioactivity of [14C]PEG400 and [3H]PEG900 after i.v. administration to rats and dogs were 85.1%, 82.5%, 85.1%, and 81.9%, respectively. Shaffer et al. (1950) found that 86% of PEG1000 polymers and 96% of PEG6000 polymers were excreted in urine within 12 hours after i.v. administration to humans. Friman and Svanvik (1997) found that almost 100% of PEG1000 was excreted in urine after i.v. administration to rats. Urinary excretion of PEG polymers is also size dependent, as it occurs by passive glomerular filtration. Yamaoka and coworkers’ (1994) study in mice indicated that urinary clearance decreased significantly when the molecular mass of PEG polymers exceeded 20 kDa. Biliary excretion is another excretion pathway for PEG polymers. It is also molecular mass dependent. Yamaoka et al. (1994) found that liver clearance decreased with the increase of PEG molecular mass when the PEG molecular mass was less than 50 kDa, whereas liver clearance increased with the increase of PEG molecular mass when the PEG molecular mass was larger than 50 kDa. PEG polymers with smaller and larger molecular mass have greater hepatobiliary clearance. However, for PEG polymers with very small molecular mass, their bile excretion is not high. Shaffer et al. (1950) found that PEG400 is not excreted into bile after i.v. administration to dogs. Friman et al. (1995) found that the bile excretion of PEG900 was high in patients with T-tube cholecystectomy and that the concentrations of PEG900 in bile samples were 31 times higher than those detected in human plasma samples. They also observed high bile/plasma ratio of PEG450, PEG900, PEG2500, and PEG4000 in pigs and cats after i.v. administration (Friman et al., 1988; 1990). Carpenter et al. (1971) found that 61% of PEG4000 was excreted into urine and that 20% of PEG4000 was detected in feces after i.v. administration to rats. Their data indicated that biliary excretion plays a lesser role for elimination of PEG4000 compared with urinary excretion. Generally speaking, both the kidney and liver are important for elimination of PEG in vivo. Urinary excretion is the major elimination route of PEG polymers. As for oral administration, fecal excretion is the main excretion pathway of PEG polymers. For example, Pelham et al. (2008) studied pharmacokinetics of PEG3350 in humans after oral administration. The oral absolute bioavailability of PEG3350 in healthy individuals was only 0.17%; 93% of the PEG3350 was excreted in the feces and only 0.19% to 0.25% of the PEG3350 was excreted in urine.

**Pharmacokinetics of PEG-PLA.** Polyactic acid (PLA), also known as polylactide, is a kind of polyester polymer derived from the polymerization of lactic acid as the main raw material. PLA is too hydrophobic and has certain defects as an excipient for hydrophilic drugs, but the properties of PLA can be improved by connecting with hydrophilic PEG. The molecular formula of PEG-PLA is H(OCH(CH2)CO)∝O(CH2CH2O)∝H. PEG-PLA block copolymers such as diblock polymers (PEG-PLA) and triblock polymers (PLA-PEG-PLA) have a wide range of effects, which can be used to increase the drug load of hydrophobic drugs, prolong blood circulation time, and improve drug bioavailability and efficacy (Xiao et al., 2010; Chen et al., 2014; Ghasedi et al., 2018; Goudarzi et al., 2018; Amani et al., 2019; Girard et al., 2021; Massadeh et al., 2021). Although in vivo fate studies of triblock PLA-PEG-PLA polymers have not been reported, pharmacokinetic studies of diblock PEG-PLA polymers in rats after intravenous administration have previously been reported (Shi et al., 2019; Meng et al., 2021). Specifically, Meng et al. (2021) studied the pharmacokinetics of PEG-PLA copolymers in rats by liquid chromatography-tandem mass spectrometry technique. They found that the concentration of PEG-PLA was high in plasma and extracellular fluid after i.v. administration. Most of the PEG-PLA polymers were eliminated from plasma within 48 hours. Small amounts of PEG-PLA were excreted into bile (<0.8%). PEG-PLA polymers can be metabolized into PEG and lactic acid; 86% of PEG-PLA polymers were metabolized into PEG and then excreted in urine. For tissue distribution study, the concentrations of PEG-PLA synthetic polymers were higher in liver, kidney, spleen, and fat and lower in muscle and brain. PEG-PLA has a low permeability to the brain, possibly due to the blood-brain barrier. In addition, Meng (2019), also studied the inhibitory effects of PEG-PLA synthetic polymers on human CYP450s. They found that PEG-PLA had a weak inhibitory effect on CYP2C8, CYP2C9, CYP2D6, and CYP3A4 in the range of 0.075–250 μM,
whereas it had no inhibitory effect on CYP1A2, CYP2B6, and CYP2C19. Shi et al. (2019) characterized the pharmacokinetics of mPEG2000-PLA2500 in Sprague Dawley rats after i.v. injection of 5 mg/kg. The extrapolated concentration (Cn) of mPEG2000-PLA2500 in rat plasma was 25.3 ± 2.78 µg·ml⁻¹ after i.v. administration. The area under the plasma concentration-time curve (AUCₙ₋₀) of mPEG2000-PLA2500 was 44.9 ± 2.95 µg·h·ml⁻¹. The in vivo elimination of mPEG2000-PLA2500 was found to be fast. Its plasma elimination half-life (t₁/₂) was only 1.31 ± 0.155 h. Moreover, Stonik and coworkers (2001) also studied the pharmacokinetics of PEG-PLA polymers in rats. They found that PEG-PLA copolymers with small molecular mass were quickly cleared from the systemic circulation after i.v. administration. High molecular mass PEG-PLA copolymers could reduce liver intake by half and increase blood circulation time after i.v. administration. PEG-PLA particles were mainly distributed in liver parenchymal cells.

**Pharmacokinetics of PEG-PCL.** PCL is a biodegradable hydrophobic aliphatic polyester formed by ring-opening polymerization of 2-caprolactone monomer (da Silva et al., 2009). The biocompatibility of drug-loaded PCL polymer is poor due to its strong hydrophobicity (Zhang et al., 2006). The introduction of hydrophilic polymer PEG to construct the amphiphilic PEG-PCL copolymer can compensate for this deficiency. PCL(A) modified with PEG(B) in different proportions can be divided into AB diblock, ABA or BAB triblock, star block, multiblock, and graft polymer (Liu et al., 2008). The drug delivery systems based on PEG-PCL, PCL-PEG-PCL and PEG-PCL-PEG present good potential benefits (Gou et al., 2009; Feng et al., 2012; Han et al., 2013; Luo et al., 2016; Lee and Jeong, 2020; Singh et al., 2020; Di Tranì et al., 2021; Yang et al., 2021). However, recent studies suggested that PEG-PCL micelles may be involved in nanocarrier-drug interaction by influencing the activity of cytochrome P450 (Qiu et al., 2018; Li et al., 2019). In addition, Zhang et al. (2021) found that mPEG₁₂₅-PCL₄ micelles can reduce the plasma concentration of metformin by inhibiting the uptake function of organic cation transporters in rats, indicating that there are potential risks of drug interaction between PEG-PCL carrier system and organic cation drugs. Therefore, revealing the in vivo fate of PEG-PCL micelles is crucial for its medical application. To date, pharmacokinetic differences in vivo between small molecular drugs and PEG-PCL loaded particles have been extensively studied (Feng et al., 2012; Wang et al., 2014; Manjili et al., 2018; Kurd et al., 2019). However, in vivo fate studies of triblock polymers such as PCL-PEG-PCL or PEG-PCL-PEG have not been reported, and only a few studies have focused on the in vivo fate of PEG-PCL monomers (Liu et al., 2007; Kao et al., 2013; Sun et al., 2018).

Specifically, Liu et al. (2007) studied the pharmacokinetics of mPEG₅₀₀₀₀-b-PCL₅₀₀₀₀ micelles and mPEG₅₀₀₀₀-b-PCL₅₀₀₀₀ monomer in mice by isotope labeling method. Their results of the interaction between mPEG₅₀₀₀₀-b-PCL₅₀₀₀₀ copolymer monomer and plasma protein showed that the copolymer monomer did not bind to plasma protein. Therefore, the existence of plasma protein would not likely affect the critical micelle concentration of mPEG₅₀₀₀₀-b-PCL₅₀₀₀₀ significantly. After i.v. administration of mPEG₅₀₀₀₀-b-PCL₅₀₀₀₀ monomer (lower than critical micelle concentration), due to its low molecular mass and amphipathicity, the monomer had a faster distribution to tissues compared with micelles. The concentrations of mPEG₅₀₀₀₀-b-PCL₅₀₀₀₀ monomer in tissues within 1 hour from high to low were as follows: liver, kidney, and spleen. Compared with monomer, mPEG₅₀₀₀₀-b-PCL₅₀₀₀₀ micelles had a longer blood circulation time and a slower elimination rate.

Kao et al. (2013) studied the in vivo pharmacokinetics of¹³¹I benzyl PEG-PCL micelles and ¹³¹I benzyl PEG-PCL monomer in mice after i.v. injection by isotope labeling method. The radiation accumulation of monomer and micelle in tissues reached the highest values after 1 hour, and significant radiation retention was detected in reticuloendothelial system tissues. At the same time, the PEG-PCL monomers and micelles had a large radioactive accumulation in urine, indicating that they were excreted mainly through the kidneys. Sun et al. (2018) explained why PEG-PCL monomer has a faster blood clearance compared with PEG-PCL micelles. This is due to the fact that intact PEG-PCL micelles are not easily captured by Kupffer cells, although the exposed hydrophobic fragments of PEG-PCL monomers can be quickly identified and captured by Kupffer cells.

In summary, at present, there is no information about the absorption of PEG-PCL in vivo. The administration route used in current pharmacokinetic studies of PEG-PCL polymers is intravenous injection. PEG-PCL polymer monomer has a rapid plasma elimination and faster distribution to reticuloendothelial system tissues such as liver and spleen compared with micelle. PEG-PCL mainly excretes out of the body in urine. Although there is no study to reveal the metabolism process of PEG-PCL in vivo right now, we speculate that the ester bonds between PEG and PCL would break in vivo. PEG-PCL can be metabolized into PEG and PCL under the action of esterase.

**Pharmacokinetics of Poloxamers.** Poloxamers, also called Pluronic or Tetroneics, are amphiphilic nonionic triblock copolymers composed of polyoxyethylene (PEO) and polyoxypropylene (PPO) (PEOₙ–PPOₙ–PEOₙ). The triblock copolymers can self-assemble into micelles. The hydrophobic core (PPO) of such micelles can carry different drugs, and the hydrophilic shell (PEO) guarantees the micelles’ stability (Kabanov et al., 2002). Poloxamer polymers are widely used in drug delivery (Das et al., 2010; Sahu et al., 2011; Basak and Bandyopadhyay, 2013; Chen et al., 2013; Nguyen et al., 2017). In the past, poloxamers were traditionally considered ‘inert.’ However, recent studies suggest that poloxamers may have biologic effects (Blonder et al., 1999; Ren et al., 2009). Specifically, Blonder et al. (1999) demonstrated that poloxamer407 would result in hyperlipidemia. Ren et al. (2009) found that poloxamer188 may have an inhibitory effect on cytochrome P450 3A. The cytochrome P450 3A family is the main metabolic enzyme that accounts for more than 70% of the cytochrome P450 in the small intestine. Substances that inhibit cytochrome P450 3A activity can increase plasma concentration of the substrate drugs and may result in toxicity. These results indicate that there are potential risks of interaction between poloxamer carrier systems and drugs. Therefore, it is necessary to reveal the in vivo fate of poloxamer polymers for their medical application.

There have been some reports on the in vivo fate of poloxamer polymers. For example, Feng et al. (2021a,b) studied the pharmacokinetics and tissue distribution of poloxamer188 in rats after i.v. injection of 5 mg/kg. The pharmacokinetic parameters of poloxamer188 were as follows: AUC₀₋₉ was 3.0 ± 0.6 µg·h/ml, mean residence time (MRT) was 0.6 ± 0.1 h, clearance (CL) was 1.7 ± 0.3 l/h/kg, t₁/₂ was 2.0 ± 1.1 h, and volume of distribution (V₉) was 5.1 ± 3.2 l/kg. The results of tissue distribution study indicated that poloxamer188 could be rapidly distributed to tissues with a high clearance rate. Poloxamer188 was mainly distributed in kidney (26.8 µg/g at 0.2 h and 11.63 µg/g at 4 h). A small amount of poloxamer188 was detected in the other tissues such as stomach, liver, lung, muscle, and spleen. Poloxamer188 could not pass through the blood-brain barrier into brain. Moreover, Li et al. (2021) studied the pharmacokinetics of poloxamer124 in rats after i.v. injection of 10 mg/kg. The pharmacokinetic parameters of poloxamer124 were as follows: AUC₀₋₉ was 9.84 ± 3.02 mg·h/ml, t₀.₉ was 2.89 ± 1.14 h/kg, MRT was 0.7 ± 0.08 h, t₁/₂ was 2.06 ± 1.04 h, and CL was 0.98 ± 0.21 l/h/kg. Their results indicated that poloxamer124 could be rapidly eliminated in rat plasma after i.v. injection. Willcox et al. (1978) studied the distribution and excretion of poloxamer188 in dogs by radioisotope labeling method. After i.v. injection of ¹³¹I labeled poloxamer188, nearly
75% of poloxamer188 circulated in plasma was in free form, and 25% tended to be bound to albumin. Within 24 hours after i.v. administration, about 40% of poloxamer188 was excreted through urine and some remained in extravascular fluid and tissues. The highest concentration of poloxamer188 was found in bile after 24 hours. These results demonstrated that poloxamer188 was mainly excreted through kidney and bile. Grindel et al. (2002) studied the pharmacokinetics of purified poloxamer188 in rats, pregnant rats, pregnant rabbits, dogs, and humans by gel permeation chromatography. They found that the plasma clearance of poloxamer188 in rats and rabbits was faster than that in dogs and humans. Moreover, the plasma clearance of poloxamer188 was not related to dose or gender. Their study indicated that poloxamer188 polymers are mainly distributed in highly vascularized tissues and extracellular fluid after continuous i.v. infusion of purified poloxamer188. They also found a single metabolite HW1 of poloxamer188 in both dog plasma and human plasma. The metabolite was a block copolymer, and its molecular mass was 16 kDa. Compared with the parent compound, the clearance rate of the metabolite was slower. Their study also indicated that renal clearance accounted for 90% of total plasma clearance in humans. Urinary excretion is the major route of elimination for poloxamer polymers (Jewell et al., 1997; Grindel et al., 2002). Wang and Stern (1975) studied the distribution and excretion of poloxamer108 (P108) in rats by isotope labeling assay. Immediately after i.v. administration, P108 polymers were detected highly in kidney and lung. And 20 hours after i.v. administration, only a small part of P108 polymers remained in kidney, liver, small intestine, and carcass. P108 polymers were found in urine as unchanged form. About 94% of the initial dose of P108 polymers was excreted in urine and 6% of the initial dose of P108 was excreted in feces within 72 hours after i.v. administration. Batrakova et al. (2004) examined the effects of micelle formation on the distribution kinetics of poloxamer85 in mice by radioisotope labeling method. The results of their study indicated that the aggregation state of poloxamer85 would affect its half-life time in vivo, ranging from 60 to 90 hours. Poloxamer85 could remain in the blood for a long time in both monomer and micellar state. The micellar form of poloxamer85 had a longer circulation time and an increased exposure to the tissues compared with the unimer, but the formation of micelles could reduce the uptake of poloxamer85 in liver. In their study, after i.v. administration to mice, poloxamer85 was detectable in several tissues and its concentrations decreased in the following order: liver > spleen > kidney > lung > brain. Furthermore, they also found that the micelle formation reduced the uptake of poloxamer85 in liver but had no effect on the total clearance of poloxamer85, which suggested that the elimination of poloxamer85 depended on the renal elimination of poloxamer85 unimers rather than the rate of micelle disposition or disintegration.

In summary, so far, there is no information about the absorption of poloxamers in vivo. The route of administration used in current pharmacokinetic studies of poloxamer polymers is intravenous injection. Most poloxamers would not bind to plasma proteins when released to the blood. Poloxamers are mainly distributed in highly vascularized tissues such as liver, kidney, spleen, and lung. The primary elimination of poloxamers is renal excretion.

**Pharmacokinetics of PEG-PLGA.** Poly(lactic-co-glycolic acid) (PLGA) is a biodegradable functional organic compound from the random polymerization of two monomers: lactic acid and glycolic acid. PLGA also has strong hydrophobicity. Due to its poor biocompatibility with hydrophilic drugs, its application is limited. However, it can be combined with PEG to form the coblock polymer PEG-PLGA, and the connection with hydrophilic PEG can change the properties of PLGA. PEG-PLGA block copolymers are widely used in drug delivery systems for improving the drug bioavailability, prolonging the half-life of drugs in vivo, and so do (Ahmed and Badr-Eldin, 2020). So far, only a few studies have reported the biologic fate of PEG-PLGA in vivo. For instance, Avgoustakis and coworkers (2003) studied the biologic distribution of mPEG-PLGA in mice. mPEG-PLGA was labeled with radioactive labeling, and 2 minutes after i.v. administration, significant radioactivity was detected in liver and muscle. The amount of radioactivity in blood was about 75.8%. Over time, the radioactivity of mPEG-PLGA was slowly eliminated from the blood circulation and accumulated in liver and spleen. There was little change in other tissues. The excretion of mPEG-PLGA in intestine and urine increased with time, and the excretion of mPEG-PLGA was 6%–8% of the total at the end of sampling. The effects of different PLGA-mPEG ratios on biologic distribution were also studied. When the PLG-mPEG ratio ranged from 256 to 153, mPEG content increased, and blood clearance rate decreased. The ratio was further reduced from 153 to 61 and 34, and blood clearance increased. Panagti et al. (2001) studied the biologic distribution of mPEG-PLGA nanoparticles in mice by radioactive labeling assay. A large proportion of mPEG-PLGA was found in liver and spleen after i.v. injection. About 70%–80% of the proportion was detected in blood, liver, and spleen, and about 20%–30% of the proportion was detected in the other tissues. The half-life of mPEG-PLGA was about 7 hours. The distribution of mPEG-PLGA was not affected by dose.

Generally speaking, PEG-PLGA polymers are mainly distributed in liver and spleen and excreted through the bile and urine after intravenous administration. Although there is no detailed study to reveal the metabolism process of PEG-PLGA in vivo, we speculate that the ester bonds between PEG and PLGA would break in vivo. PEG-PLGA can be metabolized into PEG, lactic acid, and glycolic acid under the action of esterase.

**Pharmacokinetics of DSPE-PEG.** DSPE-PEG is a PEGylated phospholipid. It can be used as a carrier material for nanoformulations to extend the circulation time and improve the biocompatibility and stability of the drug in vivo. DSPE-PEG is an amphoteric macromolecule with good water solubility and has received extensive attention in recent years. Although DSPE-PEG is widely used, its safety and bioactivity in vivo have not been studied in detail. It has been reported that after injection of pharmaceutical preparations containing DSPE, the human body will have adverse reactions, such as increased blood pressure, difficulty breathing, chest pain, etc. (van den Hoven et al., 2013). Su et al. (2019) studied pharmacokinetics of DSPE-PEG2000 and DSPE-PEG5000 in rats after i.v. injection with a dose of 25 mg/kg. The pharmacokinetic parameters of DSPE-PEG5000 in rats after i.v. administration were: AUC0−t was 271.8 ± 14 μg/ml*h, MRT0−t was 2.064 ± 0.069 h, Vd was 0.198 ± 0.038 l/kg, C0 was 104.8 ± 13.6 μg/ml, t1/2 was 1.54 ± 0.339 h, and CL was 0.09 ± 0.005 l/h/kg. The pharmacokinetic parameters of DSPE-PEG2000 in rats after i.v. administration were: AUC0−t was 104.9 ± 13.4 μg/ml*h, MRT0−t was 1.027 ± 0.034 h, Vd was 0.327 ± 0.062 l/kg, C0 was 83.6 ± 4.2 μg/ml, t1/2 was 0.981 ± 0.078 h, and CL was 0.23 ± 0.026 l/h/kg. The results of tissue distribution study indicated that DSPE-PEG2000 and DSPE-PEG5000 were mainly distributed in lung, fat, liver, and spleen. The highest concentration of both DSPE-PEG2000 and DSPE-PEG5000 were detected in lung. Furthermore, both DSPE-PEG2000 and DSPE-PEG5000 could be detected in brain, indicating that they can pass through the blood-brain barrier. Su (2019) also studied the inhibitory effects of both DSPE-PEG2000 and DSPE-PEG5000 on CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6 and 3A4. The IC50 values of both DSPE-PEG2000 and DSPE-PEG5000 were greater than 100 μg/ml for these CYP450 enzymes. These results indicated that both DSPE-PEG2000 and DSPE-PEG5000 did not significantly inhibit the activities of CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, and 3A4. DSPE-PEG can be metabolized into DSPE and PEG by esterase in vivo and excreted as PEG in urine. The cumulative
excretion of DSPE-PEG2000 at 0–168 hours after i.v. administration was 24.21% in urine and 3.82% in feces. The cumulative excretion of DSPE-PEG5000 at 0–168 hours after i.v. administration was 40.37% in urine and 8.79% in feces.

In summary, up to the present, the route of administration used in current pharmacokinetic studies of DSPE-PEG polymers is intravenous injection; there is no information about the absorption of DSPE-PEG in vivo right now. DSPE-PEG polymers are mainly distributed in lung, fat, liver, and spleen and are excreted through the bile and urine after intravenous administration. DSPE-PEG can be metabolized into PEG and DSPE under the action of esterase in vivo.

**Current Challenges and Future Perspectives**

This review describes the absorption, distribution, metabolism, and excretion of block copolymers containing polyethylene glycol. The main focus of this paper is the in vivo fate of these PEG-related copolymers after their release from nanocarriers. Safety and efficacy are two important considerations for the materials of PEGylated nanomedicine, as the consideration for the materials of PEGylated nanomedicine, as the polymers used in nanomedicine should accumulate in liver, spleen, lung, kidney, and other tissues and eventually cause toxic reactions. The polymers used in nanomedicine should be biodegradable and easily excreted. Furthermore, some PEG-related copolymers and their metabolites are biologically active and may cause toxic or adverse effects. For example, the metabolites of polylactic acid may cause hypercalcemia and acidosis. Furthermore, PEG-related polymers and their metabolites may also influence the biologic functions of relevant transporters and metabolic enzymes such as P-glycoprotein and CYP450s, resulting in drug-nanocarrier interactions and potential adverse reactions. Therefore, biologic inertness is another required feature of pharmaceutical polymers used in nanomedicine. Therefore, a thorough understanding of the in vivo fate of nanodrug pharmaceutical preparations and their excipients is very important for the safety and effectiveness of their clinical application. However, most of the current studies only focus on the in vivo pharmacokinetics of the free drug of nanomedicine and pay little attention to the excipients, especially the polymers. The research and comprehensive understanding of in vivo fate of the nanocarriers is not adequate. This is also the reason why numerous tested nanomedicines like Ferrogene and Resovist have been withdrawn from clinical use. Although a large number of studies have been carried out on nanocarriers, more studies are needed to deeply understand their pharmacokinetic behavior in vivo. More advanced and modern biotechnologies such as bioimaging and liquid chromatography-tandem mass spectrometry techniques should be used to strengthen the research on the transport mechanism of nanocarriers in vivo, so as to ensure their reasonable design, safety, and effectiveness. We believe this review will contribute to the understanding of the pharmacokinetics of block copolymers containing PEG used in nanocarrier drug delivery systems.

**Authorship Contributions**

Wrote or contributed to the writing of the manuscript: Yin, Pang, Shan, Gu.

**References**


Blonder JM, Burd L, Fufts JC, and Rosenthal GJ (1999) Dose-dependent hyperlipidemia in rabbits and may cause serious toxicity and adverse reactions when released in vivo. For example, high molecular mass PEG-related copolymers can ensure their reasonable design, safety, and effectiveness. We believe this review will contribute to the understanding of the pharmacokinetics of block copolymers containing PEG used in nanocarrier drug delivery systems.