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Review

Pharmacokinetics of Therapeutic Monoclonal Antibodies Used in Oncology

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Abstract. *This review presents the clinical pharmacokinetics of the marketed therapeutic monoclonal antibodies used in oncology. Aspects regarding absorption, tissue distribution, elimination as well as factors influencing pharmacokinetics, pharmacodynamics and kinetic interactions are also discussed.*

Monoclonal antibodies constitute a source of therapeutic agents used in the management of various diseases (1-6). In clinical oncology, the rationale for using monoclonal antibodies is to preferentially eradicate cancer cells by specific targeting and, in corollary, to gain in tolerance when compared with conventional agents or polyclonal antibodies (serotherapy). The therapeutic potential of monoclonal antibodies was rapidly recognized after the original report of Kohler and Milstein in 1975 (7) (*i.e.* large scale production of antibodies with a defined specificity) and the first paper dealing with the treatment of a cancer patient with a monoclonal antibody was published in 1980 (8). Nevertheless, the first monoclonal antibody for the treatment of cancer (edrecolomab) was only approved in 1994, in Germany, and was followed by rituximab in November 1997 in the United States (9). To date, worldwide nine monoclonal antibodies used in the treatment of cancer are commercially available (Table I).

Most of the pharmacological studies have focussed on the mechanisms of action (remaining often unresolved) and the cytotoxic properties of the monoclonal antibodies. Few works have described in full the clinical pharmacokinetics of therapeutic monoclonal antibodies. The lack of kinetic data, as well as the absence of knowledge concerning, for instance, the elimination process, is emphasized by the empiricism sometimes observed in the modalities of treatment. The purpose of this paper is to review the kinetic characteristics of the monoclonal antibodies approved for the treatment of cancer.

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General pharmacokinetics

Structure of the monoclonal antibodies. Therapeutic monoclonal antibodies are homogeneous immunoglobulins G (IgG) of murine or murine/human origin (chimeric, humanised). These proteins are bifunctional and are composed of two domains: the Fab domain that binds the antigen site and the Fc domain that interacts with effector cytotoxic cells and that is also involved in the transport across epithelial cells and in the regulation of serum concentrations. Chimeric and humanised antibodies display a human Fc domain. The molecular weight of monoclonal antibodies is high, around 150,000D, when compared with those of commonly used drugs (less that 1,000D). Chemotherapeutic monoclonal antibodies are either naked or conjugated to anticancer drugs (gemtuzumab ozogamicin) or radioisotopes (ibritumomab tiuxetan, tositumomab). The goal of conjugation is to increase the antitumoral activity of the antibody preferentially on targeted tissues by adding that of combined cytotoxic entities.

Absorption. With respect to the official labelling, all monoclonal antibodies are administered by the intravenous route. Some papers have described the activity of alemtuzumab given subcutaneously in patients with B-cell chronic lymphocytic leukemia (B-CLL) (10-16). The kinetics of subcutaneous alemtuzumab (30 mg 3 times weekly for up to 18 weeks) have been investigated in 20 untreated patients with B-CLL (16). When compared with values obtained in 30 other patients treated by the intravenous route, alemtuzumab plasma concentrations were judged similar, but the cumulative dose to reach the potentially lympholytic

concentration (1 mg/l) was higher (mean 551 mg *versus* 90 mg). The highest concentrations ranged between 0.6 and 24.8 mg/l (mean: 5.4) and between 2.8 and 26.4 mg/l (mean: 10.7) after subcutaneous and intravenous injections, respectively (16). The concentrations appeared to increase during the 12 weeks of subcutaneous treatment when considering an example of a patient profile (*i.e.* no steady state was attained) (16). The elimination parameters could not be determined (lack of terminal points) and it is not known if the subcutaneous route has an impact on the terminal half-life. Regarding a patient with myeloma, Gasparetto *et al.* (17) observed 2 absorption peaks (2 and 6 h) after subcutaneous administration of alemtuzumab. Overall, the bioavailability of subcutaneous alemtuzumab remains unknown.

Subcutaneous injection could lead to improved ease of administration (self administration) and, recently, formulations of concentrated crystalline rituximab and trastuzumab enabling a low volume of injection have been developed (18). In the case of alemtuzumab, subcutaneous administration was associated with diminished acute systemic toxicity when compared with intravenous infusion. However, site skin reactions were observed during the first 2 weeks (13).

Distribution. The penetration of monoclonal antibodies in tumor tissues is determinant for their activity. The antibodies target proteins expressed on the membrane of tumors cells, except bevacizumab that binds the circulating cytokine VEGF (Vascular Endothelial Growth Factor). Once the antibodies have been administered systemically, they must leave the tumoral vessels to diffuse in the interstitial matrix and hence reach the tumoral cells. Given that monoclonal antibodies were used for diagnostic purposes and tumor imaging, an extensive literature has been devoted to the distribution process. In general, the diffusion of monoclonal antibodies is limited and appears heterogeneous. This feature is underscored by their low volume of distribution (3-5l) approaching that of the plasma. This limitation had already been observed in the mid eighties after the first clinical trials (19). Animal studies have shown that most monoclonal antibodies were concentrated in the region adjacent to vessels (*i.e.* within 40 μm or 2 cell diameters) (20). Experimental works using intravital microscopy (a methodology enabling the continuous monitoring of cellular processes in living systems) have revealed the spatial and temporal heterogeneous aspects of tumoral blood circulation (21). The heterogeneity of tumoral distribution (inter- and intra-individual) of an anti-angiogenic antibody was originally observed, *in vivo*, in 20 patients using positron emitting tomography (22).

The distribution depends on the tumoral physiology and the characteristics of the antibody (affinity, molecular weight). Parameters relative to vascularization (permeability, blood supply), the difference of pressure between the tumoral capillary and the interstitial fluid, the extracellular matrix (particularly the content and the organisation of the collagen), the type and the localization of the tumor appear to determine the penetration of an antibody (23-28). Tumors display increased vascular permeability when compared with normal tissues. Unfortunately, this permeability is coupled with elevated interstitial pressure, thus hindering the extravasation of drugs (29, 30). Furthermore, as stated before, the blood supply is heterogeneous and can be impaired due to the compression of the tumoral vessels by cancer cells limiting the efficient and homogeneous delivery of drugs (31). In this regard, Jain *et al.* have shown that elevated interstitial pressure observed in tumors limits their penetration (26). In addition, they observed in xenografted mice that the interstitial diffusion of a non-specific IgG was prevented in tumors with high collagen levels (27).

Concerning the monoclonal antibody, one of the factors that may limit the diffusion in tumor tissues is the affinity of the macromolecule for the tumoral antigen and the intensity of the antigenic expression (32-35). This constitutes the concept of the "binding site barrier" proposed by Weinstein *et al.* at the end of the eighties. According to their experiments performed in guinea pigs bearing allografts of primitive (34) and metastatic tumors (35), the binding of antibodies to antigens at the periphery of the tumor limits and delays the diffusion. The lack of affinity for the tumoral antigens (IgG control of the same molecular weight) results in a more homogeneous distribution in the tumor. Increasing the dose of the monoclonal antibody partially enhances the tumoral penetration (34, 35). The decrease in affinity (by 100) for the tumoral antigen can lead to a better diffusion (36). Clinically, the problem of tumoral accessibility has been advanced by some investigators reporting the poor response of bulky lymph nodes to alemtuzumab in patients with B-CLL (37, 38). Given their limited diffusion, the therapeutic potential of monoclonal antibodies is enhanced when they are combined with a conventional chemotherapy (39, 40), in the adjuvant setting (subclinical tumor) (41) or when they are linked to cytotoxic agents (calicheamicin derivative) (42) or radioisotopes (iodine 131, yttrium 90) (43).

Plasma IgG are considered to be excluded from the central nervous system (CNS) given their size. Experimental studies have shown that IgG were transported across the blood brain barrier, from brain to blood, but not the reverse (*i.e.* from blood to brain) (44, 45). As will be discussed below, the efflux appears to be mediated by the transporter FcRn expressed in brain capillary endothelial cells (45). Diffusion in the CNS has been examined for trastuzumab and rituximab by determining their levels in the

Table I. *Presentation of the therapeutic monoclonal antibodies used in oncology.*

Monoclonal antibody	Type	Conjugated	Status	Target	Approved indications
Rituximab	IgG1, chimeric	No	Approved	CD20	Follicular NHL, Large cell NHL
Ibritumomab Tiuxetan	IgG1, murine	Yttrium 90	Approved	CD20	Relapsed low-grade/follicular NHL
Tositumomab	IgG2a, murine	Iodine 131	Approved USA	CD20	Relapsed follicular NHL
Alemtuzumab	IgG1, humanised	No	Approved	CD52	Refractory B cell CLL
Gemtuzumab Ozogamicin	IgG4, humanised	Derivative of calicheamycin	Approved USA	CD33	Relapsed AML
Trastuzumab	IgG1, humanised	No	Approved	HER2	Metastatic breast cancer
Cetuximab	IgG1, chimeric	No	Approved	EGFR	Relapsed metastatic colorectal cancer
Edrecolomab	IgG2a, murine	No	Approved Germany	Ag17-1a	Adjuvant treatment of colorectal cancer
Bevacizumab	IgG1, humanised	No	Approved USA	VEGF	Metastatic colorectal cancer

cerebrospinal fluid (CSF) as a surrogate. Trastuzumab concentrations have been measured in the CSF and the serum of a patient with meningeal carcinomatosis, following intravenous infusion (46). The concentration in the CSF was very low (0.21 mg/l *versus* 70.32 mg/l in the serum). According to another case report, the CSF concentration of rituximab administered intravenously (800 mg) to a patient with CNS involvement of a lymphoma was around 1% of the blood concentration (47). However, the treatment that associated 12 rituximab infusions and 5 intrathecal chemotherapies was active for the authors. The CSF concentration of rituximab (375 mg/m^2) was 0.35 mg/l in a patient with CNS lymphoma 7 days after intravenous infusion (48). The simultaneous serum concentration was not measured. In a general way, systemic treatment of cerebral tumors with monoclonal antibodies is likely to be ineffective because of poor diffusion across the blood brain barrier. An approach to increase the cerebral concentrations of rituximab has been to deliver the antibody by the intraventricular route *via* an Ommaya reservoir. One day after an intraventricular injection of 25 mg in a patient with CNS lymphoma, the concentration in the CSF of rituximab was 10 mg/l (48). After 4 intraventricular administrations (total dose 80 mg), the lymphoma cells were cleared from the CSF, indicating that the concentration was sufficient. Unfortunately, no response was observed in the parenchymal tumor mass (48). Extended kinetic data of intrathecal rituximab are available in the cynomolgus monkey. After inventricular injections of 2 to 5 mg, the CSF concentrations appeared to decrease biphasically with a terminal half-life of 5 h (49).

Elimination. Very few data are available on the elimination pathways of therapeutic monoclonal antibodies. By analogy to endogenous IgG, monoclonal antibodies could be metabolized in the vascular compartment by endothelial cells. IgG levels in rodents and probably in humans are regulated by the FcRn receptor (50). The FcRn receptor is known to possess two main functions: the transport of IgG (particularly the transfer of IgG across the maternofetal barrier) known as transcytosis and the control of IgG catabolism (50, 51). Expression of FcRn has been reported in endothelial cells, monocytes and epithelial cells of various human tissues (52) According to the hypothesis of Brambell *et al.* (53) and the model of Junghans and Anderson (54), the circulating IgG is internalized in low pH endosomes, wheras binding of the Fc domain with FcRn is promoted. The IgG is then recycled to the cell surface and released. The IgG in excess (not bound to the saturable FcRn) undergoes degradation in lysosomes (50, 55). This theory could explain the long half-life of IgG (3 weeks, except for 7 days for the subclass IgG3 in humans) relative to other plasma proteins (56) and the dose-dependent catabolism linked to the saturable protection. Mice knocked out for a deletion of the light chain of FcRn exhibit a shorter catabolic half-life for IgG than wild-type animals (0.47 *versus* 4.9 days) (54). In addition, the impact of FcRn in the disposition of a monoclonal antibody has been shown in the mouse (57). Nevertheless, comparative kinetics of chemotherapeutic monoclonal antibodies in FcRn-deleted and wild-type mice have not be reported. The behavior of monoclonal antibodies bound to target cells is unknown. Kennedy *et al.* (58) suggested that circulating B lymphoma cells bound to rituximab *via* the CD20 antigen could be sequestered by phagocytic cells where the complex antigen/antibody is removed, releasing into the circulation the CD20-depleted cancer cell. This could explain the acute loss of the membrane target (CD20) from the circulating lymphocytes after rituximab treatment in patients with CLL and supports the possible degradation of rituximab in phagocytic cells.

The renal excretion of chemotherapeutic monoclonal antibodies is poorly documented. The expression of FcRn in proximal renal cells (59) could suggest the transfer of IgG in urine by transcytosis. The bidirectional transport of IgG in human renal cells has been shown, *in vitro* (59). Renal insufficiency does not seem to have an impact on trastuzumab elimination (60) and successful administration of rituximab (*i.e.* at full dose) has been reported in 3 patients with renal failure (2 being on hemodialysis) (61,

Table II. *Pharmacokinetic data for rituximab. Results as mean (SD).*

No. of patients	Dose (mg/m^2)	Duration of study (days)	Assay	C_{max} (mg/l)	V_d	Cl (ml/min)	$t_{1/2}$ (hours)	Reference
9	375	4	Elisa	500(135)	ND	ND	225(102)	81
3	375	NR	Elisa	254(148)	ND	1.9(2.4)	33(21)	80
	375 (4th dose)			433(273)	ND	0.7(1)	76(43)	
14	375 (1st dose	7	Elisa	205(59)	ND	0.64(0,3)	76.6(31)	79
	375 (4th dose)	90		464(119)	ND	0.15(0,05)	205(95)	
4	250	90	Elisa	64(21)	Vss:10.7(2.7)	2.8(5.1)	560(607)	82
8	375	90	Elisa	92(34)	Vss:11.1(3.2)	0.7(1)	387(189)	
37	375 (1st dose)	7	Elisa	581	ND	ND	ND	83
	375 (8th dose)	180		1177				
26	375(9th dose)	365	Elisa	463 (109)	ND	ND	ND	84

C_{max}, Concentration peak ; V_d, volume of distribution ; Cl, Clearance ; $t_{1/2}$, terminal half-life ; NR, not reported ; ND, not determined; V_{ss}, volume of distribution at steady state.

62). In addition, rituximab is not removed by hemodialysis (63). Urinary excretion of some radiolabelled antibodies (64), and particularly ibritumomab tiuxetan (65), has been quantified using a radioactive assay. This methodology is generally hampered by lack of specificity since it can not discriminate the intact antibody from the unconjugated radioisotope. Urinary recovery of ibritumomab tiuxetan labelled with yttrium 90 (Y-90) accounted for 5.9% of the dose over 7 days, when determined with a radioactive assay (65). Preclinical data suggest a good retention of Y-90 in the chelator tiuxetan with no demonstrable loss over 4 days (66). This suggests that the urinary radioactivity reflects the catabolic fate and not the dissociation of the radionucleotide from the conjugate in the plasma. On the other hand, it can not exclude the presence of the labelled antibody. Clinically, the administration of ibritumomab tiuxetan labelled with Y-90 has been judged feasible at therapeutic dosage in a patient with chronic renal failure (67). Similarly, the urine excretion of trastuzumab labelled with indium 111 (In-111) has been estimated in 11 patients after a single injection (68). It represented 25% of the injected dose over 7 days.

Pharmacokinetic parameters

Some kinetic studies suffer from a lack of information regarding the assay, the sampling process and the calculation of the parameters. Furthermore, the data are often incomplete and not cleary presented. Concentrations of monoclonal antibodies in biological fluids are generally determined by enzyme-linked immunosorbent assay (ELISA). Radioimmunoassay (RIA) and flow cytometry have also been used. Concerning antibodies conjugated to radionuclides, pharmacokinetic characteristics are derived from the blood or organ radioactivity content. The sampling duration or the number of time-points often appear insufficient to correctly estimate the elimination parameters, given their potential long half-life (3 weeks, by analogy to endogenous IgG). Hence, some data must be interpreted with caution. In general, monoclonal antibodies exhibit a low volume of distribution (3-5l), approaching the serum/plasma volume, a low systemic clearance (around 0.5-4 ml/min) and a terminal half-life varying between 2 days and 28 days. The half-life of murine antibodies is generaly shorter than that of antibodies containing a human Fc domain (*i.e.* chimeric, humanised) (69). This could be due to a lesser affinity of the murine Fc domain for the human FcRn (70). Indeed, preclinical data have indicated that increasing the affinity of the antibody for FcRn could lead to a longer half-life (71, 72).

Rituximab

Rituximab is a chimeric anti-CD20 monoclonal antibody actually approved in the treatment of indolent and agressive forms of B-cell non-Hodgkin lymphoma (NHL). It has also been investigated in other CD20-positive B-cell malignancies such B-CLL (73, 74) and in autoimmune diseases (75). The CD20 antigen is expressed on the surface of normal and malignant mature B lymphocytes. Data from *in vitro* and *in vivo* studies suggest various mechanisms of action for rituximab such as antibody-dependent cellular cytotoxicity (ADCC), complement-mediated cytotoxicity (CMC), as well as induction of apoptosis (76, 77). Rituximab is administered by intravenous infusion at the dosage of 375 mg/m^2 weekly for 4 to 8 courses when given alone, or every 3 weeks when combined with chemotherapy. Three assays based on ELISA and flow cytometry for the determination of rituximab in human plasma have been published in full (78).

Table III. *Pharmacokinetic data for alemtuzumab. Results as mean (SD).*

No. of patients	Dose (mg)	Duration of study (days)	Assay	C_{max} (mg/l)	Vss (l) calculated for 70 kg	Cl (ml/min)	$t_{1/2}$ (days)	Reference
30	30 thrice weekly	84	Immunofluorescence flow cytometry	10.7(range 2.8-26.4)	12.95	ND	6.1	16
20	30 SC thrice weekly	126	Immunofluorescence flow cytometry	5.4 (range 0.6-24.8)	ND	ND	ND	16
11	10/day (5 days)	30	Immunofluorescence flow cytometry	2.5 (0.9)	ND	ND	21	97
5	10/day (10 days)	30	Immunofluorescence flow cytometry	6.1(2.3)	ND	ND	15	97
10	20/day (5 days)	30	Immunofluorescence flow cytometry	13.7(range 7.5-16.6)	ND	ND	8 (range 4-32)	98

C_{max}, Concentration peak ; V_{ss}, volume of distribution at steady state; Cl, Clearance ; $t_{1/2}$, terminal half-life ; ND, not determined; SC, subcutaneous.

The most complete kinetic data are derived from a phase III study that included 166 patients with low-grade NHL who were receiving rituximab at 375 mg/m^2 as single therapy (79) (Table II, (79-84)). The pharmacokinetic parameters were determined in 14 patients after the first and the fourth infusion over sampling periods of 7 and 90 days, respectively (Table II). The mean value of peak serum concentration (C_{max}) increased from 205.6 mg/l (standard deviation or SD: 59.9) after the first cycle to 464.7 mg/l (SD: 119) after the fourth cycle. The mean systemic clearances were 0.64 ml/min (SD: 0.3) and 0.15 ml/min (SD: 0.05) after the first and fourth courses, respectively (79). The terminal half-lives were estimated after the two administrations and were 76.6 h (SD: 31.1) and 205 h (SD: 95). In addition, serum rituximab concentrations were measured in 147 patients over 6 months. At 3 months post-treatment, rituximab was still detectable with a median value of 20 mg/l.

Ibritumomab tiuxetan

Ibritumomab tiuxetan is a radioimmunotherapeutic agent composed of a murine anti-CD20 antibody (ibritumomab) covalently linked to a chelator (tiuxetan) radiolabelled with Y-90 for therapy or In-111 for imaging (85). The concept of radioimmunotherapy is to target ionizing radiation to radiosensible tumors (lymphoma cells herein) by means of monoclonal antibodies. Radiolabelling can partially circumvent the problems of tumoral diffusion observed with naked antibodies and appears interesting for the treatment of bulky, poorly vascularized tumors (86, 87). The cytotoxicity of ibritumomab tiuxetan is attributable to the dose of radiation and to the intrinsic activity of the murine antibody carrier (87). Y-90 delivers beta emission capable of reaching cells 5 mm away from the antibody (corresponding approximately to 100-200 cell diameters) (86). Ibritumomab tiuxetan is approved for the treatment of patients with follicular NHL refractory to rituximab and of patients with relapsed or refractory low-grade, follicular or transformed B-cell NHL. The treatment consists of a single course (dose: 0.4 or 0.3 mCi/kg depending on the platelet count, maximum 32 mCi) preceded by rituximab infused at 250 mg/m^2, to deplete circulating blood B cells and improve the distribution of the conjugate. Prior to treatment (1 week), imaging is performed to evaluate the radiation absorbed dose to healthy organs and bone marrow (*i.e.* to ensure that the exposure is within acceptable limits) with In-111 ibritumomab tiuxetan. The gamma emitter In-111 is more suitable for imaging purposes than Y-90 (beta emitter) and has no therapeutic emission.

Pharmacokinetics of Y-90 ibritumomab tiuxetan have been estimated from In-111 measurements in blood samples of 72 patients, obtained over a period of 7 days (65). The median biological half-life of the antibody was 47 h (range: 22-140). It is shorter than that of the chimeric form (rituximab), probably attributable to a lesser affinity of the murine Fc domain for the human FcRn receptor. The elimination process has also to account for the exponential decrease of Y-90 beta-emitting radiation (half-life of 64 h). As suggested, a rapid clearance of the conjugate (both in terms of antibody and activity) is desirable to avoid excessive exposure. Urinary excretion of Y-90 ibritumomab tiuxetan was determined in 10 patients over 7 days. As stated before, it represented 5.9% (range: 3.2-8.5) of the injected dose when determined by Y-90 radioactivity (65).

Tositumomab

Tositumomab is a murine anti-CD20 antibody radiolabelled with iodine-131 (I-131) that has been approved for the treatment of NHL, with or without transformation, refractory to rituximab and that has relapsed following chemotherapy (87). When compared with the metal Y-90, I-131 links directly in the antibody and does not require a chelate like tiuxetan. Furthermore, I-131 produces beta and gamma emissions with a half-life of 193 h (8 days) (87). High energy gamma emissions necessitate restrictive radiation procedures to protect the patient's family members and healthcare workers. In addition, the range

Table IV. *Pharmacokinetic data for gemtuzumab ozogamicin. Results as mean (SD).*

No. of patients	Dose (mg/m^2)	Duration of study (days)	Assay	C_{max} (mg/l)	Vss (l)	Cl (ml/min)	$t_{1/2}$ (hours)	Reference
59	9	10	Elisa	2.9(1.3)	20.9(21.5)	4.4(3.8)	72.4(42)	104
49	9 (2nd dose)	28	Elisa	3.7(1.3)	9.9(8.8)	2.2(2.5)	93.7(67.4)	104
14(children)	6	10	Elisa	1.7(1.1)	13.7(14)	10.1(22.3)	43.1(22.7)	105
14(children)	6 (2nd dose)	10	Elisa	1.9(1.1)	12.5(13.1)	5.3(8.1)	49.4(25.6)	105
2(children)	7.5	10	Elisa	3.1	6.3	2	40	105
1(child)	7.5 (2nd dose)	10	Elisa	3	14.5	1.5	33.4	105
14(children)	9	10	Elisa	3.4(1)	6.5(5.5)	2.6(3.8)	63.7(44.3)	105
9(children)	9 (2nd dose)	10	Elisa	4.6(2.1)	3.9(2.1)	3.5(7.5)	57.8(33.4)	105

C_{max}, Concentration peak ; V_{ss}, volume of distribution at steady state ; Cl, Clearance ; $t_{1/2}$, terminal half-life.

of beta emission from I-131 is shorter than that of Y-90 (0.4 mm *versus* 5 mm). The mechanism of cytotoxic action involves the intrinsic activity of the murine antibody vector (ADCC, induction of apoptosis) and the dose of radiation (88, 89). Contrasting with rituximab, tositumomab shows very weak CMC activity (77). The treatment consists of a single course (patient-specific mCi dose adjusted to an absorbed total body radiation dose of 75 cGy) preceded by a dosimetric step with the same isotope. Unlike ibritumomab tiuxetan that is administered at a dose based on body weight, tositumomab requires individual dosimetry (*i.e.* assessment of patient-specific dose) given the highly variable (5-fold) clearance rate of I-131 (90). Prior to the injections of the radioconjugate (dosimetric and therapeutic), 450 mg of unlabelled tositumomab is infused to improve biodistribution.

Pharmacokinetic data of tositumomab are sparse. The mean effective half-life has been estimated to be 59.3 h (range: 24.6-88.6 h) using a radioactive assay (91). It has to be mentionned that the radioactive methodology is not specific for estimating the concentrations of the conjugate given the dehalogenation of iodine 131. The assay evaluates the activity of the radiolabelled antibody and of the free radionuclide, but excludes the naked antibody which is potentially cytotoxic. However, the half-life of tositumomab appears shorter than that of rituximab labelled with I-131 when estimated by the same methodology (*i.e.*, radioactive assay), reflecting the rapid elimination of murine antibodies when compared with their chimeric or humanised counterparts (92).

Alemtuzumab

Alemtuzumab (or CAMPATH-1H) is a humanised monoclonal antibody against the CD52 antigen abundantly expressed on lymphocytes and monocytes (93, 94). It was first developed in a murine form (Campath-1M then Campath-1G) 25 years ago, as a lymphosuppressive agent. According to the labelling, alemtuzumab is used for the treatment of refractory B-CLL as single agent and is administered at a fixed dose (30 mg), 3 times a week for 12 weeks. Alemtuzumab is also developed as an immunosuppressive agent with a different dosage regimen. The mechanisms of action appear to include immunological processes (ADCC, CMC) and induction of apoptosis (94).

Analytical methods have been published in full (95, 96), but the kinetic data in patients with B-CLL have only become available recently (16). Alemtuzumab was given intravenously to 30 patients (30 mg 3 times weekly) for up to 12 weeks. Serum samples were taken before and after the dose (15 min). The highest peaks and trough concentrations shown a wide variation ranging from 2.8 mg/l to 26.4 mg/l and from 0.5 mg/l to 18.3 mg/l, respectively (Table III) (16). The estimated mean terminal half-life was 6.1 days when determined in 16 patients. The kinetic properties of alemtuzumab given as an immunosuppressive have been partially documented in patients receiving allogeneic stem cell transplantation (Table III) (97, 98). The antibody was administered at 10 mg/day for 5 or 10 days to 11 and 5 patients, respectively (97). The pharmacokinetic profiles were determined after the last injection over a sampling period of 30 days. In both groups, alemtuzumab serum concentrations decreased in a biphasic manner with estimated terminal half-lives of 21 and 15 days (97).

Gemtuzumab ozogamicin

Gemtuzumab ozogamicin is an immunoconjugate consisting of a humanised anti-CD33 monoclonal antibody that is covalently linked to a derivative of the antitumor antibiotic calicheamicin (N-acetyl gamma calicheamicin dimethyl hydrazide) (99, 100). The formulation contains about 50% of the conjugate, the remainder being the naked antibody (101). The CD33 antigen is found on the surface of maturing normal and leukemic myeloid cells but is absent in stem cells, lymphoid cells and non

Table V. *Pharmacokinetic data for trastuzumab. Results as mean (SD).*

No. of patients	Dose	Duration of study (days)	Assay	C_{max} (mg/l)	Vd (l) calculated for 70kg	Cl (ml/min) calculated for 70kg	$t_{1/2}$ (days)	Reference
45	100mg	7	Elisa	ND	ND	ND	8.3 (5)	109
6	1mg/kg (single dose)	21	Elisa	19.1 (2.7)	3.6 (0.4)	16.4 (3.7)	2.7 (0.4)	110
3	2mg/kg (single dose)	21	Elisa	43.4 (8.5)	3.7 (1.4)	12.9 (1.6)	3.1(2)	110
3	4mg/kg (single dose)	21	Elisa	72.4 (17.2)	5.2 (0.9)	7.5 (1.2)	8.8 (1.3)	110
6	8mg/kg (single dose)	21	Elisa	169.6 (24.9)	4.9 (0.5)	6.5 (2.2)	10.4 (3)	110
476	2mg/kg	NR	NR	110	2.95	0.15	28.5	60
15	6mg/kg (12th dose)	21	Elisa	237	ND	0.14	18.3	107

C_{max}, Concentration peak ; V_d, volume of distribution ; Cl, Clearance ; $t_{1/2}$, terminal half-life ; NR, not reported ; ND, not determined.

Table VI. *Pharmacokinetic data for cetuximab. Results as mean (SD).*

No. of patients	Dose (mg/m^2)	Duration of study (days)	Assay	C_{max} (mg/l)	Vd (l) calculated for 70kg	Cl (ml/min) calculated for 70kg	$t_{1/2}$ (days)	Reference
7	20	28	Biocore assay	ND	4.3	3.6	ND	115
6	50	28	Biocore assay	ND	3.2	1.3	ND	115
10	100	28	Biocore assay	ND	3.2	0.9	ND	115
3	200	28	Biocore assay	ND	ND	0.5	ND	115
3	400	28	Biocore assay	ND	ND	0.4	ND	115
7	250 (3rd injection)	7	Elisa	130 (31)	Vss 3.5 (0.7)	0.6 (0.15)	4.4 (1.3)	117
8	50 (single injection)	21	NR	19.9	Vss 6.4	3	1.1	118
8	100 (single injection)	21	NR	54.7	Vss 4.4	1.4	1.6	118
7	250 (single injection)	21	NR	158.1	Vss 5.3	0.8	3	118
8	400 (single injection)	21	NR	205.1	Vss 4.4	0.65	3.1	118
7	500 (single injection)	21	NR	243.2	Vss 5.9	0.65	5.5	118

C_{max}, Concentration peak ; V_d, volume of distribution ; Cl, Clearance ; $t_{1/2}$, terminal half-life ; NR, not reported ; ND, not determined ; Vss, volume of distribution at steady state.

hematopoietic tissues (99). The binding of the conjugate to the leukemic cells is followed by internalization and release of the calicheamicin derivative after lysosomal hydrolysis. After reduction, the reactive derivative causes double-stand breaks in DNA leading to eventual cell death (102). The cytotoxic action could also be attributed to the carrier since a humanised antibody directed to CD33 (HuM195) has been shown to exhibit clinical antileukemic activity *via* presumptive ADCC (103). Gemtuzumab ozogamicin is indicated as single agent therapy for the treatment of patients with acute myeloid leukemia (AML) in first relapse who are 60 years of age or older and who are not candidates for cytotoxic chemotherapy. It is administered at 9 mg/m^2 as 2 infusions separated by 2 weeks.

The pharmacokinetics have been characterized in 59 patients with AML by measuring the plasma concentrations of the targeting antibody (representating the conjugated and naked forms) as well as the total and unconjugated forms of calicheamicin, after the two injections (104) (Table IV). After the first dose, the peak plasma concentration of the antibody was 2.86 mg/l (SD: 1.35), the clearance was 4.4 ml/min (SD: 3.8) and the terminal half-life was 72.4 h (SD: 42). Following the second dose, the clearance decreased to 2.2 ml/min (SD: 2.5) and the half-life increased to 93.7 h (SD: 67.4). Total calicheamicin derivative concentrations were shown to decrease in a similar way to those of the antibody, indicating that the anticancer agent remains conjugated until binding to the CD33+ cells. The terminal half-lives for total calicheamicin were 45.1 h (SD: 25.2) and 61.1 h (SD: 45.4) after the first and second injections, respectively. The plasma concentrations of unconjugated calicheamicin (*i.e.* the toxic entity) were very low after both injections when compared with those of total calicheamicin (5 μg/l *versus* 80 μg/l in regard to the concentration peaks) (104).

The kinetic characteristics have been examined in 29 children (age range 1-16 years) with AML in first relapse (105). Gemtuzumab ozogamicin was administered twice at

Table VII. *Pharmacokinetic data for edrecolomab. Results as mean (SD).*

No. of patients	Dose (mg)	Duration of study (days)	Assay	C_{max} (mg/l)	Vd (l) calculated for 70kg	Cl (ml/min) calculated for 70kg	$t_{1/2}$ (hours)	Reference
10	400	7	RIA	150 (SEM:22)	2.7	2.6 (SEM:0.3)	15.1	121
10	200	7	Elisa	55 (SE:5)	ND	ND	25.9 (SE:1.4)	122
2	500	1	Elisa	132 (SE:7)	ND	ND	19.8 (SE:1)	122
4	500	4	RIA	100	5.1(1)	3.5 (0.8)	16.8	123

C_{max}, Concentration peak ; V_d, volume of distribution ; Cl, Clearance ; $t_{1/2}$, terminal half-life ; SE, standard error; ND, not determined.

3 dose levels (6, 7.5 or 9 mg/m^2), the infusions being separated by 14 to 28 days. The elimination parameters of gemtuzumab ozogamicin were comparable to those observed in adults (Table IV) (105). Similar to adult patients, the exposures to the conjugate increase after the second infusion, by 63 and 77% for the dose levels 6 and 9 mg/m^2, respectively.

Trastuzumab

Trastuzumab is a humanised antibody targeted against the extracellular domain of the transmembrane oncoprotein HER2 (also known as erbB2 or neu) (60, 106). HER2 encoded by the gene of the same denomination belongs to the epidermal growth factor receptor (EGFR, HER) family that includes 4 known members (HER1, HER2, HER3, HER4). The overexpression of HER2 is found in 20-30% of breast cancers and is associated with aggressive tumors and a poorer prognosis (106). Trastuzumab is approved for the treatment of metastatic breast cancer overexpressing HER2 in first-line with paclitaxel or docetaxel and as single agent in second- or third-line. Given alone or in combination, trastuzumab is administered weekly, by intravenous infusion, at a loading dose of 4 mg/kg and then at 2 mg/kg until disease progression. Trastuzumab has also been evaluated when given every 3 weeks (8 mg/kg then 6 mg/kg) (107). The mechanism of action is partially known. Various processes have been suggested such as ADCC, stimulation of HER2 endocytosis, removal of the receptor from the cell surface and anti-angiogenic effects (60, 108).

The pharmacokinetic data of trastuzumab are presented in Table V (60, 107, 109, 110). The first study was conducted in 45 patients at a fixed dose (100 mg) but could not accurately estimate the terminal half-life (109). The sampling period was short (7 days) and only included 2 points. Tokuda *et al.* (110) evaluated the kinetic profile of trastuzumab administered on a body weight basis, at various dosages (1 mg/kg, 2 mg/kg, 4 mg/kg and 8 mg/kg) over a sampling period of 21 days. With regards to the current therapeutic dosage (2 mg/kg), the terminal half-life was short, around 3 days, when determined in 3 patients after the first dose. Calculated for a woman of 70 kg, the mean volume of distribution was 3.7l and the mean clearance was 0.5 ml/min. Recent data, published partly in an abstract form (111) and based on a population pharmacokinetic approach (two-compartment model), indicate a slower elimination of trastuzumab (administered weekly at 2 mg/kg) with a terminal half-life of 28.5 days (confidence interval, 25.5-32.8 days) and a clearance of 0.15 ml/min. These data are now included in the official labelling. Hence, the trastuzumab half-life appears to approximate the half-life of endogenous IgG. This prompted investigators to test trastuzumab with a longer dosing interval. Leyland-Jones *et al.* (107) have studied trastuzumab pharmacokinetics when given every 3 weeks at 6 mg/kg in 15 patients. The terminal half-life estimated after 12 courses was 18.3 days, with a sampling period of 21 days.

Cetuximab

Cetuximab is a chimeric monoclonal antibody that binds the extracellular portion of EGFR or HER1 (112, 113). The receptor HER1 is present in healthy tissues and its overexpression in various malignancies has been found to be correlated with poor prognosis (114). Cetuximab prevents the binding of endogenous ligands (such as Epidermal Growth Factor, Transforming Growth Factor alpha) to HER1, affecting the activation of the downstream signaling network and leading to the reduction of cell proliferation and the promotion of apoptosis (112). In addition, cetuximab induces down-regulation of HER1, can exhibit ADCC and appears to decrease the production of angiogenic and metastatic factors (112). Cetuximab has been investigated in the treatment of various cancers, given alone or in association with drugs or radiation therapy. Currently, it is approved in combination with irinotecan for the treatment of patients with colorectal cancer who no longer respond to treatment with irinotecan. It can be used as a monotherapy if patients do not tolerate the camptothecin analog. Given alone or in combination with

Table VIII. *Pharmacokinetic data for bevacizumab. Results as mean (SD).*

No. of patients	Dose (mg/kg)	Duration of study (days)	Assay	C_{max} (mg/l)	Vd (l) calculated for 70 kg	Cl (ml/min) calculated for 70 kg	$t_{1/2}$ (days)	Reference
12	3 (8th dose)	30	Elisa	ND	ND	ND	13	129
5	1 (single dose)	28	Elisa	27.5	ND	0.17	ND	127
4	3 (single dose)	28	Elisa	77	ND	0.16	ND	127
5	10 (single dose)	28	Elisa	251	ND	0.14	ND	127
491	NR	NR	NR	ND	2.66	0.14	19.9	130

C_{max}, Concentration peak ; V_d, volume of distribution ; Cl, Clearance ; $t_{1/2}$, terminal half-life ; NR, not reported ; ND, not determined.

irinotecan, cetuximab is administered by intravenous infusion once a week at a loading dose of 400 mg/m^2 and then at 250 mg/m^2 until progression.

Kinetic data relative to two phases I studies of cetuximab given alone and associated with cisplatin or radiotherapy have been reported but are not very informative (115, 116) (Table VI). In one investigation, where the dose of cetuximab was escalated from 20 mg/m^2 to 100 mg/m^2, the clearance was shown to decrease from 3.6 ml/min to 0.9 ml/min (115). One investigation, published in an abstract form (117), has been devoted to cetuximab kinetics in its current dosage regimen (400 mg/m^2 then 250 mg/m^2 weekly). The terminal half-life estimated in 7 patients over a sampling period of 7 days was 4.4 days (SD: 1.3). Recent data have been obtained from 39 patients receiving cetuximab, as single infusion, at doses ranging from 50 mg/m^2 to 500 mg/m^2 (118). As reported previously, the clearance decreased from 3 ml/min to 0.65 ml/min.

Edrecolomab

Edrecolomab was the first monoclonal antibody to be approved for the treatment of cancer. It is a murine antibody that recognizes a cell-surface glycoprotein, known as 17-1A or Ep-CAM, which is expressed on epithelial tissues and on various carcinomas (119, 120). Edrecolomab is thought to act by ADCC and CMC (119). It is approved in Germany, as post-operative adjuvant therapy for patients with Dukes' stage C colorectal cancer. Edrecolomab is administered, as single agent, by intravenous infusion at 500 mg after surgery, followed by 4 doses of 100 mg given every 4 weeks. Edrecolomab is currently the only antibody to be used in the adjuvant setting.

The clinical pharmacokinetics of edrecolomab are presented in Table VII (121-123). No data are available in patients with colorectal cancer who received the antibody as adjuvant therapy. The kinetic profile has been examined in 25 patients with metastatic gastrointestinal cancer who received the antibody at 400 mg (corresponding roughly to the first dose) for 1 to 4 weekly infusions. Concentrations were determined by RIA after each infusion in various groups of patients. The kinetic properties were comparable after each of the 4 infusions. Hence, after the second dose, the terminal half-life was 15.1 h (standard error or SEM: 1.8), the volume of distribution was 2.7l (calculated for a patient of 70 kg) and the clearance was 2.5 ml/min (SEM: 0.3) when estimated in 10 patients (121) over a sampling period of 7 days. The pharmacokinetics have been determined in patients who received the antibody in repeated doses (200-500 mg) with various dosing intervals (1 day-6 weeks). The estimated terminal half-life ranged from 19.8 h to 28.6 h (122).

Bevacizumab

Contrasting with other chemotherapeutic antibodies that directly target tumor cells, bevacizumab binds the circulating growth factor VEGF, an endothelial cell mitogen and an angiogenesis inducer released by a variety of tumor cells. Bevacizumab is a humanised monoclonal antibody that neutralizes various active isoforms of VEGF preventing their interactions with specific receptors and, hence, the formation of tumoral blood vessels (124). The inhibition of tumor growth has been observed with the murine version in preclinical models (125). In addition, the antivascular effects of bevacizumab in terms of reduction of tumor blood perfusion and blood volume have been shown in 4 out of 5 patients with rectal cancer analyzed (126). Bevacizumab administration leads to a rise in endogenous total serum VEGF concentrations attributable to a decrease in the clearance of the circulating growth factor after complexation with the antibody (127, 128). Bevacizumab is being investigated given alone and combined with chemotherapy, immunotherapy and radiotherapy in the management of various solid tumors (124). Currently, it is indicated for first-line treatment of patients with metastatic colorectal cancer in combination with 5-fluorouracil-based chemotherapy. Bevacizumab is administered by intravenous infusion at 5 mg/kg every 14 days until disease progression.

Published kinetic data are mainly derived from two phases I studies (Table VIII) (127, 129, 130). In the first

investigation, the antibody was administered alone as a weight-based dose ranging from 0.1 mg/kg to 10 mg/kg in groups of 4-5 patients (127). The clearance was low (around 0.16 ml/min) and stable in the 1-10 mg/kg (therapeutic) range. In the second study, bevacizumab was combined with various anticancer drugs (doxorubicin, carboplatin+ paclitaxel, 5-fluorouracil+folinic acid) and was infused at the dose of 3 mg/kg, weekly, in 12 patients (129). The terminal half-life was estimated to be 13 days. In addition, the kinetic data obtained in 491 patients have been pooled for a population approach (130). The analysis, published in an abstract form, reported a volume of distribution of 2.66l, a clearance of 0.14 ml/min and a terminal half-life of 19.9 days (130). Results obtained from two phases II studies including 69 patients suggest that minimum steady state concentrations (determined at 3 months) are similar when bevacizumab is given at 5 mg/kg every 2 weeks (the approved regimen) or 7.5 mg/kg every 3 weeks (131).

Factors influencing pharmacokinetics

Tumor burden. Significant variations in pharmacokinetic parameters of monoclonal antibodies appear imputable to changes in antigen expression (*i.e.* to the evolution or to the variability of the tumor burden). As seen previously, gemtuzumab ozogamicin clearance has been shown to decrease by 2-fold between the 2 doses (104). Furthermore, a relationship was established between the percentage of variation of peripheral blasts cells (the target of gemtuzumab ozogamicin) and the changes in the areas under the serum time curve (AUC) of the antibody (104). Hence, the increase of systemic exposition to gemtuzumab ozogamicin could be related to the disappearance of circulating leukemic targets.

Similarly, Berinstein *et al.* (79) found in 14 patients a significant decrease of rituximab clearance, which in turn resulted in the increase of the terminal half-life between the first and the fourth perfusions. Although they did not evaluate in parallel the variation of the CD20 antigen expression, they attributed the delay in elimination of the antibody to the decrease of the peripheral target cells count. An inverse correlation was also reported between the count of blood B cells at baseline and rituximab concentrations determined at various times in 113 patients with NHL. Likewise, they found that the anti-CD20 antibody serum concentrations correlated inversely with the tumor bulk estimated before treatment (79). Igarashi *et al.* (132) indicated that 30 patients without extranodal disease displayed significantly higher serum levels than 36 patients with extranodal sites (81.2 mg/l *versus* 57 mg/l). Hence, some authors have suggested adjustment of the dosage of rituximab according to the tumor burden (133). Nevertheless, Mangel *et al.* (84) have found that rituximab levels in patients with minimal disease (consolidative immunotherapy after autologous stem cell transplantation) were similar to those observed in the study of Berinstein. As stated by the authors, the relationship between tumor burden and pharmacokinetics of rituximab appears more complex than previously reported.

Wiseman *et al.* (65) have shown an inverse relationship between the estimated effective half-life of Y-90 ibritumomab tiuxetan and tumor size at baseline in 70 patients with NHL. Finally, a population kinetic study indicated that tumor burden could affect trastuzumab clearance (111).

Shed target. The extracellular domain (ECD) of the receptor HER2 is cleaved by proteolysis and can be found in the serum of patients with breast cancer. Some authors have suggested that circulating serum HER2 ECD affect the kinetic profile of trastuzumab given alone (109) or combined with cisplatin (134). Patients with serum shed ECD above 0.5 mg/l display a shorter half-life for trastuzumab than patients with ECD less than 0.5 mg/l [2.9 days (SD: 3.2) *versus* 9.2 days (SD: 5.3) given alone and 4 days (SD: 2.6) *versus* 11 days (SD: 4.4) combined with cisplatin]. As discussed above, the estimation of the half-life was based on a short sampling duration including only two points. The relationship between the half-life and the clinical response was not established. Recent data have shown that high baseline serum ECD (>15 μg/l) is associated with a significantly better response to trastuzumab and a trend toward a longer overall survival (135).

Free circulating CD52 (sCD52) has been detected in the plasma of 117 patients with LLC (136). In addition, the levels were shown to correlate positively with the severity of the disease. Interestingly, plasma alemtuzumab concentrations of a patient with low levels of sCD52 were higher than that of a patient with high levels of sCD52. Five patients with a complete response had significantly lower levels of sCD52 than those of 9 non-responders. Unfortunately, associated alemtuzumab concentrations were not reported. Furthermore, *ex vivo* experiments indicated that plasma sCD52 can diminish the binding of alemtuzumab to cells (136). These preliminary results suggest that sCD52 could affect alemtuzumab kinetics and hence therapeutic response.

Circulating endogenous IgG. The serum concentration of endogenous IgG could theoretically affect the catabolic rate of monoclonal antibodies. Hence, high levels of IgG, such as observed in patients with IgG-myeloma, are associated with high catabolic rates probably linked to the saturation of the FcRn receptors (56). Currently, no monoclonal antibody is approved for the treatment of multiple myeloma. Rituximab has been tested in CD20+ myeloma patients and has shown a limited activity (74). In addition, a

therapeutic potential has been suggested for alemtuzumab given the expression of CD52 on plasma cells from certain patients with myeloma (137). It remains to be evaluated whether high serum concentrations of IgG have an impact on a monoclonal antibody clearance.

Gender. A population kinetic study for bevacizumab, published in an abstract form, has shown that men have a 26% faster clearance than women after adjusting the weight (130). Nevertheless, this result has no impact on the current dosage (5 mg/kg man or woman) according to the official labelling.

Body weight, body surface area. Curiously, given their relative common kinetic characteristics, dosing of monoclonal antibodies is heterogeneous. The prescribed dose is either fixed (alemtuzumab, edrecolomab) or calculated using estimated body surface area (rituximab, gemtuzumab ozogamicin, cetuximab) or weight (trastuzumab, bevacizumab). Population pharmacokinetic studies (published in an abstract form) indicate that the weight may influence the clearance of bevacizumab and that of trastuzumab (111, 130). It remains unclear whether body surface area significantly affects the pharmacokinetics of rituximab, cetuximab and gemtuzumab ozogamicin. In addition, it is not proven that dosing normalized to weight or body surface area significantly reduces the variability of elimination parameters when compared to a fixed dose and in corollary affects the clinical results. Fixed dosing would simplify the administration of monoclonal antibodies.

Pharmacodynamics

Pharmacodynamic relationships (*i.e.* linking pharmacokinetics parameters to clinical response for a given dosage) have been searched for some monoclonal antibodies. Regarding gemtuzumab ozogamicin, no relationship could be found between the plasma concentrations of the antibody (AUC) measured after the first injection and the antileukemic activity determined in 59 patients in first relapse (104).

Rituximab serum levels have been shown to correlate with therapeutic response in 137 pretreated patients with low-grade or follicular NHL (79). Hence, residual and peak concentrations of the anti-CD20 antibody given as monotherapy were statistically higher in responders than in non-responders when measured at various times from the first infusion to 3 months post-treatment. After the fourth (last) infusion rituximab levels were 502.8 mg/l for the 67 responders *versus* 412.4 mg/l for 74 non-responders. Three months after treatment, concentrations were still evaluable and were 25.4 mg/l in 62 responders compared with 5.9 mg/l in 42 non-responders (79). A relationship between rituximab serum levels and response has been searched in 66 relapsed patients with indolent NHL and mantle cell lymphoma who received the antibody as single agent (132). Contrasting with the study of Berinstein *et al.*, there was no relationship between rituximab levels determined before the third infusion and therapeutic response. Nevertheless, a longer progression-free survival was associated with serum levels above 70 mg/l (132). In a small study including 12 patients with refractory aggressive NHL and treated by rituximab administered as single agent, the AUC of the responders (n=7) was statistically higher (1.6-fold) than that of the non-responders (n=5) (138). Collectively, higher serum concentrations of rituximab appear to be associated with less advanced disease which can be translated by a lower expression of the binding target and ultimately to a better therapeutic response.

Regarding ibritumomab tiuxetan, no significant link could be established between pharmacokinetics and response, except a longer effective half-life in responders (28 h *versus* 25 h) (65).

Drug-drug pharmacokinetic interactions

Chemotherapeutic monoclonal antibodies are sometimes combined with conventional anticancer agents in order to enhance the antitumoral activity. Antibody-drug kinetic interactions have been explored with cetuximab, trastuzumab and bevacizumab in studies mostly reported in an abstract form. Neither the antibody, nor the anticancer drug alters the disposition of each other. This is not surprising since the likehood of a kinetic interaction with a monoclonal antibody appears low. Indeed, current chemotherapeutic monoclonal antibodies have not been shown to interact with metabolic systems (*i.e.* the cytochrome P450 superfamily), orphan nuclear receptors (*i.e.* pregnane X receptor or PXR, constitutive androstane receptor or CAR) or drug transporters (*i.e.* P-glycoprotein or P-gp) that constitute the principal sources of kinetic interactions. It has to be stressed that ozogamicin (the calicheamycin derivative linked to gemtuzumab) is suspected to be transported by P-gp (139).

Trastuzumab. The impact of anticancer drugs on trastuzumab elimination has been investigated in a population pharmacokinetic study that included the data obtained in 476 patients with breast cancer. The results, published in an abstract form, reported that neither the association of an anthracycline (not specified) plus cyclophosphamide nor paclitaxel influenced the clearance of the antibody (140).

The effects of trastuzumab on epirubicin pharmacokinetics have been evaluated in 7 patients with breast cancer to assess if the increased cardiotoxicity of anthracyclines combined with the antibody could be due to an interaction (141). The anthracycline was given every 3 weeks at 75 mg/m^2 combined with docetaxel. Trastuzumab was infused

at 4 mg/kg after the anticancer drugs at the first cycle and then at 2 mg/kg weekly just before epirubicin, when associated. Epirubicin and epirubicinol (main metabolite) plasma concentrations were determined at the first and at the sixth cycle (*i.e.* after 15 injections of trastuzumab). Pharmacokinetics of epirubicin and epirubicinol were not altered by trastuzumab (141).

Similarly, the safety and the kinetic impact of trastuzumab (4 mg/kg) given with high-dose chemotherapy (cyclophosphamide 1.875 g/m^2/day, 3 days; carmustine, 600 mg/m^2 and cisplatin 55 mg/m^2/day, 3 days) have been examined in 24 patients with advanced breast cancer (142). The median AUCs of the 3 alkylating agents were comparable to those determined in 476 historical controls (without trastuzumab). With regard to cardiac events, no apparent increased toxicity imputable to the addition of the antibody was observed when compared with previously treated patients (142).

Trastuzumab has been evaluated in combination with chemotherapy in patients with HER2-overexpressing advanced non-small cell lung cancer (143). Neither cisplatin nor gemcitabine pharmacokinetics (total body clearance, terminal half-life) were modified by co-administration of trastuzumab when determined in 11 and 15 patients, respectively (143).

Cetuximab. Data obtained from a phase I study suggested that the systemic clearance of cetuximab (100 mg/m^2) determined in 10 patients was similar to that estimated in 3 patients who received cisplatin (100 mg/m^2) one hour after the antibody (0.95 and 0.97 ml/min, respectively for a patient of 70 kg) (112).

An interaction has been explored between cetuximab and irinotecan since the two drugs are combined in the official regimen (117). The pharmacokinetic profile of irinotecan (350 mg/m^2) was not modified by cetuximab given at 400 mg/m^2 and then 250 mg/m^2 for 2 weekly doses in 6 patients. Furthermore, irinotecan (350 mg/m^2) had no impact on cetuximab concentrations when determined in 7 patients at the third infusion of the antibody (117).

Increasing the weekly dose of infusional 5-fluorouracil from 1.5 g/m^2 to 2 g/m^2 did not alter the kinetics of cetuximab given weeky in 19 patients with metastatic colon carcinoma (144). Finally, docetaxel (75 mg/m^2 every 3 weeks) did not modify the kinetics of the antibody given weekly in 47 patients with advanced non-small cell lung cancer (145).

Bevacizumab. Bevacizumab is used in the treatment of metastatic colorectal cancer associated with irinotecan and 5-fluorouracil. A population pharmacokinetic study, published in an abstract form, has reported that the combination of the two anticancer drugs did not influence the clearance of the antibody (130).

Conclusion

Overall, the clinical kinetics of chemotherapeutic monoclonal antibodies discussed above are characterized by a low volume of distribution approaching that of plasma and a slow elimination. Antibodies of murine origin exhibit a shorter terminal half-life that antibodies containing a human Fc domain. However, the elimination pathways of these agents remain to be determined.

Elimination parameters appear to partly support the dosing intervals of some antibodies (rituximab, cetuximab, bevacizumab) administered weekly or biweekly. Regarding trastuzumab, the estimation of a longer half-life (28.5 days) has not currently reconsidered the weekly dosing scheme. Alemtuzumab pharmacokinetics and particularly the impact of sCD52 require further studies in patients with CLL . This could alter or optimize the current arbitrary schedule of administration (3 injections per week). Radiolabelled monoclonal antibodies display a rapid elimination, probably due to their murine origin. Even though they are administered as a single dose, it is desirable to reduce the exposure given the toxicity of radioisotopes. Gemtuzumab ozogamicin kinetics are well documented. The half-life of the conjugate appears short for a humanised antibody (less than 3 days). The dosing interval (14 days) has been selected to allow complete elimination before the second dose in order to minimize accumulation and hence toxicity.

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