Cancer Chemother Pharmacol (2006) 57: 819–825
DOI 10.1007/s00280-005-0088-2

ORIGINAL ARTICLE

Rogier R. Press · Tessa Buckle · Jos H. Beijnen
Olaf van Tellingen

The effect of P-glycoprotein and cytochrome P450 3a on the oral bioavailability of vinorelbine in mice

Received: 7 March 2005 / Accepted: 25 July 2005 / Published online: 15 September 2005
© Springer-Verlag 2005

Abstract *Purpose*: This study was designed to determine the effects of P-glycoprotein (P-gp) and cytochrome P450 3a metabolism on the oral bioavailability of the vinca alkaloid Vinorelbine (Navelbine; VRL). *Methods*: Pharmacokinetics of VRL were determined in FVB wild-type and mdr1a/1b (−/−) mice after oral and intravenous administration of 10 mg/kg VRL with or without oral ritonavir (5 mg/kg) prior to VRL. Serial blood samples were drawn for a period of up to 48 hours using mice with a cannulated jugular vein. Feces was collected for a period of 96 hours. VRL was determined by ion-exchange HPLC in combination with fluorescence detection. *Results*: The oral bioavailability in wild-type was 16.0 ± 1.4% (mean ± SE) and was not significantly higher in mdr1a/1b (−/−) mice (17.9 ± 0.7%). Both after intravenous and oral administration, the AUC was not significantly different between wild-type and mdr1a/1b(−/−) mice. When RTV was co-administered the AUC of intravenous VRL increased significantly by 30% (p = 0.012). Because RTV increased the AUC of oral VRL by 83% the oral bioavailability was increased to 22.5 ± 2.3% (p = 0.016). The fecal recovery of unchanged VRL was about 34 and 6% of the dose in wild-type and mdr1a/1b(−/−) mice, respectively, and was not altered by RTV. *Conclusion*: This study shows that P-gp has little effect on the disposition and oral bioavailability of VRL. A substantial fraction of an oral dose of VRL is absorbed from the gut of wild-type mice. Consequently, first-pass metabolism is the most important factor for explaining the modest oral bioavailability, but the results with RTV suggest that cyp3a plays only a modest role in metabolic breakdown in mice. Apparently, other routes of metabolic elimination are more important. These results suggest that also in patients the oral bioavailability may not gain substantially from the co-administration of a potent P-gp and/or Cyp3a inhibitor.

Keywords P-glycoprotein · Navelbine · Ritonavir · High-performance liquid chromatography · Pharmacokinetics

R. R. Press · T. Buckle · O. van Tellingen (✉)
Department of Clinical Chemistry, The Netherlands
Cancer Institute, Antoni van Leeuwenhoek Hospital,
Plesmanlaan 121, 1066 CX, Amsterdam, The Netherlands
E-mail: o.v.tellingen@nki.nl
Tel.: +31-20-5122792
Fax: +31-20-6172799

J. H. Beijnen
Department of Pharmacy and Pharmacology,
Slotervaart Hospital/The Netherlands Cancer Institute,
Louwesweg 6, 1066 EC, Amsterdam, The Netherlands

J. H. Beijnen
Drug Toxicology, Faculty of Pharmacy,
Utrecht University, Utrecht, The Netherlands

Introduction

Vinorelbine (5′-nor-anhydro-vinblastine) tartrate-salt (Navelbine; VRL) is a semisynthetic vinca alkaloid derivate that has established clinical activity against non-small cell lung and advanced breast cancer [9, 10]. It exerts its action by binding to microtubules, thus interfering in the mitotic spindle formation and leading to cell cycle arrest in G2-M phase. VRL is structurally different from the other vinca alkaloids through a modification in the catharanthine ring moiety. This causes VRL to disrupt mitotic versus axonal microtubules more selective, which is believed to explain the reduced clinical neurotoxicity relative to the other vinca alkaloids [6, 20].

Vinca alkaloids including VRL are substrates of P-glycoprotein (P-gp) [1, 19, 23], a membrane associated drug efflux pump that confers multidrug resistance to tumor cells in vitro and possibly also to tumors in patients [18]. Although, P-gp was discovered in tumor cells, it is expressed in a variety of normal tissues with an excretory or barrier function such as the biliary canulicular membrane in the liver, the enterocytes lining the intestinal wall, the endothelial cells forming the blood

brain barrier, the adrenal gland, the testis and the placenta. Consequently, P-gp is involved in the disposition of many drugs [12] and could be of importance for the therapy with VRL. VRL is marketed as an intravenous formulation and as an oral (soft capsule) formulation [15]. The oral route is preferred over intravenous administration in terms of cost-effectiveness and patient convenience [4, 8] and was shown to be efficacious [11]. However, drug efflux in the gut wall by transport proteins such as P-gp and first-pass metabolism by cytochrome P4503A may limit the oral bioavailability [5]. Previous studies with oral taxanes have shown that P-gp plays a major role in the bioavailability of oral paclitaxel [24], but is less important in case of docetaxel where the effect of CYP3a was much more pronounced [3]. The oral bioavailability of VRL in patients was $27 \pm 12\%$ of the dose, leaving room for improvement [22].

The aim of the current study was to evaluate to what extent P-gp and CYP3a reduce the oral bioavailability of VRL in mice. For this purpose we established the pharmacokinetics of VRL in FVB wild-type and mdr1a/1b (−/−) mice receiving VRL as single agent or in combination with the potent CYP3a inhibitor ritonavir.

Materials and methods

Materials and reagents

VRL (10 mg/ml) originated from Centre de Recherche Pierre Fabre (Paris, France). Deacetylvinblastine sulphate (DVBL) was obtained from the faculty of Pharmacy (University of Utrecht, The Netherlands). Ritonavir (RTV; 80 mg/ml, NORVIR) oral formulation was from Abbott (Abbott Park, IL, USA). All other reagents were purchased from Merck (Darmstadt, Germany) and were of analytical quality, except for acetonitrile, which was of HPLC grade. Blank human plasma was obtained from healthy donors (Bloodbank, Amsterdam, The Netherlands). Saline was purchased from Braun (Emmer Compascuum, The Netherlands). Water, purified by the Milli-Q plus system (Millipore, Bedford, MA, USA) was used throughout. Bovine serum albumin (BSA) originated from Roche Diagnostics GmbH (Mannheim, Germany).

Plasma pharmacokinetic studies

The experiments were carried out in FVB wild-type and mdr1a/1b (−/−) mice aged between 9 and 14 weeks. Female mice were used arbitrarily. They had free access to food and water prior to and during the experiments. Animals were maintained and handled in accordance with institutional guidelines based on Dutch law. For serial blood sampling, the jugular vene of the mice was cannulated as described previously [2] two days prior to the start of the experiment. During the experiments the mice were housed separately. The cannule was connected to the lid of the cage with a swiffle that allowed the mouse to move freely. The cannule was filled with heparine solution (5 mg/ml in saline) to prevent clogging.

VRL has been administered orally by gavage as a saline-solution of 1 mg/ml at the dose level of 10 mg/kg. Separate cohorts of mice received VRL as an intravenous bolus injection at a dose level of 10 mg/kg. The effect of RTV on the pharmacokinetics of VRL was studied in wild-type mice. A dose of 5 mg/kg of RTV was administered orally 30 min prior to an oral or i.v. dose of 10 mg/kg VRL.

Serial blood samples were drawn for over a period of 48 h after drug administration at approximately $t =$ 15, 30 min and 1, 2, 4, 8, 24, 32 and 48 h (oral route) and $t =$ 2, 5, 30 min and 1, 2, 8, 24, 32, 48 h (i.v. route). After every sample collection, an equal amount of donor blood from an FVB mouse was returned to the mouse. About 120 to 150 µl blood samples were drawn at each time point except for the last sample where a larger (300 µl) sample was drawn. Animals were killed immediately after the last sampling. Blood samples were centrifuged for 10 min (3,500 g, 4°C) and plasma was separated and stored at −20°C until analysis.

Fecal excretion study

Fecal excretion of VRL was studied after oral administration. Cohorts of wild-type and mdr1a/1b(−/−) mice were housed in Ruco Type M/1 metabolic cages (Valkenswaard, The Netherlands) where they had free access to food and water. They were accustomed to the cages for a period of three days. Both genotypes received VRL in combination with either oral RTV 5 mg/kg or the RTV-vehicle 30 min prior to VRL. Feces was collected in 24 h fractions for up to 96 h. Feces samples were homogenized in 10 volumes of 40 g/l BSA solution in water using a Polytron PT1200 homogenizer (Kinematika AG, Switzerland) and were stored at 20°C until analysis.

Drug analysis

Plasma samples were thawed at room temperature. The plasma samples of 50 µl (150 µl for the 48-h sample) were made up with blank human plasma to a final volume of 500 µl in a glass tube with a PTFE covered screw cap and 50 µl of internal standard (100 ng/ml DVBL in acetonitrile) was added, followed by 3 ml of diethyl ether. The tubes were shaken vigorously for 5 min and were centrifuged for 5 min at 1,500 g (4°C). The glass tubes were placed in an alcohol bath with solid carbon dioxide (−70°C). Immediately after the aqueous phase was frozen, the upper organic layer was decanted into a clean polypropylene tube and evaporated to dryness under a gentle stream of nitrogen at 37°C. The residue was dissolved in 200 µl acetonitrile by sonication for 3 min. An aliquot of 100 µl was subjected to

ion-exchange high-performance liquid chromatography (IE-HPLC) as described previously [27]. Chromatographic analyses were performed using an HPLC system consisting of a high-precision pump model 300 C (Gyncotek, Gemering, Germany), an FP-1520 fluorescence detector (Jasco, Hachioju City, Japan) and a Model 360 autosampler (Kontron, Basel, Switzerland). Samples were analyzed on a glass column (200×3 mm) packed with 5 µm Spherisorb Si material (Chrompack, Middelburg, The Netherlands). The mobile phase comprised a mixture of acetonitrile-10 mM tri-sodium citrate buffer (adjusted to pH 3.0 with hydrochloric acid) (87/13, v/v) and contained 5 mM tetrabutyammoniumbromide. The column was eluted at ambient temperature at a flow rate of 0.4 ml/min. The excitation monochromator was set at 270 nm and emission was monitored using a 360 nm long-pass filter; bandwidth was 40 nm. Integration was done using the Winner on Windows (WOW) software (Spectra Physics, San Jose, CA, USA). The ratio of peak areas of VRL and internal standard was used for quantitative calculations.

Samples containing 100 µg/ml VRL in plasma were prepared by diluting the VRL stock solution (10 mg/ml) in blank human plasma. This stock was used to prepare three calibration standards of 1, 10 and 100 ng/ml VRL in plasma, that were stored at −20°C. A calibration curve was constructed in plasma (final concentrations 1, 2, 5, 10, 20, 50, 100 ng/ml) in duplicate with each series just prior to analysis. The calibration curves were calculated by weighted least squares linear regression analysis. Quality control (QC) samples were analyzed together with the samples and the calibration curve. A different stock of 100 µg/ml was used to prepare the QCs. Three QCs containing 1, 10 and 100 ng/ml VRL were prepared in plasma and were stored at −20°C. Prior to analysis they were thawed and analyzed in duplicate. The lower limit of quantitation (LOQ) of the assay was 1 ng/ml using 500 µl of sample. However, where less than 500 µl of sample was available, the LOQ increased proportionally.

Pharmacokinetic calculations

Pharmacokinetic parameters were calculated using the software program MW\Pharm (MediWare, Groningen, The Netherlands [21]. Intravenous curves were fitted using a 3-compartment open model. The AUC after oral administration was calculated by non-compartmental methods using the linear trapezoidal rule without extrapolation to infinity. Statistical analyses were done using SPSS (version 11.5, SPSS, Chicago, IL, USA). The calculation of the oral bioavailability (F) and of the differences in F between subgroups was calculated by 3-way analyses of variance after logarithmic transformation of the AUC values.

Results

In this study we have used a cannulation method for the collection of serial blood samples, thus allowing the construction of a full plasma concentration–time curve from one mouse. This setup reduces the number of mice required for pharmacokinetic studies and provides more information about the intra and inter-individual variation. Although, the assay is suited for the quantification of the metabolite desacetylVRL, the plasma concentrations were generally near or below the LOQ because of the limitations of the sample size.

Following an i.v. bolus injection, the plasma concentration–time curve decline smoothly in accordance to a 3-compartment open model (Figs. 1 and 2). It appears that the absence of P-gp has little effect on the

Table 1 Pharmacokinetic parameters of VRL after i.v. and oral administration (values given as mean ± SE)

Genotype	Wild-type			Mdr1a/1b (−/−)	
Route	i.v				
Co-administration	None	RTV		None	
n	9	6		7	
AUC (mean ± SE) µg h L^{-1}	3613 ± 111	4717 ± 307	$p = 0.012$	3568 ± 231	NS
T1/2 (α) h^{-1}	0.190 ± 0.032	0.194 ± 0.013	NS	0.331 ± 0.065	NS
T1/2 (β) h^{-1}	2.271 ± 0.045	1.728 ± 0.096	NS	2.310 ± 0.496	NS
T1/2 (γ) h^{-1}	11.13 ± 0.54	8.85 ± 0.39	$p = 0.009$	12.08 ± 0.35	NS
Clearance L h^{-1} kg^{-1}	2.78 ± 0.08	2.16 ± 0.14	$p = 0.001$	2.87 ± 0.18	NS
Volume of distribution L kg^{-1}	44.7 ± 2.3	28.0 ± 3.1	$p = 0.001$	50.6 ± 4.6	NS
Route	oral				
Co-administration	None	RTV		None	
n	8	6		6	
AUC (mean ± SE) 0–8 h µg h L^{-1}	589 ± 46	1080 ± 109	$p = 0.005$	627 ± 37	NS
F %	16.0 ± 1.4	22.5 ± 2.3	$p = 0.016$	17.9 ± 1.7	NS
Fecal excretion % of the dose	33.9 ± 3.7	33.9 ± 2.1	NS	5.7 ± 0.6	$p < 0.001$

VRL was given at a dose of 10 mg/kg as single drug to wild-type and mdr1a/1b(−/−) mice, whereas, wild-type mice also received VRL in combination with 5 mg/kg of oral RTV. Differences (*p*-values) are relative to wild-type mice receiving VRL as single agent

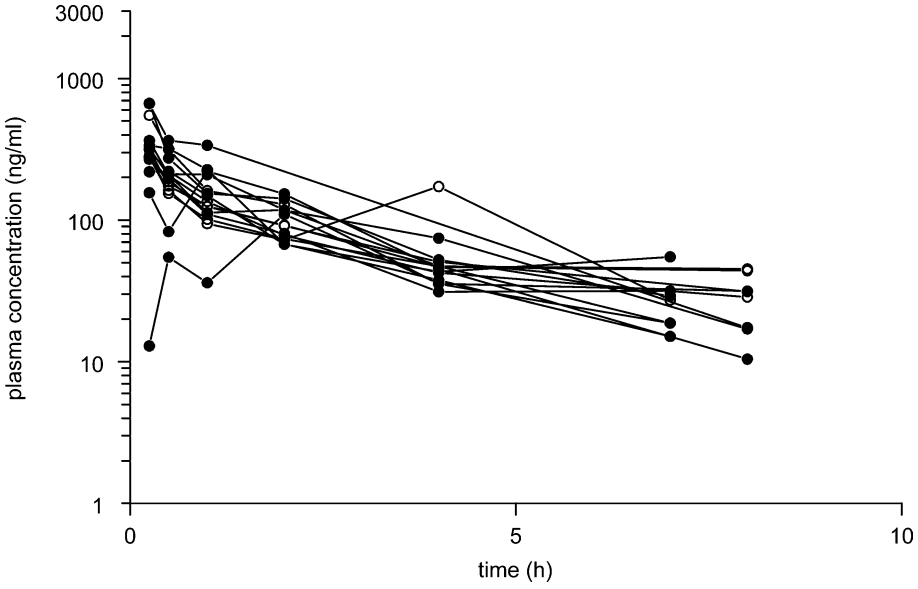

Fig. 1 Plasma concentration–time curves of intravenous VRL 10 mg/kg in wild-type (*closed symbols*) and mdr1a/1b (−/−) mice (*open symbols*)

plasma pharmacokinetics of VRL (Fig. 1). Although the plasma curves does reveal a significantly higher plasma concentration in mdr1a1b(−/−) mice at 48 h post drug administration, the apparent reduction in the elimination was too small to result in a significant change in the AUC or the terminal half-life (Table 1). Compared to the smooth curves after i.v. administration the plasma concentration–time curves after oral administration show greater fluctuations (Figs. 3 and 4). We, therefore, calculated the AUC after oral administration by non-compartmental methods. In most cases, the maximum plasma concentration (C_{max}) was reached within 15 min, indicating that a substantial fraction of the dose rapidly reaches the small intestines from where it also rapidly enters the systemic circulation. The AUC after oral administration and the oral bioavailability was also not significantly higher in mdr1a1b(−/−) mice.

The co-administration of RTV did change the plasma pharmacokinetics of VRL. The plasma concentration of VRL was higher when RTV was given, especially during the first 24 h period (Fig. 2). Overall, the plasma AUC was about 30% higher when RTV was co-administered. The distribution volume was reduced and the elimination half-life (T1/2(γ)) was significantly shorter. The AUC after oral administration was increased by about 1.8-fold when RTV was added to the regimen resulting in a significant enhancement of the oral bioavailability to 22.5 ± 2.3% ($p = 0.016$).

To determine the fraction of the dose that was absorbed from the intestines after oral administration, we have performed a fecal excretion study in wild-type and mdr1a1b(−/−) mice after oral dosing of VRL either as single drug or in combination with RTV. In wild-type mice the fecal recovery was only about 34% (Table 1), suggesting that even in the presence of P-gp at least 66%

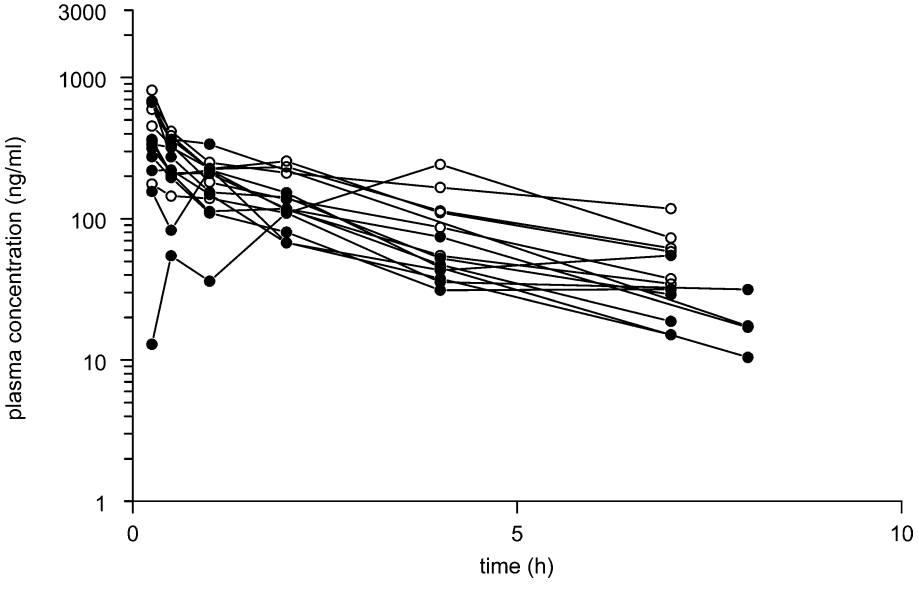

Fig. 2 Plasma concentration–time curves of intravenous VRL 10 mg/kg in wild-type mice with (*open symbols*) or without (*closed symbols*) RTV 5 mg/kg prior to VRL administration

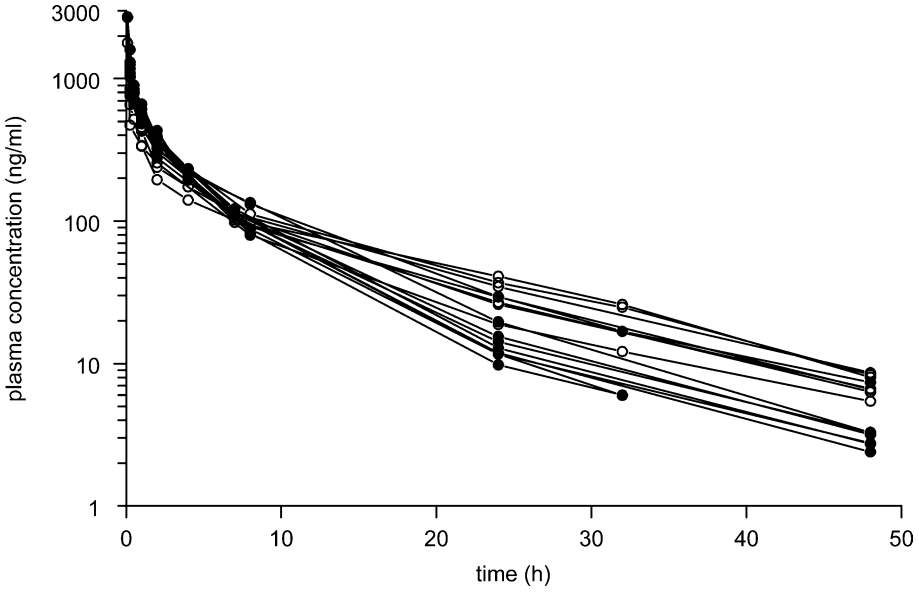

Fig. 3 Plasma concentration–time curves of oral VRL 10 mg/kg in wild-type (*closed symbols*) and mdr1a/1b (−/−) mice (*open symbols*)

of the dose was absorbed in the intestines. In mdr1a1b(−/−) mice the fecal recovery was reduced to about 6% of the dose. Although, this does indicate a role for P-gp in the intestinal uptake of VRL it implies only a rather modest enhancement of the intestinal uptake. The addition of RTV to the regimen had no effect on the fecal recovery.

Discussion

This study shows that the role of P-gp in the disposition of VRL in mice is very limited. The fecal recovery of oral VRL in wild-type mice is already low, indicating that intestinal P-gp is not acting as a very efficient barrier against the uptake of this drug. Despite the high-intestinal uptake, the oral bioavailability of VRL is low, due to substantial first-pass metabolism. The co-administration of a potent inhibitor of cyp3a mediated metabolism increased the oral bioavailability significantly, however, as the increase in the AUC of VRL after oral administration was still modest when given with RTV, cyp3a appears to be only a minor component in this first-pass metabolic breakdown.

This study was designed to improve our understanding of the role of P-gp and metabolism in the oral bioavailability of VRL. Although mice and humans are different, the pharmacokinetic handling of VRL appears to share many similarities between the two species. The oral bioavailability of VRL in patients receiving liquid-filled capsules was 27± 12% of the dose and the absorption was rapid with a mean time of maximum

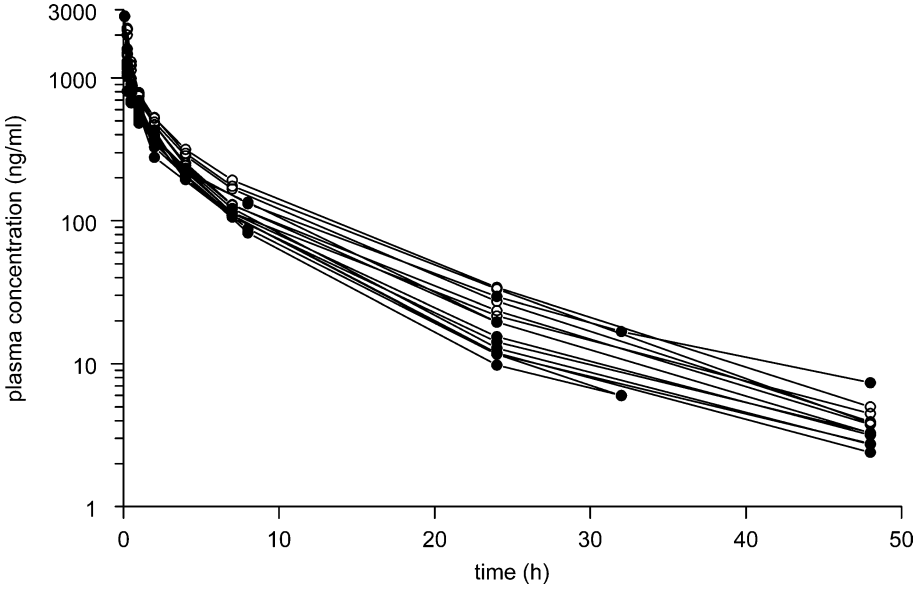

Fig. 4 Plasma concentration–time curves of oral VRL 10 mg/kg with RTV (*open symbols*) and without RTV (*closed symbols*)

plasma concentration (T_{max}) of 0.91 ± 0.22 h [22]. In our study, the C_{max} was also reached rapidly and the oral bioavailability of VRL in wild-type mice was 16 ± 1.4% of the dose. Although this is lower than in humans, it must be noted that due to the fluctuations in the oral curves, it was not possible to model these curves or to extrapolate the AUCs to infinity. As this will result in an underestimation of the oral bioavailability, the actual oral bioavailability in mice may be more close to bioavailability in humans. Since all groups of mice in this study were treated similar, the use of AUC values without extrapolation to infinity for calculation of the oral bioavailability does not preclude the comparison of the subgroups made within this analysis.

Unfortunately, there are no comprehensive clinical pharmacokinetic studies published on the excretion of unchanged VRL and on metabolism in humans, but just a few scattered reports. Interestingly though, Jehl et al [13] reported that the urinary excretion of unchanged VRL was 10.9 ± 0.7% of the dose, which is very similar to our previous results in mice [28]. Metabolism will most likely be an important elimination route in both species, although the extent by which each of the possible metabolic routes will be utilized may differ as the formation of deacetylVRL appears to a more important route in mice than in humans [13, 28].

The results of this study are to some extent comparable to our previous results with docetaxel in mice [3], where we demonstrated that the intestinal uptake of docetaxel in wild-type mice was also high, despite the fact that this compound is a good substrate of P-gp. These results with VRL and docetaxel thus confirm that P-gp is not in all cases an important component of the defense against the uptake of P-gp substrate drugs from the gut [7, 17] and are in marked contrast to the results with a closely related drug as paclitaxel [24]. The substantial uptake of VRL (or docetaxel) from the gut might be explained by the fact that a bulk amount of drug reaches into the gut and saturates the capacity of P-gp to handle this. However, it is not immediately obvious why paclitaxel would then behave so differently. In part, this difference may be caused by the rate and extent of metabolism of these compounds. Similar as with docetaxel the systemic clearance of VRL is not altered when P-gp is absent, suggesting that the metabolic degradation is far more important for the elimination of this drug than the excretion of unmetabolized drug. This is supported by previous biliary excretion experiments using mice with cannulated bile ducts. In case of docetaxel [3] and of the vinca alkaloid vinblastine [25] the amount of unchanged drug was minor compared to the amount of biliary excreted metabolites, whereas the fraction of unchanged paclitaxel versus the total amount of biliary excreted drug products was in the order of 30 to 40% [24]. Thus, whereas it is possible that also in case of paclitaxel a substantial fraction of an orally administered dose is initially absorbed from the gut, hepatobiliary excretion immediately returns a substantial fraction of unchanged drug back into the gut lumen. Then, of course, this fraction will be available for a second (and third...) round of absorption, but at that time the intra-gut luminal concentration of drug will be lower and thus less likely cause saturation of intestinal P-gp.

Another difference between VRL and docetaxel on the one hand and paclitaxel at the other appears to be the rate of uptake from the gut. Whereas, the maximum C_{max} with the two former drugs is reached within 15 min, it takes about an hour to reach the C_{max} in case of paclitaxel. This result might suggest that VRL and docetaxel more readily cross the intestinal barrier and thus also saturate intestinal P-gp more easily. However, the difference in C_{max} may also be due to the rate of metabolic degradation. Extensive entero-hepatic circulation may cause a delay the time that the maximum plasma level is reached as unchanged drug excreted in the bile will mix in the gut with drug released from the stomach. However, if all drug that is being absorbed is already metabolized during first-pass, this will not occur.

Previous in vitro results with human liver microsomes showed that CYP3A4 is mainly responsible for the metabolism of VRL [14]. Consequently, inhibition of CYP3A4 by a potent inhibitor as RTV might help to increase the oral bioavailability substantially as exemplified by the combination of RTV and saquinavir or other protease inhibitors [26]. Although, there was a significant enhancement of the oral bioavailability the result was rather modest, certainly when compared to the results with docetaxel in mice [3]. In this latter study a RTV dose of 12.5 mg/kg was used, whereas, we have reduced the RTV dose to 5 mg/kg in order to avoid the complications of delayed stomach emptying that may occur at the higher dose level. Although this dose reduction may have an impact on the magnitude of the interaction with VRL, we would still have expected a more substantial interaction albeit less than docetaxel, as RTV is among the most potent CYP3A4 inhibitors. This rather modest result may be related to a lower affinity of VRL for the murine Cyp3a than for the human CYP3A4 enzyme but it may also be that CYP3A4 may be less important for VRL elimination in vivo than anticipated as was already suggested previously [16].

The co-administration of RTV resulted in a reduction in the distribution volume, which may suggest that RTV reduces the tissue binding relative to plasma protein binding. It may, however, also be an artifact due to the fact that the distribution volume is reversely proportional to the elimination rate constant. Remarkably, the T1/2(γ), which is predominantly determined by the VRL plasma concentrations at time 24 h and later, was shorter with concomitant RTV instead of longer, as we would have expected with an inhibitor of metabolism. Although speculative, it is conceivable that RTV induces the expression of metabolic enzymes that are responsible for a more rapid elimination of VRL after 24 h. Consequently, this shorter T1/2(γ) results in an apparent reduction of the distribution volume.

In conclusion, we have established that P-gp has limited impact on the clearance or the oral bioavailability of VRL. Metabolic degradation appears to be the most important route of elimination, although the contribution of Cyp3a mediated first pass metabolism is limited. Our results suggest that the oral bioavailability in patients may also not benefit from the co-administration of a potent P-gp inhibitor. Further studies are needed to identify the route(s) of metabolic breakdown.

Acknowledgements We are grateful for the biotechnical support by Mr Ton Schrauwers and we are also indebted to Dr G. Hart for advice on statistical computations.

References

1. Adams DJ, Knick VC (1995) P-glycoprotein mediated resistance to 5′-nor-anhydro-vinblastine (Navelbine). Invest New Drugs 13:13–21
2. Bardelmeijer HA, Buckle T, Ouwehand M, Beijnen JH, Schellens JH, van Tellingen O (2003) Cannulation of the jugular vein in mice: a method for serial withdrawal of blood samples. Lab Anim 37:181–187
3. Bardelmeijer HA, Ouwehand M, Buckle T, Huisman MT, Schellens JH, Beijnen JH, van Tellingen O (2002) Low systemic exposure of oral docetaxel in mice resulting from extensive first-pass metabolism is boosted by ritonavir. Cancer Res 62:6158–6164
4. Bardelmeijer HA, van Tellingen O, Schellens JH, Beijnen JH (2000) The oral route for the administration of cytotoxic drugs: strategies to increase the efficiency and consistency of drug delivery. Invest New Drugs 18:231–241
5. Benet LZ, Izumi T, Zhang Y, Silverman JA, Wacher VJ (1999) Intestinal MDR transport proteins and P-450 enzymes as barriers to oral drug delivery. J Controlled Release 62:25–31
6. Binet S, Chaineau E, Fellous A, Lataste H, Krikorian A, Couzinier JP, Meininger V (1990) Immunofluorescence study of the action of navelbine, vincristine and vinblastine on mitotic and axonal microtubules. Int J Cancer 46:262–266
7. Chiou WL, Chung SM, Wu TC, Ma C (2001) A comprehensive account on the role of efflux transporters in the gastrointestinal absorption of 13 commonly used substrate drugs in humans. Int J Clin Pharmacol Ther 39:93–101
8. Depierre A, Freyer G, Jassem J, Orfeuvre H, Ramlau R, Lemarie E, Koralewski P, Mauriac L, Breton JL, Delozier T, Trillet-Lenoir V (2001) Oral vinorelbine: feasibility and safety profile. Ann Oncol 12:1677–1681
9. Domenech GH, Vogel CL (2001) A review of vinorelbine in the treatment of breast cancer. Clin Breast Cancer 2:113–128
10. Edelman MJ (2001) New directions in the treatment of non-small cell lung cancer: an overview. Oncologist 6:1–3
11. Freyer G, Delozier T, Lichinister M, Gedouin D, Bougnoux P, His P, Imadalou K, Trillet-Lenoir V (2003) Phase II study of oral vinorelbine in first-line advanced breast cancer chemotherapy. J Clin Oncol 21:35–40
12. Fromm MF (2003) Importance of P-glycoprotein for drug disposition in humans. Eur J Clin Invest 33 Suppl 2:6–9
13. Jehl F, Quoix E, Leveque D, Pauli G, Breillout F, Krikorian A, Monteil H (1991) Pharmacokinetic and preliminary metabolic fate of navelbine in humans as determined by high performance liquid chromatography. Cancer Res 51:2073–2076
14. Kajita J, Kuwabara T, Kobayashi H, Kobayashi S (2000) CYP3A4 is mainly responsibile for the metabolism of a new vinca alkaloid, vinorelbine, in human liver microsomes. Drug Metab Dispos 28:1121–1127
15. Leveque D, Jehl F (1996) Clinical pharmacokinetics of vinorelbine. Clin Pharmacokinet 31:184–197
16. Leveque D, Wisniewski S, Renault C, Peter JD, Le Corre P, Monteil H, Jehl F (2003) The effect of rifampin on the pharmacokinetics of vinorelbine in the micropig. Anticancer Res 23:2741–2744
17. Lin JH, Yamazaki M (2003) Clinical relevance of P-glycoprotein in drug therapy. Drug Metab Rev 35:417–454
18. Ling V (1997) Multidrug resistance: molecular mechanisms and clinical relevance. Cancer Chemother Pharmacol 40:S3–S8
19. Myer MS, Joone G, Chasen MR, van Rensburg CE (1999) The chemosensitizing potential of GF120918 is independent of the magnitude of P-glycoprotein-mediated resistance to conventional chemotherapeutic agents in a small cell lung cancer line. Oncol Rep 6:217–218
20. Ngan VK, Bellman K, Hill BT, Wilson L, Jordan MA (2001) Mechanism of mitotic block and inhibition of cell proliferation by the semisynthetic Vinca alkaloids vinorelbine and its newer derivative vinflunine. Mol Pharmacol 60:225–232
21. Proost JH, Meijer DK (1992) MW/Pharm, an integrated software package for drug dosage regimen calculation and therapeutic drug monitoring. Comput Biol Med 22:155–163
22. Rowinsky EK, Noe DA, Trump DL, Winer EP, Lucas VS, Wargin WA, Hohneker JA, Lubejko B, Sartorius SE, Ettinger DS, . (1994) Pharmacokinetic, bioavailability, and feasibility study of oral vinorelbine in patients with solid tumors. J Clin Oncol 12:1754–1763
23. Shepard RL, Cao J, Starling JJ, Dantzig AH (2003) Modulation of P-glycoprotein but not MRP1- or BCRP-mediated drug resistance by LY335979. Int J Cancer 103:121–125
24. Sparreboom A, van Asperen J, Mayer U, Schinkel AH, Smit JW, Meijer DKF, Borst P, Nooijen WJ, Beijnen JH, van Tellingen O (1997) Limited oral bioavailability and active epithelial excretion of paclitaxel (Taxol) caused by P-glycoprotein in the intestine. Proc Natl Acad Sci USA 94:2031–2035
25. van Asperen J, van Tellingen O, Beijnen JH (2000) The role of mdr1a P-glycoprotein in the biliary and intestinal secretion of doxorubicin and vinblastine in mice. Drug Metab Dispos 28:264–267
26. van Heeswijk RP, Veldkamp A, Mulder JW, Meenhorst PL, Lange JM, Beijnen JH, Hoetelmans RM (2001) Combination of protease inhibitors for the treatment of HIV-1-infected patients: a review of pharmacokinetics and clinical experience. Antivir Ther 6:201–229
27. van Tellingen O, Kuijpers A, Beijnen JH, Baselier MR, Burghouts JT, Nooyen WJ (1992) Bio-analysis of vinorelbine by high-performance liquid chromatography with fluorescence detection. J Chromatogr 573:328–332
28. van Tellingen O, Kuijpers AV, Beijnen JH, Nooijen WJ, Bult A (1993) Plasma pharmacokinetics, tissue disposition, excretion and metabolism of vinorelbine in mice as determined by high performance liquid chromatography. Invest New Drugs 11:141–150