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Short Review

Isolated Limb Perfusion with Chemotherapeutic Agents for Melanoma: A Reevaluation of Drug Dosimetry

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THE COMPARTMENTAL drug delivery system of isolated limb perfusion (ILP) was first used clinically for melanoma of the extremity by Creech *et al.* [1] over 30 years ago. Since then, ILP with chemotherapeutic agents, particularly melphalan, has become routine practice in many cancer centers [2]. The general consensus is that ILP combined with regional hyperthermia is the treatment of choice for patients having regional cutaneous metastases arising from melanoma of the extremities. However, the role of ILP in the curative treatment of localized (M.D. Anderson Stage I) melanoma of the extremity remains a controversial issue [3, 4]. Currently, the World Health Organization (WHO) and the European Organization for Research and Treatment of Cancer (EORTC) are conducting prospective randomized studies to evaluate the therapeutic efficacy of prophylactic ILP for extremity melanoma of greater than 1.5 mm thickness. These studies may allow the role of ILP in cancer chemotherapy to be better defined.

Although the potential therapeutic advantage of ILP may be significant, very little effort has been made to obtain relevant pharmacologic data to rationally optimize treatment schedules for improved antitumor responses. In this regard, it is essential to consider a fundamental pharmacological principle, which stipulates that the antitumor or toxic effects of drugs will be related to the concentration of cytotoxic agents at the cellular, subcellular, or macromolecular target sites. Since this concentration is generally difficult to determine in clinical pharmacology, biological responses have instead been equated, with excellent correlations, to plasma peak drug concentrations or area under the drug concentration × time curve (AUC). Thus, any schema of dosing must be designed to reproducibly achieve similar drug levels in the perfusate from patient to patient to permit maximal antitumor efficacy with minimal side-effects. Dosimetry in ILP, however, has also been an area of controversy, primarily because the methods for estimation of doses appear to have been derived empirically rather than being guided by pharmacological principles or data. In this report, we review previous methods of dosing in ILP, and offer a rationale for an alternative dosing schema designed to minimize variability in drug levels between patients and achieve desired optimal drug concentrations in future studies.

PRESENT METHODS FOR DOSE ESTIMATIONS IN ILP

Currently, two methods are available for estimating doses of melphalan to be used in ILP for the treatment of melanoma. In the first method, which is the most widely used, the dose is based on the total body weight of the individual [5]. The second method bases the dose on volume of extremity tissue to be perfused, and was introduced by Wieberdink *et al.* [6] to take into account the gross difference in volumes between the upper and lower limbs. Both methods, however, are far from ideal as judged by the generally large variation in the concentration of drug achieved in the perfusate in the limited

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Table 1. Peak melphalan concentrations in perfusate and systemic circulation during isolated limb perfusion

Patient No.	Artery perfused	Peak concentrations (μg/ml)	
		Perfusate	Systemic
1	Axillary	115.0	0.09
2	Popliteal	40.0	0.20
3	Popliteal	31.1	0.10
4	Femoral	6.3	0.30
5	Femoral	26.4	0.20
6	Femoral	22.8	0.50

Adapted from Minor *et al.* [7].

pharmacokinetic studies that have been performed.

Minor *et al.* [7] conducted pharmacokinetic studies of melphalan in six patients undergoing ILP with combined regional hyperthermia. The patients were administered doses of 0.75 mg/kg for axillary or popliteal perfusions, and 1.2 mg/kg for femoral perfusions. The peak concentrations of melphalan in the perfusate and in the systemic circulation from that study are summarized in Table 1, and indicate the 18-fold range (6.3–115.0 μg/ml) in peak perfusate levels. Although the time of peak perfusate concentration was not given, it is logical to assume that this would occur within a few minutes of drug addition to the perfusate. Thus, the wide variation in peak levels is probably not a result of differences between patients in the extent of leakage of drug into the systemic circulation, as has been implied by Minor *et al.* [7].

Briele *et al.* [8] also based their dose on body weight, and administered melphalan at 1.0–1.5 mg/kg (60–80 mg total dose for the upper extremity and 80–100 mg total dose for the lower limb). These authors found only minor differences between upper and lower limb perfusions in melphalan peak concentrations (59.5 vs. 56.0 μg/ml), which is in sharp contrast to the highly variable data of Minor *et al.* [7].

In contrast to the pharmacokinetic studies of Minor *et al.* [7] and Briele *et al.* [8], that of Benckhuijsen *et al.* [9] used a dose of melphalan based on regional limb volume to be perfused. Melphalan was standardized at a dose level of 10 mg/l of limb volume irrespective of whether the upper or lower limb was being perfused. Their data (see Table 2, last column) indicate almost a six-fold spread in peak levels of melphalan. Although the authors did not elaborate, there was a good correlation between the total dose given (hence limb volumes) and the corresponding peak concentrations that were achieved (see Fig. 1). The authors, however, did demonstrate an important correlation ($r = 0.83$) for melphalan between total dose and AUC. Since AUC is a measure of drug exposure of tissues, this implies that limbs with lower volumes may be under-dosed while limbs with higher volumes may experience toxicity at the standard dose of 10 mg/l. This implication is strengthened by toxicity data reported by Wieberdink *et al.* [6] and van Os *et al.* [10]. Using a dose of 9–11 mg/l, Wieberdink *et al.* have reported incidences of more severe toxic reactions with iliac compared to axillary perfusions. Similarly, of the 16 patients who had an iliac perfusion at 10 mg melphalan per liter in the van Os *et al.* study, 14 showed a grade II reaction, whereas two patients developed a more serious grade III reaction. In comparison, of the five patients with axillary perfusions, two experienced a grade I reaction, while the remaining three patients showed a grade II reaction. Although the number of axillary perfusions was small, van Os *et al.* [10] still concluded that the dose of 10 mg/l is low for axillary perfusions.

PROPOSED ALTERNATIVE METHOD FOR DOSE ESTIMATION IN ILP

The limitations of using body weight or limb volume as a basis for dose estimation in ILP have prompted us to seek an alternative procedure which would obviate large variations in peak concentrations and AUC of drug in the perfusate. For this purpose, it is essential to consider the volume of the perfusate, which indeed has received little or no attention, judging by its noticeable absence in the majority of previous reports on the technique. In ILP, the perfusate comprises the externally added fluid to the system and the blood in the patient's limb. Immediately after drug addition, the predicted concentration in the perfusate can be obtained from the following relationship:

$$\text{perfusate concentration } (\mu g/ml) = \frac{\text{mg drug added to perfusate}}{\text{volume of perfusate (ml)}} \times 1000.$$

Since the amount of drug used and the volume of externally added perfusion fluid are known, only

Table 2. Comparison of peak melphalan concentrations derived mathematically or experimentally

Level of isolation	Limb volume (l)	Dose range (mg)	Calculated limb blood volume (ml)	Externally added fluid volume in perfusate (ml)	Total perfusate volume (ml)	Theoretical concentration at T_0 (μg/ml)	Actual concentration at T_0 (μg/ml)
Brachial (2)	2.1–2.15	21–21.5	87–88	800	887–888	23.6–24.2	24 ± 1
Axillary (3)	3.0–3.2	30–32	104–107	800	904–907	33.2–35.3	37 ± 2
Femoral/ popliteal (4)	5.0–7.5	50–75	134–164	800	934–964	53.5–77.8	56 ± 8
Iliac: <12 l (11)	8.25–11.8	82.5–118	172–206	800	972–1006	84.8–117.3	93 ± 6
Iliac: >12 l (4)	13.75–16.0	137.5–160	222–240	800	1022–1040	134.5–153.8	137±9

Adapted from Benckhuijsen *et al.* [9]. The externally added fluid volume in the perfusate in the actual study ranged from 750 to 850 ml. Lack of further details regarding this volume has necessitated the use of 800 ml as an average for this exercise. Limb blood volume was calculated as $60 \times \sqrt{\text{limb volume (l)}}$. The theoretical concentration of melphalan was obtained by dividing the dose (mg) by the total perfusate volume, and is presented as a range to correlate with the dose administered as reported by the authors. Values for actual concentration at T_0 (zero time) are presented as means ± S.E.M. Numbers in parentheses indicate number of observations.

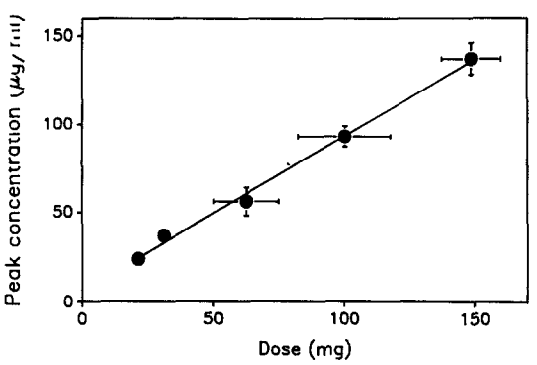

Fig. 1. Correlation between dose and peak perfusate concentration of melphalan. The dose is shown as a range, while the concentration is presented as a mean ± S.E. The line is the computer generated least-squares linear regression fit, described by the model y = mx + c: *slope* (m) = 0.87 ± 0.03 (*mean ± S.E.*), y *intercept* (c) = 6.0 ± 2.9, *correlation coefficient* (r) = 0.998. *Drawn using data of Benckhuijsen* et al. [9].

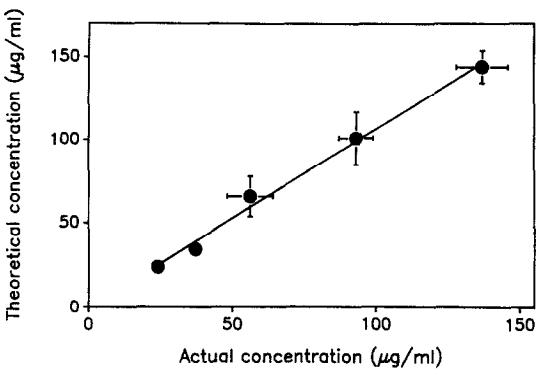

Fig. 2. Correlation between actual and theoretical concentration of melphalan in perfusate. The theoretical concentration is shown as a range, while the actual concentration is presented as a mean ± S.E. The line is a computer generated least-squares linear regression fit, described by y = mx + c: *slope* (m) = 1.08 ± 0.05 (*mean ± S.E.*), y *intercept* (c) = −0.86 ± 4.29, *correlation coefficient* (r) = 0.996. *Actual concentration data from Benckhuijsen* et al. [9]. *The raw information provided in this reported study were used to determine theoretical concentrations, as described in the text and in the legend to Table* 2.

the volume of blood in the limb is needed to use the above formula for calculation of drug concentration. Benckhuijsen *et al.* [9] have derived a relationship which is applicable to such estimation of limb blood volume:

$$\text{blood volume in limb (ml)} = 60 \times \sqrt{\text{limb volume (l)}}.$$

We have applied the above two formulae to the data of Benckhuisen *et al.* [9] for retrospective validation, the results of which are presented in Table 2 and Fig. 2. The data indicate a very good correlation between predicted melphalan concentrations and those that were actually found during perfusions in patients. As noted in Table 2, the contribution of blood volume of the limb to total perfusate volume varies from about 10% for a 2-l limb to about 22% for a 15-l limb. Thus, a large change in limb volume results in a relatively small change in total perfusate volume. This accounts for the wide variations in reported peak perfusate concentrations in ILP when doses are based on limb volume [9]. Similar logic would apply in explaining the variation in peak levels when doses are based on body weight. Once again, differences in the

volume of perfusate will be small between patients of varying weights, assuming that the volume of the externally added perfusion fluid is constant and comparatively larger than the blood volume of the limb. A main contribution to these gross variations in peak concentrations probably arises from differences in body weights. Thus, peak concentrations of drug will be lower for a smaller patient and higher for a larger patient. If, on the other hand, patients of similar sizes are used, then variations in peak drug concentrations will be small. This may provide one explanation for the small variation in perfusate drug levels in ILP of lower or upper extremities reported by Briele *et al.* [8]. Another reason for the small variation in this reported study is related to the slightly higher dose used for lower extremities (80–100 mg) compared to upper extremities (60–80 mg); the larger dose may have offset the assumed slightly greater total perfusate volume for the lower limb to affect similar perfusate concentrations in both upper and lower limb perfusions.

The validation of the above formula in accurately predicting peak drug concentrations enables us to use the following relationship to calculate the total dose of melphalan for any given patient:

$$\text{dose (mg)} = \text{desired peak level } (\mu\text{g/ml}) \times \frac{\text{volume of perfusate (ml)}}{1000}$$

(where perfusate volume = volume of externally added fluid + volume of blood in limb). Theoretically, if the volume of the externally added priming fluid is sufficiently large (>5 l) in comparison to the limb blood volume (<0.25 l), then the above formula can be simplified as follows for either upper or lower limb perfusions:

$$\text{dose (mg)} = \text{desired peak level } (\mu\text{g/ml}) \times \frac{\text{volume of external fluid (ml)}}{1000}.$$

In practice, however, the volume of the priming fluid is less than 1.5 l so that the blood volume of limbs generally cannot be ignored.

USE OF DESIRED PEAK DRUG CONCENTRATIONS IN DOSE CALCULATIONS FOR ILP

The demonstrated close correlations between dose and peak perfusate concentrations, between dose and AUC, and, by inference, between AUC and peak perfusate levels for melphalan validate the use of peak concentrations as a measure of tissue exposure to the drug. Although the application of the above relationship for dose determination would overcome variability in peak drug levels, it is important to initially establish what is the desired optimal (or maximally tolerated) peak concentration for a bolus dose and split doses of the drug, and if this concentration will change between patients of differing skin complexion and body habitus, and between perfusions of lower and upper limbs. In this latter regard, lower doses have commonly been employed for upper extremities [2] suggesting a greater sensitivity of this limb to melphalan. Similarly, a light-skinned person or a person predisposed to peripheral vascular disease (e.g. diabetes, atherosclerosis) appears to be less tolerant to the drug [11, 12], and a lower desired peak concentration may be appropriate in these individuals. It is equally important to note from the formula established for dose estimation that perfusate volume will directly affect peak drug levels that will be achieved. In only a few reported studies with melphalan, however, is the volume of perfusate quoted, and in these cases the volume of the priming fluid has varied from 750 to 1050 ml [7–9, 13], which may contribute substantially to the outcome of therapy. The composition of the perfusate used may also affect the relationship between peak perfusate concentration and AUC of the drug by altering the pharmacokinetics of the cytotoxic agent [8]. This in turn may alter responses of the normal tissue and tumor to the drug.

In conclusion, a method is presented for estimation of total dose of melphalan for use in ILP which will produce uniform targeted peak drug concentrations. The dose is calculated independent of body weight but requires assessment of limb volumes to be perfused for estimation of regional blood volume. The method is likely to be applicable to other drugs used in perfusion systems.

REFERENCES

1. Creech O Jr, Krementz ET, Ryan RF *et al.* Chemotherapy of cancer: regional perfusion utilizing an extracorporeal circuit. *Ann Surg* 1958, **148**, 616–632.
2. Krementz ET. Regional perfusion: current sophistication, what next? *Cancer* 1986, **57**, 416–432.
3. Ghussen F, Kruger I, Groth W, Stutzer H. The role of regional hyperthermic cytostatic perfusion in the treatment of extremity melanoma. *Cancer* 1988, **61**, 654–659.
4. Franklin HR, Koops HS, Oldhoff J *et al.* To perfuse, or not to perfuse? A retrospective comparative study to evaluate the effect of adjuvant isolated regional perfusion in patients with stage I extremity melanoma with a thickness of 1.5 mm or greater. *J Clin Oncol* 1988, **6**, 701–708.

5. Krementz ET, Ryan RF. Chemotherapy of melanoma of the extremities by perfusion: fourteen years of clinical experience. *Ann Surg* 1972, **175**, 900–917.
6. Wieberdink J, Benckhuijsen C, Braat RP, Van Slooten EA, Olthuis GAA. Dosimetry in isolation perfusion of the limbs by assessment of perfused tissue volume and grading of toxic tissue reactions. *Eur J Cancer Clin Oncol* 1984, **18**, 905–910.
7. Minor DR, Allen RE, Alberts D *et al.* A clinical and pharmacokinetic study of isolated limb perfusion with heat and melphalan for melanoma. *Cancer* 1985, **55**, 2638–2644.
8. Briele HA, Djuric M, Jung DT *et al.* Pharmacokinetics of melphalan in clinical isolation limb perfusion of the extremities. *Cancer Res 1985*, **45**, 1885–1889.
9. Benckhuijsen C, Varossieau FJ, Hart AAM, Wieberdink J, Noordhoek J. Pharmacokinetics of melphalan in isolated perfusion of the limbs. *J Pharmacol Exp Ther* 1986, **237**, 583–588.
10. van Os J, Koops HS, Oldhoff J. Dosimetry of cytostatics in hyperthermic regional isolated perfusion. *Cancer* 1985, **55**, 698–701.
11. McBride CM, Clark RL. Experience with l-phenylalanine mustard dihydrochloride in isolation–perfusion of extremities for malignant melanoma. *Cancer* 1971, **28**, 1293–1296.
12. Sugarbaker EV, McBride CM. Survival and regional disease control after isolation–perfusion for invasive stage I melanoma of the extremities. *Cancer* 1976, **37**, 188–198.
13. Ghussen F, Nagel K, Groth W, Muller JM, Stutzer H. A prospective randomized study of regional extremity perfusion in patients with malignant melanoma. *Ann Surg* 1984, **200**, 764–768.