4

Isolated Limb Perfusion in the Treatment of Advanced Soft-tissue Sarcomas

Joseph M. Klauser, Dina Lev-Chelouche, Isaac Meller, Moshe Inbar and Mordechai Gutman

BACKGROUND

In 1958, Creech and Krementz[1] introduced a novel method of drug delivery for patients with advanced cancer and named it isolated limb perfusion (ILP).[1] The idea was to apply the newly invented technique of cardiopulmonary bypass to regional chemotherapy. ILP entailed exposing the major blood vessels to an extremity, isolating it temporarily and perfusing the extremity via a heart–lung machine with very high doses of chemotherapeutic drugs (Figure 4.1). The authors believed it would be possible to obtain high tissue concentrations of the drug with minimal systemic exposure and hence few complications. Following the observation that heat has its own antineoplastic properties, Stehlin[2] in 1969 modified the technique to include hyperthermia.

The response rates observed with ILP in patients with metastatic melanoma confined to the limb were higher than those associated with any other known modality. This, combined with the fact that some 25% of complete responders have a 10-year disease-free interval, promoted the use of ILP. Since then, ILP has been widely recognized as a standard treatment strategy for advanced melanoma of the extremities.

Despite its effectiveness, ILP did not become widely used, and several major cancer centers did not include it in their therapeutic arsenal. There are several reasons for this. ILP is a multidisciplinary procedure. It is surgically demanding and rather long (3–4 h). It necessitates a heart–lung machine and related technician and equipment, and isotopic monitoring. Unlike the reasonable ease with which new operative techniques are implemented through the acquisition of knowledge and skills, ILP requires a thorough knowledge of vascular surgery and a large degree of coordination and dependence on other disciplines outside the realm of general surgery. In addition, the available literature on ILP showed great variability in surgical technique, drugs administered, degree of hyperthermia, indications, and response evaluation. This, coupled with the retrospective nature of most reported patient series, made it difficult to reach valid conclusions as to when and how to use ILP.

The situation has gradually changed, mainly because of two advances. First, the standardization of ILP and the conduct of multicenter studies based on principles of modern surgical oncology and consistent with the standards of the National Cancer Institute made it possible to evaluate outcomes in a uniform fashion, and hence to better define the indications for ILP. The second advance was the addition of tumor necrosis factor (TNF), an exciting new cytokine, to the treatment protocols.[3] TNF is a new drug and its potentially serious side-effects necessitated modifications to the ILP technique. Therefore, all centers began to routinely monitor ILP patients for isotope leakage and better standardization was achieved.

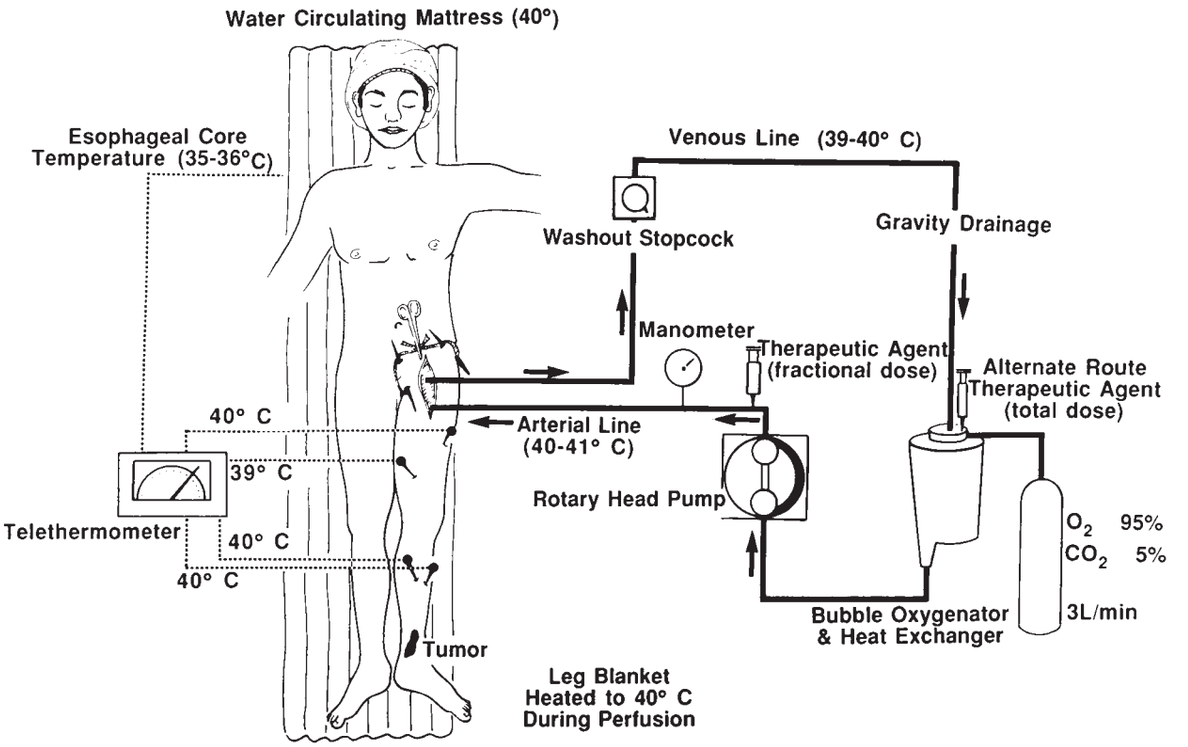


Figure 4.1 Schematic presentation of isolated limb perfusion.

TECHNICAL CONSIDERATIONS OF ISOLATED LIMB PERFUSION

Preparation

Candidates for ILP must be carefully evaluated. A metastatic work-up to rule out evidence of disease outside the extremity is indicated. The tumor to be perfused should be confined to the limb. It should not extend to the groin or axillary areas because these areas, as well as the proximal thigh, buttocks, and upper posterior arm, are relatively poorly perfused, even with proximal femur cannulation.

Ulcerated or infected tumors shuld be recognized as a septic risk. The existing bacterial colonization may turn into overt sepsis or abscess formation following the induction of rapid necrosis with TNF or chemotherapy. Cultures and preoperative antibiotics are routinely required.

Patient evaluation and preparation are similar to those preceding any major surgical procedure. Specific attention must be given to the peripheral vascular system. Evidence of peripheral vascular disease may require further evaluation by angiography. Patients with severe arteriosclerotic disease, especially those with nonpalpable distal pulses, are usually not suitable candidates for ILP.

Evaluation of the venous system for deep thrombosis (DVT) by ultrasound Doppler is important, particularly in patients who have undergone prior surgery or ILP to the affected limb. Since the adequacy of perfusion depends on the patency of the venous system, patients with DVT are poor candidates for ILP. Finally, the neurological status of the limb should be carefully recorded, particularly in patients with prior surgery, radiotherapy, or tumors adjacent to the nerves. Limb volume should be measured or calculated, because the drug dosage used during ILP is based on the volume of the extremity.[4] Limb volume can be measured using the water displacement method, in which the extremity is immersed in a calibrated cylinder filled with water. Alternatively, it can be calculated by measuring limb length and circumference at multiple points and incorporating them into a mathematical formula used for calculating the volume of a cylinder.

The patient should be fully informed of the procedure. Such explanations should emphasize short-term and particularly long-term complications, that may be associated with the procedure.

Table 4.1 Site distribution of 228 limb perfusions*

Perfusion site	*Melanoma*	*Sarcoma*
Lower limb	89	88
Iliac	61	47
Femoral	18	22
Popliteal	10	19
Upper limb	33	18
Subclavian	24	11
Brachial	9	7

*Author series 1990–98.

Selection of the Procedure

ILP can be performed through various sites (Table 4.1). For the lower limb it is done via the external iliac, common femoral or popliteal vessels; for the upper limb it is performed via the brachial, axillary or subclavian vessels. The level of perfusion should be based on the nature and extent of the tumor, as well as technical considerations such as prior surgery or radiotherapy.

Because the majority of ILP candidates are patients with melanoma of the lower limb, the most common route of perfusion is the external iliac vessels. For patients with soft-tissue sarcomas (STS), more distal sites for cannulation may be chosen that are proximal to the tumor mass. Perfusions via the popliteal or brachial vessels are considered simpler, despite the smaller diameter of the vessels, because these vessels are usually accessible with less dissection and accommodate the use of a tourniquet applied proximally on the thigh or arm to ascertain complete isolation and avoidance of systemic leakage.

Surgery

ILP entails major surgery and is performed under endotracheal general anesthesia. Given the need for systemic heparinization, epidural and regional anesthesia are not recommended.

In the operating room the entire extremity is cleaned. Four thermistor probes are inserted; two into the subcutaneous tissue and two into the muscles, in the distal and proximal parts of the extremity, respectively, to measure limb temperature during the procedure. A heating blanket is wrapped around the limb and sterile draping is applied on top of it. It is important that the limb can be manipulated and positioned during the procedure to permit the application and wrapping of the Esmark band on its root. For distal (popliteal/brachial) cannulations, a pneumatic tourniquet is applied proximal to the operative site (Figures 4.2A, 4.2B).

Lower Limb Perfusion

Iliac perfusion

An oblique incision is made in the iliac region. The fascia and muscles are sectioned and the retroperitoneum is entered. The peritoneum is retracted medially. After exposing the external iliac vessels from their origin to the inguinal ligament, the vessels are dissected circumferentially and all side branches, including collaterals situated behind the inguinal ligament (i.e. the epigastric, obturators, deep internal and external circumflex vessles), are ligated and sectioned. All branches, especially from the posterior aspect of the external iliac vein, should be ligated. These deep collaterals are not affected by external Esmark banding, and their control is crucial for minimizing leakage during perfusion. Ligation of the internal iliac vein is optional (Figure 4.2C).

Axilla

The vessels are dissected circumferentially, and all collaterals are ligated and divided. The artery and vein are then clamped proximally and cannulated (8–14F). The tips of the cannulae are directed towards the proximal portion of the arm. An Esmark tourniquet is wrapped tightly around the root of the shoulder and anchored with Steinmann pins inserted into the skin below and lateral to the breast. Because the shoulder is smaller than the root of the lower limb, isolation and control can be achieved more easily in the arm than in the leg.

Brachial perfusion

This is performed through a longitudinal incision in the medial aspect of the arm. Smaller cannulae adjusted to the vessel size are chosen. Minimal dissection and collateral ligation are required since complete isolation is easily achieved with the aid of a small pneumatic (blood pressure) tourniquet applied proximally and inflated to 300 mmHg.

Extracorporeal Circulation

The extracorporeal system consists of a roller-pump similar to that used for cardiopulmonary bypass surgery (Figure 4.2D). A single-head pump is sufficient. The pediatric surgery disposable pack, with its smaller-sized oxygenator, venous reservoir and tubing, is most suitable. A heat exchanger is required to warm the perfusate to 42°C. Priming is with 700–1000 ml of balanced electrolyte solution, one unit of packed RBCs and 1500 U heparin. Dextran (Haemacel®) can be used

to replace blood. Mild hyperthermia (39–40°C) is used; true hyperthermia (41–42°C) is rarely used because it produces severe limb toxicity.

Limb temperature is increased by heating the perfusate to 42°C and by applying the heated blanket and draping it around the limb. The temperature probes enable close monitoring.

The first stage of ILP is devoted to heating the perfusate and assessing leakage. Significant time may be required to warm skin temperature to 39°C. Leakage from the limb is dependent on the perfusion flow rate.[5] Experience with high perfusion flow rates (700–1300 ml/min and 300–600 ml/min) for the lower and upper limbs, respectively, resulted in relatively high systemic leakage rates (12.5 ± 2.9%) and consequently severe systemic toxicity. Decreasing the perfusion flow rate to 400–500 ml/min and 150–300 ml/min for the lower and upper limbs respectively, reduced leakage and systemic toxic effects. Increased pressure may contribute to leakage by opening up vessels and forcing blood through small collaterals in subcutaneous tissues, muscles, and vessels along the periosteum between the limb and major vasculature. This is likely with high flow rates that may induce venous pressure in the perfused limb that exceed that of the systemic pressure (Figure 4.2E).

Leakage Monitoring and Adjustment

The patient is continuously monitored to ensure that the perfusate does not leak into the circulatory system. Such monitoring was not routinely done prior to the TNF era, and surgeons relied simply on stable reservoir volume and their own personal experience to estimate leakage. With TNF, leakage monitoring became mandatory. Protocols using microlabeled albumin or technetium-labeled RBC are used for monitoring leakage during ILP.[6,7] After establishing perfusion with a stable flow and venous reservoir volume, the isotope is injected into the perfusate. A gamma camera is positioned over the precordial area of the head. Any increase in counts over the background signifies a systemic leak. A leak of less than 1% can be detected, allowing rapid adjustments to limit side-effects. Another indication of leakage is alterations in the calibrated venous reservoir volume, which drains the

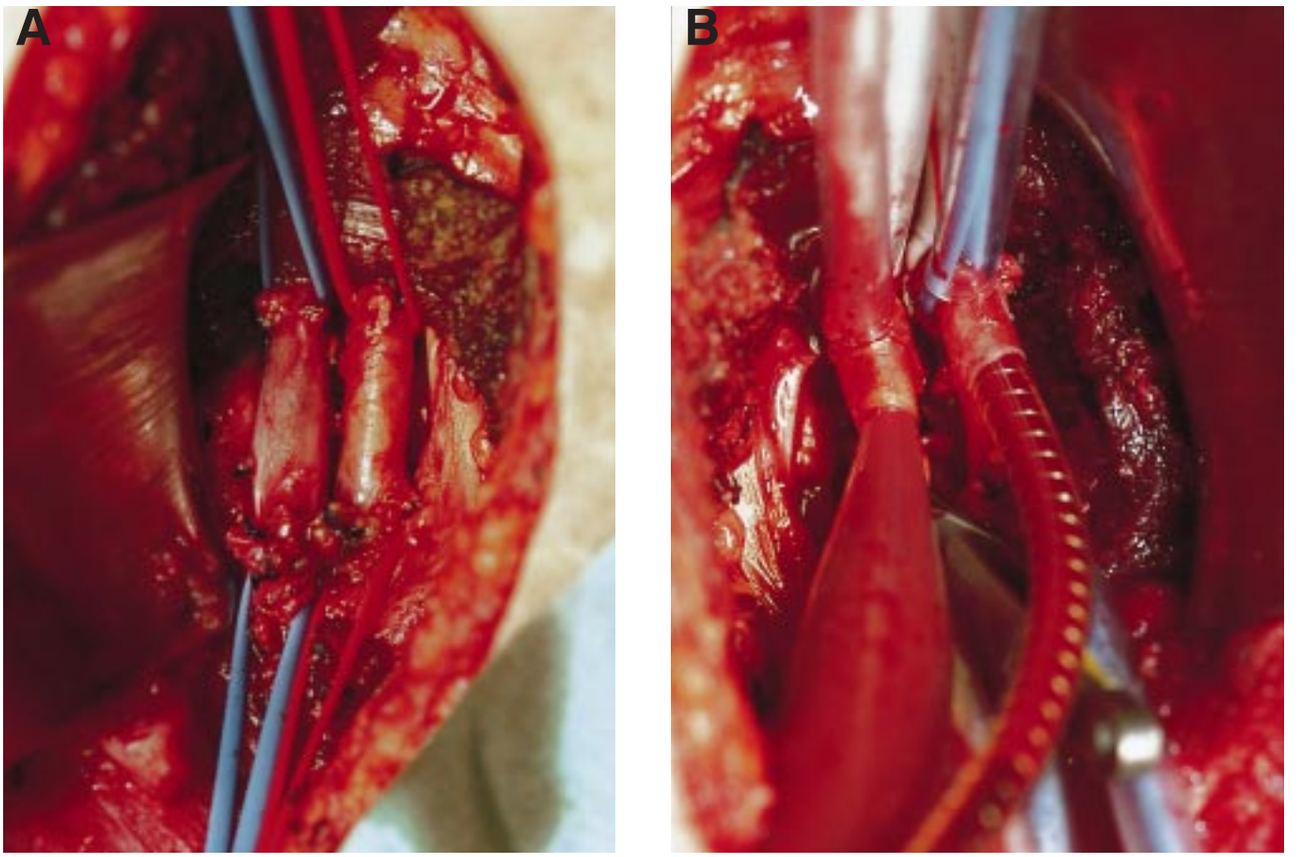
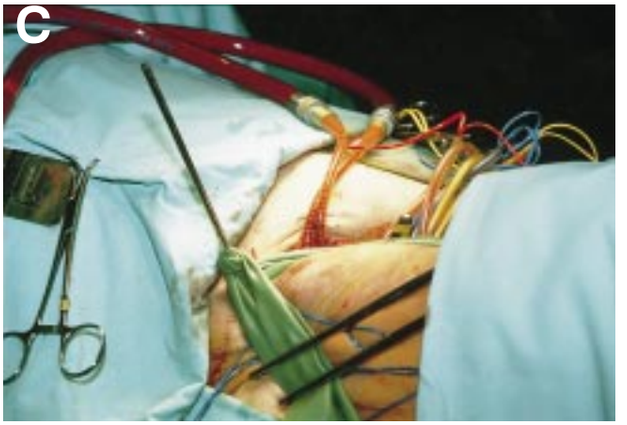
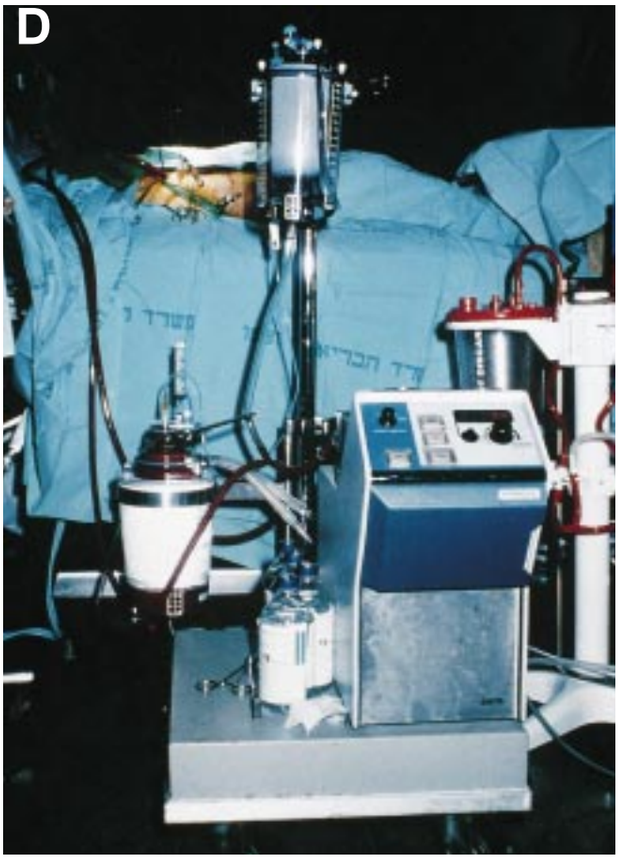


Figure 4.2A,B *(see above and following page)*





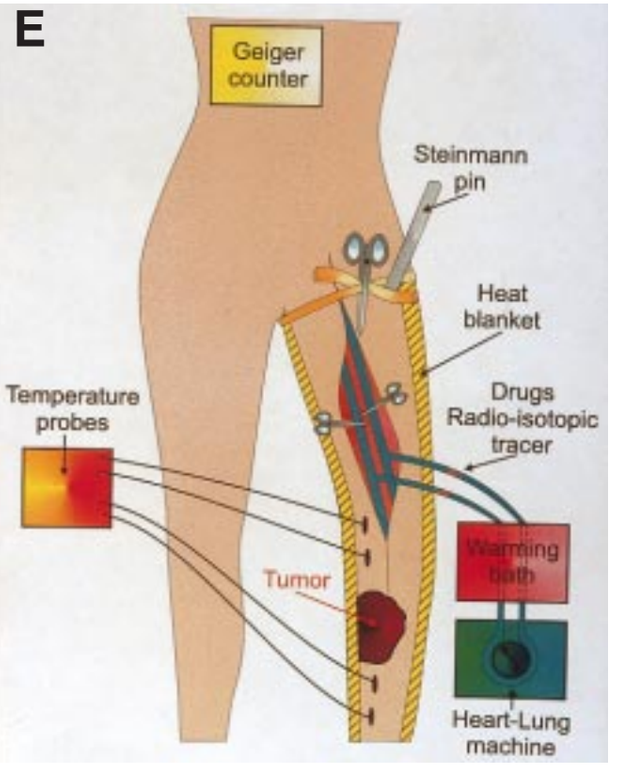


Figure 4.2 Isolated limb perfusion (ILP); stages of the procedure: (**A**) Entire limb is scrubbed, tissue thermistors are placed; (**B**) limb is wrapped with heating mattress; (**C**) wide exposure of the artery and vein, ligation of all the collaterals; (**D**) cannulation and proximal occlusion of blood vessels; (**E**) in iliac perfusions, a Steinman pin is inserted into the iliac bone to anchor the Esmark band.

venous effluent by gravity. A decrease in reservoir volume indicates a leak from the perfused limb into the systemic circulation, whereas an increase in venous reservoir volume signifies a leak from the systemic circulation into the limb.

With the information provided from monitoring, and the volume of the venous reservoir, it is possible to manipulate the perfusion rate and minimize leakage.

Rapid shifts in reservoir volume indicate a missed collateral vessel. Rapid leakage also occurs when the cannula or its side holes are placed above the level of the tourniquet. Under such circumstances the pump should be turned off and the operative field re-explored in order to allow identification and ligation of a missed collateral, repositioning of the cannulae, and readjustment of the tourniquet. Less dramatic leakage can be manipulated by reapplication of the tourniquet or adjustment of the circuit flow pressure.

Leakage from the systemic circulation to the limb is less frequent and can usually be dealt with by increasing flow rate or pressure. Systemic leaks of less than 2% can be achieved in almost 95% of patients.[5,7,8]

This improvement in isolation techniques has made ILP a safe procedure. There is no longer a need for sophisticated, invasive monitoring or intensive care. Patients are routinely transferred to the surgical ward 2–3 h following the procedure.

Termination of ILP

After the drug administration and perfusion treatment periods (e.g. 30 min for TNF alone followed by 90 min of melphalan), the circuit is interrupted and the perfusate washed from the limb with 2 L of saline and 1 L of dextran polymer or blood. The pump is then turned off, the tourniquet cuff is deflated, and the cannulae are removed. The vein is repaired and thereafter the arteriotomy is sutured, re-establishing blood flow to the limb.

DRUGS

Melphalan

This alkylating agent, a phenylalanine mustard, was originally selected for melanoma perfusion because phenylalanine is an essential precursor in melanin synthesis and is taken up preferentially by melanocytes.

Melphalan, which is an ideal drug for ILP, possesses a short half-life, low endothelial toxicity, limited cell cycle specificity, and a relatively linear dose–response relationship for cytotoxicity. The optimal dose of melphalan is 10 mg/L limb volume for the lower extremity and 13 mg/L limb volume for upper extremity perfusion. At such dosages, perfusate concentrations of melphalan are 50–100-fold higher than systemic levels, which remain less than 1 μg/ml.[9]

Given its high response rate (60–80%) with a complete response of 30–55% in melanoma patients undergoing ILP[10–12] and the chemoresistant nature of melanoma, it is not surprising that melphalan is the drug of choice for this procedure.

Other Cytotoxic Drugs

Other chemotherapeutic agents, including cisplatinum, actinomycin-D, and DTIC, have been tested in conjunction with ILP in both human and animal studies, but none has demonstrated better results than with melphalan.[2] Cisplatinum appears to be suitable for ILP, because its concentration in tumor tissue is selectively increased in the presence of mild hyperthermia. The primary concern is its considerable regional toxicity, especially neurotoxicity. Doxorubicin was also investigated in the ILP setting, mainly for nonresectable STS, but was found ineffective both alone and in combination with melphalan. Regional toxicity was unacceptable, and amputation rates were as high as 40%.[13]

Tumor Necrosis Factor

Tumor necrosis factor-α (TNF-α) was discovered in 1975 and made available for clinical use in 1985.[14] TNF-α can produce very fast and effective tumor necrosis, as has been demonstrated in a variety of tumor-bearing mice and cultured cancer cells. Attempts to duplicate the effect in humans, however, have failed. Only anecdotal cases of partial response were described in more than 800 cancer patients treated systemically with rTNF-α. This failure was attributed to the inability to administer sufficient doses of TNF because of life-threatening side-effects.[3,8] Based on data from murine tumor models, the rTNF-α dose required to achieve the antitumor effect is 10–20 times higher than the maximal tolerated systemic dose (MTD) in humans, which is approximately 200 mg/m^2. Administration of higher doses produces the hemodynamic conditions typically associated with septic shock (e.g., tachycardia, hypotension, decreased vascular resistance, increased cardiac index), as well as coagulopathy, thrombocytopenia, and metabolic impairment such as increased bilirubin and liver enzymes and decreased cholesterol levels.[15] The end result is multiorgan failure. By using rTNF-α in the ILP setting it is possible to overcome barriers raised by the systemic toxicity, and doses of 3–4 mg can be administered.[16]

The most pronounced effect of TNF *in vivo* is seen in the tumor's vasculature.[8] TNF suppresses specific adhesion molecules such as integrin-αVβ3 on the surface of endothelial cells within the tumor and on membranal receptors on macrophages and leukocytes.[17,18] Antagonists of αVβ3 interfere with adhesion-dependent signals, causing apoptosis of angiogenic endothelial cells. This TNF-induced endothelial damage is exclusive to tumor vasculature; normal vasculature is spared, as has been demonstrated in angiograms performed in sarcoma patients before and after ILP–TNF.

The administration of TNF alone, as demonstrated in mouse tumor models, human tumor xenografts in nude mice, and a pilot study in six patients, has only a transient antitumor effect. There is regrowth of the tumor after its necrosis.[19] To achieve a high and prolonged response the addition of a cytotoxic drug is mandatory.

Hyperthermia

The fact that heat is effective against cancer and tumor growth has been known since the middle of the nineteenth century. A clinical trial demonstrated a good tumor response to isolation perfusion with a heated perfusate without cytotoxic drugs.[2] Based on this work, heat was added to the melphalan perfusion system with the general belief that hyperthermic melphalan perfusion is superior to normothermic ILP. Hyperthermia has been the subject of a great deal of research that has led to several hypotheses concerning its role in tumor necrosis. Trus hyperthermia (> 41°C) decreases the

tissue pH and aerobic glycolysis. It induces cycling of tumor cells and changes the proportion of cells in the sensitive S and M phases. Cell membranes become more sensitive to active agents and the tumor cells, rendering them more vulnerable to cytotoxic drugs. There is an apparent decrease in DNA repair, possibly due to the development of oxygen-free radicals that cause breaks in the DNA strand.

Hyperthermia appears to enhance the antitumoral effect of both TNF and melphalan.[20] However, true hyperthermia cannot be used in ILP because of the risk of severe regional toxicity.[21] Mild hyperthermic conditions, commonly used, are still considered synergistic to melphalan and TNF and do not increase the regional toxicity.[22]

Table 4.2 Regional toxicity – Weiberdink classification

Grade I	No reaction
Grade II	Slight erythema/edema
Grade III	Considerable erythema/edema with some blistering
Grade IV	Extensive epidermolysis and/or obvious damage to the deep tissues, causing functional disturbances; threatening or established compartmental syndrome
Grade V	Reaction which may necessitate amputation

COMPLICATIONS OF ILP

Regional Toxicity

The Wieberdink grading system (Table 4.2) is routinely used to evaluate the regional toxic effect of ILP. Mild edema, erythema, discomfort, and a warm limb (grade I) commonly develop within 2–3 days following ILP. Moderate to severe toxicity (grade III–IV) occurs in 15–30% of patients. Patients may develop skin blisters, particularly on the palms of the hand and the soles of the feet. Full-thickness loss is rare. Pain and significant discomfort occur in 25–40% of patients and are probably related to muscle swelling, compartmental syndrome, and neurotoxicity. Limb-threatening complications with extensive tissue injury and severe edema occur in less than 10% of patients. Limb loss occurs only rarely (0.5–1.5%). Most of the complications following ILP resolve spontaneously within 2–3 weeks.[21]

Adding TNF to melphalan, in our experience and that of some others, does not appear to increase regional toxicity.[8,23] Other groups, however, have reported that the rapid onset, severity, and duration of limb toxicity are more prominent when TNF is added to melphalan. A phase I study revealed that a 6 mg dose of TNF combined with melphalan induced severe muscle and nerve toxicity but TNF alone elicited no regional toxicity.[16]

Nerve toxicity, manifested by shooting pain or paresthesias, occurs 2–3 weeks after ILP in 25–40% of patients. It usually resolves within a few months. Long-term neuropathy is rarer (1–4%).[21]

Vascular complications may also develop following ILP. Arterial complications are rare and unrelated to the drugs used in ILP. The incidence of thrombosis at the arteriotomy site is 2.5%.[21]

The incidence of DVT is significant (~10%) despite heparinization during ILP. The thrombogenic effect of the tumor, the cytotoxic drugs, and the surgical trauma, coupled with edema and increased compartmental pressure and decreased mobility, are contributing factors.

Another important category of complications is associated with the rapid necrosis induced by TNF. This is a real threat in patients with ulcerated tumors, in whom an existing bacterial colonization may turn into overt sepsis. Such was our experience with a 14-year-old boy whose ulcerated sarcoma underwent liquefaction necrosis, abscess formation, and uncontrolled sepsis, that necessitated urgent amputation. We also encountered staphylococcal sepsis originating in the infected necrotic tumor that ultimately proved fatal to a 78-year-old patient.

Systemic Side-effects

Most systemic side-effects are caused by leakage of drugs from the perfusate into the systemic circulation during ILP. Even with complete isolation and a thorough wash-out of the perfusate on completion of the ILP, the drug may remain in the tissue of the limb or its intravascular compartment and redistribute once normal systemic circulation is re-established. Systemic toxicity from melphalan perfusion is limited if systemic leakage is less than 10%.[24] Since melphalan declines from the perfusate 10 min after administration, its systemic toxicity is mainly related to early leakage. With systemic leakage of melphalan, patients typically experience some nausea and vomiting immediately following ILP. Ten to 14 days later, they develop mild, short-lived neutropenia.

The addition of TNF introduces the risk of immediate life-threatening complications. A stable and high TNF level in the perfusate and limb is present during the entire ILP; consequently, a small but continuous leak may divert a systemically significant dose of TNF. Furthermore, TNF induces the generation of secondary cytokines and mediators, which can elicit many side-

effects, some of which are similar to those observed following rTNF-α administration.[7]

The systemic side-effects of TNF may be divided into three categories: cardiovascular, metabolic, and hematologic.[5] The most immediate effect involves the cardiovascular system, and is manifest by tachycardia, hypotension, increased cardiac index, and a marked decrease in systemic vascular resistance (SVR). Unlike many types of shock, in which vasoconstriction dominates, septic shock associated with TNF leads to a pathognomonic vasodilation. There is good correlation (regression analysis = 0.8) between the severity of the decrease in SVR and the systemic levels of TNF.[5]

Also of importance is the cardiac effect of TNF. Despite the hyperdynamic state and increase in cardiac index associated with the drug, it produces an overall cardiodepressant effect that is manifest by a decrease in the left ventricular stroke work index. The metabolic effects of TNF mainly relate to helatic toxic effects and may include hyperbilirubinemia, increased levels of liver enzymes, and marked hypocholesterolemia. The hematologic systemic side-effects are manifested by leukopenia, thrombocytopenia, and coagulopathy.

With the standardization of the ILP technique and meticulous isolation, systemic leakage is minimized. This virtually eliminates side-effects and greatly simplifies the entire procedure. No special monitoring and intensive care are required, and the patient may be transferred to the surgical ward 2–3 h following the procedure.

ILP FOR EXTREMITY SOFT-TISSUE SARCOMA

As it became evident that amputation is not mandatory in patients with soft-tissue sarcoma, and that comparable survival rates can be achieved with adequate tumor resection,[25] limb preservation became a major goal. However, when the tumor is large, expanding into more than one compartment, or it is adjacent to or invading a major blood vessel or nerve, or when there is a multifocal appearance, amputation or mutilating surgery are still considered almost inevitable.

Several treatment modalities have been developed to facilitate limb-sparing in patients with advanced tumors. Neoadjuvant therapies, mainly preoperative radiation or a combined preoperative (intra-arterial and/or systemic) chemo- and radiotherapy)[26,27], have led to a significant reduction in amputation rates (which were 50% prior to 1977), although 8–15% of patients with extremity sarcoma still undergo amputation.[26]

Prior to the era of TNF-based perfusion, ILP was not a viable option for tumor reduction in STS due to poor response rates.[28] However, promising results following the introduction of high-dose TNF in ILP[3] led to a phase II multicenter study to determine the effect of ILP with TNF and melphalan in patients with nonresectable STS confined to the limb. Because resection is impossible, amputation is almost always necessary in this group.

ILP/TNF is also indicated as a palliative measure in selected patients who have a reasonable life expectancy in the presence of distant STS metastasis. In these cases, avoidance of amputation is the main goal.

The typical changes following ILP/TNF may be summarized as follows:

- In large, bulky tumors, marked softening can be noticed in the first days post-ILP, but this is not an accurate measurement for response because it may be secondary to edema following perfusion.
- The tumor mass may become smaller or completely disappear both clinically and on imaging studies. At times the mass reduction is limited, but in cases where the tumor is adherent to a major nerve (e.g. sciatic nerve) or blood vessel (e.g. popliteal vessels), even a small change in size (e.g. 2–3 cm), can make it possible to perform marginal resection and to preserve these structures.
- In ulcerated tumors that penetrate the skin, hemorrhagic necrosis can be seen even a few hours after the perfusion.

An accurate assesment of response in STS following ILP/TNF is difficult. The disappearance of the vascular bed, as demonstrated by postoperative angiography, is usually a good indicator of massive necrosis. Newer techniques, such as magnetic resonance spectroscopy and positron-emission tomography scanning,[29,30] can be useful in assessing the extent of necrosis vs. tumor viability and increase the accuracy of response assessment.

Histopathology is the most accurate method with which to assess tumor response. Typical histological changes 6–8 weeks after TNF perfusion include cystic hemorrhagic necrosis in the central area of the remaining tumor. Although spontaneous necrosis is encountered in STS, the magnitude and extent of necrosis following ILP/TNF are unique (Figure 4.8). If any viable tumor cells exist, they are usually observed in the periphery of these "cysts" but their malignant potential cannot be fully determined. Other histological changes are extensive interstitial and pericystic fibrosis, which can also be seen following neoadjuvant chemotherapy. No correlation has been found between tumor size, histological subtype, and pattern of response.

There is a notable discrepancy between the clinical and the pathological response assessments. A histological examination can upgrade the overall

response rate, since masses that remain unchanged or only partially regressed may be converted into a complete response (no viable tumor cells) following pathological examination. Since studies performed on sarcoma patients undergoing ILP with cytostatic drugs in the pre-TNF era were based only on clinical and radiological assessment, the reported response rates may have been underestimated relative to pathologically based TNF studies.

Table 4.3 ILP/TNF + mephalan in STS

Author	*Ref.*	*Year*	*Drug*	*No. of patients*	*% RR*	*% CR*
Vaglini	31	1994	TNF + melph	9	89	67
Gutman	23	1996	TNF + melph	35	94	37
Eggermont	8	1996	TNF/melph/IFN-γ	186	82	29

Result of ILP/TNF in STS

A review of worldwide experience with ILP using rTNF-α and melphalan ± IFN (Table 4.3) discloses an overall response rate of 82–100%, with a complete response of 29–67% and partial response of 22–56%. Eighteen percent or less of patients exhibited no response or a progression of disease. In 20% of patients with a partial response, a near-total response (> 95% necrosis) was found. Only the finding of several tumor cells precluded categorizing these patients as having a complete response.[8,16,23,31]

These results were obtained in a selected group of patients with very extensive disease. All were candidates for amputation. The average tumor size was 16 cm; most tumors were high-grade (85%), and 43% were recurrent.[8,23] There was also a relatively high rate of multifocal disease (23%). In this group of patients, who have a grave prognosis anyway, the value of limb preservation is even more enhanced.[32]

Limb salvage has been achieved in 85% of these patients. This high rate is the most valuable and proven benefit of ILP/TNF in advanced sarcoma patients. Complete response *per se* is not crucial; whether the tumor responds completely or partially is irrelevant, as long as it becomes amenable to resection without loss of limb function.

A tumor that has shrunk considerably may still be impossible to resect without endangering limb function. In such cases a second perfusion may be considered to achieve further tumor shrinkage. Our experience with nine PR patients with very large tumors who underwent two TNF perfusions scheduled 6–10 weeks apart, resulted in the conversion of six to complete response and two to further tumor shrinkage. Limb salvage was ultimately possible in eight of these patients.

The response to ILP/TNF is remarkable, but is it longstanding? Recurrent local disease in patients whose limbs are considered salvages after ILP/TNF ranges from 10 to 15% after 3–24 months (median follow-up 22 months).[8,23] This recurrence rate is relatively low, considering the large median tumor size, the percentage of recurrent sarcomas (40%), and the large percentage (25%) of multifocal STS in this group. Local recurrence rates could probably be further improved by the administration of postresection radiation therapy to a higher percentage of patients.

References

1. Creech O, Krementz ET, Ryan RF *et al.* Chemotherapy of cancer: regional perfusion utilizing an extracorporeal circuit. Ann Surg. 1958;148:616–31.
2. Stehlin JS. Hyperthermic perfusion with chemotherapy for cancers of the extremities. Surg Gynecol Obstet. 1969;129:305–8.
3. Lienard D, Ewaienko P, Delmotti JJ *et al.* High-dose recombinant tumor necrosis factor alpha in combination with interferon gamma and melphalan in isolation perfusion of the limbs for melanoma and sarcoma. J Clin Oncol. 1992;10:52–60.
4. Wieberdink J, Benckhuysen C, Braat RP *et al.* Dosimetry in isolation perfusion of the limbs by assessment of perfused tissue volume and grading of toxic tissue reactions. Eur Cancer Clin Oncol. 1982;18:905–10.
5. Sorkin P, Abu-Abid S, Lev D *et al.* Systemic leakage and side effects of tumor necrosis factor administered via isolated limb perfusion can be manipulated by flow rate adjustment. Arch Surg. 1995;130:1079–84.
6. Barker WC, Andrich MP, Alexander HR *et al.* Continuous intraoperative external monitoring of perfusate leak using I-131 human serum albumin during isolated perfusion of the liver and limbs. Eur J Nucl Med. 1995;22:1242–8.
7. Thom AK, Alexander HR, Andrich MP *et al.* Cytokine levels and systemic toxicity in patients undergoing isolated limb perfusion (ILP) with high-dose TNF, interferon-gamma and melphalan. J Clin Oncol. 1995;13:264–73.
8. Eggermont AMM, Schraffordt-Koops H, Klausner JM *et al.* Isolated limb perfusion with tumor necrosis factor and melphalan for limb salvage in 186 patients with locally advanced soft-tissue extremity sarcomas: the cumulative multicenter European experience. Ann Surg. 1996;224:756–65.

9. Briele HA, Dyuric M, Jung DT *et al.* Pharmacokinetics of melphalan in clinical isolation perfusion of the extremities. Cancer Res. 1985;45:1885–9.
10. Ghussen F, Kruger I, Groth W *et al.* The role of hyperthermic cytostatic perfusion in the treatment of extremity melanoma. Cancer. 1988;61:654–9.
11. Hafstrom L, Rudenstam CM, Blomquist E *et al.* Regional hyperthermic perfusion with melphalan after surgery for recurrent melanoma of the extremities. J Clin Oncol. 1991;9:2091–4.
12. Lienard D, Eggermont AM, Kroon BBR *et al.* Isolated limb perfusion in primary and recurrent melanoma: Indications and results. Semin Surg Oncol. 1998;14:202–9.
13. Klaase JM, Kroon BBR, Benckhuysen C *et al.* Results of regional isolation perfusion with cytostatics in patients with soft-tissue tumors of the extremities. Cancer. 1989;64: 616–21.
14. Aggarwal BB, Kohr WJ, Hass PE *et al.* Human tumor necrosis factor: production, purification and characterization. Biol Chem. 1985;260:2345–54.
15. Bevilcqua MP, Pober JS, Majeau GR *et al.* Recombinant tumor necrosis factor induces coagulant activity in cultured human vasculature endothelium; characterization and comparison with actions of interleukin-1. Proc Natl Acad Sci USA. 1986;83:4533–7.
16. Alexander HR, Fraker DL, Bartlett DL. Isolated limb perfusion for malignant melanoma. Semin Surg Oncol. 1996;12:416–26.
17. Cheresh DA. Death to a blood vessel, death to a tumor. Nature Med. 1998;4:395–8.
18. Ruegg C. Evidence for the involvement of endothelial cell integrin $\alpha/\beta 3$ in the disruption of the tumor vasculature induced by TNF and IFN-γ. Nature Med. 1998;4:408–14.
19. Regeness A, Muller M, Curschellas E *et al.* Antitumor effects of tumor necrosis factor in combination with chemotherapeutic agents. Int J Cancer. 1987;39:266–73.
20. Watanabe N, Niitso Y, Umeno N *et al.* Synergistic cytotoxic and antitumor effects of recombinant human tumor necrosis factor and hyperthermia. Cancer Res. 1988;48: 650–3.
21. Vrouenraets BC, Klaase JM, Kroon BB *et al.* Long-term morbidity after regional isolated perfusion with melphalan for melanoma of the limbs. The influence of acute regional toxic reactions. Arch Surg. 1995;130:43–7.
22. Klaase JM, Kroon BB, van Slooten GW *et al.* Relation between calculated melphalan peak concentrations and toxicity in regional isolated perfusion for melanoma. Reg Cancer Treat. 1992;4:309–12.
23. Gutman M, Inbar M, Shlush-Lev D *et al.* High dose tumor necrosis factor alpha and melphalan administered via isolated limb perfusion for advanced soft tissue sarcoma results in a >90% response rate and limb preservation. Cancer. 1997;79:1129–37.
24. Hoekstra HJ, Naujocks T, Schraffordt Koops H *et al.* Continuous leakage monitoring during hyperthermic isolated regional perfusion of the lower limb: techniques and results. Reg Cancer Treat. 1992;4:301–4.
25. Gaynor JJ, Tan CC, Casper ES *et al.* Refinement of clinicopathologic staging for localized soft tissue sarcoma of the extremity: a study of 423 adults. J Clin Oncol. 1992; 10:1317–27.
26. Lawrence W, Donegan WL, Natrajan N *et al.* Adult soft-tissue sarcomas; a pattern of care survey of the American College of Surgeons. Ann Surg. 1987;205:349–59.
27. Eilber FR, Eckhardt JF, Rosen G *et al.* Neoadjuvant chemotherapy and radiotherapy in the multidisciplinary management of soft-tissue sarcomas of the extremity. Surg Oncol Clin N Am. 1993;2:611–20.
28. Krementz ET, Carter RD, Sutherland CM *et al.* Chemotherapy of sarcomas of the limbs by regional perfusion. Ann Surg. 1977;185:555–64
29. Sijens PE, Eggermont AMM, Van Dijk P *et al.* 31P magnetic resonance spectroscopy as predictor for clinical response in human extremity sarcoma treated by single dose TNF-alpha and melphalan isolated limb perfusion. NMR Biomed. 1995;8:215–21.
30. Nieweg OF, Prulm J, Hoekstra HY *et al.* Positron emission tomography with fluorine-18-fluorodeoxyglucose for the evaluation of therapeutic isolated regional limb perfusion in a patient with soft-tissue sarcoma. J Nucl Med. 1994; 35:90–2.
31. Vaglini M, Belli F, Ammatuna M *et al.* Treatment of primary or relapsing limb cancer by isolation perfusion with high dose alpha-tumor necrosis factor gamma interferon and melphalan. Cancer. 1994;73:483–92.
32. Lev D, Abu-Abid S, Kollander Y *et al.* Multifocal soft tissue sarcoma limb salavage following isolated limb perfusion with tumor necrosis factor and melphalan. J Surg Oncol. 1999;70:185–98.