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Review

Intratumoural chemotherapy of lung cancer for diagnosis and treatment of draining lymph node metastasis

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Abstract

Objectives Reviewed here is the potential effectiveness of cytotoxic drugs delivered by intratumoural injection into endobronchial tumours through a bronchoscope for the treatment of non-small cell lung cancer and the diagnosis of occult or obvious cancer cell metastasis to mediastinal lymph nodes.

Key findings Intratumoural lymphatic treatment may be achieved by injection of cisplatin or other cytotoxic drugs into the malignant tissue located in the lumen of the airways or in the peribronchial structures using a needle catheter through a flexible bronchoscope. This procedure is termed endobronchial intratumoural chemotherapy and its use before systemic chemotherapy and/or radiotherapy or surgery may provide a prophylactic or therapeutic treatment for eradication of micrometastases or occult metastases that migrate to the regional lymph nodes draining the tumour area.

Conclusions To better elucidate the mode of action of direct injection of cytotoxic drugs into tumours, we review the physiology of lymphatic drainage and sentinel lymph node function. In this light, the potential efficacy of intratumoural chemotherapy for prophylaxis and locoregional therapy of cancer metastasis via the sentinel and regional lymph nodes is indicated. Randomized multicenter clinical studies are needed to evaluate this new and safe procedure designed to improve the condition of non-small cell lung cancer patients and prolong their survival.

Keywords endobronchial intratumoural chemotherapy; intratumoural cytotoxic drug injection; lung cancer; lymphatic drainage to lymph nodes; sentinel lymph node detection

Introduction

Non-small cell lung cancer (NSCLC) remains the leading cause of cancer-related mortality in both sexes worldwide.[1] At present, surgical resection offers the best chance for survival in NSCLC patients. Surgery may be curative for stage I and stage II disease. In certain conditions, patients with stage IIIA disease may also be candidates for surgical resection. However, in patients with stage IIIB disease, the tumours are usually considered unresectable unless they are downstaged by neoadjuvant radiochemotherapy or by one of the interventional endobronchial procedures. Patients with stage IV disease have distant metastases and are offered only non-surgical treatments, except for rare cases of resectable solitary metastasis in a patient who also has a resectable primary lesion.[1,2]

In general, only 25–30% of lung cancer patients are considered candidates at presentation for potentially curative resection.[1] This fact is due to early local metastatic lymph node dissemination of the tumour, which occurs during growth of most primary malignancies. Indeed, it has been stated that 20–25% of patients initially considered with clinical stage I disease are found during surgery to have mediastinal lymph node metastases.[3,4] As a general rule, if nodal involvement is recognized, the chances of long-term survival are less than 50%.[5,6] Therefore, in order to ameliorate this unfavourable outcome for patients who are eligible for surgical resection, some safety measures must be taken to manage mediastinal lymph node metastases. Procedures for dealing with lymph node metastases may be summarized as follows. (i) Classical systematic lymph node dissection during surgery. (ii) Dissection after sentinel lymph node detection and mapping. (iii) Pre- or post-surgical chemoradiotherapy. The presence of metastatic tumour cells mandates more extensive systemic therapy. The total nodal tumour burden (number of affected nodes and metastatic

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tumour volume) severely affects prognosis.[7,8] (iv) The novel use of endobronchial intratumoural chemotherapy (EITC).

Systematic lymph node dissection

It has been shown that surgical dissection of the regional lymphatics can somewhat improve postoperative survival. For that reason, surgeons try their best to achieve lymph node dissection that is as complete as possible during surgical resection for lung cancer. In spite of this, only 20–25% of lymph node metastasis may be removed by ordinary mediastinal lymph node dissection. If we consider that 20–25% of patients with clinical stage I disease have mediastinal lymph node metastases at the time of diagnosis,[3,4] with 20–25% amelioration by dissection, only 5% of patients with clinical stage I disease may actually benefit from this classical procedure.[9] New approaches are needed to improve current techniques for mediastinal lymph node dissection.

Preoperative detection and mapping of sentinel lymph nodes with labelling agents

New techniques developed in recent years have somewhat improved the detection and elimination of lymph nodes with metastasis. This detection and mapping of sentinel lymph node metastasis has employed radioactive labelling or tracer dyes preoperatively or perioperatively. By such detection, dissection of metastasis-free nodes and needless prolonged surgery has been avoided. Presently, mediastinal lymph node dissection with the help of sentinel lymph node mapping is a procedure that is used increasingly to secure more complete local control of NSLC and is accompanied by improvement in survival rates.[10–12] Unfortunately, in spite of this progress in lymph node dissection, the relapse rate after surgery, even in early stage lung cancer, due to occult regional lymph node metastases continues to remain rather high. Therefore, to improve the survival rate after surgery in early lung cancer, we need better regional lymph node mapping techniques and new therapeutic modalities for targeting occult metastases and micrometastases. There is a need for new therapeutic procedures that reduce the risk of local metastases to the regional lymph nodes.

Systemic intravenous chemotherapy and/or radiotherapy as neoadjuvant therapy before surgery in early lung cancer

As noted, the elimination of mediastinal lymph nodes with occult metastasis or micrometastasis during surgery is not always effective. For that reason, some investigators have favoured the use of systemic chemotherapy, preoperatively or postoperatively, even for stage I disease.[13] Studies showing significant improvement of survival with newer drugs such as cisplatin, paclitaxel or carboplatin in patients with stage IIIA or IIIB NSCLC[14–19] have prompted us to consider the role of neoadjuvant intravenous chemotherapy in conventionally resectable patients, that is those with stage I or II disease (T1, T2 or T3 + N0; T1, T2 or T3 + N1) and also in patients with limited stage IIIA disease and unexpected N2 disease with normal nodes at computed tomographic scan but with microscopic N2 disease at mediastinoscopy.

There are not many published studies of neoadjuvant chemotherapy versus surgery alone in patients with stage I and stage II disease. Some reports are negative[20–22] and some are positive.[23,24] A potential cause of confusion in all these studies is the inconsistent addition of preoperative and/or postoperative radiotherapy for some patients, usually in patients with residual disease remaining after resection. In any event, the response rates and prolonged survival achieved by systemic postoperative chemotherapy and radiotherapy, in addition to the treatment before surgery for patients with stage I and II disease, remains modest. Furthermore, any small gains are obtained at the cost of the significant toxicity of systemic chemotherapy. For that reason, preoperative chemoradiotherapy is not used routinely in stage I and II disease without obvious mediastinal lymph node involvement.[13]

We therefore propose that an additional benefit may be achieved by the novel preoperative therapeutic paradigm of direct preoperative intratumoural chemotherapy, to thereby treat occult metastases or micrometastases in early lung cancer. Since severe systemic toxicity limits the use of conventional chemotherapy, we suggest that local intratumoural delivery of cytotoxic drugs that produce no systemic toxicity may have a significant benefit for the eradication of occult metastases and micrometastases that migrate through the draining lymph nodes in stage I and II disease.

Endobrobronchial intratumoural chemotherapy

The results of clinical studies on EITC consisting of cytotoxic drug injections into and around endobronchial tumours through a flexible bronchoscope,[25–35] has led us to consider that application of preoperative EITC in early lung cancer should have the added potential benefit of reducing the risk of relapse by killing occult or micrometastatic cancer cells that migrate to the regional lymph nodes.

Lymphatic transport of intratumoural injected tracer compounds to the sentinel lymph nodes: translation of a diagnostic concept to a therapeutic concept

The characteristics of lymphatic drainage, as well as the incidence of lymph node metastasis have been examined extensively in recent years.[9] In light of these studies we may hypothesize that when an anticancer drug is injected into and around a tumour, the drug molecules are transported by lymphatic drainage to sentinel and regional lymph nodes and may thereby kill metastatic malignant cells without systemic drug toxicity. To support this hypothesis, in the following sections we review the findings of recent investigations on the physiology of lymphatic drainage to the sentinel lymph nodes (SLNs). These studies are persuasive in arguing for the benefits achieved by intratumoural delivery of cytotoxic drugs for eradication of occult regional lymph node metastases as well as the primary lesions in early lung cancer.

Physiology of lymphatic drainage

Recent biological and technological developments on lymphatic vascular biology have led to a better understanding of lymphatic spread of early lung cancer.[36–39] Clinical studies

on sentinel lymph node mapping in humans have shown that molecules of a tracer substance injected directly into the bronchial wall will be transported to the regional lymph nodes by lymphatic drainage within 20 to 60 min.[40] Similarly, molecules of cytotoxic drugs injected directly into the bronchial wall will be transported to the regional lymph nodes by lymphatic drainage in the same manner as the tracer compounds. Recent studies concerning lymphatic function and drainage to regional lymph nodes are considered in the following sections to help understand the potential prophylactic effect of intratumoural injection of cytotoxic drugs for the eradication of tumour cell metastasis to the regional lymph nodes.

Lymphatic system

Oxygen, nutrients and hormones are delivered to tissues by blood vessels and capillaries, which are involved in molecular transport processes with the surrounding tissues. Blood pressure causes plasma to leak continuously from the capillaries into fluid of the interstitial space. The fluid of the interstitial space drains continuously into lymphatic capillaries with low pressure. The lymph, a milky protein-rich fluid, is found in the lymphatic vasculature. The lymphatic vasculature acts as a network that drains the lymph from various body tissues and returns it to the circulating blood.

Lymphatic drainage

Lymphatic vessels drain fluid from the interstitial space of the body, returning to a central location. The lymphatic vasculature commences as highly permeable blind-ending sacs referred to as lymphatic capillaries. The lymphatic capillaries are relatively large thin-walled vessels composed of a single layer of endothelial cells. They are not sheathed by pericytes or smooth muscle cells and have little or no basement membrane. The lack of a continuous basement membrane surrounding the lymphatic capillaries facilitates entry of fluid from the interstitial space into these vessels. As a result, plasma components, macromolecules and cells, such as extravasated leukocytes, activated antigen-presenting cells, and particulate matter such as bacteria and malignant cells, enter these blind-ended lymphatic capillaries through loose valve-like openings in their walls (see Figure 1).[36–39]

Lymphatics are thin-walled, low-pressure vessels that collect fluid and cells from the interstitium and return it to the circulation via the thoracic duct. Tumour cells leave the primary tumour and spread directly into the surrounding tissue, from which they subsequently invade lymphatic vessels. Figure 1a shows a single tumour cell leaving the primary mass and entering the lymphatic system. Also, the molecules of the cytotoxic drug injected into the tumour pass into the interstitial fluid, which enters the lymphatics and subsequently is transported to lymph nodes.

Interstitial fluid, collected by the initial lymphatic capillary plexus, is transported by pre-collector lymphatic vessels. Lymph is then transported towards larger collecting lymphatic vessels. These have a smooth muscle cell layer, basement membrane and endoluminal valves, and are linked to the extracellular matrix by anchoring filaments. The latter are very thin (4–10 nm) fibrillin-containing filaments, which extend into the endothelial cell plasma membrane. Anchoring filaments prevent vessel collapse under conditions of high interstitial pressure. The contraction of smooth muscle cells, and surrounding skeletal muscles, as well as arterial pulsations, contribute to lymphatic flow, and endoluminal valves prevent backflow to maintain unidirectional lymph flow. The lymphatic vessels are designed for interstitial fluid volume regulation and absorption of dietary fat. In addition, the lymphatic system is important for immune function and for the metastatic spread of cancer.[36–39]

Lymph fluid is transported via afferent lymph ducts to a series of lymph nodes (an afferent lymphatic vessel is a vessel that enters a lymph gland or node). Afferent lymph ducts divide before passing beneath the capsule of the node into cortical sinuses of lymph nodes, then pass through a reticuloendothelial cell filter.[36] Lymph is filtered through this lymph node filter where foreign particles are taken up by antigen-presenting cells that initiate specific immune responses. Although lymph continues through the medullar sinus to the hilar region of the lymph node, tumour cells may become trapped and proliferate here or spread further to distal organs.[41] Lymph fluid then leaves the lymph nodes through efferent lymphatic vessels, carrying the lymph that has not been absorbed by the venous system in the gland. Such vessels form trunks that ultimately empty into the thoracic duct and right lymphatic duct, which in turn empty into the lymphaticovenous junctions in the jugular area. Although it is possible for diffusing intratumoural drugs to enter the blood circulation via the nodal blood supply and the vasa vasorum of the lymph ducts, this mechanism for drug transport to systemic circulation should be negligible.

Pathogenesis of lymphatic involvement in cancer

Tumour metastasis

As noted in the Introduction, distant organ metastasis is a primary factor in determining cancer patient survival. Metastatic spread is thought to occur via the body's systems for transporting fluid and cells, that is the blood vascular and lymphatic systems. Cancer cells may use these systems through expression of growth factors that can alter the normal pattern of angiogenesis and lymphangiogenesis, thus creating conduits for tumour metastasis.[36,37]

In summary, tumours have three ways of spreading: (i) by local invasion; (ii) through the bloodstream; and (iii) through the lymphatic system. However, in early lung cancer, the most important mechanism for tumour spreading is usually via the lymphatic system.

Lymph node metastasis

Metastatic tumour dissemination through the blood or lymphatic vessels occurs in most forms of human cancer, with regional lymph node metastasis often being the most important prognostic factor for early carcinoma patients.[38] Therefore, it is important to determine the potential routes of cancer metastasis via the lymphatic vasculature in early lung cancer.

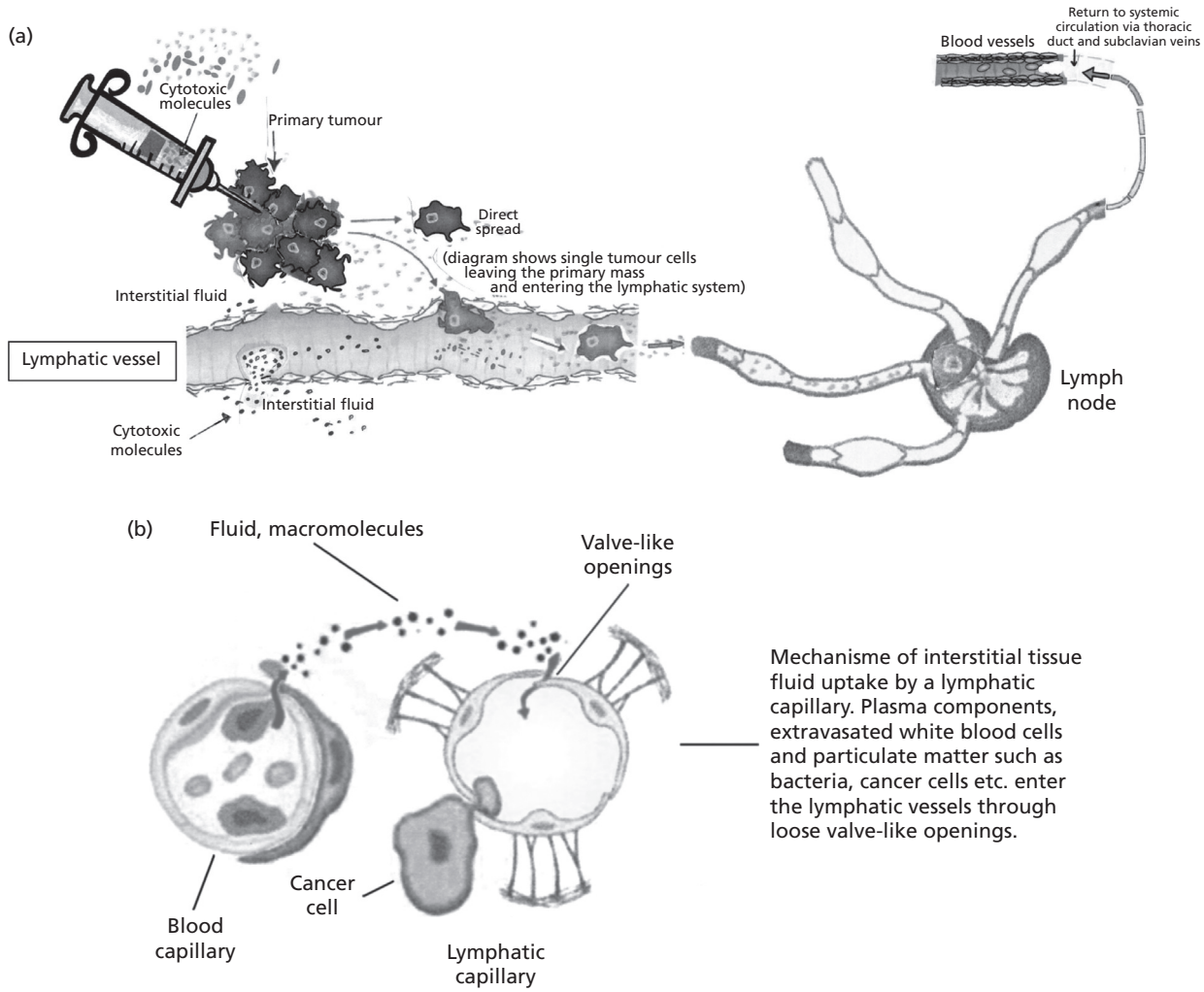

Figure 1 Schematic diagrams of lymphatic drainage. (a) Lymphatic vessel structure and potential modes of tumour cell dissemination via lymphatics and transport of intratumourally injected cytotoxic drug to the lymph nodes. Modified from: Stacker SA, Baldwin ME, Achen MG. The role of tumor lymphangiogenesis in metastatic spread. *FASEB J* 2002; 16: 922–934. (b) Mechanism of interstitial tissue fluid uptake by lymphatic capillary

Micrometastasis and occult lymph node metastasis

Micrometastasis to lymph nodes is thought to occur in most primary tumours before they are clinically detectable and therefore can be the most significant prognostic indicator in many human cancers.[42] Occult lymph node metastases (undetected by conventional methods) have been seen in up to 20% of T1 and T2 lung cancer tumours.[43,44] Positive emission tomography (PET) has demonstrated more sensitivity and specificity than computed tomographic scan for mediastinal lymph node assessment in patients with potentially operable NSCLC.[45] However, micrometastases, and especially microscopic nodal disease, might be underestimated by PET due to its limitation for detection of lesions smaller than 4–5 mm.[46,47] Therefore, it must always be understood that PET cannot detect small or micrometastases because of the lower limits of resolution of the PET camera.[46]

The following section considers the literature concerning clinical trials devoted to the detection of micrometastases in the draining lymph nodes in order to demonstrate the potential efficiency of intratumoural and peritumoural injection of cytotoxic drugs.

Detection and mapping of human sentinel lymph nodes

Based on results of clinical studies reported for the preoperative delivery of tracer substances for sentinel node detection and mapping, it is reasonable to conclude that EITC should result in transport of the cytotoxic drugs by lymphatic drainage to the regional lymph nodes in a manner similar to the transport of tracer substances. As a consequence, anticancer drugs injected into and around solid tumours may be expected to migrate to the regional nodes and thereby eradicate early micrometastases in the regional lymph nodes. Preoperative (neoadjuvant) EITC should therefore be an especially effective prophylactic treatment

in the early stages of lung cancer, with an expected improvement in postoperative patient survival.

Sentinel lymph nodes

The SLNs may be defined as the first nodes in the lymphatic basin into which the primary tumour drains. The SLN is thereby the first lymph node to receive lymphatic drainage from a tumour with the potential to contain metastatic cancer cells.[48–50] The SLNs are part of an orderly and predictable lymphatic drainage pathway that may incorporate migrating metastatic tumour cells.[36]

Identifying the sentinel lymph nodes

The SLNs for a given tumour may be determined by injecting a tracer compound at the periphery of the tumour that will migrate via the lymphatic system to the first draining node (the SLN) and identify it by staining. The tracer substance may be a vital staining dye such as isosulfan blue that can be visualized, or a radiolabelled compound that can be examined radiologically.

In 1992, Morton and colleagues[48] first described lymphatic mapping utilizing an intradermal injection technique for malignant melanoma. They identified the specific lymph nodes that receive afferent lymphatic drainage from the primary tumour site and were the first to employ this approach to identify SLNs for lymphadenectomy in patients with malignant melanoma. They proved that 'skip metastases', that is passage beyond an uninvolved sentinel node, was a rare event (<2%). Therefore, if the SLN is negative for metastatic cells, the remainder of the lymph nodes should be negative.[48] In another melanoma study, Tiffet *et al.*[49] showed that intraoperative dynamic lymphoscintigraphy correctly identified 90% of the SLNs in less than 15 min after a peritumoural injection.

Importance of injecting the tracer at the periphery of the tumour

It has been thought that the lymphatics within the tumour do not readily transport cancer cells because the elevated hydrostatic pressure within a tumour may compress these vessels.[51] Based on animal models, it is suggested that intratumoural lymphatic vessels may not be completely functional because of vessel collapse.[52] It is therefore more likely that lymphatic vessels at the tumour margin may be of primary importance for tumour cell dissemination.[51] The injection of tracer substances at the tumour margin is therefore preferred for diagnostic purposes because the tracer is thereby likely to be more easily transported via the local lymphatic vessels.[40] In the context of this review, in which we propose the use of intratumoural chemotherapy for both diagnostic and therapeutic purposes, it is important that cytotoxic drugs be injected at the periphery of the tumour as well as the interior of the tumour for intratumoural chemotherapy. For more effective EITC, it is therefore important to inject cytotoxic drugs (including a chromophoric drug such as blue mitoxantrone) into the normal bronchial mucosa around the tumour in addition to direct injection into the tumour bulk.

Sentinel lymph node metastasis in lung cancer

Intraoperative and preoperative computed tomographic guided injection techniques

The diagnostic concept for sentinel lymph node metastasis described by Morton *et al.*[48] for melanoma has the potential to be applied to lung cancer. Indeed, to reduce the need for mediastinal lymph node dissection, the availability of intraoperative sentinel lymph node dissection biopsy has been examined using radioisotope mapping in lung cancer.[9,53–55]

A sentinel lymph node labelling technique might thereby increase the accuracy of surgical lymph node staging and improve the prognostic significance of micrometastatic lymph node involvement in patients with early stage NSCLC. SLN mapping with blue dye and with radioisotopic technetium-99m has been shown as a safe way of identifying the first site of potential nodal metastases in up to 85% of lung cancer patients.[40]

In a recent study by Tiffet *et al.*,[56] SLN staining was evaluated in stage I or II peripheral NSCLC. A 2-ml volume of Patent Blue Dye-V (Laboratory Guerbet, Roissy, France) was injected in four divided doses (0.5 ml each) into each quadrant of lung tissue immediately surrounding the tumour perioperatively in a collapsed lung. The mean time from injection to examination of SLNs was 18 min (5–30 min). This study provides supportive data for the SLN and the application of SLN staining to lung cancer, with a sensitivity of 75% and a false negative of 14%. The authors suggest that preoperative bronchoscopic staining injections need to be further explored in future studies.[56]

Bronchoscopic injection of radioisotopes for detection and mapping of sentinel lymph nodes

In the studies noted above, investigators used intraoperative or perioperative techniques for the SLN detection.[57] Lardinois *et al.*,[40] in a prospective study, evaluated the feasibility of a preoperative bronchoscopic radioisotope application by transbronchial injection, followed by conventional SLN identification. The occurrence and distribution of micrometastases in relation to SLN activity was also investigated in this study. Radiolabelling with technetium-99m nanocolloid was endoscopically delivered by injection in intubated patients in the operating room. Clinical stage T1–3 N0–1 NSCLC was included in that study.

Patients were initially intubated with a single-lumen endotracheal tube. Then, bronchoscopy was performed with a fibre-optic bronchoscope through the endotracheal tube. When the tumour was bronchoscopically visible, a protected 21 gauge and 13 mm long transbronchial injection needle device was inserted through the endoscope. The needle tip was transbronchially inserted at the tumour margin. The mean time interval between endoscopic injection and first measurement of radioactivity with a hand-held gamma probe was 1.4 h (range 1–3 h). At thoracotomy, scintigraphic readings of both the primary tumour and hilar and mediastinal lymph node stations were obtained with a hand-held gamma counter. Patients underwent lung resection and mediastinal lymph adenectomy. These authors stated that the identification of SLNs was possible in 19/20 (95%) patients after bronchoscopic injection of the radiolabel. Metastatic nodal disease was

found in 9/19 patients (47%). These preliminary results suggest that SLN mapping using a preoperative bronhoscopic radioisotope technique may be easy to perform and safe. No complications related to the procedure were observed. In addition, using this technique, the diagnostic yield for identification of SLNs was greater than by the intraoperative technique, where a sentinel node could be found in 47–82% of patients.[40] This may be due to the method of injecting technetium-99m with ventilated lungs and to the fact that the migration of the radionucleide was not affected during preparation of the structures to be resected as occurs with the intraoperative technique. The bronchoscopic technique may also afford the advantage of being less invasive for the patient than the transthoracic approach.

Intratumoural injection of tracer substances for peripheral tumours not visible bronchoscopically

Lardinois *et al*.[40] states in the above-mentioned study: 'when the tumor was not visible endo-bronchially, the needle was inserted at the carina of the most distal pulmonary sub-segment that could be reached endoscopically in the proximity of the tumor according to its location on preoperative computed tomographic scan; and despite that sentinel lymph nodes were visualized'. This statement is also important for delivery of cytotoxic drugs for therapeutic purposes.

Intratumoural injection of cytotoxic drugs for treatment of possible micrometastases

The observations of Lardinois *et al*.[40] are important as they to support our hypothesis that a chemotherapeutic agent could also be transported to the sentinel and regional lymph nodes through lymphatic drainage following injection of a drug into an area of normal bronchial mucosa that drains the tumour. Indeed, they have demonstrated that any radioactive substance in solution, delivered by injection into the bronchial wall adjacent to the tumour, will reach the regional lymph nodes by lymphatic drainage. Similarly, it is reasonable to conclude that cytotoxic drugs injected directly into the bronchial wall adjacent to the tumour site will be transported to the regional lymph nodes.

Clinical evidence of cytotoxic drug transport via lymphatic drainage to the sentinel and regional lymph nodes

Animal studies have demonstrated that mitoxantrone (a blue cytotoxic drug) appears in the lymphatic nodes draining the intratumoural injection site.[58] Such first-pass transport of drug to the regional lymph nodes therefore has important implications for locoregional therapy of metastasis. Indeed, Baitchev *et al*.[59] in their clinical study in early breast cancer, have observed the first-pass transport of the blue-staining mitoxantrone to the regional lymph nodes. These authors investigated the clinical and morphological behaviour of drug diffusion and efficacy after perioperative locoregional application of mitoxantrone in patients with early breast cancer. In this study the locoregional application of mitoxantrone was carried out by injection through a 25 gauge needle at a dose of 0.5 ml (1.0 mg) at two sites into the primary tumour and surrounding breast parenchyma in 37 patients. Because of its staining property, this local mitoxantrone injection was found to be useful as a marker for the SLNs. Intraoperative inspection of mitoxantrone diffusion in breast and axillary tissues was enabled by the blue colour of the drug. Identification of reactive morphological changes in axillary nodes was determined intraoperatively by blue staining of lymph vessels identified 2–4 cm laterally from the injection sites. For 30 patients, a mean of 1.5 stained axillary nodes/patient was found.

Light microscopy of metastatasis in blue-stained lymph nodes showed changes: dilatation of marginal sinus and inflammatory alteration. Some characteristic changes for the metastatic tumour cells in SLNs due to cytotoxic agents were found, that is vacuolization of the cytoplasm and nuclei, and irregular delineation of the nuclear membrane. In some cases, small prominent hyperchrome nucleoli and isolated apoptoic corpuscles among the tumour cells were observed. The median follow-up of patients was 19.2 months (range 8–30 months). Over this period of time, no locoregional recurrence or distant metastases were observed. Baitchev *et al*.[59] concluded that 'lymphotropic loco-regional application of mitoxantrone results in diffusion of the drug in the regional lymph drainage and cytotoxic effects in axillary nodes'. This study strongly corroborates observations in early lung cancer.

Discussion

Patients presenting with stage I and II lung cancer are usually considered candidates for potentially curative resection.[1] However, in these cases, 5-year survival after surgery is not as high as might be expected. This may be explained by the existence of occult infraclinical lymph node metastases. Indeed, during operation, 20–25% of these patients are recognized as having mediastinal lymph node metastases not detected by normal preoperative diagnostic procedures.[3,4] Studies have shown that if nodal involvement is present, the prognosis for long-term survival is less than 50%.[5] On the other hand, it has been demonstrated that adequate surgical dissection of the regional lymph nodes somewhat improves survival. Based on such studies and experience, surgeons do their best to achieve a systematic and complete lymph node dissection during surgical resection. Yet conventional mediastinal lymph node dissection cures only 20–25% of lymph node metastasis at best. This means that perhaps only 5% of the patients indicated for resection may benefit from a conventional mediastinal lymph node dissection.[9] Therefore, clinical research aimed at improving the usual techniques of mediastinal lymph node dissection is important in view of the current minimal efficacy and poor survival rates.

Mediastinal lymph node dissection with the help of sentinel lymph node mapping and detection is increasingly used for local control of NSCLC, with an improvement in survival rates.[10–12] However, in spite of such technical refinements, the relapse rate after surgery, even in early stage lung cancer, due to regional lymph node micrometastasis remains quite high. As a consequence, some thoracic surgeons favour the use of systemic chemotherapy administered preoperatively or postoperatively even in stage I disease.[25] This view is supported by studies suggesting significant improvement in prolonged survival with intravenous administration of drugs such as cisplatin, paclitaxel and carboplatin in patients with

stage IIIA or IIIB NSCLC.[14–19] Based on these partially successful results with stage IIIA and IIIB patients, it has been considered that a preoperative sequence of systemic intravenous chemotherapy and/or radiotherapy administered to early stage I/II lung cancer might also be advantageous as a prophylactic measure against occult metastases. However, the response rates and prolonged survival times achieved by the addition of systemic chemotherapy and radiotherapy to the treatment protocol before surgery in patients with NSCLC stage I and II remain modest, and these small gains are obtained at the cost of significant toxicity. Thus, systemic chemotherapy and radiotherapy is not routinely used before surgery in stage I and II disease.[25] There is therefore a significant need for a more efficient and safer preoperative therapy that would eradicate infraclinical micrometastases or occult metastases in patients presenting with early lung cancer.

In view of the results of our previous clinical study demonstrating the beneficial effect on survival of preoperative delivery of cytotoxic drugs by injection into the tumour area through a bronchoscope, this minimally invasive bronchoscopic procedure should be considered a localized neoadjuvant chemotherapy procedure that can kill micrometastases or occult metastases.[60]

To support this hypothesis, we have reviewed here the physiopathology of lymphatic drainage as well as the incidence of lymph node metastasis.[9] Indeed, the results of recent investigations on the physiology of lymphatic drainage to the SLNs supports the view that intratumoural delivery of cytotoxic drugs can not only destroy the primary lesion but also enhance the eradication of regional lymph node metastases in early lung cancer.

Important corroboration of our concept for control of lymph node metastasis by preoperative EITC is afforded by the study of Lardinois *et al.*[40] using preoperative bronchoscopic injection of labelling compounds for SLN mapping. That study clearly established that preoperative bronchoscopic injection of tracer/marker agents into the tumour and adjacent normal bronchial mucous membrane results in their transport to sentinel and regional lymph nodes within 6 h. The migration of cytotoxic drugs injected similarly should follow the same pathway and kill malignant metastatic cells without systemic drug toxicity. The clinical findings of Baitchev *et al.*[59] with intratumoural injection of blue mitoxantrone in early breast cancer also supports our hypothesis that a chromophoric cancer drug such as mitoxantrone (with a molecular weight comparable with staining dyes) will be transported to the sentinel and regional lymph nodes through lymphatic drainage immediately following the bronchoscopic injection into either the tumour or the tissue adjacent to the tumour site.

Recently, Xie *et al.*[61] have published a review that indicates the importance of drug delivery to the lymphatic system in future cancer therapies. They note that we have

Table 1 Key studies relevant to this review and their major contributions

Goldberg *et al.*[26]	This comprehensive review outlines experimental and clinical studies concerning the delivery of anticancer drugs by inratumoural injection. The review indicates that the direct intratumoural administration of a cytotoxic drug can afford a 10–30 times higher concentration of drug in the tumour tissue without systemic toxicity as compared with conventional chemotherapy.
Seymour *et al.*[35]	This is a review of the medical literature on the subject of intratumoural delivery of various drugs by direct injection using a special needle catheter through a bronchoscope for diagnostic and therapeutic purposes. The review indicates that bronchoscopic delivery of drugs is very effective with no undesirable side-effects.
Celikoglu *et al.*[29]	This review summarizes studies on the efficacy of intratumoural injection of cytotoxic drugs for the treatment of lung cancer with endobronchial localization by use of a needle catheter through a flexible bronchoscope. It demonstrated that intratumoural delivery of cytotoxic drugs by direct injection necrotizes the cancer cells and facilitates opening of obstructed airways. It is shown that there is no local adverse effect of the technique and no systemic drug toxicity, a general problem for intravenous drug delivery. It is also shown that this method may be considered a complementary interventional bronchoscopic procedure that can be used in combination with other conventional local and systemic cancer therapies. The method is shown to have significant advantages compared with other interventional bronchoscopic procedures because it is also effective for patients presenting with tumour infiltration of the airways wall and for extrabronchial localization of the tumour. In addition, it is suggested that intratumoural injection of cytotoxic drugs can result in drug transport to locoregional lymph nodes for the eradication of occult metastases. This issue is an important aspect of the present review. The following two clinical investigations by Baitchev *et al.*[59] and Lardinois *et al.*[40] afford very important results support to confirm this hypothesis.
Baitchev *et al.*[59]	This clinical study clearly demonstrates that a chromophoric cancer drug (such as mitoxantrone with a molecular weight comparable with staining dyes used for sentinel lymph node mapping) is transported to the sentinel and regional lymph nodes through lymphatic drainage in early breast cancer, immediately following the intratumoural injection into either the tumour or into the tissue adjacent to the tumour site. Furthermore, it was also shown by pathological examination that the metastatic cancer cells observed in the extirpated lymph nodes were necrotized.
Lardinois *et al.*[40]	This clinical study involved preoperative sentinel lymph nodes mapping and detection by intratumoural injection of a tracer substance with a needle catheter through a flexible bronchoscope and demonstrated that the tracer was readily transported to sentinel lymph nodes via lymphatic channels. We deduced from these findings that cytotoxic drugs directly injected into tumour will also be transported through lymphatic vessels to the locoregional lymph nodes. Taking into consideration the results of Baitchev *et al.*,[59] we may conclude that the intratumoural and extratumoural bronchoscopic injection of a drug such as cisplatin will result in drug transport to the locoregional lymph nodes, thereby attacking occult metastases or micrometastases.
Xie *et al.*[61]	This recent publication emphasizes the importance of drug delivery to the lymphatic system in future cancer therapies for the eradication of occult metastases.

known for more than 40 years that the lymphatics are the first site of metastasis for most solid cancers. However, until now, there have been few studies aimed at targeting chemotherapy to lymphatic tissues. With conventional systemic chemotherapy, it is almost impossible to deliver cytotoxic drugs specifically to the tumour cells or lymphatic system without damaging healthy organs or tissues. It is therefore important to emphasize here that intratumoural chemotherapy accompanied by the intralymphatic effect affords a unique approach to the eradication of both primary lesions as well as metastasis.

Conclusions

In view of the fact that only moderate benefit is achieved for NSCLC by administration of systemic chemotherapy, it seems reasonable to propose clinical trials of preoperative locoregional intratumoural injection of chemotherapy with currently approved drugs (especially including a staining drug such as blue mitoxantrone or red daunorubicin) with the expectation that drug transport to the regional nodes will destroy (and stain) metastatic tumour cells that have migrated to the lymph nodes (see Table 1).

Considerable experience with minimally invasive bronchoscopic intratumoural delivery of cytotoxic drugs for obstructive lung cancer treatment has indicated no complications using this technique and no systemic toxic side-effects from cytotoxic drugs such as cisplatin, 5-fluorouracil and mitoxantrone.[26–35] This experience provides the ethical support for prospective clinical studies. Accordingly, we propose open single-arm clinical trials using preoperative intratumoural and peritumoural injection of cisplatin (combined with mitoxantrone as a marker). In this regard, we have effectively used the blue staining cancer drug, mitoxantrone, in EITC clinical studies for more than 10 years.[27]

Declarations

Conflict of interest

The Author(s) declare(s) that they have no conflicts of interest to disclose.

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