Drug Evaluation
Future Oncology

Lyso-thermosensitive liposomal doxorubicin: an adjuvant to increase the cure rate of radiofrequency ablation in liver cancer

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Hepatocellular carcinoma (HCC) is the fourth leading cause of cancer death worldwide. No more than 30% of HCC patients are considered suitable for curative treatment because of tumor size and severity of liver impairment, among other factors. Radiofrequency ablation (RFA) monotherapy can cure small (<3 cm) HCC tumors. An adjuvant that interacts synergistically with RFA might enable curative therapy for many HCC patients with lesions >3 cm. Lyso-thermosensitive liposomal doxorubicin (LTLD) consists of the heat-enhanced cytotoxic doxorubicin within a heat-activated liposome. LTLD is infused intravenously prior to RFA. When heated to >39.5°C, LTLD releases doxorubicin in high concentrations into the tumor and the tumor margins. The RFA plus LTLD combination has shown a statistically significant dose–response effect for time to treatment failure in a Phase I trial in which most subjects (62.5%) had tumors >3 cm. RFA plus LTLD is currently being evaluated in a 600-patient randomized, double-blind, dummy-controlled trial.

Hepatocellular carcinoma (HCC) is the fourth leading cause of cancer death worldwide [101]. Surgical resection is the mainstay of curative treatment. However, no more than 30% of HCC patients are considered suitable for surgical treatment because of tumor size, multifocal tumors, vascular invasion, presence of extrahepatic metastases and/or extensive liver impairment. The majority of patients with HCC have underlying cirrhosis, most commonly due to hepatisis B or C viral infection, which may restrict the feasibility of surgical resection even with small tumors. Liver transplantation is an alternative curative treatment, but its application is limited by a severe shortage of liver graft donors. Thermal ablation modalities such as radiofrequency ablation (RFA), microwave ablation and high-intensity focused ultrasound have emerged as important treatment options for such patients in recent years [1].

During RFA, radiofrequency waves emitted from an electrode inserted into the tumor induce vibration of ions in the cancer cells, which causes frictional heat and thermal coagulative necrosis of the cells. RFA monotherapy is a curative treatment for HCC tumors ≤3 cm. It is a safe treatment with a morbidity rate of less than 10% and a mortality rate of approximately 0.5% [2]. It is the most commonly used treatment for patients with small HCC associated with significant cirrhosis not suitable for resection. Whether it can replace resection as the treatment of small HCC in patients with good liver function remains controversial. A randomized trial has shown that RFA can achieve long-term survival rates for small HCC that are similar to resection [3], but a more recent randomized trial demonstrated that survival and recurrence rates were better with resection than RFA [4]. The efficacy of RFA is significantly influenced by tumor size. For patients with lesions too big to be treated within a single ablation zone (those with lesions >3 cm, approximately half of the HCC population), RFA is much more likely to leave viable tumor cells in the margins or clefts of overlapping ablation zones. Local recurrence rates after RFA for tumors ≤3 cm are reported to be ≤20% [5–9]; however, for tumors >3 cm, local recurrence rates of ≥40% have been observed [10,11].

Lyso-thermosensitive liposomal doxorubicin (LTLD; ThermoDox® [Celsion Corp., MD, USA]) is designed for use with RFA and consists of the heat-enhanced cytotoxic anthracycline antibiotic doxorubicin within a heat-activated liposome. This article reviews the rationale and preliminary clinical data of its combined use with RFA for liver cancer and discusses its potential role in the future oncological treatment of HCC.

Keywords
- hepatocellular carcinoma
- lyso-thermosensitive liposomal doxorubicin
- radiofrequency ablation

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Rationale

A review of 13 published trials of single-agent doxorubicin among 644 HCC patients found an objective response rate of 19% and median overall survival of 4 months [12]. In subsequent randomized trials comparing overall survival, single-agent doxorubicin has been found to be superior to no anticancer treatment [13], equivalent to combination chemotherapy with cisplatin, IFN-α-2b, doxorubicin and fluorouracil [14] and superior to single-agent nolatrexed [15]. However, systemic doxorubicin has not become a standard treatment for HCC due to its relatively high incidence of severe toxicity, including congestive heart failure and neutropenia [13–15].

Lyso-thermosensitive liposomal doxorubicin is administered intravenously but, because it is a liposome, rapidly concentrates in the liver and spleen [16]. Since LTLD is larger than free doxorubicin, it is over 1000-times less permeable across normal blood vessels than free doxorubicin, offering less potential for systemic toxicity. However, tumors have much higher microvascular permeability than normal blood vessels, so LTLD is able to accumulate in tumors [16,17]. In addition, hyperthermia has been shown to preferentially increase liposomal permeability within the microvasculature in tumor versus normal tissue. Hyperthermia has a biological effect of increasing the pore size in tumor blood vessels, and therefore enhancing the extravasation of liposomes into the tumor interstitium. Studies demonstrated that tumors that were impermeable to liposomes at 34°C had significant extravasation and thus increased permeability at 42°C. The optimal liposome size for heat-induced extravasation was found to be 100 nm (the mean diameter of LTLD). The same effect was not observed in normal tissue [18,19]. Therefore, it can be hypothesized that hyperthermia induces preferential extravasation of liposomes in tumor tissue compared with healthy tissue. The benefits of hyperthermia with thermosensitive liposomes for the delivery of chemotherapeutic agents are twofold: enhanced localization of the liposomes into the tumor interstitium, coupled with a triggered and rapid release of the drug in the tumor and the tumor vasculature.

Within 30 s of exposure to temperatures of ≥39.5°C, LTLD releases its doxorubicin contents, creating a large concentration gradient of doxorubicin around the zone of RFA-induced tumor cell necrosis. Preclinical studies found that at temperatures of ≥39.5°C, LTLD produces doxorubicin tumor concentrations up to 15-fold greater than free (nonliposomal) doxorubicin administered at the same doses [Celsion Corp., Unpublished Data]. The doxorubicin then kills any tumor cells adjacent to the ablation zone, providing more successful treatment of HCC lesions >3 cm than thermal ablation alone. *In vitro* studies have repeatedly shown enhancement of cell killing when doxorubicin is combined with hyperthermia compared with doxorubicin without hyperthermia [20–31]. This enhancement has been attributed to the ability of hyperthermia to increase intracellular retention of chemotherapeutic agents by upregulating their influx [20,26].

Overview of strategies to improve RFA treatment for HCC

Box 1 summarizes initiatives to increase the HCC cure rate by combining another therapy with RFA. This article will not deal with proposals to increase the efficacy of RFA alone (e.g., better guidance with contrast-enhanced ultrasound, fusion imaging or robotics; redesigning RFA electrodes; or improving treatment algorithms to magnify the ablation zone) [32–34]. One approach aims to improve the cure rate in multifocal disease by performing surgical resection on resectable tumors and RFA on unresectable lesions [35]. A strategy for tumors >3 cm is to perform transarterial chemoembolization (TACE) to downsize the tumors so they are more curable with RFA [36]. Thus far, randomized trial findings for this combination are modest. Morimoto *et al.* randomized 37 patients with solitary medium (3.1–5.0 cm) HCC lesions to RFA plus TACE or RFA alone [37]. After 3 years, the RFA plus TACE group had a statistically significant advantage in local tumor control, but not in overall survival. Wang *et al.* randomized 83 patients, of whom 59% had lesions >3 cm, to RFA plus TACE or RFA alone [38]. The RFA plus TACE group had a statistically significant advantage in 'quality of life', but not in recurrence rate. Yang *et al.* randomized 36 patients with large (>5 cm) HCC lesions to RFA plus TACE or RFA alone [39]. The two groups did not differ significantly in either relapse rate or overall survival. A frequent suggestion for tumors >3 cm in the literature is to perform RFA and then to administer a systemic treatment to eradicate any residual tumor. Therapy with interferon [40,41], sorafenib (or other molecularly targeted agents once available) [42–44] and vitamin A or vitamin K analogs [45] have been recommended. In the simultaneous RFA plus LTLD approach, LTLD is administered systemically but rapidly concentrates in the liver, where it acts synergistically with RFA.

Box 1. Radiofrequency ablation-based combination therapies to increase the hepatocellular carcinoma cure rate.

Surgery for resectable tumors and RFA for unresectable tumors (efficacy additive)

- RFA plus partial hepatectomy [35]

Precede RFA with local treatment to downsize tumor (efficacy additive)

- TACE plus RFA [36–39]

Follow RFA with systemic therapy to eradicate any residual tumor (efficacy additive)

- RFA plus interferon [40,41]
- RFA plus sorafenib (or newer molecularly targeted agents) [42–44]
- RFA plus vitamin analogs [45]

Simultaneous RFA and heat-enhanced, organ-specific chemotherapy (efficacy synergistic)

- RFA plus lyso-thermosensitive liposomal doxorubicin [47]

RFA: Radiofrequency ablation; TACE: Transarterial chemoembolization.

Introduction to LTLD

Chemistry

Lyso-thermosensitive liposomal doxorubicin is the first heat-activated formulation of liposomal doxorubicin. The active drug in LTLD is doxorubicin hydrochloride. Doxorubicin consists of a naphthacenequinone nucleus linked through a glycosidic bond at ring atom 7 to an amino sugar, daunosamine. The chemical formula is $C_{27}H_{29}NO_{11}$·HCl and the molecular weight is 579.99 Da. LTLD combines doxorubicin with lyso-thermosensitive liposomes that are made from three synthetic phospholipids: 1,2-dipalmitoyl-*sn*-glycero-3-phosphocholine, 1-stearoyl-2-hydroxy-*sn*-glycero-3-phosphocholine and 1,2-distearoyl-*sn*-glycero-3-phosphoethanolamine-*N*-methoxypolyethylene-glycol-2000. LTLD is manufactured as stable doxorubicin-loaded liposomes, which are stored as a frozen solution [Celsion Corp., Unpublished Data].

Pharmacodynamics

For single LTLD doses, the plasma half-life (mean ± standard deviation of 24 patients) is 18.52 ± 8.36 h. Initial evaluation of two cycles of LTLD, 21 days apart, suggests that there does not appear to be any appreciable accumulation of doxorubicin between LTLD infusions [Celsion Corp., Unpublished Data].

Pharmacokinetics & metabolism

The major portion of exposure to LTLD (~95% of the liposomal doxorubicin plasma area under the curve $[AUC]_{0-\infty}$) occurs during the first 6 h following the infusion, establishing this time period as optimal for application of RFA. Most of the free doxorubicin exposure also occurs during the first 6 h (87.9% of the free doxorubicin $AUC_{0-\infty}$). Free doxorubicin represents 43.6% of the total doxorubicin $AUC_{0-\infty}$. For both liposomal and free doxorubicin, maximum plasma concentration occurs just before the end of the 30-min infusion [Celsion Corp., Unpublished Data].

Clinical efficacy

Phase I studies

A 24-subject trial (nine with HCC and 15 with metastatic liver tumors from nine other primary sites) has been completed. A total of 15 (62.5%) of the 24 subjects had tumors >3.0 cm [Celsion Corp., Unpublished Data]. Treatment failure was operationally defined as objective disease progression and/or initiation of an alternative anticancer therapy. There was a statistically significant (p = 0.038) LTLD dose–response effect: median time to treatment failure for patients receiving at least the maximum tolerated dose of 50 mg/m^2 was 374 days, while that for patients receiving less than 50 mg/m^2 was 80 days. Time to treatment failure was significantly associated with LTLD dose but not with tumor size (≤3 cm or >3 cm), tumor type (HCC or metastatic liver cancer) or RFA type (open surgical or percutaneous) (Table 1). There were three subjects at the 60 mg/m^2 dose level. None of these three subjects experienced treatment failure; they were censored at 122, 283 and 337 days. Furthermore, there were four subjects with tumors >5 cm in the trial. Of these, the two subjects treated at <50 mg/m^2 experienced treatment failure at 25 and 93 days, respectively, while the two subjects treated at ≥50 mg/m^2 experienced treatment failure at 261 and 374 days, respectively [Celsion Corp., Unpublished Data]. These very limited data in large liver lesions are again suggestive of a LTLD dose–response relationship.

Phase II studies

Owing to promising Phase I results, the developers of LTLD worked with the US FDA and proceeded directly into Phase III testing.

Table 1. Phase I radiofrequency ablation/lyso-thermosensitive liposomal doxorubicin time to treatment failure by primary site, tumor size, radiofrequency ablation type and lyso-thermosensitive liposomal doxorubicin dose.

Factor	Failed	Censored	Median TTF (days)	p-value
Primary site				
Liver	7	2	355	0.2227
Other†	13	2	64	
Tumor size‡				
≤3.0 cm	6	2	156	0.4135
>3.0 cm	14	1	86	
RFA type				
Open surgical	6	1	188	0.4315
Percutaneous	14	3	80	
LTLD dose				
<50 mg/m^2	15	0	80	0.0380
≥50 mg/m^2	5	4	374	

Source data are from the clinical study report [Celsion Corp., Unpublished Data] *and were extracted as of 5 February 2008. Treatment failure is operationally defined as disease progression and/or initiation of an alternative anticancer therapy. This is a conservative way to assess efficacy, since the 15 patients with other primary sites might progress and/or begin alternative treatment for that primary or for a baseline metastatic site other than the liver. Patients not experiencing treatment failure are censored as of their last reported on-study date. For each factor, TTFs are computed by the product-limit method* [48] *and compared by the two-tailed log-rank test* [49]. *No adjustments are made for multiple comparisons.*

†*There were a total of nine other primary sites.*

‡*The maximum tumor diameter for one patient was not reported.*

LTLD: Lyso-thermosensitive liposomal doxorubicin; RFA: Radiofrequency ablation; TTF: Time to treatment failure.

Phase III studies

A randomized, double-blind, dummy-controlled, multicenter Phase III trial ('HEAT study') is underway, comparing RFA/LTLD with RFA alone among 600 patients with unresectable HCC. Child–Pugh class A or B patients are eligible, except that both ascites and encephalopathy are exclusionary. Eligible patients can have no more than four HCC lesions with at least one ≥3.0 cm and none >7.0 cm in maximum diameter. However, if a patient has a large lesion (5.0–7.0 cm), any other lesions must be less than 5.0 cm. In both arms, RFA may be performed percutaneously, laparoscopically **or surgically, per the investigators'** clinical judgment. Patients in the combination arm will receive a single 30-min intravenous infusion of LTLD at 50 mg/m^2, starting 15 min before RFA; the RFA-only arm patients will receive a dummy infusion. Progression-free survival is the primary end point. Secondary end points include overall survival, time to local recurrence and time to a clinically significant deterioration in patient self-reported symptoms. The National Cancer Institute of the USA has recommended the HEAT study as a priority clinical trial for HCC [46].

Lyso-thermosensitive liposomal doxorubicin is not approved for use yet. However, this pivotal trial has randomized 556 of the 600 HCC subjects at the time of writing this article. It is expected that the Phase III trial will produce data robust enough to test the hypothesis that LTLD in combination with RFA could increase the cure of medium/large HCC.

Safety & tolerability

The maximum tolerated dose of LTLD was determined to be 50 mg/m^2 in the Phase I trial, based on two dose-limiting toxicities (grade 3 alanine aminotransferase increase and grade 4 neutropenia) at a dose of 60 mg/m^2 [Celsion Corp., Unpublished Data]. LTLD was not associated with either hand–foot syndrome or with congestive heart failure. The most common LTLD grade 3+ adverse events (affecting ≥5% of patients) were aspartate aminotransferase abnormalities (40.0%), alanine aminotransferase abnormalities (32.7%), neutropenia (29.1%), leukopenia (12.7%) and lymphopenia (9.1%). Very few of the abnormalities in liver function were associated with LTLD. The most common LTLD drug-related adverse events (affecting ≥5% of patients) were alopecia (36.4%), neutropenia (32.7%), leukopenia (20.0%), decreased hemoglobin (18.2%), fatigue (14.5%), nausea (10.9%), thrombocytopenia (9.1%), decreased ejection fraction (9.1%), anorexia (7.3%), taste alteration (5.5%) and fever (5.5%) (Table 2). All of the

Table 2. Frequency listing of adverse events while on lyso-thermosensitive liposomal doxorubicin.

AE	Lyso-thermosensitive liposomal doxorubicin (n = 55)			
	Any AE, n (%)	*Grade 3 or more AE, n (%)*	*Drug-related AE, n (%)*	*Serious AE, n (%)*
Metabolic/laboratory symptoms				
AST, serum glutamic oxaloacetic transaminase	28 (50.9)	22 (40.0)	1 (1.8)	0 (0.0)
ALT, serum glutamic pyruvic transaminase	28 (50.9)	18 (32.7)	1 (1.8)	0 (0.0)
Serum albumin – low (hypoalbuminemia)	10 (18.2)	0 (0.0)	0 (0.0)	0 (0.0)
Serum sodium – high (hypernatremia)	7 (12.7)	1 (1.8)	0 (0.0)	0 (0.0)
Serum potassium – high (hyperkalemia)	6 (10.9)	2 (3.6)	0 (0.0)	0 (0.0)
Creatine phosphokinase	5 (9.1)	1 (1.8)	0 (0.0)	0 (0.0)
Alkaline phosphatase	4 (7.3)	1 (1.8)	0 (0.0)	0 (0.0)
Serum glucose – high (hyperglycemia)	3 (5.5)	1 (1.8)	0 (0.0)	0 (0.0)
Serum phosphate – low (hypophosphatemia)	3 (5.5)	2 (3.6)	0 (0.0)	0 (0.0)
Serum potassium – low (hypokalemia)	3 (5.5)	0 (0.0)	0 (0.0)	0 (0.0)
Blood and lymphatic system disorders				
Neutropenia/granulocytopenia (ANC/AGC)	18 (32.7)	16 (29.1)	18 (32.7)	0 (0.0)
Decreased hemoglobin	13 (23.6)	1 (1.8)	10 (18.2)	0 (0.0)
Leukocytes (decrease in total WBCs)	11 (20.0)	7 (12.7)	11 (20.0)	0 (0.0)
Thrombocytopenia	9 (16.4)	2 (3.6)	5 (9.1)	1 (1.8)
Lymphopenia	5 (9.1)	5 (9.1)	1 (1.8)	0 (0.0)
Gastrointestinal disorders				
Nausea	12 (21.8)	0 (0.0)	6 (10.9)	0 (0.0)
Anorexia	8 (14.5)	0 (0.0)	4 (7.3)	0 (0.0)
Vomiting	7 (12.7)	0 (0.0)	2 (3.6)	0 (0.0)
Constipation	6 (10.9)	0 (0.0)	0 (0.0)	0 (0.0)
Diarrhea	4 (7.3)	1 (1.8)	2 (3.6)	0 (0.0)
Mucositis/stomatitis oral cavity	3 (5.5)	0 (0.0)	2 (3.6)	0 (0.0)
Taste alteration (dysgeusia)	3 (5.5)	0 (0.0)	3 (5.5)	0 (0.0)
Constitutional symptoms				
Fatigue (asthenia, lethargy and malaise)	14 (25.5)	0 (0.0)	8 (14.5)	0 (0.0)
Weight loss	7 (12.7)	0 (0.0)	0 (0.0)	0 (0.0)
Fever (without neutropenia)	5 (9.1)	0 (0.0)	3 (5.5)	1 (1.8)
Insomnia	3 (5.5)	0 (0.0)	0 (0.0)	0 (0.0)
Skin and subcutaneous tissue disorders				
Alopecia	21 (38.2)	0 (0.0)	20 (36.4)	0 (0.0)
Pain				
Abdomen NOS	10 (18.2)	2 (3.6)	0 (0.0)	0 (0.0)
Joint	5 (9.1)	2 (3.6)	0 (0.0)	0 (0.0)
Back	3 (5.5)	0 (0.0)	0 (0.0)	0 (0.0)
Bone	3 (5.5)	1 (1.8)	0 (0.0)	0 (0.0)
Extremity – limb	3 (5.5)	0 (0.0)	0 (0.0)	0 (0.0)
Muscle	3 (5.5)	2 (3.6)	0 (0.0)	0 (0.0)
Chest/thorax NOS	3 (5.5)	1 (1.8)	0 (0.0)	0 (0.0)
Throat/pharynx/larynx	3 (5.5)	0 (0.0)	0 (0.0)	0 (0.0)
Hepatobiliary disorders				
Bilirubin (hyperbilirubinemia)	9 (16.4)	1 (1.8)	0 (0.0)	0 (0.0)

AEs occurring in at least 5% of patients (liver, breast or prostate cancer) treated with lyso-thermosensitive liposomal doxorubicin using targeted application of heat, either by radiofrequency ablation or microwave (as of 1 April 2008).
AE: Adverse event; AGC: Absolute granulocyte count; ALT: Alanine aminotransferase; ANC: Absolute neutrophil count; AST: Aspartate aminotransferase; NOS: Not otherwise specified; WBC: White blood cell.
Data taken from [Celsion Corp., Unpublished Data].

Table 2. Frequency listing of adverse events while on lyso-thermosensitive liposomal doxorubicin.

AE	Lyso-thermosensitive liposomal doxorubicin (n = 55)			
	Any AE, n (%)	*Grade 3 or more AE, n (%)*	*Drug-related AE, n (%)*	*Serious AE, n (%)*
Pulmonary/upper respiratory disorders				
Dyspnea (shortness of breath)	7 (12.7)	0 (0.0)	0 (0.0)	0 (0.0)
Infections				
Urinary tract NOS	5 (9.1)	1 (1.8)	1 (1.8)	1 (1.8)
Hepatobiliary infection	3 (5.5)	2 (3.6)	1 (1.8)	0 (0.0)
Renal/genitourinary disorders				
Urinary retention	5 (9.1)	0 (0.0)	0 (0.0)	1 (1.8)
Bladder spasms	3 (5.5)	0 (0.0)	2 (3.6)	0 (0.0)
Incontinence, urinary	3 (5.5)	0 (0.0)	0 (0.0)	0 (0.0)
Hemorrhage				
Urinary NOS	5 (9.1)	0 (0.0)	1 (1.8)	0 (0.0)
General cardiac symptoms				
Decreased ejection fraction	5 (9.1)	0 (0.0)	5 (9.1)	0 (0.0)
Hypertension	4 (7.3)	0 (0.0)	0 (0.0)	0 (0.0)
Hypotension	4 (7.3)	0 (0.0)	0 (0.0)	0 (0.0)
Musculoskeletal symptoms				
Muscle weakness – generalized	3 (5.5)	0 (0.0)	1 (1.8)	0 (0.0)
Lymphatic disorders				
Edema – limb	4 (7.3)	0 (0.0)	0 (0.0)	0 (0.0)
Neurological disorders				
Mood alteration: depression	5 (9.1)	0 (0.0)	0 (0.0)	0 (0.0)
Dizziness	4 (7.3)	0 (0.0)	1 (1.8)	0 (0.0)

AEs occurring in at least 5% of patients (liver, breast or prostate cancer) treated with lyso-thermosensitive liposomal doxorubicin using targeted application of heat, either by radiofrequency ablation or microwave (as of 1 April 2008).
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Data taken from [Celsion Corp., Unpublished Data].

ejection fraction decreases were drug-related but none were serious or grade 3+ [Celsion Corp., Unpublished Data]. These drug-related events are consistent with the adverse event profile of systemic doxorubicin. As noted earlier, with RFA plus LTLD therapy, high concentrations of doxorubicin are deposited in tumors, but some is released to circulate as free doxorubicin.

Comparison with other agents for HCC tumors >3 cm

The clinical benefit of the RFA plus LTLD combination is hypothesized to be an increased chance of cure of medium/large HCC lesions. The clinical benefit of the standard-of-care treatments for medium/large HCC (TACE or sorafenib) is limited to only some increase in survival. Currently, the role of RFA alone as a curative treatment for HCC >3 cm remains uncertain because of high recurrence rates. By adding LTLD, there is a potential to reduce recurrence and enhance the curative efficacy of RFA for HCC >3 cm, but this has to be corroborated by the data from the ongoing Phase III trial.

All of the proposed RFA-based combination therapies to increase the HCC cure rate are hypothesized to have a mainly additive efficacy. The RFA plus LTLD combination is hypothesized to have a synergistic efficacy, since both the tumor specificity and the antitumor activity of LTLD are enhanced by the heat of RFA.

Conclusion & future perspective

As LTLD is a liposome, it rapidly concentrates in the liver, where it permeates HCC lesions and their vasculature. The heat of RFA furthers this process and very quickly releases doxorubicin in the heated area. At the same time, hyperthermia increases the cytotoxicity of doxorubicin, producing a synergistic interaction.

If its curative and synergistic potential is borne out in the Phase III HEAT study, a rational future strategy for HCC lesions >3 cm is to

employ RFA plus LTLD as a front-line therapy. TACE, sorafenib or another molecularly targeted agent, or one of the other investigational RFA-based combination therapies, can be used as second-line or third-line treatments, if needed.

Plans to study combinations of LTLD with other thermal ablative modalities such as high-intensity focused ultrasound are being contemplated. The RFA plus LTLD combination also has great potential as a front-line therapy for colorectal liver metastases, but this requires a separate clinical trial.

Financial & competing interests disclosure

N Borys is the Chief Medical Officer for Celsion Corp. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

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Executive summary

Mechanism of action

- Lyso-thermosensitive liposomal doxorubicin (LTLD; ThermoDox® [Celsion Corp., MD, USA]) is designed for use with radiofrequency ablation (RFA) and consists of the heat-enhanced cytotoxic anthracycline antibiotic doxorubicin within a heat-activated liposome.
- LTLD is administered intravenously but, because it is a liposome, rapidly concentrates in the liver and spleen. Since tumors have much higher microvascular permeability than normal tissues, LTLD accumulates in liver tumors.
- When LTLD is heated by RFA, a high concentration of doxorubicin is released into the tumor and tumor margins.
- Doxorubicin's cytotoxic mechanism of action and its ability to bind to DNA and inhibit nucleic acid synthesis is enhanced by hyperthermia.

Pharmacokinetic properties

- Approximately 95% of the liposomal doxorubicin plasma area under the curve occurs during the first 6 h following infusion, establishing this time period as optimal for application of RFA. The plasma half-life (mean ± standard deviation) is 18.52 ± 8.36 h.

Clinical efficacy

- A statistically significant LTLD dose–response effect for time to treatment failure was found in Phase I.

Safety & tolerability

- Dosage and administration:
  - Adult: 50 mg/m^2 LTLD infused intravenously over 30 min, with RFA initiated within 15 min of starting the infusion. All RFA procedures should be completed within 3 h.
  - Pediatric: The safety and efficacy of LTLD has not been studied in children.
- Precautions:
  - Administer steroids and multiple histamine receptor blockade beginning 24 h before LTLD to prevent acute infusion reaction.
- Side effects:
  - Hematological: neutropenia is the dose-limiting toxicity; leukopenia, lymphopenia, anemia and thrombocytopenia have been reported.
  - Cardiovascular: decreased ejection fraction affects <10% of subjects; unlike free (nonliposomal) doxorubicin, congestive heart failure is not reported.
  - Gastrointestinal: nausea affects approximately 10% of subjects.
  - Dermatological: alopecia affects approximately one out of three subjects; unlike some other liposomal doxorubicin formulations, hand–foot syndrome is not reported.
  - Other: fatigue affects approximately 15% of patients.

Regulatory affairs

- LTLD is an investigational drug now in Phase III study for treatment of hepatocellular carcinoma lesions >3.0 cm in size.

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