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Clinical Trial Results

NGR-hTNF and Doxorubicin as Second-Line Treatment of Patients with Small Cell Lung Cancer

Vanesa Gregorc,[a] Raffaele Cavina,[b] Sivia Novello,[c] Francesco Grossi,[d] Chiara Lazzari,[a] Enrica Capelletto,[c] Carlo Genova,[d] Giulia Salini,[e] Antonio Lambiase,[e] Armando Santoro[b]

[a]Department of Oncology, IRCCS San Raffaele Hospital, Milan, Italy; [b]Department of Oncology, Humanitas Cancer Center, Rozzano, Italy; [c]Department of Oncology, University of Turin, AOU San Luigi, Orbassano, Italy; [d]UOS Tumori Polmonari, Ospedale Policlinico San Martino, Genova, Italy; [e]MolMed, Milan, Italy

Trial Information

- **ClinicalTrials.gov Identifier:** NCT00483509
- **Sponsor(s):** MolMed
- **Principal Investigator:** Vanesa Gregorc
- **IRB Approved:** Yes

Lessons Learned

- NGR-hTNF was safely combined with doxorubicin, showing a promising antitumor activity in unselected patients with relapsed small cell lung cancer.
- Similar antitumor activity was observed in platinum-sensitive and platinum-resistant patient cohorts.

Abstract

Background. Relapsed small cell lung cancer (SCLC) patients have limited treatment options and poor outcomes. NGR-hTNF is a vascular-targeting agent, which increases intratumoral chemotherapy penetration and T-lymphocyte infiltration.

Methods. Twenty-eight patients relapsing after at least one platinum-based regimen with a treatment-free interval shorter ($n = 16$; platinum-resistant) or longer ($n = 12$; platinum-sensitive) than 3 months received NGR-hTNF 0.8 μg/m^2 plus doxorubicin 75 mg/m^2 every 3 weeks. The primary endpoint of this single-arm phase II trial was progression-free survival (PFS), and safety, response rate, and survival were secondary endpoints.

Results. The most common grade 3–4 toxicities were neutropenia (53%) and anemia (21%). Median PFS was 3.2 months for all patients, 2.7 months for platinum-resistant patients, and 4.1 months for platinum-sensitive patients. Seven patients had partial responses (25%), including four (25%) with platinum-resistant and three (25%) with platinum-sensitive relapse. Mean changes from baseline in tumor burden (after two, four, and six cycles) did not differ between platinum-resistant (−9%, −29%, and −32%) and platinum-sensitive (−11%, −20%, and −43%) cohorts. Overall survival was associated only with baseline lymphocyte counts, with median survival times of 13.1 and 5.2 months for lymphocyte counts above or below the median, respectively.

Conclusion. NGR-hTNF plus doxorubicin showed manageable toxicity and promising activity in patients with relapsed SCLC. ***The Oncologist*** 2018;23:1133–e112

Discussion

SCLC is characterized by high response rates to first-line platinum/etoposide-based chemotherapy. However, despite initial chemosensitivity, nearly all patients eventually experience relapse, which has historically been classified as platinum-resistant or platinum-sensitive according to a treatment-free interval shorter or longer than 3 months [1,2]. Salvage chemotherapy with topotecan yielded modest survival improvements in relapsed SCLC [3,4].

Since its discovery, tumor necrosis factor alpha (TNF) has shown powerful antitumor activity, but its early-stage development was hampered by severe toxicities, the maximum tolerated dose being 10-fold lower than the estimated effective dose. To increase the therapeutic index, NGR-hTNF was developed by conjugating TNF with the tumor-homing peptide NGR (asparagine-glycine-arginine), which selectively binds a CD13 isoform expressed by newly formed tumor blood vessels [5,6].

In preclinical models, NGR-TNF was 10-fold more active than untargeted TNF, with activity mostly noticed at low doses. Furthermore, a sequence- and time-dependent synergism between NGR-hTNF and chemotherapy was observed

Correspondence: Vanesa Gregorc, M.D., Department of Oncology, IRCCS San Raffaele Hospital, Via Olgettina 60, Milan, Italy. Telephone: +39-3397299898; e-mail: vanesa.gregorc@hsr.it Received February 7, 2018; accepted for publication April 27, 2018; published Online First on August 3, 2018. http://dx.doi.org/10.1634/theoncologist.2018-0292

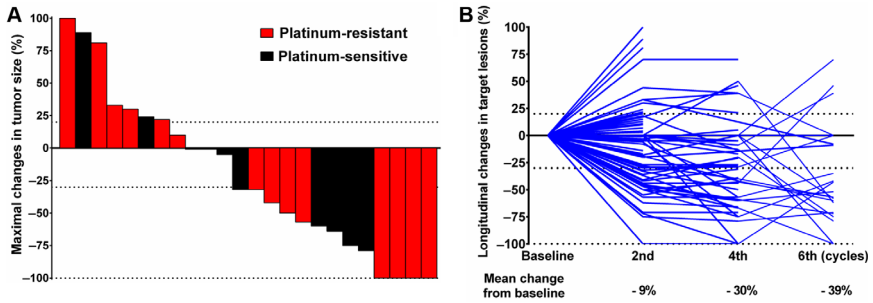


Figure 1. Waterfall plot of maximum post-treatment decrease in the longest sum of tumor diameter (**A**; *n* = 24 patients) and on-treatment changes in all target lesions (**B**; *n* = 79 target lesions).

when the former was administered 2 hours before the latter [7,8]. Phase I trials selected NGR-hTNF 0.8 μg/m^2 as the optimal dose in monotherapy [9] and in combination with doxorubicin [10], based on dynamic imaging changes, soluble TNF-receptors kinetics, and tolerability. In this single-arm phase II trial, the addition of NGR-hTNF 0.8 μg/m^2 to doxorubicin 75 mg/m^2 was associated with a manageable toxicity profile and similar antitumor activity in platinum-resistant and platinum-sensitive patients. Median progression-free survival was 3.2 months (95% confidence interval [CI] 1.8–4.7; 28 events) for all patients (*n* = 28), 2.7 months (1.8–3.6) for the platinum-resistant cohort (*n* = 16), and 4.1 months (2.4–5.8) for the platinum-sensitive cohort (*n* = 12). By radiologic tumor assessment, seven patients had partial response (PR; 25%; 95% CI 11–45) and eight stable disease (SD; 29%), for an overall disease control rate of 54% (95% CI 34–73). Among patients with PR and SD, median progression-free times were 6.3 months (range 2.7–7.9) and 4.6 months (range 3.2–7.0), respectively. There were four PR (25%) and four SD (25%) in the platinum-resistant cohort and three PR (25%) and four SD (33%) in the platinum-sensitive cohort. Maximum percentage change in target lesion burden for individual patients are plotted in Figure 1A and on-treatment changes for all target lesions in Figure 1B. Reductions in tumor burden from baseline were noted in 14 (58%) of 24 patients who had at least one postbaseline assessment.

Consistently with NGR-hTNF and doxorubicin synergism shown in immunocompetent mice but not in nude mice [11], this study showed an association between overall survival and baseline lymphocyte count, with median survival of 13.1 months and 5.2 months in patients with counts above or below median (1.2/mL), respectively.

In this regard, the drug ability to increase the intratumoral T-cell infiltration [12], a prerequisite for response to immune checkpoint blockade [13], should facilitate the combination of NGR-hTNF with immune checkpoint inhibitors, to be evaluated in a randomized phase II setting.

Trial Information	
Disease	Lung cancer—SCLC
Stage of Disease/Treatment	Metastatic/advanced
Prior Therapy	One prior regimen
Type of Study - 1	Phase II
Type of Study - 2	Single arm
Primary Endpoint	Progression-free survival
Secondary Endpoint	Overall response rate
Secondary Endpoint	Overall survival
Secondary Endpoint	Safety
Additional Details of Endpoints or Study Design	
According to a two-stage Simon's optimal trial design (p0 = 35%, p1 = 60%, α = 10%, and β = 10%), the planned sample size was 27 patients, with 16 to be enrolled in the first stage. Study treatment was considered worthy of additional testing if 7 and 13 patients were progression free at 18 weeks after first and second stages, respectively.	
Investigator's Analysis	Active and should be pursued further

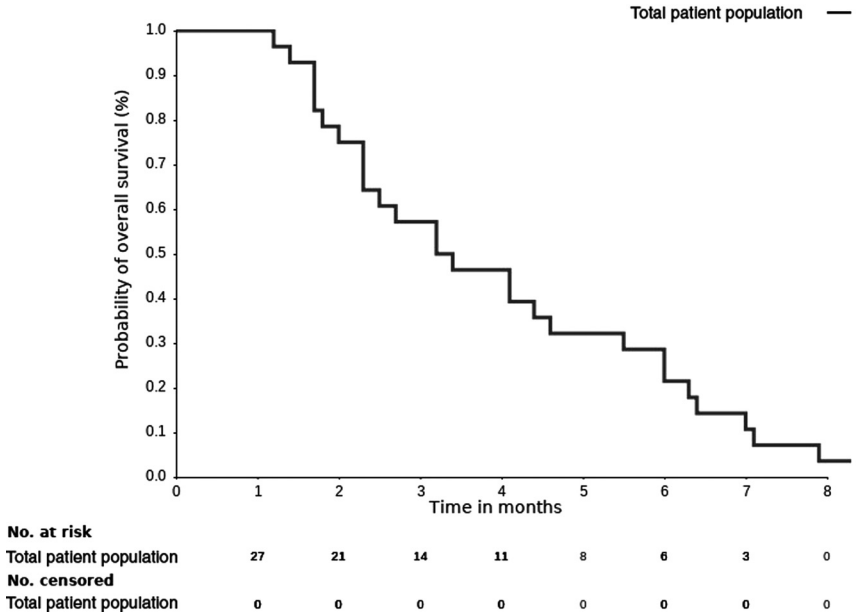
Drug Information	
Drug 1	
Generic/Working Name	NGR-hTNF
Trade Name	Zafiride
Company Name	MolMed
Drug Type	Biological
Drug Class	Angiogenesis—antivascular

Dose	0.8 mcg/m^2
Route	IV
Schedule of Administration	Every 3 weeks until progressive disease
Drug 2	
Generic/Working Name	Doxorubicin
Trade Name	Adriblastine
Company Name	Pfizer
Drug Type	Other
Drug Class	Anthracycline
Dose	75 mg/m^2
Route	IV
Schedule of Administration	Every 3 weeks up to 550 mg/m^2

Patient Characteristics		
Number of Patients, Male	19	
Number of Patients, Female	9	
Stage	Metastatic or advanced	
Age	Median (range): 63 (41–76)	
Number of Prior Systemic Therapies	Median (range): 1 (1–3)	
Performance Status: ECOG	0 — 14 1 — 12 2 — 2 3 — Unknown —	
Other	**Best response to prior therapy, n (%)**	
	Complete response (CR)	2 (7%)
	Partial response (PR)	11 (39%)
	Stable disease (SD)	6 (21%)
	Progressive disease (PD)	9 (32%)
	Treatment-free interval, months	
	Median	2.9
	Range	0.4–10.3

Primary Assessment Method	
Number of Patients Screened	28
Number of Patients Enrolled	28
Number of Patients Evaluable for Toxicity	28
Number of Patients Evaluated for Efficacy	28
Evaluation Method	RECIST 1.0
Response Assessment CR	*n* = 0 (0%)
Response Assessment PR	*n* = 7 (25%)
Response Assessment SD	*n* = 8 (29%)
Response Assessment PD	*n* = 10 (36%)
Response Assessment OTHER	*n* = 3 (11%)
(Median) Duration Assessments PFS	3.2 months, CI: 1.8–4.7
(Median) Duration Assessments OS	5.6 months, CI: 5.3–5.9
(Median) Duration Assessments Response Duration	6.3 months

Kaplan-Meier (Time Units: Months)



Time of scheduled assessment and/or time of event	No. progressed (or deaths)	No. censored	Percent at start of evaluation period	Kaplan-Meier %	No. at next evaluation/No. at risk
1.2	1	0	100.00	96.43	27
1.4	1	0	96.43	92.86	26
1.7	3	0	92.86	82.14	23
1.8	1	0	82.14	78.57	22
2.0	1	0	78.57	75.00	21
2.3	3	0	75.00	64.29	18
2.5	1	0	64.29	60.71	17
2.7	1	0	60.71	57.14	16
3.2	2	0	57.14	50.00	14
3.4	1	0	50.00	46.43	13
4.1	2	0	46.43	39.29	11
4.4	1	0	39.29	35.71	10
4.6	1	0	35.71	32.14	9
5.5	1	0	32.14	28.57	8
6	2	0	28.57	21.43	6
6.3	1	0	21.43	17.86	5
6.4	1	0	17.86	14.29	4
7	1	0	14.29	10.71	3
7.1	1	0	10.71	7.14	2
7.9	1	0	7.14	3.57	1
8.4	1	0	3.57	0.00	0

Adverse Events					
Adverse event	**Grade 1 *n* (%)**	**Grade 2 *n* (%)**	**Grade 3 *n* (%)**	**Grade 4 *n* (%)**	**All grades *n* (%)**
Neutropenia	—	2 (7)	2 (7)	13 (46)	17 (61)
Anemia	2 (7)	8 (29)	5 (18)	1 (3)	16 (57)
Chills	11 (39)	4 (14)	—	—	15 (54)
Fatigue	1 (3)	7 (25)	5 (18)	—	13 (46)
Nausea	7 (25)	5 (18)	—	—	12 (43)
Lymphopenia	2 (7)	6 (21)	4 (14)	—	12 (43)
Thrombocytopenia	—	5 (18)	4 (14)	1 (3)	10 (36)
Appetite loss	5 (18)	2 (7)	3 (11)	—	10 (36)
Pyrexia	8 (29)	—	—	—	8 (29)
Mucositis	3 (11)	2 (7)	1 (3)	—	6 (21)
Dyspnea	2 (7)	2 (7)	1 (3)	—	5 (18)
Vomiting	3 (11)	2 (7)	—	—	5 (18)
Constipation	3 (11)	2 (7)	—	—	5 (18)
Cough	3 (11)	1 (3)	—	—	4 (14)
Diarrhea	4 (14)	—	—	—	4 (14)
Alopecia	1 (3)	2 (7)	—	—	3 (11)
Conjunctivitis	2 (7)	1 (3)	—	—	3 (11)
Sinus tachycardia	3 (11)	—	—	—	3 (11)
Dry skin	3 (11)	—	—	—	3 (11)
Feeling cold	3 (11)	—	—	—	3 (11)
Urinary infection	3 (11)	—	—	—	3 (11)

Study-emergent adverse events in ≥10% of cases of safety population ($n = 28$), irrespective of treatment relationship, classified by preferred term and worst grade per patient.

Abbreviation: —, no data.

Assessment, Analysis, and Discussion	
Completion	Study completed
Investigator's Assessment	Active and should be pursued further

Patients eligible for this multicenter, single-arm, phase II trial were aged 18 years or older and had to have pathologically proven small cell lung cancer (SCLC), radiologically documented disease progression after at least one platinum/etoposide-based regimen, a performance status (PS) of 0–2, adequate bone marrow, hepatic and renal function, and measurable disease according to RECIST (version 1.0). A 4-week washout period for both radiotherapy and chemotherapy and 2 weeks for surgery were required before treatment initiation. Exclusion criteria included active brain metastases, significant cardiac dysfunction, including a left ventricular ejection fraction less than 55%, uncontrolled hypertension, and serious systemic disease or infection.

The primary study endpoint was progression-free survival, defined as the time from baseline to disease progression or death, whichever occurred first. Secondary endpoints included response rate, defined as the proportion of patients with complete response (CR) or partial response (PR), with radiologic assessments done at baseline and every other cycle (6 weeks); disease control rate, defined as the percentage of patients who had a best response of CR or PR or stable disease (SD); overall survival, defined as the time from baseline to death; and evaluation of toxicity using the Common Terminology Criteria for Adverse Events (version 3.0).

NGR-hTNF 0.8 μg/m^2 was given intravenously as a 1-hour infusion followed by doxorubicin 75 mg/m^2 as a 15-minute intravenous infusion 2 hours after NGR-hTNF dosing. Maximum cumulative doxorubicin dose was capped at 550 mg/m^2, whereas NGR-hTNF was continued until disease progression or intolerable toxicity occurred. For retreatment on next cycle, all the reported toxicities had to be recovered to grade 1 or resolved. For patients unable to meet retreatment criteria, a 1–3-week delay for both drugs was allowed. No formal dose reduction for NGR-hTNF was planned. If chills occurred during NGR-hTNF infusion, premedication with paracetamol was recommended for subsequent cycles. Doxorubicin dose modifications were applied according to the summary of product characteristics.

All analyses were based on the intent-to-treat principle. Time-to-event outcome variables were estimated by the Kaplan-Meier method. Exploratory Cox regression models assessed associations between overall survival and baseline characteristics, including age (> vs. ≤ median), sex, PS (0 vs. 1–

2), number of prior regimens (1 vs. 2–3), best response to prior therapy (CR/PR vs. SD/PD), treatment-free interval (> vs. ≤3 months), and baseline lymphocyte count (> vs. ≤ median).

Among 28 enrolled patients, treatment discontinuations resulted from PD in 22 (79%), symptomatic deterioration in 5 (18%), and toxicity in 1 (3%; grade 3 anemia and mucositis after three cycles). All patients received at least one dose of study drugs and were assessable for toxicity. In total, 114 cycles of NGR-hTNF (median 3; range 1–10) and 102 cycles of doxorubicin (median 3; range 1–7) were administered. Thirteen patients (46%) received at least four cycles and nine patients (32%) at least six courses. All treatment cycles were given at the planned dose, except for six patients (21%) requiring doxorubicin dose reductions for hematologic toxicity over 13 cycles (13%). Five patients (18%) discontinued doxorubicin when the lifetime cumulative dose (550 mg/m^2) was reached.

Treatment-emergent adverse events, regardless of cause, were reported by 27 patients (96%). Grade 3–4 neutropenia occurred in 15 patients (53%), febrile neutropenia in 2 patients (7%), anemia in 6 patients (21%), and thrombocytopenia in 5 patients (18%). Six patients (21%) had cardiac events (atrial hypertrophy, atrial fibrillation, sinus tachycardia, bundle branch block, pericardial effusion, and mitral valve incompetence). A minority of all the adverse events (27 of 349; 8%) were considered related to NGR-hTNF infusion, being mostly represented by transient, mild-to-moderate chills in 15 patients (53%). No serious adverse events related to NGR-hTNF were reported.

After the first study stage ($n = 16$), seven (44%) patients were alive and progression free at 18 weeks. An additional 11 patients were enrolled in the second study stage (total $n = 27$), with 13 (48%) being alive and progression free at 18 weeks. Median progression-free survival was 3.2 months for all patients ($n = 28$). Seven patients had PR (25%) and eight SD (29%). Response rates were similar in patients pretreated with one (25%) or more than one (25%) regimen. Among 24 patients with at least an on-treatment assessment, the baseline target lesion burden was significantly greater in platinum-resistant than in platinum-sensitive patients (mean 13.1 vs. 6.8 cm, respectively). After two, four, and six cycles, mean percentage changes from baseline in target lesion burden did not differ between platinum-resistant (−9%, −29%, and −32%, respectively) and platinum-sensitive (−11%, −20%, and −43%, respectively) cohorts.

The 1-year overall survival rate was 30% (95% confidence interval 13–47; 27 events) for all patients, 27% (5–50) for platinum-resistant and 33% (7–60) for platinum-sensitive cohorts. By multivariate Cox regression analyses, the baseline lymphocyte count was the only factor significantly associated with overall survival. In patients with baseline lymphocyte counts higher or lower than the median value (1.2/mL), median overall survival times were 13.1 months versus 5.2 months among all patients, 15.7 months versus 5.2 months in platinum-resistant patients, and 5.6 months versus 4.6 months in platinum-sensitive patients, respectively.

In conclusion, this single-arm phase II trial showed a safe toxicity profile and promising activity of NGR-hTNF and doxorubicin combination in unselected patients with relapsed SCLC. Overall results seem promising, especially considering that more than half of cases presented with platinum-resistant relapse and one third with multiple prior treatment lines. The 1-year survival rate appears in line with that reported in relapsed SCLC with topotecan [3,4] or immune checkpoint inhibitors [14,15]. Indeed, a phase I/II trial testing immune checkpoint blockade with the programmed cell death protein 1 inhibitor nivolumab alone or combined with two doses of the cytotoxic T-lymphocyte-associated antigen 4 inhibitor ipilimumab showed response and 1-year survival rates of 10% and 33% in the monotherapy cohort and 19%–23% and 35%–43% in the combination cohorts, respectively [14]. Another early-stage trial with the programmed death-ligand 1 inhibitor pembrolizumab reported response and 1-year survival rates of 33% and 38%, respectively [15]. However, a phase III study of ipilimumab with or without chemotherapy among newly diagnosed SCLC patients was negative [16].

Finally, the mechanism of action of NGR-hTNF, which increases the intratumoral lymphocyte infiltration [12], might facilitate its combination with immune checkpoint inhibitors, which require high levels of tumor-infiltrating lymphocytes [13], with benefit to be assessed in a randomized phase II setting.

Disclosures

Giulia Salini: MolMed (E); **Antonio Lambiase:** MolMed (E).
The other authors indicated no financial relationships.

(C/A) Consulting/advisory relationship; (RF) Research funding; (E) Employment; (ET) Expert testimony; (H) Honoraria received; (OI) Ownership interests; (IP) Intellectual property rights/inventor/patent holder; (SAB) Scientific advisory board

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