OPEN

Safety and Immunogenicity of MAGE-A3 Cancer Immunotherapeutic with or without Adjuvant Chemotherapy in Patients with Resected Stage IB to III *MAGE-A3*-Positive Non–Small-Cell Lung Cancer

Jean-Louis Pujol, MD, PhD, Johan F. Vansteenkiste, MD, PhD,† Tommaso Martino De Pas, MD,‡ Djordje Atanackovic, MD,§ Martin Reck, MD, PhD,¶ Michiel Thomeer, MD, PhD,‖ Jean-Yves Douillard, MD, PhD,# Gianpiero Fasola, MD,** Vanessa Potter, MD, PhD,†† Paul Taylor, MD,‡‡ Lionel Bosquée, MD,§§ Robert Scheubel, MD,¶¶ Silvija Jarnjak, MD,‖‖ Muriel Debois, MSc,‖‖ Pedro de Sousa Alves, PhD,‖‖ Jamila Louahed, PhD,‖‖ Vincent G. Brichard, MD, PhD,‖‖ and Frédéric F. Lehmann, MD, PhD‖‖*

Introduction: To assess the safety and immunogenicity of MAGE-A3 immunotherapeutic in patients with stage IB–III *MAGE-A3*-positive non–small-cell lung cancer (NSCLC) who were or were not undergoing standard cisplatin/vinorelbine chemotherapy.

Methods: This open, prospective, multicenter, parallel-group phase I study (NCT00455572) enrolled patients with resected (cohorts 1–3) or unresectable (cohort 4) *MAGE-A3*-positive NSCLC. MAGE-A3 immunotherapeutic (300 μg recombinant MAGE-A3 formulated with AS15) was administered (eight doses, 3 weeks apart) concurrent with (cohort 1), after (cohort 2), or without (cohort 3) standard-adjuvant chemotherapy, or after standard radiotherapy and/or chemotherapy (cohort 4).

Results: Sixty-seven patients received greater than or equal to 1 dose of MAGE-A3 immunotherapeutic. Grade 3/4 adverse events (AEs) were reported for 16 out of 19 (84%), 2 out of 18 (11%), 5 out of 18 (28%), and 1 out of 12 (8%) patients in cohorts 1, 2, 3, and 4, respectively. Many grade 3/4 AEs in cohort 1 (e.g., neutropenia) were typical of chemotherapy. Six patients, including three in cohort 1, reported study treatment–related grade 3/4 AEs (injection-site reactions or musculoskeletal/back pain, which resolved within 5 days). One patient (in cohort 4) died, but this and the other serious adverse events were not study treatment related. MAGE-A3-specific antibody responses to immunotherapy were induced in all patients evaluated in all cohorts. MAGE-A3-specific $CD4^+$ T-cell responses

*Thoracic Oncology Unit, Arnaud de Villeneuve Hospital/CHRU Montpellier, Montpellier, France; †Department of Pneumology, University Hospital, KU Leuven, Leuven, Belgium; ‡Medical Oncology, European Institute of Oncology, Milan, Italy; §Division of Hematology and Hematologic Malignancies, University of Utah Huntsman Cancer Institute, Salt Lake City, Utah; ¶Department of Thoracic Oncology, Hospital Grosshansdorf, Grosshansdorf, Germany; ‖Department of Respiratory Medicine, Ziekenhuis Oost Limburg, Genk and LCRP Oncology Cluster, University of Hasselt, Hasselt, Belgium; #Department of Medical Oncologie, ICO R. Gauducheau, St-Herblain, France; **Department of Oncology, University Hospital S. Maria della misericordia, Udine, Italy; ††Nottingham University Hospitals NHS Trust, Nottingham, United Kingdom; ‡‡Northwest Lung Centre, University Hospitals of South Manchester, Manchester, United Kingdom; §§Thoracic Oncology-Pneumology Service, André Renard Clinic, Herstal, Belgium; ¶¶Clinic of Thoracic Surgery, Waldburg-Zeil Clinic, Wangen im Allgäu, Germany; and ‖‖GSK Vaccines, Rixensart, Belgium.

Disclosure: All authors participated in the design, or implementation, or analysis and interpretation of the study results and in the development of this manuscript. All authors had full access to the data and gave final approval before submission. The corresponding author was responsible for submission of the publication. All authors completed the ICMJE Form for Disclosure of Potential Conflicts of Interest and declared that the following interests are relevant to the submitted work. Dr. Debois, Dr. Jarnjak, Dr. de Sousa Alves, Dr. Louahed, Dr. Brichard, and Dr. Lehmann were employees of GSK group of companies at the time of the study. Dr. Jarnjak, Dr. de Sousa Alves, Dr. Louahed, Dr. Brichard, and Dr. Lehmann report ownership of GSK stock. Dr. Vansteenkiste reports his institution (KUL, Leuven, Belgium) receiving payments from GSK for consultancy. Dr. Reck reports receiving payments from AstraZeneca, Boerhinger-Ingelheim, Bristol-Myers Squibb, Hoffman-La-Roche, Lilly, MSD, Novartis, and Pfizer for consultancy or lectures. Dr. Douillard reports being a board member at GSK, AstraZeneca, and Boerhinger-Ingelheim, reports receiving payments from GSK for travel and other study-related purposes, and for consultancy, and reports receiving payments from AstraZeneca and Boerhinger-Ingelheim for lectures and consultancy. Dr. Fasola reports being a board member at Amgen and reports receiving payments from GSK, AstraZeneca, and Novartis for lectures. Dr. Potter reports receiving payments from GSK for consultancy. Dr. Taylor reports receiving payments from GSK for travel related to the study. Dr. Bosquée reports his institution (ULB, Brussels, Belgium) receiving payments from GSK for consultancy. Dr. Pujol, Dr. De Pas, Dr. Atanackovic, Dr. Thomeer, and Dr. Scheubel report no conflicts of interest.

GlaxoSmithKline Biologicals SA sponsored the clinical trial and covered the costs associated with the development and publishing of the manuscript, including scientific writing assistance.

Address for correspondence: Pr Jean-Louis Pujol, Respiratory Illnesses Service, Arnaud de Villeneuve Hospital/CHRU Montpellier, Avenue du Doyen Giraud, 34295 Montpellier CEDEX, France. E-mail: jl-pujol@chu-montpellier.fr.

DOI: 10.1097/JTO.0000000000000653

Copyright © 2015 by the International Association for the Study of Lung Cancer. This is an open-access article distributed under the terms of the Creative Commons Attribution-Non Commercial-No Derivatives License 4.0 (CCBY-NC-ND), where it is permissible to download and share the work provided it is properly cited. The work cannot be changed in any way or used commercially.

ISSN: 1556-0864/15/1010-1458

to immunotherapy were detected in 4 out of 11 (36%), 4 out of 15 (27%), 2 out of 8 (25%), and 5 out of 6 (83%) evaluated patients in cohorts 1, 2, 3, and 4, respectively; and $CD8^+$ T-cell responses were only detected in four patients.

Conclusion: In resected and unresectable NSCLC patients and irrespective of whether standard chemotherapy was concurrent or not, MAGE-A3 immunotherapeutic is well tolerated and induces MAGE-A3-specific immune responses.

KeyWords: Adjuvant chemotherapy, Immunotherapy, Immunostimulant, MAGE-A3, Non–small cell lung carcinoma, Vaccine

(*J Thorac Oncol.* 2015;10: 1458–1467)

Non–small-cell lung cancer (NSCLC) represents approximately 85% of all lung cancer cases.[1,2] Complete resection is the mainstay of treatment for early (stage I and II) and some locally advanced (stage IIIA) NSCLC. The major limitation remains recurrence of disease, with 5-year survival ranging between 73% (stage IA) and 24% (stage IIIA).[3]

Chemotherapy with cisplatin (CDDP)-based doublets is the standard treatment after surgery for stage IIA–IIIA NSCLC with an absolute 5-year survival benefit of 5.4%.[4,5] The absolute 5-year survival benefit for CDDP and vinorelbine (VNR) chemotherapy has been estimated at greater than or equal to 8.6%.[6,7] However, the lung adjuvant cisplatin evaluation (LACE) meta-analysis did not identify a survival benefit for CDDP-based adjuvant chemotherapy in stage IB disease,[5] although a survival benefit was suggested for patients with large tumors in the CALGB9633 study.[8] Hence, the efficacy of adjuvant chemotherapy remains limited and only for selected patients. For unresectable stage III NSCLC, platinum-based chemotherapy combined with thoracic radiotherapy remains a recommended treatment and provides marginal 5-year survival benefit over radiotherapy alone.[9–11] Therefore, even with these advances, new approaches are required to improve the prognosis for NSCLC patients.

Cancer immunotherapy represents a different approach and encompasses a broad range of strategies to induce or boost immune-mediated tumor-cell destruction (active immunotherapy), or to counteract mechanisms by which tumor cells evade or suppress immune-mediated destruction (passive immunotherapy).[12–16] Several different strategies have entered late-stage clinical development,[12,16,17] including active immunotherapies[18–23] and immune-checkpoint inhibitors.[24–26] Immune-checkpoint inhibitors include the anticytotoxic T lymphocyte-associated antigen (CTLA-4), and programmed death receptor-1 (PD-1), and anti-PD-L1 compounds. These compounds target tumor-mediated inhibition of cytotoxic T-cell activity; preliminary evidence for these compounds being administered as monotherapies or in combination with chemotherapy or targeted agents has revealed promising activity with manageable toxicity.[27,28]

MAGE-A3 cancer immunotherapeutic is an active immunotherapy that has been evaluated in NSCLC and melanoma patients.[29–32] It contains recombinant full-length MAGE-A3 protein formulated with the immunostimulant AS15. MAGE-A3 is a cancer/testis antigen[33] and is considered to be tumor-specific because the only normal cells (spermatogonia and trophoblasts) in which it is expressed are unable to present MAGE-A3 epitopes because of the absence of class I and II human leukocyte antigens.[34–38] *MAGE-A3* expression has been identified in 24% to 45% of NSCLCs (stages I–IV) and seems relatively more prevalent in squamous cell carcinomas than in other histological types.[36,39,40]

The intention of combining active immunotherapy with radiotherapy and/or chemotherapy would be to provide additive or synergistic clinical benefits without jeopardizing schedule and dosage of standard chemotherapies or radiotherapies. Moreover, the earlier administration of active immunotherapy, that is, concurrent with rather than after chemotherapy, would permit the earlier development of an immune response. Theoretically, chemotherapy or radiotherapy may increase the susceptibility of residual tumor cells to immune-mediated attack by inhibiting regulatory-T cells or by invoking stress or immunogenic cell death responses in the same tumor cells and resulting in the exposure/release of tumor-associated antigens and the potential for tumor-antigen spreading.[41–45] Conversely, chemotherapy, and corticosteroids often used concurrently as antiemetics, may detrimentally affect the response to active immunotherapy through their potentially immunosuppressive side-effects.[46] Therefore, the objective of this phase I study was to evaluate, in patients with *MAGE A3*-positive NSCLC, the safety and immunogenicity (MAGE-A3-specific T-cell and antibody responses) of MAGE-A3 immunotherapeutic in different treatment settings that do or do not include chemotherapy. Two of the four treatment settings were also evaluated in the concurrently performed MAGE-A3 as Adjuvant Non-Small Cell LunG CanceR ImmunoTherapy (MAGRIT) trial initiated in October 2007.[31]

PATIENTS AND METHODS

Study Design

The clinical study (NCT00455572) was an open, non-randomized phase I/II study conducted at 18 centers in five European countries (Belgium, France, Germany, Italy, and United Kingdom) and Canada. The study was conducted in accordance with good clinical practice guidelines and all applicable regulatory requirements, including the Declaration of Helsinki (1996). The protocol was approved by the review boards at all participating institutions, and all patients gave written informed consent.

Eligible patients had pathological stage IB, II, or III (but not III/N2–3[47]) NSCLC, which was *MAGE A3*-positive (determined by reverse-transcriptase polymerase chain reaction [PCR] on paraffin-embedded primary tumor sample; Supplementary Methods, Supplemental Digital Content 1, http://links.lww.com/JTO/A890). Patients had adequate bone-marrow reserve and renal and hepatic function (Supplementary Methods, Supplemental Digital Content 1, http://links.lww.com/JTO/A890). In patients with resected tumors, resection was anatomical, involving at least a lobe and with a level of lymph node sampling corresponding to the standard procedures at the center (Supplementary Methods, Supplemental Digital Content 1, http://links.lww.com/JTO/A890). Patients who had concomitant or previous malignancies at other sites (unless the malignancy had been effectively treated), history of anaphylaxis or severe allergic reaction; concurrent severe medical problems,

Copyright © 2015 by the International Association for the Study of Lung Cancer

psychiatric or addictive disorders, and patients who required concomitant treatment with systemic corticosteroids, or any other immunosuppressive agents were not eligible (Supplementary Methods, Supplemental Digital Content 1, http://links.lww.com/JTO/A890, for other standard eligibility criteria).

Patients with resected tumors were entered into cohort 1, standard CDDP/VNR chemotherapy concurrently delivered with MAGE-A3 immunotherapeutic; cohort 2, chemotherapy (≥2 cycles) before MAGE-A3 immunotherapeutic; or cohort 3, MAGE-A3 immunotherapeutic only. Patients with unresectable stage III tumors were entered into cohort 4, chemotherapy (≥2 cycles) and radiotherapy before MAGE-A3 immunotherapeutic. Patients in cohort 1 or 2 had Eastern Cooperative Oncology Group scores less than or equal to 1 at screening, whereas patients in cohort 3 or 4 had Eastern Cooperative Oncology Group scores less than or equal to 2 at screening. Patients in cohort 4 had stable disease or objective responses (confirmed by computed tomography scan) after standard radiotherapy/chemotherapy.

Study Treatment

The treatment plan included eight doses (3 weeks apart) of MAGE-A3 immunotherapeutic (the study treatment; GSK Vaccines, Rixensart, Belgium; Fig. 1*A*). The first dose was within 4 to 12 weeks after resection (cohorts 1 and 3), 2 to 4 weeks after adjuvant chemotherapy (cohort 2), or 2 to 6 weeks after radiotherapy/chemotherapy (cohort 4). In cohort 1, the first four doses of MAGE-A3 immunotherapeutic and chemotherapy were administered concurrently, with MAGE-A3 immunotherapeutic being administered on day 8 of each chemotherapy cycle. In cohort 2, MAGE-A3 immunotherapeutic was administered 2 to 4 weeks after completion of all chemotherapy cycles. Chemotherapy consisted of four consecutive cycles, at 3-week intervals: CDDP (80 mg/m^2) on day 1; VNR (30 mg/m^2) on days 1 and 8 (Supplementary Methods, Supplemental Digital Content 1, http://links.lww.com/JTO/A890, for dose modification rules for chemotherapy). MAGE-A3 immunotherapeutic was injected intramuscularly (one dose = 0.5 ml) in the deltoid or lateral region of the thigh, alternating left and right side for subsequent doses. MAGE-A3 immunotherapeutic contained recombinant MAGE-A3 protein (300 μg)[48] and GSK's proprietary immunostimulant, AS15 (Supplementary Methods, Supplemental Digital Content 1, http://links.lww.com/JTO/A890. for the preparation of MAGE-A3 immunotherapeutic for injection).

Endpoints

The safety endpoints were the occurrence of adverse events (AEs), including abnormal hematological and biochemical laboratory values and potential immune-mediated disorders, and serious adverse events (SAEs), primarily from the first dose to 30 days after the last dose of the MAGE-A3 immunotherapeutic. The immunogenicity endpoints were MAGE-A3-specific antibody seropositivity and T-cell responses to treatment primarily after the fourth dose.

Safety Assessment

All AEs including SAEs were evaluated by the investigator and were categorized according to Medical Dictionary for Regulatory Activities (MedDRA) terminology (version 12.0; MedDRA MSSO, McLean, VA). Each AE/SAE was graded for intensity according to the International Common Terminology Criteria for Adverse Events (CTCAE; version 3.0; National Cancer Institute, Bethesda, MD) and was assessed for its relationship with the MAGE-A3 immunotherapeutic treatment. SAEs were identified in accordance with the standard definition (Supplementary Methods, Supplemental Digital Content 1, http://links.lww.com/JTO/A890).

Immunogenicity Assessments

MAGE-A3-specific antibody concentrations were determined using an adapted enzyme-linked immunosorbent assay methodology[32] with reference to a standard curve of positive control samples (values are reported as enzyme-linked immunosorbent assay units per milliliter). Peroxidase-linked anti-human IgG (CAPPEL Inc., PA) was used to detect antibodies bound to recombinant MAGE-A3 antigen. Seropositivity was defined as an antibody concentration greater than or equal to 27 enzyme-linked immunosorbent assay units per milliliter, and a humoral response to treatment defined by seroconversion or by greater than or equal to twofold increase in concentration compared with pretreatment. MAGE-A3-specific CD4$^+$ and CD8$^+$ T-cell responses were assessed by in vitro peptide-stimulation of peripheral blood mononuclear cells (PBMCs) using intracellular-cytokine staining and flow cytometry. Frozen stocks of PBMCs were prepared from blood samples using routine procedures. For each sample, freshly thawed PBMCs were seeded (2×10^5 cells/round-bottom well, in a 96-well plate) into 24 wells and incubated in culture medium (Roswell Park Memorial Institute medium plus 10% horse serum) supplemented with interleukin-2 (10 IU/ml) and interleukin-7 (20 ng/ml) and containing a peptide pool (15 mers with 10 amino-acid overlaps) spanning the entire sequence of MAGE-A3 protein at 1 μg/ml/peptide. A predefined cutoff of less than 60% viability invalidated a thawed PBMC sample for further analysis. Each well was split in two on day 7, pooled back down to 12 wells on day 13. On day 14, each well was split in two, and 12 wells were incubated for 6 hours with the culture medium containing the peptide pool (test), and the other 12 duplicate wells were incubated with the culture medium without the peptide pool (control); with Brefeldin A being added after 2-hour incubation. Intracellular cytokine staining was performed with the following antibodies anti-CD3^{PercP}, anti-CD4$^{PE\text{-}cy7}$, anti-CD8$^{APC\text{-}cy7}$, anti-interferon (anti-IFN)-γ^{FITC}, and anti-tumor necrosis factor^{PE} (anti-TNF^{PE}), and fluorescence-activated cell sorting was then used to detect and enumerate CD4$^+$ or CD8$^+$ T cells that were also IFN-γ^+ TNF-α^+ (at least 3000 T cells were required to validate the well). A ratio of the percentages of IFN-γ^+ TNF-α^+ (CD4$^+$ or CD8$^+$) T cells/all (CD4$^+$ or CD8$^+$) T cells in the peptide-stimulated (test) well versus the peptide-unstimulated (control) well was calculated for each pair of duplicate wells. A T-cell immunogenicity score was determined as the geometric mean of the ratios in the pairs of duplicate wells. A PBMC sample was valid when at least 7 of 12 of these ratios were used to determine the T-cell immunogenicity score. A MAGE-A3-specific (CD4$^+$ or CD8$^+$) T-cell response was identified if the patient's T-cell immunogenicity score was above the cut-off score (CD4$^+$ = 8.4; or CD8$^+$ = 3.2). The cut-off scores were

Copyright © 2015 by the International Association for the Study of Lung Cancer

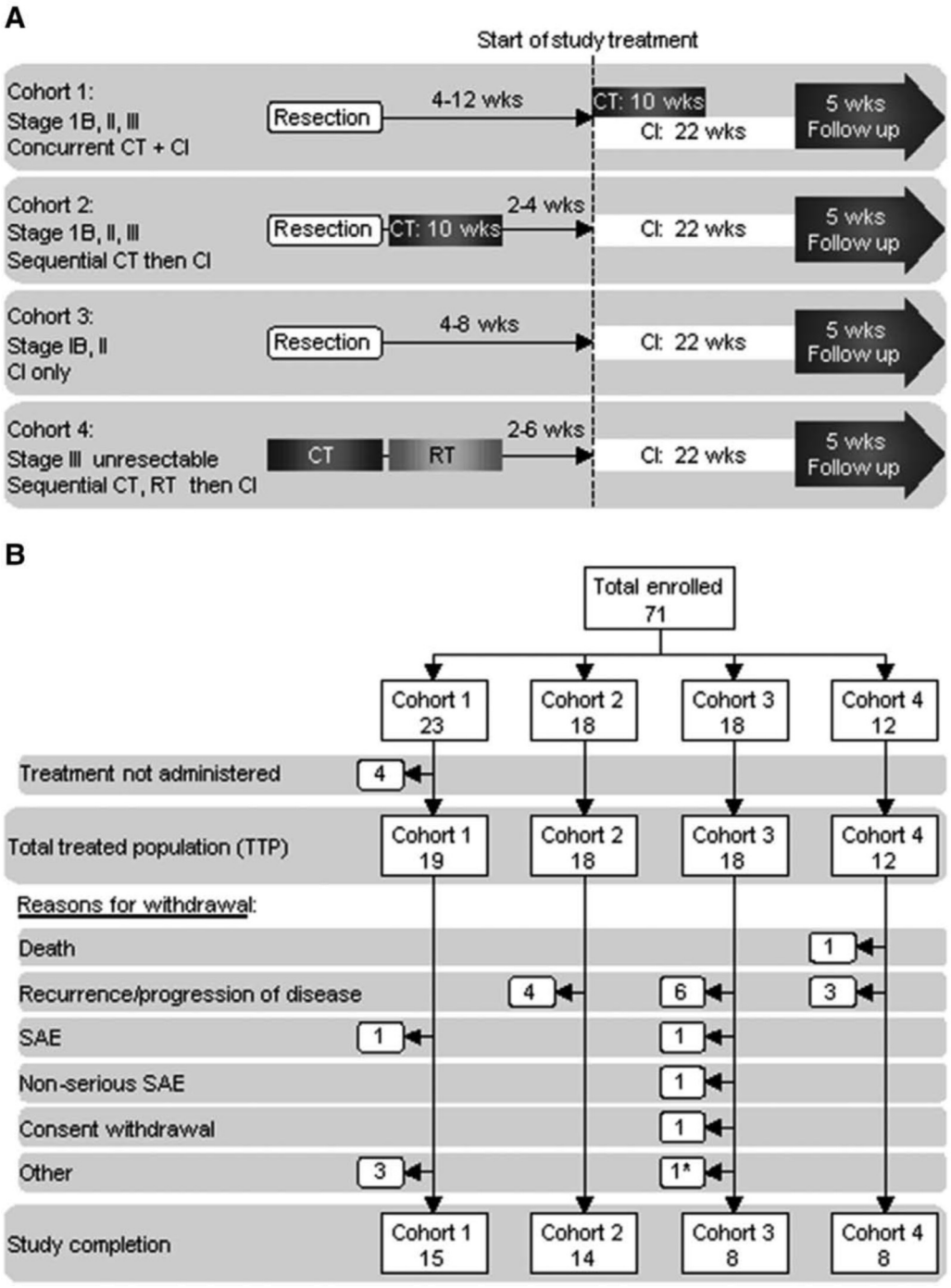

FIGURE 1. A, Study design. B, The allocation and participation of patients during the course of the study. A, The treatment plan included eight doses (3 weeks apart) of MAGE-A3 cancer immunotherapeutic (CI). In cohort 1, CI was integrated into the chemotherapy (CT) treatment schedule such that the first four doses of CI were administered on the eighth day of each of the four chemotherapy cycles. B, All patients in the total treated population (TTP) were included for the analyses of safety. The reasons for withdrawal from the study are described to the left. *Note that this one patient in cohort 3 received the full eight-dose regimen but did not attend the concluding visit. Note that the three "other" reasons for withdrawal in cohort 1 included reduced compliance, the diagnosis of a different type of cancer, and a comorbidity related to a suspected cytochrome polymorphism. Cohort 1, concurrent chemotherapy + immunotherapy; cohort 2, sequential chemotherapy then immunotherapy; Cohort 3, immunotherapy only; Cohort 4 (unresectable non–small-cell lung cancer), sequential chemotherapy, radiotherapy, then immunotherapy.

Copyright © 2015 by the International Association for the Study of Lung Cancer

derived using the same methodology but with PBMCs from 24 healthy Caucasian donors; and calculated as the upper 95th percentile of 24 T-cell immunogenicity scores. A MAGE-A3-specific T-cell response to treatment was identified if the T-cell immunogenicity score postdose 4 or postdose 8 was above the cutoff and was greater than or equal to fourfold higher than pretreatment.

Statistics

This was the first study of MAGE-A3 immunotherapeutic in a target population of MAGE A3-positive NSCLC patients who were receiving, or had recently received, chemotherapy after tumor resection. The sample size was, therefore, typical of studies of this kind and was based on general experience rather than any formal estimate or hypothesis. It was anticipated that out of 18 patients enrolled in a cohort, 15 patients would be evaluated for immunogenicity. Appropriate descriptive statistics were applied to demographic and other baseline characteristics. SAS (version 9.2, SAS Institute Inc. Cary, NC) was used for all computations.

RESULTS

Patients and Treatment

From May 2007 to February 2013, 428 patients were screened. Of those patients with available and valid tumor-PCR results, 33% (106/320) had *MAGE-A3*-positive tumors. From those 106 patients, 71 were enrolled in cohorts 1, 2, 3, and 4 (23, 18, 18, and 12, respectively; Fig. 1). Sixty-seven patients received at least one dose of MAGE-A3 immunotherapeutic (19, 18, 18, and 12 in cohorts 1, 2, 3, and 4, respectively) and were included in the total treated population, in which the study endpoints were evaluated.

Patients were aged from 37 to 81 years (median 61 years; Table 1). The majority of patients were male (78%). As expected, more patients in cohort 3 (12/18) had stage IB tumors than in cohorts 1 (3/19) and 2 (3/18).

All patients in cohorts 2 and 4 received at least two cycles of prior chemotherapy. All patients in cohort 4 except one received prior radiotherapy. The full eight doses of MAGE-A3 immunotherapeutic were received by 46 out of 67 (69%) patients. These included 15 out of 19 (79%), 14 out of 18 (78%), 9 out of 18 (50%), and 8 out of 12 (68%) patients in cohorts 1, 2, 3, and 4, respectively (Fig. 1). In cohort 1, 16 out of 19 (84%) patients received at least two cycles of concomitant chemotherapy and for those who completed the study, 9 out of 15 (60%) received the full 80 mg/m^2 doses of CDDP, and 1 out of 15 (7%) received carboplatin instead of CDDP. The largest dose reduction of CDDP was by 25% to 60 mg/m^2, which occurred in 2 out of 15 (13%) patients.

The most frequent reason for early discontinuation from the study was the recurrence of disease, and this occurred in cohort 2 (4/18; 22%), cohort 3 (6/18; 33%), and cohort 4 (3/12; 25%), but not in cohort 1 (Fig. 1B). Other reasons for discontinuations included one patient who died (cohort 4; because of a bronchial hemorrhage); two patients who withdrew because of treatment-unrelated SAEs (cohorts 1 and 3; pneumonia and cardiac failure, respectively); one patient who withdrew because of a nonserious AE (cohort 3; influenza-like illness); one patient who withdrew consent (cohort 3); one patient who was lost to follow-up (cohort 3); and three patients who withdrew for other reasons (cohort 1; Fig. 1).

Safety

All patients in cohorts 1 and 2 and most patients in cohort 3 (17/18; 94%) and cohort 4 (11/12, 92%) reported AEs (Table 2 and Fig. 2). Most patients (67–94%) in each cohort reported AEs in the general disorders and administration site conditions category. In cohort 1, 14 out of 19 (74%) patients reported gastrointestinal disorders (mainly nausea,

TABLE 1. Patient Characteristics

		All	Cohort 1	Cohort 2	Cohort 3	Cohort 4
Enrolled patients		71	23	18	18	12
TTP		67	19	18	18	12
Median age (range), yr		61 (37–81)	59 (37–75)	61 (47–69)	68 (51–81)	62 (48–73)
Sex (%)	Female	15 (22)	5 (26)	3 (17)	2 (11)	5 (42)
	Male	52 (78)	14 (74)	15 (83)	16 (89)	7 (58)
ECOG performance status	0	23	3	8	4	8
	1	43	16	10	13	4
	2	1	0	0	1	0
Stage	Stage IB	18	3	3	12	0
	Stage II (IIA, IIB)	17 (3, 14)	6 (1, 5)	9 (2, 7)	2 (0, 2)	0
	Stage III (IIIA, IIIB)	32 (21, 11)	10 (9, 1)	6 (5, 1)	4 (3, 1)	12 (4, 8)
Histology type	Squamous	29	9	10	9	1
	Nonsquamous	38	10	8	9	11
Type of surgery	Lobectomy	42	13	13	16	0
	Pneumonectomy	13	6	5	2	0

Cohort 1, concurrent chemotherapy + immunotherapy; cohort 2, sequential chemotherapy then immunotherapy; cohort 3, immunotherapy only; cohort 4 (unresectable NSCLC), sequential chemotherapy, radiotherapy, then immunotherapy; NSCLC, non–small-cell lung cancer; TTP, total treated population.

Copyright © 2015 by the International Association for the Study of Lung Cancer

TABLE 2. Summary of Patient Numbers Reporting AEs/SAEs from the First Dose to 30 Days After the Last Dose of the MAGE-A3 Immunotherapeutic

Adverse Events		Cohort 1 (N = 19), n (%)	Cohort 2 (N = 18), n (%)	Cohort 3 (N = 18), n (%)	Cohort 4 (N = 12), n (%)
Any	All grades	19 (100)	18 (100)	17 (94)	11 (92)
	Grade 3	5 (26)	2 (11)	3 (17)	1 (8)
	Grade 4	11 (58)	0	2 (11)	0
	Grade 5	0	0	0	1 (8)
	SAEs[a]	14 (74)	0	4 (22)	3 (25)
Related to CI treatment	All grades	14 (74)	17 (94)	16 (89)	7 (58)
	Grade 3	3 (16)	1 (6)	2 (11)	0
	Grade 4	0	0	0	0
	Grade 5	0	0	0	0
	SAEs	0	0	0	0

[a]Two patients in cohort 1 and one patient in Cohort 3 reported SAEs that occurred during the study period but either before the first MAGE-A3 immunotherapeutic dose or more than 30 days after the last MAGE-A3 immunotherapeutic dose.

AEs, adverse events; SAEs, serious adverse events; CI, cancer immunotherapeutic; cohort 1, concurrent chemotherapy + immunotherapy; cohort 2, sequential chemotherapy then immunotherapy; cohort 3, immunotherapy only; cohort 4 (unresectable NSCLC), sequential chemotherapy, radiotherapy, then immunotherapy; NSCLC, non–small-cell lung cancer.

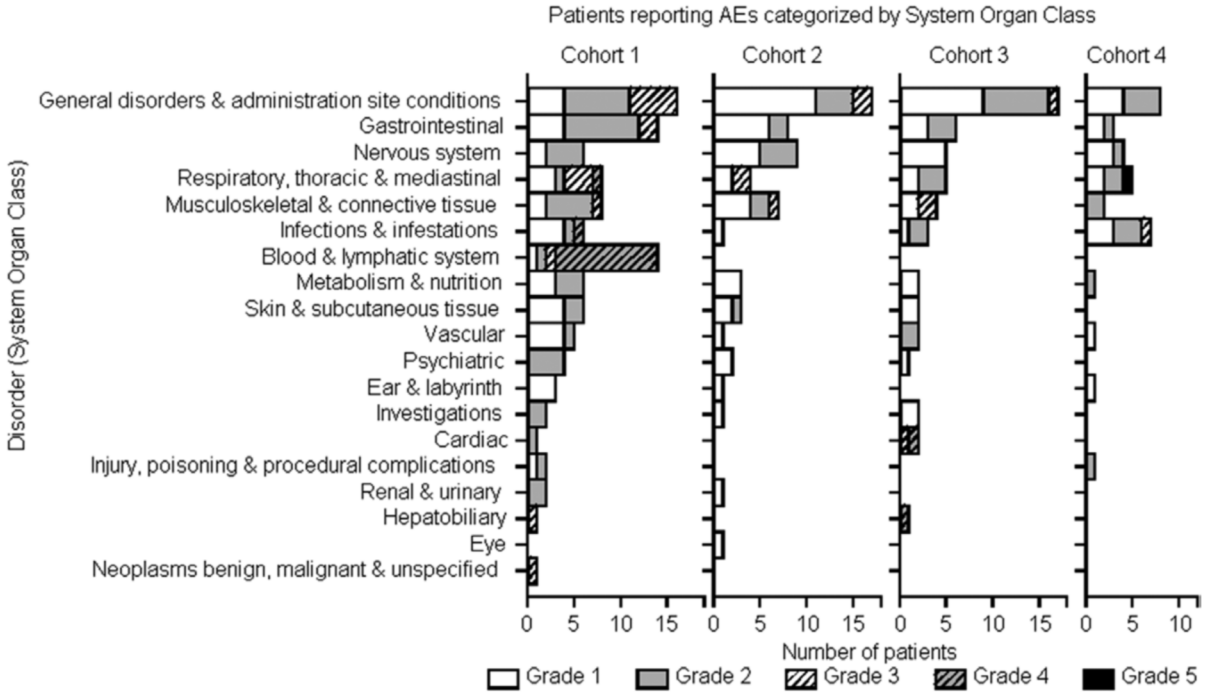

FIGURE 2. The number of patients reporting adverse events (AEs) from the first dose to 30 days after the last dose of the MAGE-A3 immunotherapeutic, categorized by MedDRA System Organ Class, in the total treated population (TTP). For a patient reporting an AE with the same System Organ Class on more than one occasion, only the highest graded AE is indicated. Note that the category general disorders and administration site conditions includes the preferred terms; administration site pain, administration site reaction, asthenia, chills, fatigue, gait disturbance, hypothermia, influenza-like illness, injection-site coldness, injection-site erythema, injection-site hematoma, injection-site inflammation, injection-site oedema, injection-site pain, injection-site pruritus, injection-site rash, injection-site reaction, injection-site swelling, malaise, mucosal inflammation, pyrexia, and sense of oppression. Cohort 1, concurrent chemotherapy + immunotherapy; cohort 2, sequential chemotherapy then immunotherapy; cohort 3, immunotherapy only; cohort 4 (unresectable non–small-cell lung cancer), sequential chemotherapy, radiotherapy, then immunotherapy.

constipation, vomiting, and diarrhea) and 14 out of 19 (74%) patients reported blood and lymphatic disorders (mainly neutropenia and anemia), whereas in cohorts 2, 3, and 4, 8 out of 18 (44%), 6 out of 18 (33%), and 3 out of 12 (25%) patients, respectively, reported gastrointestinal disorders, and no patients reported blood and lymphatic disorders (Fig. 2). Moreover, in cohort 1, most (33/39, 85%) of the gastrointestinal disorders and all (28/28, 100%) of the blood and lymphatic

Copyright © 2015 by the International Association for the Study of Lung Cancer

disorders were reported during the phase when chemotherapy was concomitant with immunotherapy (i.e., up to 7 days after MAGE-A3 immunotherapeutic dose 4) and were typical reactions to chemotherapy.

One grade 5 AE, the fatal bronchial hemorrhage (unrelated to study treatment), occurred 20 days after dose 1 in cohort 4. Grade 3/4 AEs were reported for 16 out of 19 (84%), 2 out of 18 (11%), 5 out of 18 (28%), and 1 out of 12 (8%) patients in cohorts 1, 2, 3, and 4, respectively (Table 2). The most frequent of these grade 3/4 AEs in cohort 1 was neutropenia. No other abnormal laboratory values were reported during the study.

Study treatment–related AEs were graded 1 to 3 (mostly 1 or 2) and reported for 14 out of 19 (74%), 17 out of 18 (94%), 16 out of 18 (89%), and 7 out of 12 (58%) patients in cohort 1, 2, 3, and 4, respectively (Table 2). Most of the general disorders and administration site conditions were study treatment related. Other categories of related AEs (eg, gastrointestinal, nervous system) were only reported for a minority of patients (≤4) in each cohort. The related grade 3 AEs were reported for 3 out of 19 (16%), 1 out of 18 (6%), and 2 out of 18 (11%) patients in cohorts 1, 2 and 3, respectively, and all resolved within 5 days. These AEs were injection-site related or musculoskeletal/back pain. No related grade 3 AEs were reported in cohort 4.

Thirty-six SAEs were reported for 21 patients: 28 SAEs for 14 out of 19 (74%) patients in cohort 1, 5 SAEs for 4 out of 18 (22%) patients in cohort 3, and 3 SAEs, including the fatal one, for 3 out of 12 (25%) patients in cohort 4. No patients reported a SAE in cohort 2. No SAE was study treatment related. Thirty-one SAEs were graded 3 or above.

One potential immune-mediated disorder, a grade 1 Raynaud's phenomenon considered as study treatment unrelated, was reported 19 days postdose 2 for a patient in cohort 1.

Immunogenicity

Immunogenicity results were not obtained from all patients because of missing or invalid samples. Before immunotherapy, 6 out of 65 patients (9%) were seropositive for MAGE-A3-specific antibodies and were in cohort 1 (1/19, 5%), cohort 2 (4/18, 22%), and cohort 4 (1/12, 8%), but not in cohort 3 (0/16). After dose 4, all patients evaluated were seropositive (12, 15, 12, and 9 patients in cohorts 1, 2, 3, and 4 respectively; Fig. 3A) and were humoral responders to immunotherapy.

Before immunotherapy, MAGE-A3-specific $CD4^+$ T-cell responses were detected in 5 out of 49 (10%) patients: 23% (3/13), 6% (1/17) 0% (0/11), and 12% (1/8) patients in cohorts 1, 2, 3, and 4, respectively. MAGE-A3-specific $CD8^+$ T-cell responses were detected in 2 out of 49 patients (5%), both of whom were in cohort 2 (i.e., 2/17, 6%). Out of these seven patients in whom T-cell responses were detected, only one patient was also antibody seropositive.

After dosing, MAGE-A3-specific $CD4^+$ T-cell responses to immunotherapy were induced in 36% (4/11), 27% (4/15), 25% (2/8), and 83% (5/6) patients in cohorts 1, 2, 3, and 4, respectively (Fig. 3B). MAGE-A3-specific $CD8^+$ T-cell responses to immunotherapy were induced in four patients; 1 out of 11 (9%) in cohort 1, 1 out of 15 (7%) in cohort 2, 0 out of 8 (0%) in cohort 3, and 2 out of 6 (33%) in cohort 4.

DISCUSSION

Historically, the potential immunosuppressive activity of chemotherapy, and its use with antiemetic corticosteroids, has been viewed as a barrier for its use in combination with immunotherapy.[46] However, recent evidence has suggested that chemotherapy and/or radiotherapy may promote immune-mediated tumor destruction suggesting potential synergistic actions with immunotherapy.[41,46,49] In this study, the immunogenicity and safety of MAGE-A3 immunotherapeutic treatment in stage IB to III NSCLC patients seemed not to be detrimentally affected by concurrent or prior CDDP/VNR-based chemotherapy or prior radiotherapy, supporting the use of MAGE-A3 immunotherapeutic in the chemotherapy and radiotherapy settings. Although patients were recruited from many centers in Europe and Canada, the strength and generalizability of the conclusions are limited by the study design: a nonrandomized trial of a small patient population without inferential statistics.

The safety profile of the MAGE-A3 immunotherapeutic treatment was in line with what has been observed with previous studies, with mainly grade 1/2 AEs categorized as general disorders and administration site conditions and typical of immunization.[29,30] Grade 3/4 AEs were only frequently observed in cohort 1 and were typical of chemotherapy (eg, neutropenia).[6] The tolerance to chemotherapy in cohort 1 seemed unaffected by the concurrently administered immunotherapy probably reflecting the different modes of action of the two treatments.

Before immunotherapy, a small minority of patients displayed MAGE-A3-specific immune responses (9% antibody, 10% $CD4^+$ T cell, and 5% $CD8^+$ T cell) in line with a previous study where 9% patients were seropositive.[30] Therefore, although the tumors were *MAGE-A3* positive by reverse-transcriptase PCR, this positive expression had not necessarily translated into a spontaneous immune response. No association was identified between seropositivity and cell-mediated immune reactivity. Yet there was the suggestion that the prior chemotherapy or radiotherapy administered in cohorts 2 and 4 had elevated the pretreatment frequency of seropositive patients to 17% (6/30) compared with the other adjuvant-setting cohort (5%, 1/19) and to the detection of MAGE-A3-specific $CD8^+$ T-cell responses in cohort 2.

After four doses of MAGE-A3 immunotherapeutic, MAGE-A3-specific antibody responses to immunotherapy were detected in all patients. Although MAGE-A3-specific antibodies may not directly be involved in eradicating tumor cells, they may contribute; because antibody-mediated opsonization of MAGE-A3 has been found to promote cross-presentation to naïve T cells.[50] This potential mechanism may be relevant if MAGE-A3 is released during chemotherapy-related immunogenic tumor-cell death.[41]

MAGE-A3-specific $CD4^+$ T-cell responses to immunotherapy were detected in 29% (10/34) of patients with resected tumors (cohorts 1–3) and in 83.3% of patients with unresectable tumors (cohort 4). The corresponding $CD8^+$ T-cell responses to immunotherapy were 6% (2/34; cohorts 1–3) and 33% (cohort 4). The higher prevalence and magnitude of these responses in cohort 4 suggested that the presence of tumor tissue combined with its prior exposure to

 Copyright © 2015 by the International Association for the Study of Lung Cancer

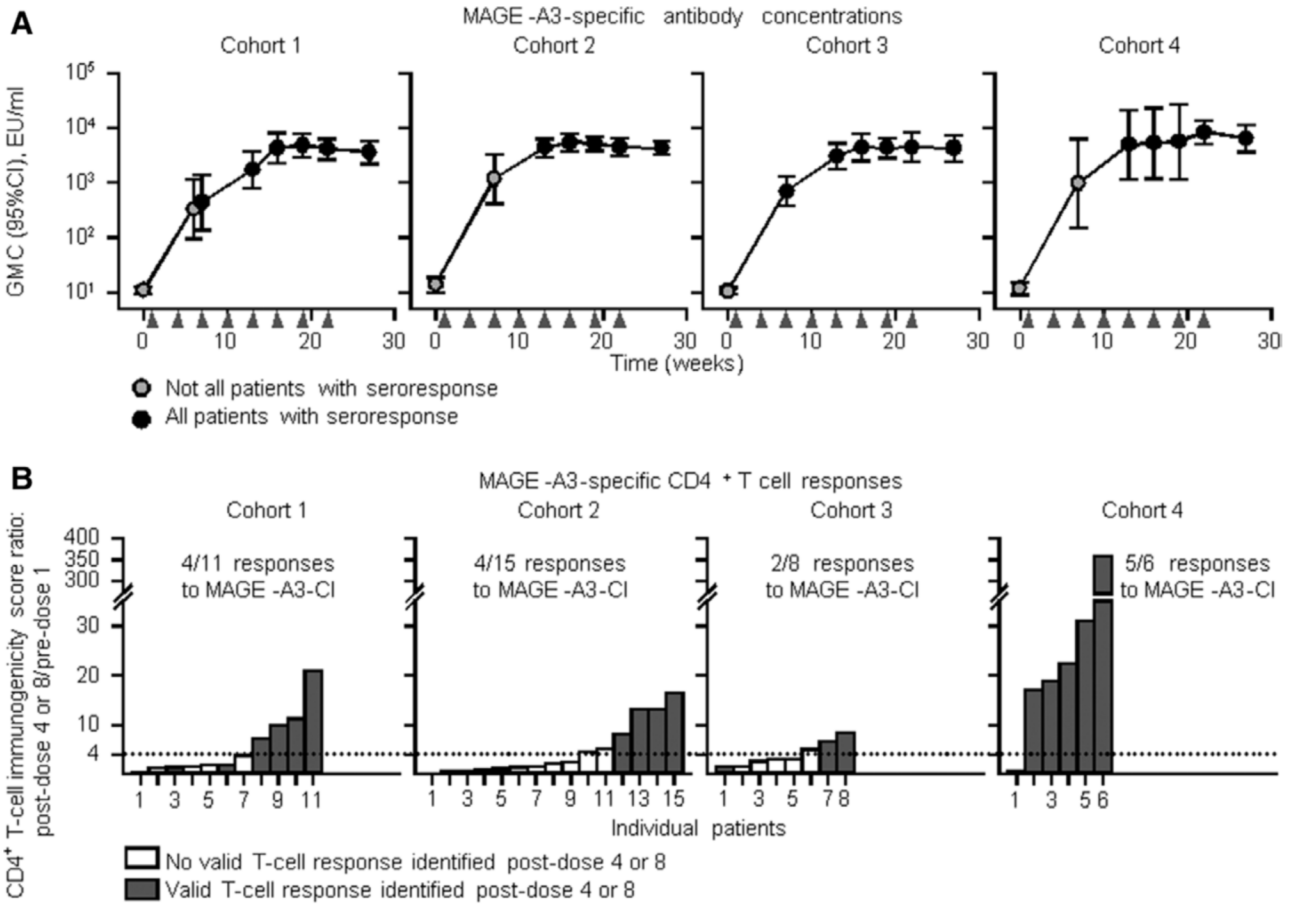

FIGURE 3. MAGE-A3-specific immunogenicity in the total treated population (TTP). A, Geometric mean MAGE-A3-specific antibody concentrations in each cohort, measured in serum samples taken before the first dose (at week 0) of the MAGE-A3 immunotherapeutic through week 30. Gray triangles on the *x*-axis indicate the timings of each of the eight MAGE-A3 immunotherapeutic doses. Error bars describe 95% confidence intervals (95% CIs). At weeks 0, 7, 13, 16, 19, 22, and 27; N = 19, 12, 15, 15, 15, 15, and 15 in cohort 1, respectively; N = 18, 17, 15, 16, 14, 12, and 13 in cohort 2, respectively; and N = 17, 14, 12, 11, 11, 9, and 9, in cohort 3, and N = 12, 10, 9, 8, 8, 8 and 7, in cohort 4, respectively. At week 6 in cohort 1, N = 13. B, MAGE-A3-specific ratios of $CD4^+$ T-cell immunogenicity scores (based on the frequencies of interferon (IFN)-γ^+ tumor necrosis factor (TNF)-α^+ $CD4^+$ T cells in peptide-stimulation cultures of peripheral blood mononuclear cells [PBMCs] in vitro) for individual patients in each cohort, typically calculated as the higher of the two T-cell immunogenicity scores posttreatment divided by the T-cell immunogenicity score pretreatment. The postimmunotherapy scores were evaluated 3 weeks postdose 4 or 5 weeks postdose 8, except for two patients in cohort 1 and one patient in cohort 4. In these patients, the scores were evaluated in PBMC samples taken before the patients' early discontinuation (at least after dose 1). Bars in gray or white describe ratios for samples in which posttreatment MAGE-A3-specific T-cell responses were or were not identified (i.e., T-cell immunogenicity score was above or below the cutoff), respectively. A MAGE-A3-specific $CD4^+$ T-cell response *to* immunotherapy was induced if the T-cell immunogenicity score posttreatment was above the cutoff and was greater than or equal to fourfold higher (dotted line) than the T-cell immunogenicity score pretreatment. Cohort 1, concurrent chemotherapy + immunotherapy; cohort 2, sequential chemotherapy then immunotherapy; cohort 3, immunotherapy only; cohort 4 (unresectable non–small-cell lung cancer), sequential chemotherapy, radiotherapy, then immunotherapy.

radiotherapy/chemotherapy (potentially promoting immunogenic-cell death) may have supported the development of T-cell responses to immunotherapy.[41,43,44]

In the recent placebo-controlled MAGRIT phase III trial,[31] the treatment of stage IB, II, and IIIA NSCLC patients with MAGE-A3 immunotherapeutic in the equivalent adjuvant settings used for cohorts 2 and 3 did not increase disease-free survival. However, the MAGRIT trial was not designed to evaluate cell-mediated immunity. In cohorts 2 and 3 in this study, 4 out of 18 and 6 out of 18 patients had relapsed by the concluding visit, respectively, whereas in cohort 1, no patients had relapsed. Moreover, cohorts 1 and 2 contained more patients with stages II and III tumors than cohort 3. However, given the limitation of the study design, further investigations would be required to evaluate whether the concurrent administration of chemotherapy with immunotherapy resulted in better clinical outcomes. Also given the recent results of MAGRIT and the trials evaluating immune-checkpoint inhibitors, the induction of tumor-specific immune responses may be insufficient for improving clinical outcomes in the absence

Copyright © 2015 by the International Association for the Study of Lung Cancer

of releasing the immune blockade.[24–28] Nevertheless, this study demonstrated that administering active immunotherapy and chemotherapy together is feasible. The earlier development of a tumor-specific immune response should be advantageous in a disease setting where relapse tends to be relatively rapid postsurgery.

ACKNOWLEDGMENTS

The authors thank all the clinical trial participants, the investigators, the nurses, clinicians, laboratory personnel and other staff members for conducting this study, and in particular Dr. Reiner Bonnet, Dr. Elisabeth Quoix, Dr. Yves Pachecot, and Dr. Alberto Sobrero. Matthew Morgan (MG Science Communications, Rixensart, Belgium) provided scientific writing services and Véronique Duquenne (XPE, Pharma and Science, Wavre, Belgium) coordinated the manuscript's development.

REFERENCES

1. Peters S, Adjei AA, Gridelli C, et al. Metastatic non-small-cell lung cancer (NSCLC): ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. *Ann Oncol* 2012;23 (Suppl 7):vii56–vii64.
2. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. *CA Cancer J Clin* 2011;61:69–90.
3. Goldstraw P, Crowley J, Chansky K, et al.; International Association for the Study of Lung Cancer International Staging Committee; Participating Institutions. The IASLC Lung Cancer Staging Project: proposals for the revision of the TNM stage groupings in the forthcoming (seventh) edition of the TNM Classification of malignant tumours. *J Thorac Oncol* 2007;2:706–714.
4. Vansteenkiste J, De Ruysscher D, Eberhardt WE, et al. Early and locally advanced non-small-cell lung cancer (NSCLC): ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. *Ann Oncol* 2013;24 (Suppl 6):vi89–vi98.
5. Pignon JP, Tribodet H, Scagliotti GV, et al.; LACE Collaborative Group. Lung adjuvant cisplatin evaluation: a pooled analysis by the LACE Collaborative Group. *J Clin Oncol* 2008;26:3552–3559.
6. Douillard JY, Rosell R, De Lena M, et al. Adjuvant vinorelbine plus cisplatin versus observation in patients with completely resected stage IB-IIIA non-small-cell lung cancer (Adjuvant Navelbine International Trialist Association [ANITA]): a randomised controlled trial. *Lancet Oncol* 2006;7:719–727.
7. Douillard JY, Tribodet H, Aubert D, et al.; LACE Collaborative Group. Adjuvant cisplatin and vinorelbine for completely resected non-small cell lung cancer: subgroup analysis of the Lung Adjuvant Cisplatin Evaluation. *J Thorac Oncol* 2010;5:220–228.
8. Strauss GM, Herndon JE 2nd, Maddaus MA, et al. Adjuvant paclitaxel plus carboplatin compared with observation in stage IB non-small-cell lung cancer: CALGB 9633 with the Cancer and Leukemia Group B, Radiation Therapy Oncology Group, and North Central Cancer Treatment Group Study Groups. *J Clin Oncol* 2008;26:5043–5051.
9. Sause W, Kolesar P, Taylor S IV, et al. Final results of phase III trial in regionally advanced unresectable non-small cell lung cancer: Radiation Therapy Oncology Group, Eastern Cooperative Oncology Group, and Southwest Oncology Group. *Chest* 2000;117:358–364.
10. Pfister DG, Johnson DH, Azzoli CG, et al.; American Society of Clinical Oncology. American Society of Clinical Oncology treatment of unresectable non-small-cell lung cancer guideline: update 2003. *J Clin Oncol* 2004;22:330–353.
11. Spira A, Ettinger DS. Multidisciplinary management of lung cancer. *N Engl J Med* 2004;350:379–392.
12. Brahmer JR. Harnessing the immune system for the treatment of non-small-cell lung cancer. *J Clin Oncol* 2013;31:1021–1028.
13. Hall RD, Gray JE, Chiappori AA. Beyond the standard of care: a review of novel immunotherapy trials for the treatment of lung cancer. *Cancer Control* 2013;20:22–31.
14. Mellman I, Coukos G, Dranoff G. Cancer immunotherapy comes of age. *Nature* 2011;480:480–489.
15. Chen DS, Mellman I. Oncology meets immunology: the cancer-immunity cycle. *Immunity* 2013;39:1–10.
16. Decoster L, Wauters I, Vansteenkiste JF. Vaccination therapy for non-small-cell lung cancer: review of agents in phase III development. *Ann Oncol* 2012;23:1387–1393.
17. Limacher JM, Spring-Giusti C, Bellon N, Ancian P, Rooke R, Bonnefoy JY. Therapeutic cancer vaccines in the treatment of non-small-cell lung cancer. *Expert Rev Vaccines* 2013;12:263–270.
18. Tyagi P, Mirakhur B. MAGRIT: the largest-ever phase III lung cancer trial aims to establish a novel tumor-specific approach to therapy. *Clin Lung Cancer* 2009;10:371–374.
19. Quoix E, Ramlau R, Westeel V, et al. Therapeutic vaccination with TG4010 and first-line chemotherapy in advanced non-small-cell lung cancer: a controlled phase 2B trial. *Lancet Oncol* 2011;12:1125–1133.
20. Wu YL, Park K, Soo RA, et al. INSPIRE: A phase III study of the BLP25 liposome vaccine (L-BLP25) in Asian patients with unresectable stage III non-small cell lung cancer. *BMC Cancer* 2011;11:430.
21. Butts C, Socinski MA, Mitchell PL, et al.; START trial team. Tecemotide (L-BLP25) versus placebo after chemoradiotherapy for stage III non-small-cell lung cancer (START): a randomised, double-blind, phase 3 trial. *Lancet Oncol* 2014;15:59–68.
22. Nemunaitis J, Dillman RO, Schwarzenberger PO, et al. Phase II study of belagenpumatucel-L, a transforming growth factor beta-2 antisense gene-modified allogeneic tumor cell vaccine in non-small-cell lung cancer. *J Clin Oncol* 2006;24:4721–4730.
23. Neninger Vinageras E, de la Torre A, Osorio Rodríguez M, et al. Phase II randomized controlled trial of an epidermal growth factor vaccine in advanced non-small-cell lung cancer. *J Clin Oncol* 2008;26:1452–1458.
24. Lynch TJ, Bondarenko I, Luft A, et al. Ipilimumab in combination with paclitaxel and carboplatin as first-line treatment in stage IIIB/IV non-small-cell lung cancer: results from a randomized, double-blind, multicenter phase II study. *J Clin Oncol* 2012;30:2046–2054.
25. Brahmer JR, Tykodi SS, Chow LQ, et al. Safety and activity of anti-PD-L1 antibody in patients with advanced cancer. *N Engl J Med* 2012;366:2455–2465.
26. Topalian SL, Hodi FS, Brahmer JR, et al. Safety, activity, and immune correlates of anti-PD-1 antibody in cancer. *N Engl J Med* 2012;366:2443–2454.
27. Pardoll DM. The blockade of immune checkpoints in cancer immunotherapy. *Nat Rev Cancer* 2012;12:252–264.
28. Langer CJ. Emerging immunotherapies in the treatment of non-small cell lung cancer (NSCLC): the role of immune checkpoint inhibitors. *Am J Clin Oncol* 2015;38:422–430.
29. Kruit WH, Suciu S, Dreno B, et al. Selection of immunostimulant AS15 for active immunization with MAGE-A3 protein: results of a randomized phase II study of the European Organisation for Research and Treatment of Cancer Melanoma Group in Metastatic Melanoma. *J Clin Oncol* 2013;31:2413–2420.
30. Vansteenkiste J, Zielinski M, Linder A, et al. Adjuvant MAGE-A3 immunotherapy in resected non-small-cell lung cancer: phase II randomized study results. *J Clin Oncol* 2013;31:2396–2403.
31. Vansteenkiste J, Vanakesa T, De Pas T, et al. MAGRIT, a double-blind, randomized, placebo-controlled Phase III study to assess the efficacy of the recMAGE-A3 + AS15 cancer immunotherapeutic as adjuvant therapy in patients with resected MAGE-A3-positive non-small cell lung cancer (NSCLC). *Ann Oncol* 2014;25:409–416.
32. Vantomme V, Dantinne C, Amrani N, et al. Immunologic analysis of a phase I/II study of vaccination with MAGE-3 protein combined with the AS02B adjuvant in patients with MAGE-3-positive tumors. *J Immunother* 2004;27:124–135.
33. Cheng YH, Wong EW, Cheng CY. Cancer/testis (CT) antigens, carcinogenesis and spermatogenesis. *Spermatogenesis* 2011;1:209–220.
34. Blaschitz A, Hutter H, Dohr G. HLA Class I protein expression in the human placenta. *Early Pregnancy* 2001;5:67–69.
35. Jassim A, Ollier W, Payne A, Biro A, Oliver RT, Festenstein H. Analysis of HLA antigens on germ cells in human semen. *Eur J Immunol* 1989;19:1215–1220.

 Copyright © 2015 by the International Association for the Study of Lung Cancer

36. Gure AO, Chua R, Williamson B, et al. Cancer-testis genes are coordinately expressed and are markers of poor outcome in non-small cell lung cancer. *Clin Cancer Res* 2005;11:8055–8062.
37. Jungbluth AA, Silva WA Jr, Iversen K, et al. Expression of cancer-testis (CT) antigens in placenta. *Cancer Immun* 2007;7:15.
38. Apps R, Murphy SP, Fernando R, Gardner L, Ahad T, Moffett A. Human leucocyte antigen (HLA) expression of primary trophoblast cells and placental cell lines, determined using single antigen beads to characterize allotype specificities of anti-HLA antibodies. *Immunology* 2009;127:26–39.
39. Linder A, Budihardjo-Welim H, Velehorschi W, et al. Prognostic value of MAGE-A3 and PRAME gene expression in non-small-cell lung cancer (NSCLC). *J Clin Oncol* 2012;30:abstr 7056.
40. Shigematsu Y, Hanagiri T, Shiota H, et al. Clinical significance of cancer/testis antigens expression in patients with non-small cell lung cancer. *Lung Cancer* 2010;68:105–110.
41. Zitvogel L, Galluzzi L, Smyth MJ, Kroemer G. Mechanism of action of conventional and targeted anticancer therapies: reinstating immunosurveillance. *Immunity* 2013;39:74–88.
42. McCoy MJ, Lake RA, van der Most RG, Dick IM, Nowak AK. Post-chemotherapy T-cell recovery is a marker of improved survival in patients with advanced thoracic malignancies. *Br J Cancer* 2012;107:1107–1115.
43. Formenti SC, Demaria S. Combining radiotherapy and cancer immunotherapy: a paradigm shift. *J Natl Cancer Inst* 2013;105:256–265.
44. Gelbard A, Garnett CT, Abrams SI, et al. Combination chemotherapy and radiation of human squamous cell carcinoma of the head and neck augments CTL-mediated lysis. *Clin Cancer Res* 2006;12:1897–1905.
45. Corbière V, Chapiro J, Stroobant V, et al. Antigen spreading contributes to MAGE vaccination-induced regression of melanoma metastases. *Cancer Res* 2011;71:1253–1262.
46. Zhang T, Herlyn D. Combination of active specific immunotherapy or adoptive antibody or lymphocyte immunotherapy with chemotherapy in the treatment of cancer. *Cancer Immunol Immunother* 2009;58:475–492.
47. Mountain CF. Revisions in the international system for staging lung cancer. *Chest* 1997;111:1710–1717.
48. Marchand M, Punt CJ, Aamdal S, et al. Immunisation of metastatic cancer patients with MAGE-3 protein combined with adjuvant SBAS-2: a clinical report. *Eur J Cancer* 2003;39:70–77.
49. Baxevanis CN, Perez SA, Papamichail M. Combinatorial treatments including vaccines, chemotherapy and monoclonal antibodies for cancer therapy. *Cancer Immunol Immunother* 2009;58:317–324.
50. Moeller I, Spagnoli GC, Finke J, Veelken H, Houet L. Uptake routes of tumor-antigen MAGE-A3 by dendritic cells determine priming of naïve T-cell subtypes. *Cancer Immunol Immunother* 2012;61:2079–2090.

Copyright © 2015 by the International Association for the Study of Lung Cancer