Lung Cancer 117 (2018) 64–69

Contents lists available at ScienceDirect

Lung Cancer

journal homepage: www.elsevier.com/locate/lungcan

Prognostic role of the VeriStrat test in first line patients with non-small cell lung cancer treated with platinum-based chemotherapy

Francesco Grossi[a], Carlo Genova[a], Erika Rijavec[a], Giulia Barletta[a], Federica Biello[a], Maria Giovanna Dal Bello[a], Krista Meyer[b], Joanna Roder[b], Heinrich Roder[b], Julia Grigorieva[b,*]

[a] Lung Cancer Unit, IRCCS AOU San Martino—IST Istituto Nazionale per la Ricerca sul Cancro, Largo R. Benzi 10, 16132 Genova, Italy
[b] Biodesix, 2970 Wilderness Place, Boulder, CO 80301, USA

ARTICLE INFO

Keywords:
Non-small cell lung cancer
VeriStrat
Chemotherapy
Biomarkers
Prognosis
First line therapy

ABSTRACT

Objectives: VeriStrat® is a blood-based test that utilizes matrix-assisted laser desorption/ionization time-of-flight (MALDI ToF) mass spectrometry to assign a binary classification of VeriStrat Good or VeriStrat Poor that is associated with treatment outcomes in cancer patients. A number of other studies have shown an association between VeriStrat status and clinical outcomes in second and subsequent lines of therapy. The prognostic properties of VeriStrat were demonstrated in the placebo arms of two randomized studies in non-small cell lung cancer (NSCLC): TOPICAL and BR.21; the predictive properties of the test were shown in a prospective randomized phase III study PROSE in the second line treatment of NSCLC with erlotinib versus chemotherapy. Motivated by these observations, we sought to extend the clinical utility of VeriStrat to standard first line chemotherapy and evaluated the performance of the test in a number of clinical studies of patients treated with platinum-based regimens.

Materials and methods: We examine the performance of VeriStrat in three independent clinical trials where the test classification was acquired for prospectively collected baseline samples from 481 patients treated with platinum-based chemotherapy in first line.

Results: Across these trials, 66–70% of patients were classified as VeriStrat Good; patients classified as VeriStrat Good had significantly longer progression-free survival and overall survival than VeriStrat Poor patients, with hazard ratios ranging from 0.36 to 0.72 and 0.26 to 0.51, respectively. These results demonstrated that VeriStrat is a strong prognostic test in NSCLC patients treated with platinum-based regimens in the first line.

Conclusion: VeriStrat provides valuable clinical information that may be used to support patient-physician conversations regarding prognosis and treatment options, and to identify a subset of patients who might benefit from other treatment strategies, possibly in the framework of clinical trials.

1. Introduction

NSCLC is one of the major causes of cancer-related death worldwide and remains a challenge to treat because patients are typically diagnosed at advanced stages where 5-year survival is less than 5% [1]. With the exception of patients whose tumors harbor sensitizing *EGFR* mutations, *ALK* or *ROS-1* translocations, platinum doublets with third generation agents such as taxanes, gemcitabine, vinorelbine and pemetrexed with or without bevacizumab or necitumumab, are the standard of care in the first line setting [2]. Benefit from these treatments varies, with median time to progression around 4–6 months and median survival around 8–14 months, depending on regimen and clinical characteristics of patients [3,4].

The most promising recent advances in the treatment of NSCLC are associated with immunotherapies, especially immune checkpoint inhibitors, which have provided significant improvements in terms of outcomes and durable responses for a proportion of patients in second and subsequent lines of therapy [5]. More recently, checkpoint therapies have shown superiority to standard platinum regimens in treatment-naïve patients with high expression (on at least 50% of tumor cells) of PD-L1 [6]. Most recently, the FDA granted approval for the anti-PD-1 agent pembrolizumab in combination pemetrexed and

Abbreviations: ALK, anaplastic lymphoma kinase; CPH, Cox proportional hazard; EGFR, epidermal growth factor receptor; ERCC1, excision repair cross-complementation group 1 protein; HR, hazard ratio; MALDI ToF, matrix-assisted laser desorption/ionization time-of-flight; NSCLC, non-small cell lung cancer; OS, overall survival; PD-1, programmed cell death protein-1; PD-L1, programmed death-ligand 1; PFS, progression-free survival; PS, performance status; RRM1, ribonucleotide reductase subunit M1; TKI, tyrosine kinase inhibitor

* Corresponding author at: Biodesix, PO Box 774872, Steamboat Springs, CO 80477, USA.
E-mail address: julia.grigorieva@biodesix.com (J. Grigorieva).

https://doi.org/10.1016/j.lungcan.2017.12.007
Received 25 May 2017; Received in revised form 23 September 2017; Accepted 12 December 2017
0169-5002/ © 2017 The Authors. Published by Elsevier Ireland Ltd. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/BY-NC-ND/4.0/).

Table 1
Baseline clinical characteristics of patients by VeriStrat test classification Italian, NExUS, and eLung cohorts.

		Italian			NExUS[a]			eLung		
		VS Good n (%)	VS Poor n (%)	*p* value	VS Good n (%)	VS Poor n (%)	*p* value	VS Good n (%)	VS Poor n (%)	*p* value
Age	Range	44–76	46–80	.264	25–78	35–77	.993	35–86	50–82	.864
		66	66		59	60		66	67	
Gender	Male	34 (68)	17 (65)	1.000	169 (61)	84 (66)	.376	90 (63)	40 (66)	.874
	Female	16 (32)	9 (35)		107 (39)	43 (34)		52 (37)	21 (34)	
Smoking	Never	4 (8)	3 (11)	.685	56 (20)	15 (12)	.048	N/A	N/A	N/A
History	Ever	46 (92)	23 (89)		220 (80)	112 (88)				
Histology	Non-Squamous	50 (100)	26 (100)	–	276 (100)	127 (100)	–	99 (70)	28 (46)	.002
	Squamous	0	0		0	0		43 (30)	33 (54)	
ECOG PS	0	15 (30)	5 (19)	.414	127 (46)	43 (34)	.023	58 (41)	14 (23)	.017
	1	33 (66)	21 (81%)		149 (54)	84 (66)		84 (59%)	47 (77)	
	2	2 (4)	0		0	0		0	0	
Stage	IIIB	0	0	–	30 (11)	15 (12)	.003	7 (5)	1 (2)	.440
	IV	50 (100)	26 (100)		246 (89)	112 (88)		135 (95)	60 (98)	
Treatment	Carbo/Pem	28 (56)	15 (58)	1.00	–	–	–	–	–	–
	Cis/Pem	22(44)	11 (42)							
	Cis/Gem[b]				136 (100)	66 (100)	–			
	Carbo/Pacli/Cet	–	–	–	–	–	–	52 (37)	27 (44)	.205
	Carbo or Cis/Gem/Cet							56 (39)	26 (43)	
	Carbo or Cis/Pem/Cet[c]							34 (24)	8 (13)	

Carbo, carboplatin; Cet, cetuximab; Cis, cisplatin; Gem, gemcitabine; Pacli, paclitaxel; Pem, pemetrexed; N/A, not available; VS, VeriStrat.

[a] Total study population (both treatment arms) that had VeriStrat Good or Poor classifications. Since the studies were randomized, we assume that the proportions in the individual arms are similar to the whole population.

[b] Treatment arm discussed in the paper.

[c] In non-squamous patients.

carboplatin in first line treatment of non-squamous NSCLC, regardless of PD-L1 expression. However, many patients still have little benefit from immunotherapy, moreover, some may experience hyperprogression and be harmed by it [7].

Given these challenges, it appears that standard chemotherapy will remain an important option for many patients who do not harbor "actionable" mutations. Choosing an optimal front-line chemotherapy strategy for NSCLC remains challenging, and many candidate biomarkers, such as thymidylate synthase for pemetrexed-based regimens [8] and excision repair cross-complementation group 1 (ERCC1) protein and ribonucleotide reductase subunit M1 (RRM1) for platinum chemotherapy and gemcitabine, respectively [9–11], have been investigated. However, none of these studies resulted in the development of a validated test for broad clinical use. Challenges to validation and adoption include, among others, insufficient method reproducibility, paucity of validating clinical trials, tumor heterogeneity, and differential biomarker expression in primary and metastatic sites [12], as well as insufficiency of single-molecule biomarkers for representation of complex biological processes.

In contrast, VeriStrat is a true multivariate blood-based test that measures multiple circulating components representative of the complexity of the host-tumor interaction that underlies the significant variability in outcomes in NSCLC patients. The test, initially created for assessing clinical outcome of patients receiving EGFR TKI therapy in NSCLC, utilizes MALDI ToF mass spectrometry to assign VeriStrat Good or VeriStrat Poor classifications to serum or plasma samples [13]. VeriStrat was developed by comparing mass spectra from pre-treatment samples from patients who experienced either long-term stable disease or early progression on gefitinib therapy (reference set) and identifying eight features (i.e. peaks), differentiating these two outcome groups. The algorithm is based on a k-nearest neighbors (kNN) classification scheme, which evaluates mass spectra from a patient's sample with respect to the intensity of eight peaks of those in the reference set. The peaks are comprised of multiple peptides, including serum amyloid A1 (SAA1)[31]; however, the protein content of VeriStrat signature has not been fully identified yet.

VeriStrat has been validated in numerous independent studies which, across various tumor types and treatments, demonstrated that in the majority of treatment regimens patients classified as VeriStrat Good have better outcomes than patients with a VeriStrat Poor classification, raising the question whether VeriStrat is prognostic independent of treatment or predictive for relative benefit of one treatment over another [14–21].

The prognostic role of a test can only be truly assessed in the absence of active treatment, and retrospective analyses of the TOPICAL [22] and NCIC BR.21 [23] studies, comparing efficacy of erlotinib to that of best supportive care (BSC), addressed this question, providing information on the role of VeriStrat in BSC arms. In both trials, in the absence of active treatment VeriStrat Good patients had better outcomes than VeriStrat Poor patients with HRs for OS of 0.54 (95% confidence interval (CI) 0.41–0.73, p-value < .001) in the TOPICAL and 0.44 (95%CI 0.31–0.63, p-value < .001) in the BR.21 studies [19,24]. The predictive property of VeriStrat was confirmed in the prospective phase III PROSE study, which demonstrated that VeriStrat is both prognostic of overall outcomes and predictive of differential benefit from single agent chemotherapy versus erlotinib (p-value = .017 for interaction of treatment with VeriStrat classification in OS) [25].

In this paper we examine the results of three recently reported studies where VeriStrat was applied to samples from advanced NSCLC patients treated with platinum-based chemotherapy in the front line [26–28]. These studies included well-characterized populations of patients recruited in the scope of clinical trials, which were treated with platinum-based chemotherapy, reflecting common clinical practice.

We believe that demonstration of the prognostic role of VeriStrat in the first line setting has significant clinical utility; it may facilitate identification of patients who do not receive meaningful benefit from standard first line chemotherapy regimens, making alternative treatments, including clinical trials, a viable option.

2. Materials and methods

2.1. Trials and participants

Previously presented results for three cohorts of treatment-naïve NSCLC patients (N = 481) receiving platinum-based chemotherapy and

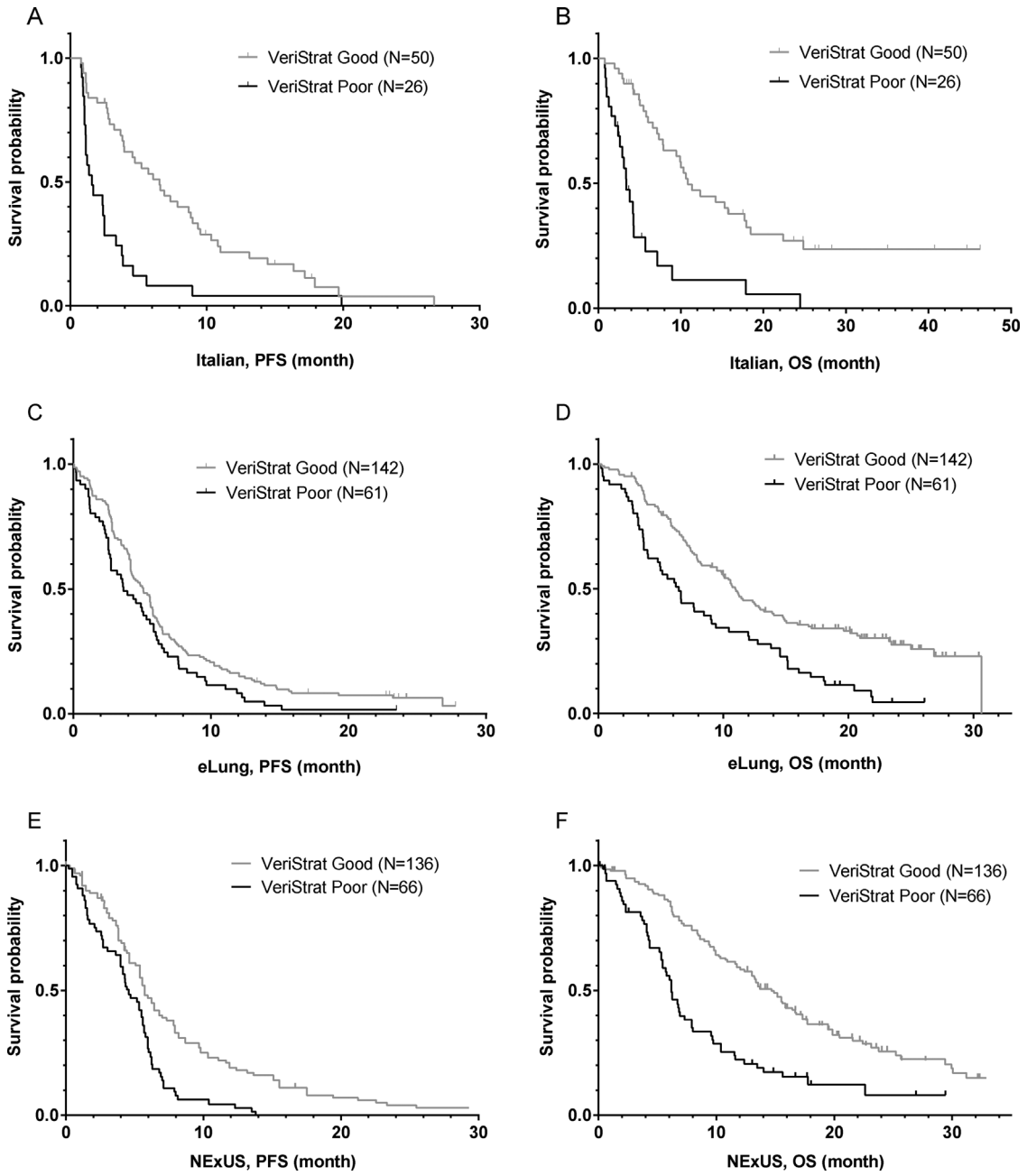

Fig. 1. Comparison of PFS and OS for patients in VS Good and Poor groups: A, B Italian cohort; C, D eLung cohort; E, F NExUS cohort.

for which plasma or serum samples were collected in a pre-planned manner in the frame-work of registered clinical trials were summarized and compared with respect to VeriStrat classification and outcomes.

All patients in the trials provided informed consent and the trials were approved by the ethics committees of the respective institutions.

2.1.1. NExUS cohort

The NExUS trial was a prospective, randomized phase III study of gemcitabine (1250 mg/m^2 on days 1 and 8) plus cisplatin (75 mg/m^2 on day 1) in combination with sorafenib (400 mg twice a day) versus gemcitabine and cisplatin plus placebo, administered until disease progression or unacceptable toxicity. All patients had stage IIIB or IV NSCLC and an ECOG PS of 0 or 1 [29]. Baseline plasma samples from 419 of the 722 non-squamous patients were available for the retrospective VeriStrat analysis. The results of the original VeriStrat analysis for both arms of the trial were presented at the ESMO Congress, 2012 [26]; it was shown that VeriStrat Poor patients benefit significantly more than VeriStrat Good patients, in terms of PFS, from the addition of sorafenib to cisplatin and gemcitabine. In this paper we focus on the role of VeriStrat in the 202 patients treated with cisplatin and gemcitabine, because it is an approved standard therapy for NSCLC in first line.

The trial was registered at ClinicalTrials.gov (NCT00449033).

2.1.2. Italian cohort

The observational Italian study evaluated the role of VeriStrat in first line treatment of 76 non-squamous NSCLC patients with the standard platinum-based combination (carboplatin, AUC 5 or cisplatin, 75 mg/m^2) with pemetrexed (500 mg/m^2) q 21 days, using pre-treatment serum samples [28]. All patients had stage IV disease; the majority of them had ECOG PS of 0 or 1. The choice between carboplatin and cisplatin was made by the treating physician based on creatinine clearance and other clinical characteristics, such as age and comorbidities. After four cycles of chemotherapy, eligible patients could

Table 2
Progression-free and overall survival of NSCLC patients by VeriStrat classification.

		Italian	NExUS	eLung
PFS	Median, month (95% CI)			
	VeriStrat Good	6.5 (3.9–8.8)	5.7 (5.5–6.9)	5.1 (4.2–5.7)
	VeriStrat Poor	1.6 (1.1–2.5)	4.6 (4.1–5.7)	3.6 (2.7–5.3)
	HR (95% CI)	0.36 (0.22–0.61)	0.51 (0.37–0.71)	0.72 (0.53–0.97)
	Log-rank p value	< .001	< .001	.032
OS	Median, month (95% CI)			
	VeriStrat Good	10.8 (7.8–17.7)	14.7 (12.5–16.9)	10.9 (9.5–12.9)
	VeriStrat Poor	3.4 (2.4–4.3)	6.3 (5.6–8.1)	6.4 (4.0–9.0)
	HR (95% CI)	0.26 (0.12–0.55)	0.41 (0.30–0.58)	0.51 (0.37–0.71)
	Log-rank p value	< .001	< .001	< .001

start single-agent maintenance with pemetrexed. The study continues to recruit patients with squamous histology, who are treated with a combination of a platinum agent and gemcitabine.

This trial was registered at ClinicalTrials.gov (NCT02055144).

2.1.3. eLung cohort

In the eLung trial, chemotherapy-naïve advanced NSCLC patients were randomized to one of three regimens of platinum doublets: carboplatin (AUC 6, day 1) plus paclitaxel (200 mg/m^2, day 1), or carboplatin (AUC 6, day 1) or cisplatin (75 mg/m^2) plus gemcitabine (1000 mg/m^2, days 1 and 8), or carboplatin (AUC 6, day 1) or cisplatin (75 mg/m^2) plus pemetrexed (500 mg/m^2, in non-squamous patients) and concurrent cetuximab (loading dose 400 mg/m^2, followed by 250 mg/m^2), q 21 days, administered for four to six cycles, followed by cetuximab maintenance (500 mg/m^2) q 2 weeks; 206 out of 601 of these patients had serum available for testing; 203 received VeriStrat classification. The results for the combined arms were presented at the ASCO congress, 2013 [27].

The trial was registered at ClinicalTrials.gov (NCT00828841).

2.2. Samples and VeriStrat testing

Plasma or serum samples were collected according to the respective trial protocols prior to commencement of treatment and stored frozen. Samples were anonymized and shipped in batches to Biodesix (Boulder, CO).

VeriStrat testing was carried out on available serum or plasma samples by Biodesix (Boulder, CO), blinded to all clinical, treatment, and outcome data. The procedure, equivalent to the commercial VeriStrat test, is described elsewhere [19].

2.3. Statistical analysis

For the Italian and eLung cohorts, the VeriStrat classifications were sent to the respective principal investigators, who unblinded the results and sent the associated clinical and outcome data to Biodesix for statistical analyses after having received test results. Statistical analysis for the NExUS trial was performed by an independent statistician.

Survival analyses were carried out using SAS Enterprise Guide 5.1 (Cary, NC, USA) and GraphPad Prism6 (La Jolla, CA. USA). Differences between groups were assessed using log-rank p values in PRISM and CPH p values in SAS. HRs were calculated using Mantel-Haenszel and CPH models using PRISM and SAS, respectively; p-values for association of categorical variables were calculated by Fisher's exact test, using SAS or PRISM. Kaplan-Meier plots were generated using PRISM. Comparison of age between VeriStrat groups was performed using the unpaired *t*-test.

3. Results and discussion

3.1. Cohort characteristics and comparisons

The cohorts of patients treated with platinum-based chemotherapy had similar proportions of VeriStrat Poor classification (30–34%). The Italian cohort had a lower prevalence of patients with PS = 0 (versus PS > 0) than the NExUS cohort (Fisher's test p-value .010); the differences in PS between Italian and eLung cohorts, and between eLung and NExUS cohorts were not statistically significant (Fisher's test p-values .156 and .115, respectively). All patients in the Italian cohort were diagnosed with stage IV disease, while 11% in the NExUS cohort and 4% of patients in the eLung cohort had stage IIIB disease; also, while both Italian and NExUS patients had non-squamous histology, 27% of the eLung patients had squamous disease (Table A.1).

Taken together, the cohorts serve as a broad representation of patients with advanced NSCLC eligible for platinum-doublet therapy. Median age and performance status of the patients, along with other clinical factors, were in-line with commonly reported statistics, and patients with both squamous and non-squamous histology were represented.

Similarly to previous studies, a higher proportion of VeriStrat Poor classifications were often associated with known poor prognostic factors, such as advanced stage, squamous histology, and worse performance status (Table 1). However, numerous multivariate analyses of OS and PFS adjusted for these prognostic factors have previously shown that VeriStrat is an independent predictor of outcomes [15,16,19,25].

3.2. VeriStrat association with outcomes

Results of comparisons between VeriStrat Good and Poor classifications are summarized in Fig. 1 and Table 2. VeriStrat Good patients have significantly better outcomes than VeriStrat Poor patients, with

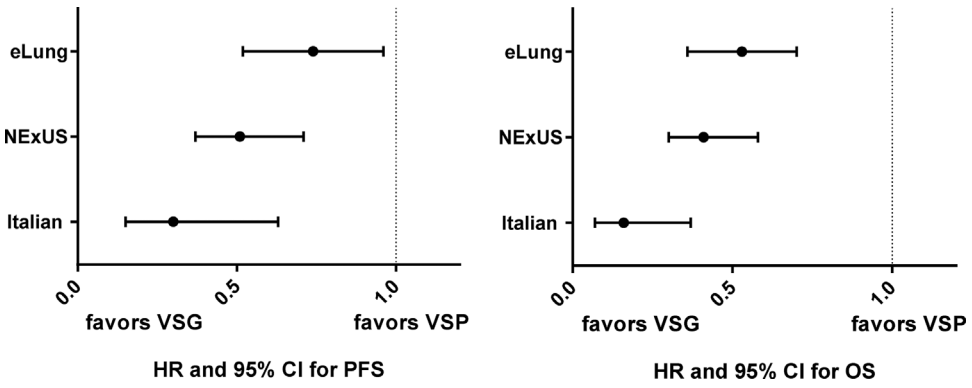

Fig. 2. Forest plots by VeriStrat classification for PFS and OS.
VSG, VeriStrat Good; VSP, VeriStrat Poor.

HRs of 0.36 ($p < .001$), 0.51 ($p < .001$), and 0.72 ($p = .032$) for PFS; HRs for OS between VeriStrat Good and Poor classifications were 0.26 ($p < .001$), 0.41 ($p < .001$), and 0.51 ($p < .001$) in the Italian, NExUS, and eLung cohorts, respectively. VeriStrat remained statistically significant in multivariate analyses adjusted for various clinical characteristics, such as PS, histology, stage, gender and other ($p < .001$ for PFS and OS in the Italian study, $p < .001$ for PFS and OS in NExUS, and $p = .071$ (trend) and .007 for PFS and OS, respectively, in the eLung study, as shown in Table A.2). The magnitude of the prognostic effect, which was especially pronounced in the Italian cohort, is illustrated by the forest plots of HRs in Fig. 2.

Though limited by differences in patient population, regimens, and treatment after progression, some numerical observations about PFS and OS in the VeriStrat-stratified groups of patients can be made. VeriStrat Good patients in the Italian, NExUS and eLung cohorts had comparable median PFS (6.5, 5.7, and 5.1 months, respectively), while VeriStrat Poor patients in the Italian cohort had much shorter median PFS than NExUS and eLung patients (1.6 months versus 4.6 and 3.7 months).

Median OS in VeriStrat Good patients in the Italian and eLung cohorts was similar (10.8 months and 10.9 months, respectively), and slightly longer in the NExUS group (14.7 months), but in the VeriStrat Poor cohorts did not exceed 6.5 months, and in the Italian cohort it was close to the outcome reported for patients on placebo from the TOPICAL trial (3.1 months versus 2.9 months [24]). The latter observation is especially noteworthy when considering that patients in the TOPICAL trial were elderly and deemed unfit for chemotherapy due to poor PS or other co-morbidities [22], while the patients in the chemotherapy studies were younger and had better prognostic characteristics. These results, if confirmed in a randomized study, could indicate that for VeriStrat Poor patients, who comprised a significant proportion (30–35%) of first line NSCLC patients in these cohorts, pemetrexed-platinum combinations provide no clinical benefit and should be avoided.

The results, however, do not directly address the issue of whether VeriStrat in the platinum-based therapy demonstrated purely prognostic or combination of prognostic and predictive properties, which is directly related to the question of whether it is feasible to overcome the dismal prognosis of VeriStrat Poor patients. However, promising preliminary data were obtained from the analysis of a small cohort of patients who were treated with gemcitabine [15] and in the second arm of the NExUS study, where VeriStrat Poor patients were treated with cisplatin/gemcitabine plus sorafenib [26]; in both cases, VeriStrat Poor patients had outcomes similar to those of VeriStrat Good patients, and the interactions of VeriStrat classifications with the comparative regimens were statistically significant. Further, in the P06162 trial the addition of the experimental agent ficlatuzumab to gefitinib was shown to improve outcomes in VeriStrat Poor patients, as compared to gefitinib alone [30]. Predictive properties of VeriStrat were also convincingly demonstrated in the prospective randomized phase III study PROSE in the second line. Thus, the test is both predictive and prognostic, and while we cannot demonstrate its predictive properties when patients are treated with similar therapies, as were described here, other approaches may lead to differential benefit relative to platinum-based chemotherapy. Since many of these options are not FDA-approved, a VeriStrat Poor status may identify a subset of patients for whom clinical trial enrollment and/or using broad molecular profiling in search of better therapeutic options should be considered.

The results presented here have several limitations: Two out of the three statistical analyses were undertaken by Biodesix; however, performing of the test blinded to clinical and outcome data and the ability of the study principal investigators to perform independent analyses of the data mitigated the risk of bias. Also, a significant proportion of patients in NExUS and eLung studies did not provide serum or plasma sample for testing, as is often the case when biomarker evaluation is added to the study protocol later in the course of a study.

The biological underpinnings of VeriStrat are not fully understood yet; it has been shown that some mass spectral peaks detected in samples classified as VeriStrat Poor strongly correlate with acute phase reactants such as serum amyloid A 1 (SAA1) [31] and C-reactive protein (unpublished data), which are known to be poor prognostic factors [32–34]. SAA, widely known as an acute phase reactant, is an important pro-inflammatory immunomodulator that induces the secretion of pro-inflammatory cytokines, such as tumor necrosis factor-α (TNF-α), interleukin-1β (IL-1β), and interleukin-8 (IL-8); it also acts as a chemoattractant for human monocytes, neutrophils and T cells, stimulates an M2-like alternative macrophage phenotype, and activates multiple cancer-related downstream pathways [35–37]. All these effects on the immune system may have influence on clinical outcomes. However, SAA1, and its truncated forms, constitute just some of the components of the VeriStrat peaks, which are likely correlated with other proteins that have not been identified yet, and no relevant SAA1 concentration cut-off was established for clinical use so far. Furthermore, there are other prognostic factors and molecules, such LDH or CRP that may be correlated with VeriStrat classification [38,39], but were not investigated directly in these studies, though it would be informative to include them in the multivariate analyses. However, while these proteins were shown to be associated with prognosis, no cut-offs for them were validated independently of the training sets, and there is no alternative validated test for platinum-based chemotherapy that is used in broad clinical practice. VeriStrat, to the contrary, was validated in many independent studies.

Finally, numerical comparison of outcomes is limited by the differences between cohorts of patients and their subsequent treatments, and can be used only in a qualitative sense and as a basis for future studies. However, the consistency of the results in three independent cohorts is a strong validation of the applicability of VeriStrat for platinum-based chemotherapy in first line.

4. Conclusion

The results discussed here may be viewed as a further confirmation of the prognostic role of the VeriStrat test in NSCLC patients. As such, VeriStrat may be valuable as an additional stratification parameter in the design of clinical trials, along with other prognostic factors, such as performance status, stage, and histology. In clinical practice, a better understanding of prognosis is critical for making informed treatment decisions, including enrollment in clinical trials, and is an important element of the patient-physician discussion of therapy options and best treatment strategies. This would be especially critical for patients classified as VeriStrat Poor, including those unfit to receive standard chemotherapies. Further studies to find treatment alternatives that improve outcomes for these patients are warranted.

Conflict of interest statement

Julia Grigorieva, Krista Meyer, Joanna Roder, and Heinrich Roder are employees of Biodesix and hold company options.

Francesco Grossi was compensated as a speaker for Eli Lilly, Bristol-Myers-Squibb, and received fees for participating in sponsored meetings and advisory boards of Bristol-Myers-Squibb.

Carlo Genova was compensated as a speaker for Bristol-Myers-Squibb, Astra Zeneca, and Boehringer-Ingelheim.

Other authors report no conflict of interests.

Role of funding source

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.lungcan.2017.12.007.

References

[1] N. Howlader, A.M. Noone, M. Krapcho, J. Garshell, D. Miller, S.F. Altekruse, C.L. Kosary, M. Yu, J. Ruhl, Z. Tatalovich, A. Mariotto, D.R. Lewis, H. Chen, E.J. Feuer, K.A. Cronin, SEER Cancer Statistics Review, 1975-2012, (2015) http://seer.cancer.gov/csr/1975_2012/ . (Accessed 15 August 2015).

[2] National Cancer Institute, Non-Small Cell Lung Cancer Treatment –for Health Professionals, (2016) http://www.cancer.gov/types/lung/hp/non-small-cell-lung-treatment-pdq#link/_4 . (Accessed 15 January 2016).

[3] J. Goffin, C. Lacchetti, P.M. Ellis, Y.C. Ung, W.K. Evans, First-line systemic chemotherapy in the treatment of advanced non-small cell lung cancer: a systematic review, J. Thorac. Oncol. 5 (2) (2010) 260–274.

[4] D. Moro-Sibilot, E. Smit, J. de Castro Carpeno, K. Lesniewski-Kmak, J. Aerts, R. Villatoro, K. Kraaij, K. Nacerddine, Y. Dyachkova, K.T. Smith, K. Taipale, A.C. Girvan, C. Visseren-Grul, P.A. Schnabel, Outcomes and resource use of non-small cell lung cancer (NSCLC) patients treated with first-line platinum-based chemotherapy across Europe: FRAME prospective observational study, Lung Cancer 88 (2) (2015) 215–222.

[5] B. Melosky, Q. Chu, R. Juergens, N. Leighl, D. McLeod, V. Hirsh, Pointed progress in second-line advanced non-small-cell lung cancer: the rapidly evolving field of checkpoint inhibition, J. Clin. Oncol. 34 (14) (2016) 1676–1688.

[6] M. Reck, D. Rodríguez-Abreu, A.G. Robinson, R. Hui, T. Csőszi, A. Fülöp, Pembrolizumab versus chemotherapy for PD-L1-positive non-small-cell lung cancer, N. Engl. J. Med. 375 (19) (2016) 1823–1833.

[7] S. Champiat, L. Dercle, S. Ammari, C. Massard, A. Hollebecque, S. Postel-Vinay, et al., Hyperprogressive disease (HPD) is a new pattern of progression in cancer patients treated by anti-PD-1/PD-L1, Clin. Cancer Res. 23 (8) (2016) 1920–1928.

[8] M.C. Nicolson, D.A. Fennell, D. Ferry, K. O'Byrne, R. Shah, V. Potter, G. Skailes, S. Upadhyay, P. Taylor, V. Andre, T.S. Nguyen, S.P. Myrand, C. Visseren-Grul, M. Das, K.M. Kerr, Thymidylate synthase expression and outcome of patients receiving pemetrexed for advanced nonsquamous non-small-cell lung cancer in a prospective blinded assessment phase II clinical trial, J. Thorac. Oncol. 8 (7) (2013) 930–939.

[9] K. Azuma, Y. Komohara, T. Sasada, Y. Terazaki, J. Ikeda, T. Hoshino, K. Itoh, A. Yamada, H. Aizawa, Excision repair cross-complementation group 1 predicts progression-free and overall survival in non-small cell lung cancer patients treated with platinum-based chemotherapy, Cancer Sci. 98 (9) (2007) 1336–1343.

[10] W.S. Kwon, S.Y. Rha, Y.H. Choi, J.O. Lee, K.H. Park, J.J. Jung, T.S. Kim, H.C. Jeung, H.C. Chung, Ribonucleotide reductase M1 (RRM1) 2464G > A polymorphism shows an association with gemcitabine chemosensitivity in cancer cell lines, Pharmacogenet. Genom. 16 (6) (2006) 429–438.

[11] L.P. Martin, T.C. Hamilton, R.J. Schilder, Platinum resistance: the role of DNA repair pathways, Clin. Cancer Res. 14 (5) (2008) 1291–1295.

[12] C. Gomez-Roca, C.M. Raynaud, F. Penault-Llorca, O. Mercier, F. Commo, L. Morat, L. Sabatier, P. Dartevelle, E. Taranchon, B. Besse, P. Validire, A. Italiano, J.C. Soria, Differential expression of biomarkers in primary non-small cell lung cancer and metastatic sites, J. Thorac. Oncol. 4 (10) (2009) 1212–1220.

[13] F. Taguchi, B. Solomon, V. Gregorc, H. Roder, R. Gray, K. Kasahara, M. Nishio, J. Brahmer, A. Spreafico, V. Ludovini, P.P. Massion, R. Dziadziuszko, J. Schiller, J. Grigorieva, M. Tsypin, S.W. Hunsucker, R. Caprioli, M.W. Duncan, F.R. Hirsch, P.A. Bunn Jr., D.P. Carbone, Mass spectrometry to classify non-small-cell lung cancer patients for clinical outcome after treatment with epidermal growth factor receptor tyrosine kinase inhibitors: a multicohort cross-institutional study, J. Natl. Cancer Inst. 99 (11) (2007) 838–846.

[14] C.H. Chung, E.H. Seeley, H. Roder, J. Grigorieva, M. Tsypin, J. Roder, B.A. Burtness, A. Argiris, A.A. Forastiere, J. Gilbert, B. Murphy, R.M. Caprioli, D.P. Carbone, E.E. Cohen, Detection of tumor epidermal growth factor receptor pathway dependence by serum mass spectrometry in cancer patients, Cancer Epidemiol. Biomarkers Prev. 19 (2) (2010) 358–365.

[15] T.E. Stinchcombe, J. Roder, A.H. Peterman, J. Grigorieva, C.B. Lee, D.T. Moore, M.A. Socinski, A retrospective analysis of VeriStrat status on outcome of a randomized phase II trial of first-line therapy with gemcitabine, erlotinib, or the combination in elderly patients (age 70 years or older) with stage IIIB/IV non-small-cell lung cancer, J. Thorac. Oncol. 8 (4) (2013) 443–451.

[16] O. Gautschi, A.C. Dingemans, S. Crowe, S. Peters, H. Roder, J. Grigorieva, J. Roder, F. Zappa, M. Pless, M. Brutsche, F. Baty, L. Bubendorf, S.F. Hsu Schmitz, K.J. Na, D. Carbone, R. Stahel, E. Smit, VeriStrat has a prognostic value for patients with advanced non-small cell lung cancer treated with erlotinib and bevacizumab in the first line: pooled analysis of SAKK19/05 and NTR528, Lung Cancer 79 (1) (2013) 59–64.

[17] J.L. Kuiper, J.S. Lind, H.J. Groen, J. Roder, J. Grigorieva, H. Roder, A.M. Dingemans, E.F. Smit, VeriStrat((R)) has prognostic value in advanced stage NSCLC patients treated with erlotinib and sorafenib, Br. J. Cancer 107 (11) (2012) 1820–1825.

[18] D.P. Carbone, J.S. Salmon, D. Billheimer, H. Chen, A. Sandler, H. Roder, J. Roder, M. Tsypin, R.S. Herbst, A.S. Tsao, H.T. Tran, T.P. Dang, VeriStrat classifier for survival and time to progression in non-small cell lung cancer (NSCLC) patients treated with erlotinib and bevacizumab, Lung Cancer 69 (3) (2010) 337–340.

[19] D.P. Carbone, K. Ding, H. Roder, J. Grigorieva, J. Roder, M.S. Tsao, L. Seymour, F.A. Shepherd, Prognostic and predictive role of the VeriStrat plasma test in patients with advanced non-small-cell lung cancer treated with erlotinib or placebo in the NCIC Clinical Trials Group BR. 21 trial, J. Thorac. Oncol. 7 (11) (2012) 1653–1660.

[20] D. Renouf, W. Parulekar, J. Grigorieva, D. Tu, M. Moore, Assessment of the association of the VeriStrat test with outcomes in patients (pts) with advanced pancreatic cancer (PC) treated with gemcitabine (G) with or without erlotinib (E) in the NCIC CTG PA.3 phase III trial, 2013 ASCO Annual Meeting, Chicago, J. Clin. Oncol. (2013).

[21] J. Roder, H. Roder, S.W. Hunsucker, J. Grigorieva, C.E. Ellis, A. Florance, R. Gagnon, L. O'Rourke, S. Johnston, Retrospective analysis of study EGF30008 by mass spectrometry based assay (VeriStrat), San Antonio Breast Cancer Symposium, San Antonio, TX, USA, 2011.

[22] S.M. Lee, I. Khan, S. Upadhyay, C. Lewanski, S. Falk, G. Skailes, E. Marshall, P.J. Woll, M. Hatton, R. Lal, R. Jones, E. Toy, D. Chao, G. Middleton, S. Bulley, Y. Ngai, R. Rudd, A. Hackshaw, C. Boshoff, First-line erlotinib in patients with advanced non-small-cell lung cancer unsuitable for chemotherapy (TOPICAL): a double-blind, placebo-controlled, phase 3 trial, Lancet Oncol. 13 (11) (2012) 1161–1170.

[23] F.A. Shepherd, J. Rodrigues Pereira, T. Ciuleanu, E.H. Tan, V. Hirsh, S. Thongprasert, D. Campos, S. Maoleekoonpiroj, M. Smylie, R. Martins, M. van Kooten, M. Dediu, B. Findlay, D. Tu, D. Johnston, A. Bezjak, G. Clark, P. Santabarbara, L. Seymour, Erlotinib in previously treated non-small-cell lung cancer, N. Engl. J. Med. 353 (2) (2005) 123–132.

[24] S.M. Lee, S. Nash, Y. Ngai, A. Hackshaw, Prognostic and predictive value of the VeriStrat classifier in chemo-naive NSCLC patients treated with erlotinib or placebo (TOPICAL trial), 16th World Conference on Lung Cancer, Denver, CO, 2015.

[25] V. Gregorc, S. Novello, C. Lazzari, S. Barni, M. Aieta, M. Mencoboni, F. Grossi, T.D. Pas, F. de Marinis, A. Bearz, I. Floriani, V. Torri, A. Bulotta, A. Cattaneo, J. Grigorieva, M. Tsypin, J. Roder, C. Doglioni, M.G. Levra, F. Petrelli, S. Foti, M. Vigano, A. Bachi, H. Roder, Predictive value of a proteomic signature in patients with non-small-cell lung cancer treated with second-line erlotinib or chemotherapy (PROSE): a biomarker-stratified, randomised phase 3 trial, Lancet Oncol. 15 (7) (2014) 713–721.

[26] J. Vansteenkiste, L. Paz-Ares, T. Eisen, D. Heigener, R. Eberhardt, M. Thomas, C. Zhou, A. Santoro, C. Lathia, H. Roder, A plasma proteomic signature predicts outcomes in a Phase 3 study of gemcitabine (G) + cisplatin (C) ± sorafenib in first line Stage IIIB or IV NSCLC, 2012 ESMO Congress, Vienna, Austria, Ann. Oncol. (2012) p. ix407.

[27] P. Miller, M. Walker, K. Tauer, J. Atkins, K. Sivarajan, V. Patel, B. Bastos, K. Meyer, J. Roder, L. Uyeda, S. Salazar, C. Langer, M. Socinski, D. Spigel, L. Schwartzberg, Serum and tumor biomarkers predict outcome in the eLung trial, a multicenter, randomized phase IIb study of standard platinum doublets (PD) plus cetuximab (CET) as first-line treatment of advanced non-small cell lung cancer (NSCLC), 2013 ASCO Annual Meeting, J. Clin. Oncol. (2013).

[28] F. Grossi, E. Rijavec, C. Genova, G. Barletta, F. Biello, C. Maggioni, G. Burrafato, C. Sini, M.G. Dal Bello, K. Meyer, J. Roder, H. Roder, J. Grigorieva, Serum proteomic test in advanced non-squamous non-small cell lung cancer treated in first line with standard chemotherapy, Br. J. Cancer 116 (1) (2017) 36–43.

[29] L.G. Paz-Ares, B. Biesma, D. Heigener, J. von Pawel, T. Eisen, J. Bennouna, L. Zhang, M. Liao, Y. Sun, S. Gans, K. Syrigos, E. Le Marie, M. Gottfried, J. Vansteenkiste, V. Alberola, U.P. Strauss, E. Montegriffo, T.J. Ong, A. Santoro, Phase III, randomized, double-blind, placebo-controlled trial of gemcitabine/cisplatin alone or with sorafenib for the first-line treatment of advanced, nonsquamous non-small-cell lung cancer, J. Clin. Oncol. 30 (25) (2012) 3084–3092.

[30] T.S. Mok, S.L. Geater, W.C. Su, E.H. Tan, J.C. Yang, G.C. Chang, M. Han, P. Komarnitsky, F. Payumo, J.E. Garrus, S. Close, K. Park, A randomized phase 2 study comparing the combination of ficlatuzumab and gefitinib with gefitinib alone in asian patients with advanced stage pulmonary adenocarcinoma, J. Thorac. Oncol. 11 (10) (2016) 1736–1744.

[31] E. Milan, C. Lazzari, S. Anand, I. Floriani, V. Torri, C. Sorlini, V. Gregorc, A. Bachi, SAA1 is over-expressed in plasma of non small cell lung cancer patients with poor outcome after treatment with epidermal growth factor receptor tyrosine-kinase inhibitors, J. Proteom. 76 (2012) 91–101.

[32] H. Biran, N. Friedman, L. Neumann, M. Pras, R. Shainkin-Kestenbaum, Serum amyloid A (SAA) variations in patients with cancer: correlation with disease activity, stage, primary site, and prognosis, J. Clin. Pathol. 39 (7) (1986) 794–797.

[33] W.C. Cho, T.T. Yip, W.W. Cheng, J.S. Au, Serum amyloid A is elevated in the serum of lung cancer patients with poor prognosis, Br. J. Cancer 102 (12) (2010) 1731–1735.

[34] P. Findeisen, M. Zapatka, T. Peccerella, H. Matzk, M. Neumaier, D. Schadendorf, S. Ugurel, Serum amyloid A as a prognostic marker in melanoma identified by proteomic profiling, J. Clin. Oncol. 27 (13) (2009) 2199–2208.

[35] B. Mieke De, G. Mieke, W. Ji Ming, S. Jacques Van, O. Ghislain, S. Sofie, D. Jo Van, Structure and expression of different serum amyloid a (SAA) variants and their concentration-dependent functions during host insults, Curr. Med. Chem. 23 (17) (2016) 1725–1755.

[36] I.N. Baranova, A.C.P. Souza, A.V. Bocharov, T.G. Vishnyakova, X. Hu, B.L. Vaisman, M.J. Amar, Z. Chen, A.T. Remaley, A.P. Patterson, P.S.T. Yuen, R.A. Star, T.L. Eggerman, Human SR-BII mediates SAA uptake and contributes to SAA pro-inflammatory signaling in vitro and in vivo, PLoS One 12 (4) (2017) e0175824.

[37] F.P. Ribeiro, C.J. Furlaneto, E. Hatanaka, W.B. Ribeiro, G.M. Souza, M.A. Cassatella, A. Campa, mRNA expression and release of interleukin-8 induced by serum amyloid A in neutrophils and monocytes, Mediat. Inflamm. 12 (3) (2003) 173–178.

[38] D.S. Lee, K.R. Park, S.J. Kim, M.J. Chung, Y.H. Lee, J.H. Chang, J.H. Kang, S.H. Hong, M.S. Kim, Y.S. Kim, Serum lactate dehydrogenase levels at presentation in stage IV non-small cell lung cancer: predictive value of metastases and relation to survival outcomes, Tumour Biol.: J. Int. Soc. Oncodev. Biol. Med. 31 (1) (2016) 619–625.

[39] S. Shrotriya, D. Walsh, N. Bennani-Baiti, S. Thomas, C. Lorton, C-Reactive protein is an important biomarker for prognosis tumor recurrence and treatment response in adult solid tumors: a systematic review, PLoS One 10 (12) (2015) e0143080.