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Real-World Utilization of Biomarker Testing for Patients with Advanced Non—Small Cell Lung Cancer in a Tertiary Referral Center and Referring Hospitals

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The continued introduction of biomarkers and innovative testing methods makes already complex diagnosis in patients with stage IV non—small-cell lung cancer (NSCLC) even more complex. This study primarily analyzed variations in biomarker testing in clinical practice in patients referred to a comprehensive cancer center in the Netherlands. The secondary aim was to compare the cost of biomarker testing with the cost of whole-genome sequencing. The cohort included 102 stage IV NSCLC patients who received biomarker testing in 2017 or 2018 at the comprehensive cancer center. The complete biomarker testing history of the cohort was identified using linked data from the comprehensive cancer center and the nationwide network and registry of histopathology and cytopathology in the Netherlands. Unique biomarker-test combinations, costs, turnaround times, and test utilization were examined. The results indicate substantial variation in test utilization and sequences. The mean cost per patient of biomarker testing was 2259.92 ± 1217.10 USD, or 1881.23 ± 1013.15 EUR. Targeted gene panels were most frequently conducted, followed by IHC analysis for programmed cell death protein ligand 1. Typically, the most common biomarkers were assessed within the first tests, and emerging biomarkers were tested further down the test sequence. At the cost of current biomarker testing, replacing current testing with whole-genome sequencing would have led to cost-savings in only two patients (2%). *(J Mol Diagn 2021, 23: 484—494; https://doi.org/10.1016/j.jmoldx.2021.01.004)*

The use of biomarker testing[1,2] for the prediction of treatment response and disease progression has made the diagnostic pathway of advanced non—small cell lung cancer (NSCLC) increasingly complex.[3] Moreover, this pathway is expected to become even more complex in the near future with the introduction of new biomarkers and innovative testing methods, such as whole-genome sequencing (WGS) and the evaluation of circulating tumor DNA using liquid biopsies.[4] For response prediction and for selecting the optimal treatment,[5] biomarker testing needs to be completed before treatment initiation. Hence, the turnaround time of biomarker testing directly influences the time at which a patient can be started on treatment.[6] In practice, multiple biomarker tests, such as targeted gene panels and immunohistochemistry (IHC) analysis are often conducted, which can result in unnecessary delays if there is an unplanned cascade of tests

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along the pathway. In case of substantial delays, priority could be given to initiating a suboptimal therapy even before the results of biomarker testing are received.[7] While the relationship between delays and survival is unclear due to confounding by indication,[8] a large-scale study found an association between time to treatment and mortality across all tumor stages in NSCLC.[9]

Testing the biomarkers with the highest prevalence first maximizes the likelihood of finding an actionable target as early as possible and minimizes the number of tests conducted. Current clinical practice guidelines recommend routine testing for biomarkers such as *EGFR*, *ALK*, *BRAF* V600E, and *ROS1*, to predict response to targeted therapy, and programmed cell death protein ligand (PD-L)-1 to predict response to immunotherapy.[10,11] The prevalences of biomarkers vary across genes and patient subgroups. A high PD-L1 expression level is present in a subgroup of 22% of NSCLC patients[12] while 14% of patients in Europe harbor an *EGFR* mutation.[13] Across all NSCLC patient subgroups, 2% to 7% of patients harbor an *ALK* translocation, 3% to 5% of patients harbor a *BRAF* V600E mutation, and *ROS1* rearrangements can be detected in 1% to 2% of patients.

A wide variety of techniques and platforms are available to test for these biomarkers. Single-gene tests such as Sanger sequencing, IHC, and a range of *in situ* hybridization tests are, in most cases, less expensive[14] and have a shorter turnaround time compared to multigene methods such as next-generation sequencing (NGS). While NGS, and particularly WGS, can increase efficiency by substituting all other tests used for biomarker testing, WGS is more expensive compared to other biomarker tests,[15–18] which is one of the reasons that it is not yet widely used in clinical practice. However, most patients undergo multiple biomarker tests, and whether WGS is also more expensive compared to the total cost per patient of biomarker testing is currently unclear.

Nonetheless, relatively little detailed information about the use of biomarker testing in clinical practice, including its contribution to the total cost of the diagnostic pathway, is available. This information would add essential information, as previous budget-impact studies of biomarker testing in lung cancer have reported aggregated measures, such as health care resource utilization.[19–21] Additionally, other relevant information, such as how tests are sequenced,[22] test techniques used,[6,23] and the actual costs of these test sequences, is not yet fully known.

This study, therefore, aimed to provide a complete overview of biomarker testing, potentially spanning multiple treatment lines, in a cohort with stage IV NSCLC in the Netherlands. The entire cohort was referred to a comprehensive cancer center (CCC), but also data on biomarker testing prior to referral were included in the current study. More specifically, estimates of utilization, sequence, turnaround time, and total cost of biomarker testing are provided.

Materials and Methods

Methods

Exploratory data analysis and process mining techniques were used for this investigation into the biomarker-testing pathway. Process mining is a set of techniques that exploit the information contained in event logs, which describe activities in terms of when they were executed and who was involved with the activity. Process mining allows for the discovery of the actual ordering of care processes and for the evaluation of the characteristics of testing, such as turnaround times and costs. More specifically, for each patient, biomarker tests were ordered based on the times at which they were recorded. R software package version 3.5.2 (*https://www.r-project.org*, last accessed June 5, 2020)[24] was used for the analysis, and R software package bupaR version 0.4.2[25] was used for process mining.

Data Sources

Eligible patients attended a large tertiary referral site, a CCC, and were identified using linked pathology data from the referring hospital to ensure analysis of the complete diagnostic pathway, resulting in one event log that contained highly granular information on the types and timings of the activities conducted for each patient. PALGA (the nationwide network and registry of histopathology and cytopathology in the Netherlands) was used for extraction of the biomarker-testing history at other hospitals for the present patient cohort. Thus, this cohort was unique because of the access to diagnostics used by the referring hospital and CCC.

Data Cleaning and Enrichment

Duplicate activities, that is, tests with either a duplicate start or completion time, assumed to be reporting errors were removed. Activities not executed for reasons such as insufficient tumor material available were also excluded ($n = 51$; 5.5%). This led to the exclusion of data from nine patients (8.1%). The event log was enriched with data on the costs of the biomarker tests as reported in a previous microcosting study from the Netherlands in which 24 pathology laboratories participated.[15] For tests for which no cost data were available, reimbursement tariffs from 2017 were retrieved from the Dutch Healthcare Authority [*https://puc.overheid.nl/nza/doc/PUC_13010_22* (in Dutch), last accessed May 22, 2020].[26]

Patient Selection

The cohort consisted of patients with stage IV NSCLC who underwent IHC or molecular diagnostics at the CCC. Only patients who underwent biomarker testing at the CCC

between January 1, 2017, and December 31, 2018, were included. Patients who underwent biomarker testing at the CCC before or beyond this period were excluded. Data from the CCC spanned until August 2019, so there was reasonable confidence that all relevant activities within each patient episode were captured. This limited time interval was applied to minimize interpatient heterogeneity in data on tests received, caused by the implementation of new testing techniques over time, while retaining a patient cohort with an acceptable size. In total, data from 102 patients were included.

Biomarker Testing in the Comprehensive Cancer Center

The CCC is a nonteaching and nonacademic, specialized center. The CCC frequently organizes and participates in clinical trials. Biomarker testing is indicated for all patients with stage IV NSCLC. The oncologist requests biomarker testing, and in most cases, requests a specific biomarker test. The oncologist also specifies whether the biomarker testing is for an initial diagnosis or resistance analysis. The pathologist, together with the pulmonologist or oncologist, decides which genes will be tested for, while the molecular pathologist determines which technique or test will be used for each biomarker. Biomarker tests were conducted sequentially according to the NSCLC biomarker-testing strategy that the CCC had in place during the study period. With the identification of an actionable target, no further testing was undertaken, given that actionable targets rarely overlap.[27] The CCC conducted all testing in-house.

In this case, the CCC was a tertiary referral hospital; thus, almost all of the patients treated at the CCC had previously undergone diagnostics, and potentially also treatment, elsewhere. Reasons for referral to the CCC included enrollment in a clinical trial, case complexity, and having exhausted treatment options at the referring hospital. Although most patients referred to the CCC had undergone diagnostics and treatment previously, it is possible that not all relevant biomarkers for an initial diagnosis were tested at the referring hospital. Additionally, testing at the CCC is sometimes conducted to establish the eligibility of patients for enrollment in clinical trials. Therefore, biomarker testing at the CCC may have been more elaborate, and thus more expensive, compared to testing at nonspecialized centers and nonacademic hospitals. In most cases, physicians at the CCC trust the results of tests conducted elsewhere, minimizing the need for retesting the same biomarkers. Given the sequential nature of the test strategy, the test sequence conducted at the CCC was dependent on the tests conducted at other hospitals.

Validation with Clinicians

The findings were iteratively validated with a lung pathologist (K.M.) and pulmonologist (E.S.) employed at the CCC. First, during the initial stages of the analysis, discussions improved the understanding of the large degree of variation in the tests utilized and in the test sequences. Second, once the analysis was completed and the results of the study were presented to the clinicians, it became clear that the department of pathology was responsible for the order of the individual tests, and whether they were conducted in parallel or sequentially. Once all of the individual tests included in an order were completed, the results were sent to the requesting oncologist or pulmonologist. Therefore, how individual tests are sequenced is typically not known to the oncologist or pulmonologist. After discussing the results of the current study, both of the clinicians were confident that the results reflected their experience in daily clinical practice.

Results

Patient Population and Health Care Utilization

Table 1 describes the final patient cohort. The cost per patient reported in Table 1 includes the costs of all biomarker tests conducted. Additionally, the mean ± SD total cost per patient in those who underwent biomarker testing at other centers was 2550.91 ± 1221.51 USD, or 2124.87 ± 1017.50 EUR. In these patients the mean estimated cost of biomarker tests conducted at the CCC was 1778.44 ± 1197.39 USD, or 1481.42 ± 997.41 EUR.

Biomarker tests conducted at the CCC and at the referring centers are summarized in Tables 2 and 3, respectively. (All genes included in the assays and hotspot panel are listed in Supplemental Table S1). In some patients, the same test was

Table 1 Characteristics of the Patient Population

Characteristic	Value (N = 102)
Age, median (IQR), years	58.8 (12.6)
Sex, *n* (%)	
Female	51 (50.0)
Male	51 (50.0)
Stage, *n* (%)	
4	37 (36.3)
4A	23 (22.5)
4B	42 (41.2)
Histologic examination,* *n* (%)	
Adenocarcinoma	75 (73.5)
Squamous cell	12 (11.8)
Other specified carcinomas	9 (8.8)
Unspecified carcinomas (NOS)	6 (5.9)
Tests, median *n* (IQR)	7 (4)
Cost per patient of biomarker testing,† means ± SD	2258.42 ± 1216.29 USD;1881.23 ± 1013.15 EUR
Patient received biomarker testing also at other center(s), *n* (%)	49 (48.0)

IQR, interquartile range; NOS, not otherwise specified.
*Classification of histologic examination is based on ICD-O codes.[28]
†Calculated by dividing the sum of all biomarker test costs by the number of patients in the cohort.

Table 2 Descriptives of Biomarker Tests Conducted at the CCC

Biomarker	Test technique or platform	Absolute frequency	Unique patients tested, *n* (%)	Turnaround time, median (IQR), days	Cost, EUR	Cost, USD
MET exon 14 deletion	RT-PCR	85	78 (77.2)	8.1 (4.1)	275.24	330.43
EGFR, *HER2*	Multiplex fragment analysis	77	73 (72.3)	8.3 (5.0)	436.26*	523.73
PD-L1	IHC	70	70 (69.3)	NA	93.74	112.53
Assay	TSACP MiSeq (Illumina, San Diego, CA)†	44	41 (40.6)	11.8 (3.8)	258.96	310.88
	Path version 2D‡	23	17 (16.8)	86.9 (100)	993.67§	1192.90
	Archer FusionPlex MiSeq (Illumina)¶	2	2 (2.0)	9.1 (9.1)	993.67§	1192.90
	Total	69	54 (53.5)	12.8 (9.2)	417.57	501.23
ALK	IHC	54	54 (53.5)	NA	101.88	122.31
	FISH	5	3 (3.0)	7.7 (4.8)	134.48	161.44
	Total	59	55 (54.5)	7.7 (4.8)	102.16	122.64
ROS1	IHC	50	50 (49.5)	NA	101.88	122.31
	FISH	8	6 (5.9)	9.9 (3.1)	134.48	161.44
	Total	58	51 (50.5)	9.9 (3.1)	102.69	123.28
Hotspot panel	Sequenom MassARRAY	51	46 (45.5)	7.8 (3.2)	436.26*	523.73
MET	FISH	33	29 (28.7)	9.1 (4.8)	134.48	161.44
	DISH	8	8 (7.9)	NA	436.26*	523.73
	IHC	1	1 (1.0)	NA	97.81	117.42
	Total	42	34 (33.7)	9.1 (4.8)	151.18	181.49
NTRK	IHC	41	41 (40.6)	NA	97.81	117.42
RET	FISH	38	34 (33.7)	10.9 (7.4)	134.48	161.44
HER2	IHC	12	12 (11.9)	NA	97.81	117.42
	DISH	7	7 (6.9)	NA	436.26*	523.73
	Sanger sequencing	3	2 (2.0)	10.3 (6.4)	71.19	85.46
	FISH	1	1 (1.0)	10.9	134.48	161.44
	Total	23	14 (13.9)	10.6 (3.6)	178.51	214.30
FGR1	FISH	5	5 (5.0)	17.8 (4.6)	134.48	161.44
EGFR	FISH	3	2 (2.0)	9.2 (5.9)	134.48	161.44
	Sanger sequencing	2	1 (1.0)	NA	71.19	85.46
	Total	5	3 (3.0)	9.2 (5.9)	115.01	138.07
EGFR T790M	HRM sequencing	4	4 (4.0)	5.9 (2.0)	97.62	117.19
NRAS	Sanger sequencing	1	1 (1.0)	55 (0.0)	60.58	72.73
	HRM sequencing	1	1 (1.0	7.8 (0.0)	74.56	89.51
	Total	2	2 (2.0)	31.4 (23.6)	67.57	81.12
NRAS exon 4	Sanger sequencing	2	2 (2.0)	9.2 (2.4)	60.58	72.73
TP53	Sanger sequencing	2	2 (2.0)	9.2 (0.0)	65.40	78.51
KRAS	HRM sequencing	1	1 (1.0)	8.2 (0.0)	97.62	117.19
	Sanger sequencing	1	1 (1.0)	6.8 (0.0)	67.33	80.83
	Total	2	2 (2.0)	7.5 (0.7)	82.47	99.01

The table includes only biomarkers that were tested more than once. All assays consist of at least the following genes: *ALK*, *EGFR*, *BRAF*, *KRAS*, and *MET*. All genes included in the assays and hotspot panel are listed in Supplemental Table S1.

CCC, comprehensive cancer center; DISH, dual *in situ* hybridization; FISH, fluorescence *in situ* hybridization; HRM, high-resolution melt; NA, not applicable; RT-PCR, reverse transcription PCR; TSACP, TruSeq Amplicon—Cancer Panel.

*Maximum reimbursed amount for simple molecular diagnostics in 2017.[26]

†Forty-eight—gene DNA assay.

‡Twenty-nine—gene DNA assay.

§Maximum reimbursed amount for complex molecular diagnostics in 2017.[26]

¶Fourteen-gene RNA assay.

conducted more than once, as indicated by the difference between the absolute frequency and the number of unique patients tested in Table 2. In cases in which IHC showed a low PD-L1 expression level, a retest with a different antibody was conducted (14 patients). The CCC used antibody clones 22C3 and SP142 for IHC analysis of PD-L1 expression level. The antibody clones used elsewhere were unknown. Of the entire cohort, 94 (92.2%) underwent testing with a gene assay using either NGS or Sequenom MassARRAY (Agena Bioscience, San Diego, CA), and 82 (80.4%) underwent IHC analysis for PD-L1 expression level. Although turnaround times with IHC tests are not available from Table 2, these tests typically have a relatively short turnaround time of up to several days.

Table 3 Descriptives of Biomarker Tests Conducted at Referring Centers

Biomarkers	Test technique or platform	Absolute frequency	Unique patients tested, *n* (%)	Cost, EUR	Cost USD
Assay	Ion AmpliSeq (Thermo Fisher Scientific, Waltham, MA)*	44	34 (69.4)	296.45	355.89
	TSACP MiSeq (Illumina)†	4	4 (8.2)	258.96	310.88
	Total	48	36 (73.5)	258.96	310.88
ALK	IHC	22	21 (42.9)	101.88	122.31
	FISH	7	6 (12.2)	134.48	161.44
	Technique unknown	4	3 (6.1)	436.26‡	523.73
	Total	33	25 (51.0)	114.53	137.49
PD-L1	IHC	29	25 (51.0)	93.74	112.53
ROS1	IHC	11	11 (22.4)	101.88	122.31
	FISH	6	6 (12.2)	134.48	161.44
	Technique unknown	2	2 (4.1)	436.26‡	523.73
	Total	58	51 (50.5)	102.69	123.28
KRAS	Technique unknown	7	6 (12.2)	436.26‡	523.73
	Sanger sequencing	1	1 (2.0)	67.33	80.83
	Idylla (Biocartis, Mechelen, Belgium)	1	1 (2.0)	257.74	309.42
	Total	9	8 (16.3)	425.53	510.85
EGFR	Technique unknown	8	8 (16.3)	436.26‡	523.73
	Sanger sequencing	1	1 (2.0)	71.19	85.46
	Total	9	8 (16.3)	425.53	510.85
RET	FISH	5	5 (10.2)	134.48	161.44
	IHC	1	1 (2.0)	97.81	117.42
	Total	6	6 (12.2)	133.07	159.75
MET	FISH	3	3 (6.1)	134.48	161.44
HER2	IHC	1	1 (2.0)	97.81	117.42
	Sanger sequencing	1	1 (2.0)	71.19	85.46
	Technique unknown	1	1 (2.0)	436.26‡	523.73
	Total	3	3 (6.1)	201.75	242.20

The table includes only biomarkers that were tested more than once. All assays consisted of at least the following genes: *ALK*, *EGFR*, *BRAF*, *KRAS*, and *MET*. All genes included in the assays and hotspot panel are listed in Supplemental Table S1.

FISH, fluorescence *in situ* hybridization; TSACP, TruSeq Amplicon—Cancer Panel.

*Fifty-gene DNA assay.

†Forty-eight—gene DNA assay.

‡Maximal reimbursed amount for simple molecular diagnostics in 2017.[26]

Unique Biomarker-Test Combinations

Including testing both at the CCC and referring centers, 99 unique biomarker-test combinations were found in 102 patients. Thus, almost none of the patients underwent exactly the same tests in the same order. Figure 1 shows all of the unique biomarker-test combinations, ordered chronologically, in the entire cohort. Figure 1 does not show which biomarker tests were conducted sequentially and which in parallel. The degree of test uniformity across patients was higher at the beginning of the test sequences, compared to the tests conducted at a later stage in the test sequences. The number of tests conducted per patient also showed a substantial degree of variation across patients. Most patients were tested first for biomarkers that were recommended by leading clinical practice guidelines,[10,11] while emerging biomarkers such as *MET*, *NRAS*, and *RET* were typically tested at a later stage, to determine eligibility for clinical trials. Overall, 69 of 102 patients were eventually tested with a targeted gene panel, and in 19 of these patients, it was the first test conducted. In some cases, the same gene was tested twice, back to back. For example, the second test was *in situ* hybridization for confirming the positive IHC result. Furthermore, when *ALK*, PD-L1, *ROS1*, and in most cases *NTRK* were tested one after another, using IHC as a part of the same workflow.

Zooming in on the 3 weeks of the test sequence (Figure 2) indicates that the tests were completed at different times in each patient. Moreover, in most patients, more than one test was completed, even within this relatively short interval.

Distribution of Cost per Patient

Figure 3 presents the distribution of the total cost per patient of the biomarker tests conducted at both the CCC and other centers. Figure 3 shows a typical right-skewed distribution, meaning that in several patients the costs were much higher than the mean. Patients undergoing testing of a relatively low cost received a relatively low number of tests. Given that WGS may replace all other biomarker tests conducted,

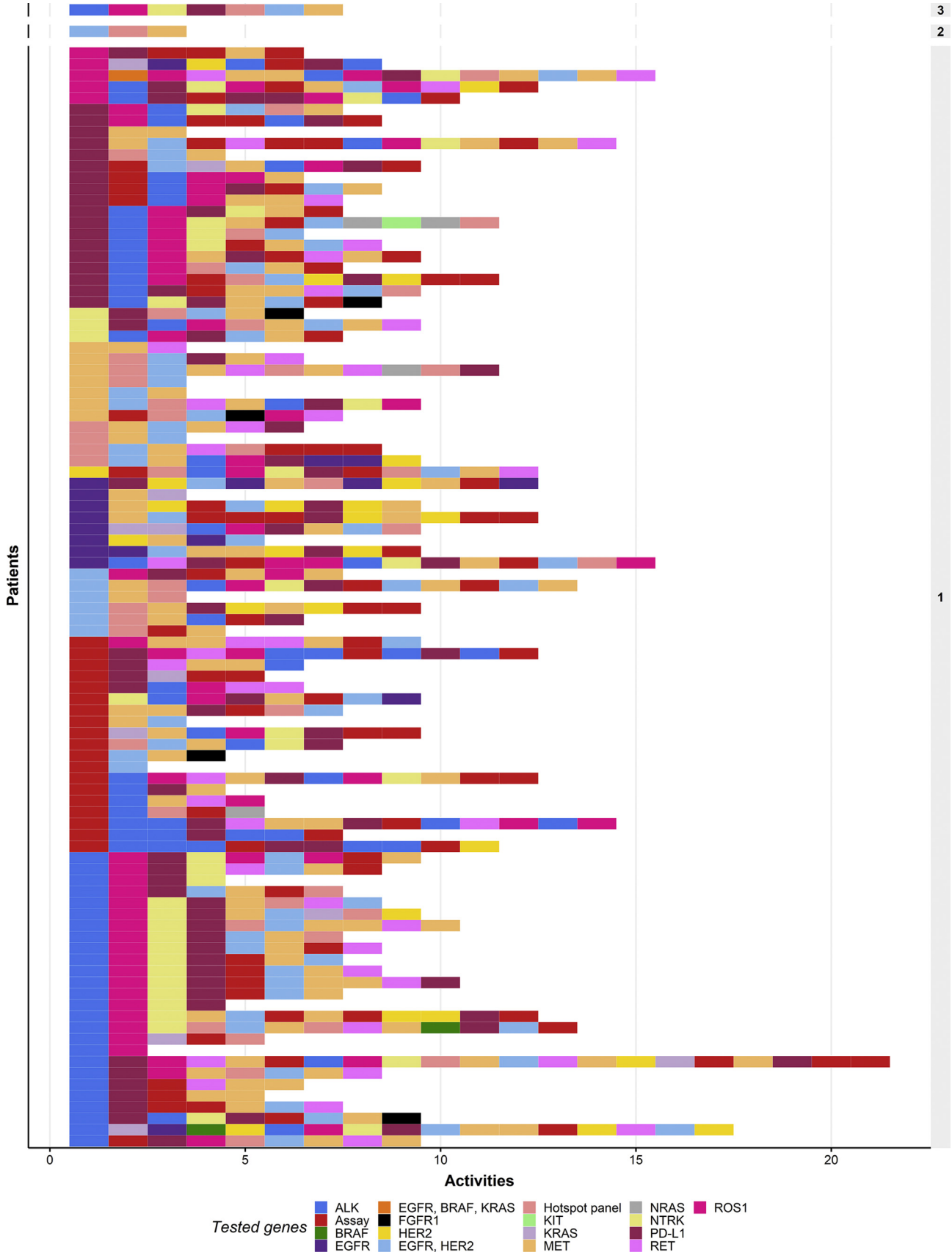

Figure 1 Unique biomarker test combinations for all individual patients included in the patient cohort. The tests were ordered chronologically. Each row represents the biomarker-test combination for one patient. Numbers shown on the right indicate the number of patients who received the same biomarker test combination.

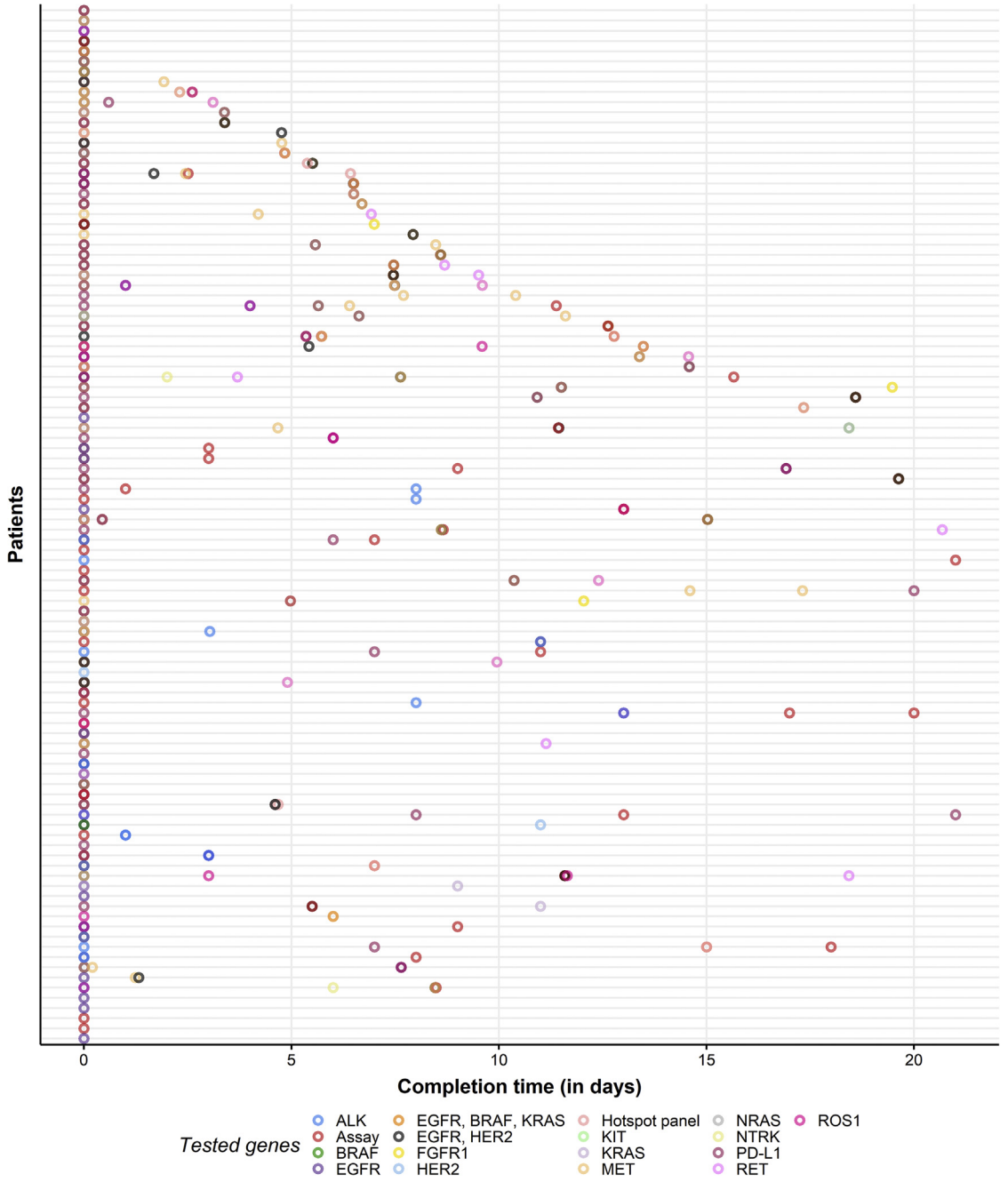

Figure 2 Distribution of biomarker tests over time, zoomed in on the first 3 weeks after completion of the first biomarker test. Each row represents one patient. Each dot represents one biomarker test. Patients are ordered by the total duration of their care pathway and may continue beyond the 3 weeks shown here.

the number of patients who would have incurred lower costs had they received WGS as the only test can be derived. The cost of WGS may be different in other countries and may continue to decrease over the years. Therefore, Figure 2 includes multiple hypothetical cost levels for WGS.

Discussion

This study provides further insight into the biomarker tests used in patients with stage IV NSCLC, based on complete biomarker-testing history, conducted at either the CCC or other centers. The patient cohort was described using clinical and other patient characteristics. The cost level at which WGS would be equally or less expensive compared to the cost per patient observed in the present cohort was examined. The median age of the cohort was lower compared to that of the total population of patients with stage IV NSCLC,[29] potentially due to the fact that eligibility of younger patients for treatment is higher, resulting in increased biomarker testing. Compared to the total population of patients with stage IV NSCLC, the cohort contained relatively more patients with adenocarcinoma, with possible

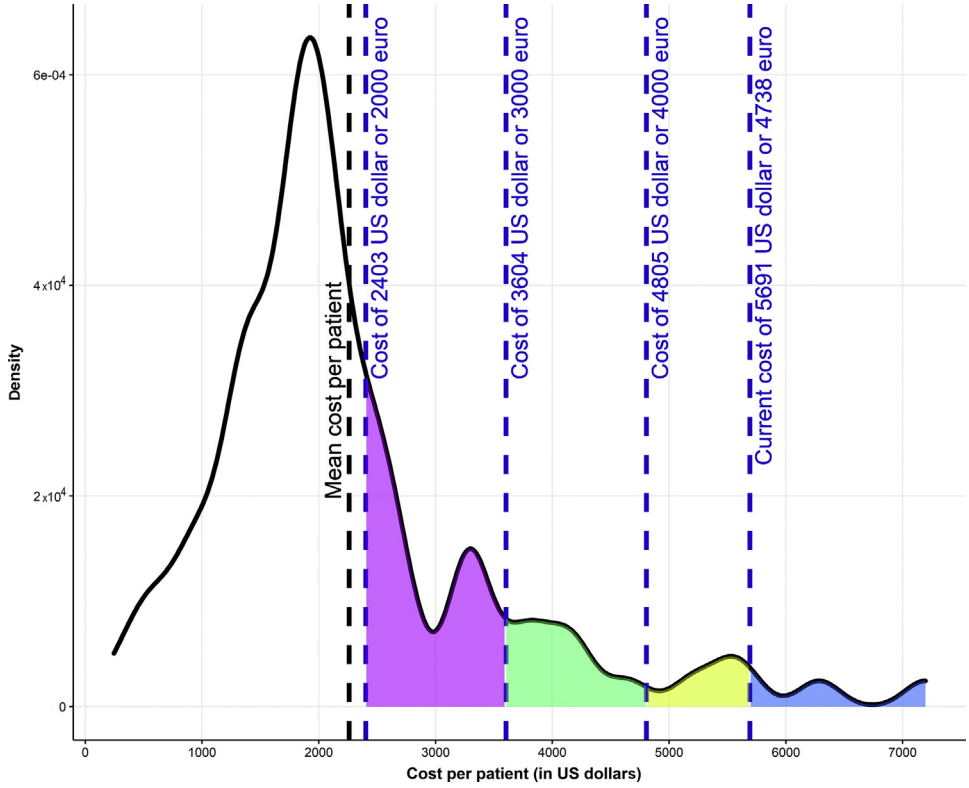

Figure 3 Distribution of the total cost per patient for biomarker testing. The **black dashed line** indicate the mean costs per patient. **Blue dashed lines** indicate the current price of whole-genome sequencing (WGS), a hypothetical cost of €2000, €3000, and €4000 per patient, respectively. Shaded areas represent the number of patients for whom WGS may have been equally expensive or less expensive at each respective price level. Purple, 17 patients (16.7%); green, 7 patients (6.9%); yellow, 3 patients (2.9%); blue, 2 patients (2.0%).

overrepresentation, which could have been caused by the fact that these patients typically have a higher probability of harboring biomarkers.[30]

The results illustrate the sequential nature of these tests and differences in testing capabilities across referring and referral centers. They show 99 unique biomarker-test combinations in 102 patients, including tests conducted at both the CCC and referring centers. The mean cost per patient of biomarker testing was 2258.42 ± 1216.29 USD, or 1881.23 ± 1013.15 EUR, of which, on average, 1778.44 USD, or 1481.42 EUR (75%) was incurred at the CCC, a marked increase from 1369.77 USD, or 1141 EUR, reported by Van Amerongen et al[22] in 2015, also based on data from the CCC. The median number of biomarker tests per patient in the cohort was substantially higher compared to the number of tests per patient assumed by Van Amerongen et al,[22] and may have been a cause of the increase in cost per patient. This increase in cost had no direct financial consequences to the patients, as these costs are reimbursed through basic health insurance in the Netherlands. However, it does increase the budget impact of biomarker testing. Figure 3 shows a long-tailed distribution, which highlights that a relatively small number of patients incurred a substantially higher total cost of biomarker testing.

The data in Figures 1 and 2 indicate that, in most patients, the most common biomarkers were tested within the first few tests, and that emerging biomarkers were typically tested later in the test sequence. An exception is *NTRK* fusion, an emerging biomarker with a relatively low prevalence,[31] that in the CCC is tested for with IHC. In the CCC, *NTRK* is often tested in the same workflow as *ALK*, PD-L1, and *ROS1*, and is therefore tested at a relatively early stage in the test sequence. Given the sequential nature of the strategy used for biomarker testing, whether additional tests are conducted is partly dependent on the results of previous tests. Further testing is also dependent on the availability of tumor material. An additional source of variation among test sequences is the highly dynamic landscape of biomarker testing, illustrated by the monthly or bimonthly changes in the test protocol of the CCC.

Testing for the most prevalent biomarkers first maximizes the likelihood of finding an actionable target as early as possible and minimizes the number of tests conducted. Testing for the most prevalent biomarkers first is especially relevant in settings in which obtaining enough biopsy material is challenging. While some patient subgroups have a higher likelihood of harboring biomarkers,[14] it is difficult to predict which patients will require a high number of tests to find a positive result. Even so, 69 of 102 patients eventually underwent testing with a targeted gene panel, and in 19 of these patients, it was the first test received. Additionally, 15 patients were tested more than once with the same gene assay. This finding was not unexpected, given that the same panel used for initial testing was also used for resistance testing in the CCC. Evidence from a decision-analytical model suggested that using NGS as the initial test can lead to cost-savings compared to a sequential

approach.[32] However, some biomarkers, such as PD-L1 expression, are currently not testable with NGS, and NGS requires a large amount of tumor material, which leads to a higher failure rate compared to IHC.[33] In general, careful management of tumor material and techniques that facilitate the testing of many genes concurrently while using a limited amount of tumor material is advisable.

The number of patients in whom biomarker testing would have been equally or less expensive had their entire test sequence been replaced with WGS was also analyzed. Depending on the assumed cost level of WGS, this number ranges from 2 patients (2.0%) at the current cost level of approximately 5691 USD, or 4700 EUR per patient, to 29 patients (28.4%) at a hypothetical cost of 2403 USD, or 2000 EUR, per patient. However, studies from other countries have reported different price levels,[16–18] and others have predicted future decreases in costs.[15] Therefore, it is likely that this number will change soon. The costs of other tests are also dynamic, so the costs of testing need to be compared regularly. Moreover, it is likely that testing for progression or treatment resistance would still be required after WGS, which would lead to higher costs. The costs of tests for treatment resistance and progression were excluded from the cost comparison in Figure 3, as the costs of those tests were unknown. Nonetheless, the downstream value that more comprehensive molecular diagnostics provide by improving the treatment decision is potentially much higher than a reduction in the costs of testing.[34]

One of the strengths of this article was the level of detail reported on the conducted tests. This was the first comprehensive report to have included tested genes, utilized techniques, costs, and turnaround times on the entire sequence of tests in patients with stage IV NSCLC. Another strength was that the sources of data used in this study were not confined to one center. Obtaining data from multiple centers was especially significant, given that the test sequence was also dependent on the tests previously conducted at other centers. Thus, the test sequence should be evaluated in its entirety. Moreover, the application of process-mining techniques in reporting sequences of biomarker testing is novel, and this was the first attempt. While process mining has been previously applied to discover care pathways,[35] only a few studies have analyzed care pathways in lung cancer,[36–38] all of which have proposed a novel method of conducting process mining without providing an empirical application. Although not all process-mining methods are useful in this context, process mining offers a valuable approach to describing care pathways.

This study also had some limitations. First, the generalizability of the results may be limited, given that the CCC may use a more elaborate test strategy for establishing the eligibility of patients for enrollment in clinical trials compared to other nonacademic and nonspecialized hospitals. Additionally, the cost of testing is specific to each setting, so the same tests in other centers may have been more or less costly.[39] The cost estimates used primarily were likely accurate representations of the national average, as they were based on cost data from 24 laboratories in the Netherlands.[14] Nonetheless, generalizing biomarker-test costs to centers in other countries remains challenging. Second, the size of the patient cohort was relatively small. However, after validation of the results, it was determined that they reflected the heterogeneity observed in clinical practice. Third, no costs were known for some test techniques. The impact of this limitation was minimized by using reimbursed tariffs. Fourth, the turnaround times of tests conducted with IHC or tests conducted at referring centers could not be calculated, as only the completion times for these tests were reported.

With the introduction of new biomarkers and testing techniques, testing strategies will likely become even more complex. Perhaps the value of WGS should be seen in light of the reduction in the complexity of the diagnostic pathway, as it is unlikely that the cost of WGS will be competitive. The value of reducing the complexity of the diagnostic pathway is an aspect of the value that WGS may provide but has not yet been explored in detail. It could be an exciting avenue for future research.

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Supplemental Data

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