CURRENT ONCOLOGY
A Canadian Cancer Research Journal

Improving molecular testing and personalized medicine in non-small-cell lung cancer in Ontario

C. Lim MD,* H.S. Sekhon MD MSc PhD,† J.C. Cutz MSc MD,‡ D.M. Hwang MD PhD,§ S. Kamel-Reid PhD,§|| R.F. Carter PhD DVM,‡‡ G. da Cunha Santos MD PhD,§ T. Waddell MD MSc PhD,** M. Binnie MD,†† M. Patel MD,‡‡ N. Paul MD,§§ T. Chung MD,§§ A. Brade MD CM PhD,|||| R. El-Maraghi MD,## C. Sit,*** M.S. Tsao MD,§ and N.B. Leighl MD MMSc*

ABSTRACT

Background Although molecular testing has become standard in managing advanced nonsquamous non-small-cell lung cancer (NSCLC), most patients undergo minimally invasive procedures, and the diagnostic tumour specimens available for testing are usually limited. A knowledge translation initiative to educate diagnostic specialists about sampling techniques and laboratory processes was undertaken to improve the uptake and application of molecular testing in advanced lung cancer.

Methods A multidisciplinary panel of physician experts including pathologists, respirologists, interventional thoracic radiologists, thoracic surgeons, medical oncologists, and radiation oncologists developed a specialty-specific education program, adapting international clinical guidelines to the local Ontario context. Expert recommendations from the program are reported here.

Results Panel experts agreed that specialists procuring samples for lung cancer diagnosis should choose biopsy techniques that maximize tumour cellularity, and that conservation strategies to maximize tissue for molecular testing should be used in tissue processing. The timeliness of molecular reporting can be improved by pathologist-initiated reflex testing upon confirmation of nonsquamous NSCLC and by prompt transportation of specimens to designated molecular diagnostic centres. To coordinate timely molecular testing and optimal treatment, collaboration and communication between all clinicians involved in diagnosing patients with advanced lung cancer are mandatory.

Conclusions Knowledge transfer to diagnostic lung cancer specialists could potentially improve molecular testing and treatment for advanced lung cancer patients.

Key Words Non-small-cell lung cancer, biomarkers, quality of care, knowledge translation

INTRODUCTION

Lung cancer remains the leading cause of cancer-related mortality in Canadians, with a 5-year survival rate of approximately 18%[1]. Non-small-cell lung cancer (NSCLC) typically presents at an advanced stage[2], and biomarker-directed therapy has greatly altered the approach to treating advanced NSCLC[3]. Compared with conventional chemotherapy regimens, tyrosine kinase inhibitors targeting the epidermal growth factor receptor (EGFR) and anaplastic lymphoma kinase (ALK) in select patient populations have been associated with improved clinical outcomes, including response and quality of life; a favourable toxicity profile; and improved progression-free and even overall survival[4–14].

Routine testing for *EGFR* mutations and *ALK* rearrangement has now become standard in the management of advanced nonsquamous NSCLC[15–18]; however, not all jurisdictions have succeeded in implementing guideline-based testing recommendations in a timely manner. In a

Correspondence to: Natasha B. Leighl, Princess Margaret Cancer Centre, University Health Network, 5-105, 610 University Avenue, Toronto, Ontario M5G 2M9.
E-mail: Natasha.Leighl@uhn.ca ■ **DOI:** https://doi.org/10.3747/co.24.3495

Canadian *EGFR* testing program initiated in 2010, molecular testing was estimated to have been initiated for only 38% of eligible NSCLC patients, and of the tests initiated, 12% were not performed because tumour specimens were insufficient[18]. This gap in knowledge translation is believed to be multi-factorial, arising from a lack of awareness on the part of non-oncology clinicians diagnosing lung cancer, and a lack of dedicated funding and laboratory infrastructure support for routine molecular testing[19,20].

Despite the incorporation of molecular testing into Cancer Care Ontario's lung cancer diagnostic pathway in 2012[2], there remains, for members of the lung cancer diagnostic team, a need for knowledge dissemination about molecular testing in lung cancer and for concrete guidance in implementing the guidelines. Cancer Care Ontario's mandate for performance improvement requires knowledge transfer through a coordinated program to engage clinicians by formal and informal means. Experience from quality improvement initiatives in Canadian oncology settings has affirmed the importance of clinician engagement and multidisciplinary education[21,22]. We therefore developed a knowledge translation intervention to educate diagnostic lung cancer specialists about clinical relevance, sampling techniques, and laboratory processes to improve the uptake and application of molecular testing in advanced lung cancer. The intervention was developed with a local context in mind, although the insights gained are broadly applicable to oncology settings in Canada and worldwide. Zer *et al.*[23] described the effect of this intervention with respect to improving clinician knowledge of best practices in tissue acquisition and handling and in expedited molecular testing. The present report summarizes key insights from this multidisciplinary education program addressing use of molecular testing for lung cancer in Ontario, including sample acquisition and processing, patient selection, interdisciplinary communication, and steps toward overcoming barriers to implementation.

METHODS

Study Objectives
The intervention was designed to increase the awareness of clinicians involved in lung cancer diagnosis—specifically, those acquiring and processing diagnostic samples—about current best practices and guidelines for molecular testing and personalized therapy in NSCLC. Additional goals included using collaborative discussion in specialty-specific working groups to identify barriers to routine biomarker testing and to develop strategies for overcoming those barriers. By engaging knowledge users and seeking their insight into gaps in the current knowledge base, we sought to identify action steps that could be used to overcome barriers to the implementation of molecular testing guidelines in lung cancer.

Ethics and Funding
The Princess Margaret Cancer Centre institutional research ethics board reviewed and approved the study. The study was conducted with the support of the Ontario Institute for Cancer Research and Cancer Care Ontario through funding provided by the Government of Ontario.

Study Design and Target Population
A multidisciplinary panel of physician experts—including key provincial leaders in pathology, interventional thoracic radiology, respirology, thoracic surgery, radiation oncology, and medical oncology—was assembled to develop specialty-specific education programs. Panel members reviewed published clinical guidelines and guidelines in development[15,22]. Recommendations were summarized and adapted to the local Ontario context. Educational content was developed based on the summarized recommendations, supplemented by specialty-specific literature review and local experience. Panel members reviewed the education materials, resolved disagreements thorough discussion, and then delivered the programs at provincial and national specialty meetings and in selected provincial health regions. In addition to a formal lecture delivered by 2–4 multidisciplinary speakers, participants were invited to interact during the session to provide feedback and to identify barriers to and solutions for implementing guideline recommendations specific to their individual practice, institution, and regional area.

RESULTS

Between May and October 2013, 10 education workshops were held across Ontario, registering 315 attendees in total. Participants included Ontario specialists from multiple disciplines involved in the diagnosis and treatment of lung cancer, including respirologists, pathologists, radiologists, thoracic surgeons, radiation and medical oncologists, pharmacists, oncology nurses, and pathology laboratory technologists. The specialty-specific recommendations addressing sample acquisition and processing, patient selection, interdisciplinary communication, and solutions for addressing barriers to implementation are summarized in the subsections that follow. An ideal NSCLC molecular diagnostic pathway is summarized in Figure 1.

Sample Acquisition

Respirology Perspectives
Flexible bronchoscopy has commonly been performed in the initial work-up for suspected lung cancer. The technique is widely available, has the potential to rule out other diagnoses, and can lead to a rapid diagnosis of lung cancer, including pathologic subtype. However, bronchoscopy alone may not yield sufficient tissue for molecular testing. Tumour cellularity in bronchial wash specimens[24]—and also in bronchial biopsy samples[25,26]—is often relatively low. The worst results with respect to *EGFR* mutation detection rates have been reported for bronchial washings or brushings and sputum[27]. Bronchial biopsy specimens can also show crush artifact, impairing downstream immunohistochemistry (IHC) or fluorescence *in situ* hybridization.

Multiple strategies can be used to increase the potential tumour yield. Increasing the size of biopsy forceps and obtaining at least 3 endobronchial biopsies makes a successful histologic diagnosis more likely[28]. Combining multiple techniques including bronchial biopsy and endobronchial ultrasonography with transbronchial needle aspiration could also improve diagnostic yield[29,30]. In

```
┌─────────────────────────────┐
│        Sample obtained by   │
│  Respirologist / Interventional │
│  Radiologist / Thoracic Surgeon │
└─────────────────────────────┘
              ↓
1-2 days
┌─────────────────────────────┐
│  Pathologist to diagnose non-  │        Reflex testing by
│  squamous lung cancer, evaluate │        pathologist at time of
│  tumor cellularity and adequacy │        diagnosis of non-
└─────────────────────────────┘        squamous lung
              ↓                          cancer
1-7 days
┌─────────────────────────────┐
│  Sample delivery to molecular  │
│        testing center          │
└─────────────────────────────┘
              ↓
1-2 days
┌─────────────────────────────┐
│  Pathologist to evaluate HE slide │
└─────────────────────────────┘
              ↓
1-2 days
┌─────────────────────────────┐
│   Lab performance of the assay  │
└─────────────────────────────┘
              ↓
7-10 days
┌─────────────────────────────┐
│  Results reported to Clinician  │
└─────────────────────────────┘
Total: target ≤ 14   ↓
days
┌─────────────────────────────┐
│  Oncologist consultation and    │        Result available at
│      treatment decision          │        consultation
└─────────────────────────────┘
```
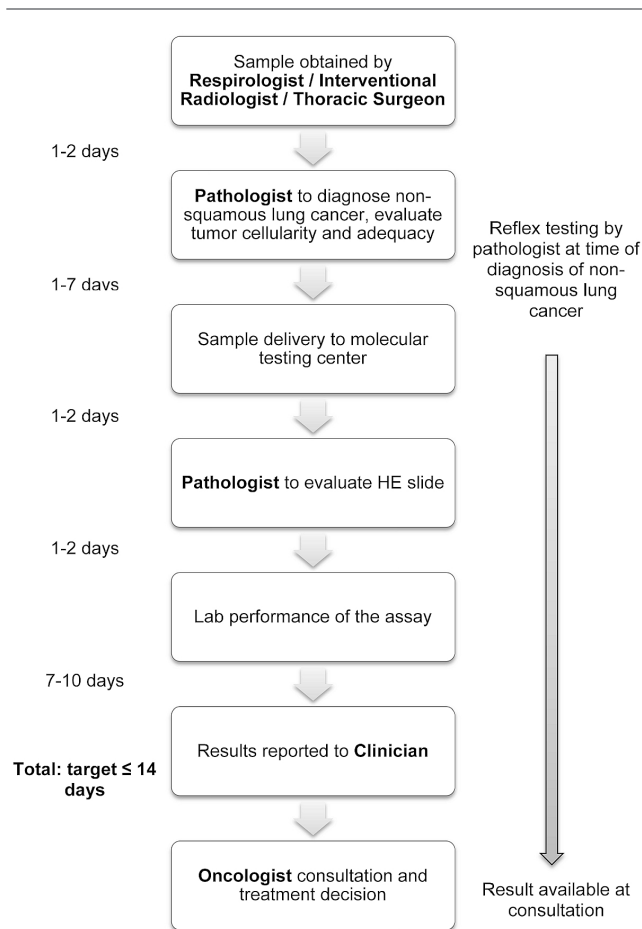
FIGURE 1   Ideal non-small-cell lung cancer molecular diagnostic pathway. HE = hematoxylin and eosin.

addition, rapid on-site evaluation (ROSE) of sample quality by a cytopathologist or a qualified cytotechnologist could improve patient safety by reducing the number of additional procedures and reducing complications; it can also optimize laboratory workflow[31–34]. In settings in which ROSE is not available, 3 aspirations or 2 aspirations plus 1 tissue core per suspected lymph node station for cytology preparation and cell block are recommended[35]. An increased extent of tissue sampling for maximizing tissue yield has to be weighed against patient tolerance and the longer procedure and local anesthesia times required.

Interventional Thoracic Radiology Perspectives

Percutaneous transthoracic biopsies are frequently used for peripheral lesions not accessible by bronchoscopy, and tissue yield is strongly influenced by the gauge of the biopsy needle used. Although the use of smaller biopsy needles can mitigate the risk of post-procedure complications including pneumothorax, hemoptysis, and hemorrhage, that practice could compromise tumour cellularity in the specimen obtained. Although the optimal needle gauge for obtaining core biopsies remains unclear, standard 20-gauge needles are frequently used[36]. Coaxial needle technique permits acquisition of multiple samples and allows for both core biopsy and cytology specimens to

be obtained while minimizing pleural punctures and potentially lowering the risk of pneumothorax[37,38].

To improve diagnostic accuracy, reduce the need for repeat procedures, and optimize patient safety, ROSE of fine-needle aspirate cytology specimens is preferred[39,40]. In settings in which ROSE is not available, 3–4 core biopsies are recommended (assuming that the lesion is of sufficient volume). When multiple core biopsies are obtained, the likelihood of unsuccessful molecular analysis because of insufficient tumour cellularity, necrotic tissue, or crush artifact is reduced.

Although fine-needle aspiration and core biopsies both yield appropriate tissue for genomic testing, as well as *ALK* by IHC and fluorescence *in situ* hybridization, it is important to appreciate that certain emerging predictive tests might be more appropriate for one sample type compared with another. A current example is expression of PD-L1, which is predictive of benefit from PD-1 axis inhibitors[41]. The relevant test has been validated in surgical pathology and core needle biopsy specimens, but not yet in cytology specimens. This situation is expected to evolve with time, however.

Thoracic Surgery Perspectives

Although surgical resection for NSCLC has historically focused primarily on early-stage disease, thoracic surgeons have an important role to play beyond surgical resection for NSCLC patients[42]. Surgeons are among the first diagnostic specialists to evaluate lung cancer patients in Canada, even at an advanced stage given the growing use of rapid diagnostic programs[43]. Resection specimens provide a larger volume of tumour tissue for downstream molecular testing.

Testing in the early-stage setting is encouraged[15]. Although therapy with tyrosine kinase inhibitors is not currently indicated in the adjuvant setting[44], tumour recurrence remains common in NSCLC, and early molecular testing is crucial for guiding therapy in a timely manner upon recurrence. In the setting of diagnosing unresectable NSCLC or metastatic recurrence, thoracic surgeons play a key role in selecting appropriate biopsy techniques to maximize tumour cellularity and facilitate molecular testing.

Pathology Perspectives

Pathologists are essential in the diagnosis, both pathologic and molecular, of lung cancer. In addition to determining malignancy, lung origin, and pathologic subtype, pathologists evaluate diagnostic specimens to ensure that samples meet the criteria for molecular analysis. Whether cytology or biopsy specimens are obtained, tumour cellularity is the key determinant of the likelihood of successful molecular testing[45–48]; both techniques can potentially yield adequate material for diagnostic molecular tests[49]. With respect to cytology specimens, preparation of a cell block is still preferred[24,50], but other specimens are also suitable for analysis[27]. It is important to recall that molecular testing should still be attempted in specimens in which it is unclear whether the sample will meet all technical requirements. In such cases, the suboptimal quality of the sample should be indicated in the report, with consideration of repeat testing if unsuccessful or if a better-quality sample becomes available.

Solutions to Implementation Barriers

Although ROSE is associated with improved diagnostic accuracy and patient safety, implementing it as part of routine practice requires the coordinated efforts of procedural specialists and pathologists, and also financial and infrastructure support[24]. The availability of ROSE in interventional and endoscopy suites in Ontario is limited by human resource and financial constraints. Potential solutions to overcoming those barriers, which have had success in other jurisdictions, include expanding the role of cytotechnologists[51] and implementing tele-cytopathology assessment[52]. Additionally, direct and timely feedback from pathologists to clinicians about specimen adequacy and tumour cellularity for both core biopsy and cytology samples—for example, from endobronchial ultrasonography or bronchoscopy—can lead to continuous performance improvement.

Sample Processing

Respirology, Interventional Thoracic Radiology, and Thoracic Surgery Perspectives

After biopsy or resection, submitting diagnostic samples to the pathology laboratory in appropriate transport media is crucial to avoid compromising sample quality for molecular assays. Preferred samples include biopsy specimens submitted in buffered formalin, thus allowing for preparation of formalin-fixed paraffin-embedded blocks; cytology specimens fixed in alcohol can also be used[50]. Heavy-metal and acidic fixatives should be avoided, and decalcification of bone samples could compromise sample viability for molecular testing[50].

Pathology Perspectives

Pathologists are the primary enablers of molecular testing. Their role in directing tissue processing and initiating routine molecular analysis in a certified diagnostic laboratory at the time of diagnosis are critical to success[16,50]. According to current guidelines, to avoid delays, samples should be sent for molecular testing within a maximum of 3 working days from diagnosis[22]. Turnaround time for molecular results should be no more than 14 days, with access to expedited testing for urgent cases.

A key challenge is tissue conservation. For small biopsy samples, tissue conservation strategies are crucial and should focus on minimizing the amount of tissue used for diagnostic work-up (including slides for routine IHC) and maximizing tissue designated for molecular testing[53]. Tissue is lost with each successive refacing of the block, and to minimize tissue wastage, cuts should therefore be limited. Pathologists should exercise judgment in prioritizing molecular testing when limited tissue is available. One potential strategy is to cut 15–20 unstained slides upon initial processing, reserving 15 slides for molecular testing and any additional IHC tests[53]. Another approach is to spread multiple biopsy cores or fragments over separate blocks for sequential consumption.

A selective approach to IHC workup should be followed, using a minimal panel of markers for subtyping analysis. However, limitations of that approach include unnecessary testing if the diagnosis is not lung cancer or if the histologic subtype is not recommended for molecular testing. For example, stains for thyroid transcription factor 1 and p63 (or p40) are commonly used for differentiating lung adenocarcinoma and squamous cell carcinoma by IHC[28,54]. Unless there is strong clinical or pathology suspicion of extrathoracic tumour metastasis to lung, extensive routine IHC staining to determine extrathoracic tumour origin is discouraged[53].

Specimens with high tumour cellularity, even if they are small, are preferred for molecular testing over those with low tumour cellularity[45,47]. Whenever possible, the entire block and unused precut slides, rather than recut slides, should be sent for molecular testing. When multiple specimens are available, pathologists should choose the best specimen for molecular analysis in consultation with the treating clinician[22]. If the optimal specimen for testing is unclear, all available specimens should be sent to the molecular diagnostic laboratory with an accompanying description of fixatives and preparation techniques used and of the highest tumour cellularity noted in the tissue block.

Solutions to Implementation Barriers

In Ontario, *EGFR* and *ALK* testing are centralized at designated molecular testing centres. The geographic distribution of referring centres relative to the designated laboratories requires transportation of samples and reporting of results between the molecular testing centres and the submitting institutions. Those additional factors can lead to delays in testing and reporting results. Further complications arise because of the time and cost associated with storing, maintaining, retrieving, and processing archival tissue samples for molecular testing. Reflex testing upon lung cancer diagnosis—and standardized regional protocols for tissue transportation and handling, and timely reporting of results—would help to overcome those barriers. Additionally, summarized feedback from the molecular testing centres to clinicians about tissue characteristics and adequacy for molecular analysis can assist with continuous performance improvement—for example, with such operator-dependent procedures as endobronchial ultrasound-guided biopsy.

Selecting Patients for Molecular Testing

Medical and Radiation Oncology Perspectives

At a minimum, all patients with advanced nonsquamous NSCLC—regardless of sex, ethnicity, or smoking status—should undergo molecular testing for *EGFR* and *ALK*[17,50]. Historically, oncologists were involved in completing the staging for patients newly diagnosed with advanced NSCLC and in initiating molecular testing. However, precious time is lost if biomarker results are not available to the oncologist at the initial consultation, leading to delays in treatment, suboptimal first-line therapy decisions, and even missed opportunities for targeted treatment[55,56].

Pathology Perspectives

Molecular testing should be ordered at the time of an advanced nonsquamous lung cancer diagnosis or at the time of recurrence or progression for patients initially presenting with earlier-stage disease without prior testing. Testing early-stage resection specimens is also encouraged, and molecular testing should be initiated even if clinical staging

data are incomplete at diagnosis[50]. The implementation of pathologist-initiated reflex testing engages pathologists in decisions about diagnostic sample adequacy and recommendations for repeat biopsy, if indicated. In addition, the reflex testing strategy allows pathologists to prioritize sample processing for molecular diagnostics, eliminates the need to re-review slides when molecular testing is requested, and thus minimizes the time from sample submission to result reporting[57]. All of those benefits can lead to more efficient molecular testing and higher rates of success. However, potential drawbacks of reflex testing include the analysis of small diagnostic biopsy specimens rather than resection specimens and the currently undefined role of molecular test results obtained before definitive surgery.

Solutions to Implementation Barriers
Economic restrictions associated with the routine funding of molecular testing in Canadian public health care systems have largely limited testing to advanced NSCLC confirmed by clinical staging. The uptake of reflex testing has also been constrained by limited awareness on the part of some community-based pathologists and diagnostic specialists who are not affiliated with molecular testing centres or who are not involved in the subsequent treatment of lung cancer patients. Reflex testing provides timely results and bypasses the need for an oncology consultation (with associated delays) before molecular testing is initiated[24]. Oncologists, pathologists, and the specialists acquiring diagnostic tissue must therefore collaborate to facilitate reflex biomarker testing and rapid turnaround time for result reporting. Dedicated government funding (including for pathology services and infrastructure), in conjunction with a streamlining of diagnostic algorithms to emphasize reflex testing, can improve molecular testing practices and patient outcomes. Advocacy and lobbying efforts to address such limitations resulted in Ontario's Ministry of Health approving routine funding for *EGFR* analysis in lung cancer as of September 2014.

Interdisciplinary Communication

Multidisciplinary Perspectives
The need for molecular testing adds complexity to diagnostic algorithms for lung cancer and requires active involvement from physicians across multiple disciplines[46]. Interventional clinicians who obtain diagnostic samples are responsible for providing clinical information to the pathology lab—including the suspected primary tumour site, any known prior molecular analysis, and available staging information—to alert pathologists of the anticipated need for molecular testing. Use of a structured requisition to assist pathologists with key clinical information can further improve the testing process.

Pathologists should provide timely and direct feedback to interventional clinicians about the adequacy of tissue sampling and molecular testing success rates[22]. Providing performance data at the hospital and individual level relative to provincial standards could also encourage performance improvement[21]. When molecular testing is not successful, it is imperative that pathologists notify the appropriate clinicians so that treatment decisions are not

delayed and arrangements for repeat biopsy can be made. Such notifications might require additional infrastructure support and communication channels linking pathologists and clinicians.

Solutions to Implementation Barriers
Maintaining a bi-directional flow of information and continuous feedback is a critical requirement to facilitate ongoing process and quality improvement. Multidisciplinary diagnostic clinics or tumour-site conferences could potentially facilitate improved communication between clinicians across multiple disciplines who are involved in the diagnosis and treatment of lung cancer patients[58]. At a provincial level, establishing a centralized molecular testing registry accessible to all clinicians could facilitate treatment planning, expedite starting treatment, and reduce requests for repeat testing from different institutions.

Our education intervention established a multidisciplinary network of Ontario specialists that will serve as a foundation for further collaborative knowledge translation efforts. Although specific communication protocols might differ between individual institutions, it is clear that close collaboration between all clinicians involved in managing patients with advanced lung cancer will contribute to timely, coordinated molecular testing.

FUTURE DIRECTIONS

Rapid advances in lung cancer treatment demand continuous re-evaluation of current practices, predictive testing, and dissemination of updated best practices (Figure 2). An anticipated challenge is funding and incorporating next-generation molecular testing platforms into routine practice to allow for extensive molecular profiling beyond the context of a clinical trial. Another challenge will be facilitating repeat tumour sampling to identify resistance mutations when resistance to initial targeted therapy develops. Evaluation of circulating tumour DNA in peripheral blood is an example of a method that could potentially improve patient access to repeat molecular testing.
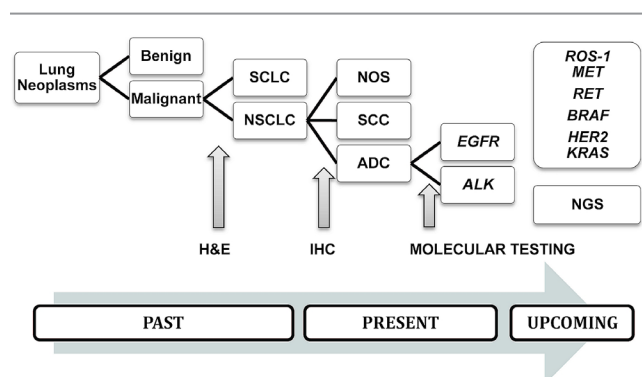

FIGURE 2 Historical lung cancer diagnosis pathway. Adapted with permission from Sekhon *et al.,* 2013[59]. H&E = hematoxylin and eosin; SCLC = small-cell lung cancer; NSCLC = non-small-cell lung cancer; IHC = immunohistochemistry; NOS = not otherwise specified; SCC = squamous cell carcinoma; ADC = adenocarcinoma; NGS = next-generation sequencing.

CONCLUSIONS

We implemented a locally developed and tailored knowledge dissemination strategy to address knowledge gaps for Ontario specialists about the importance of and requirements for molecular testing in lung cancer. Key themes that emerged from the intervention included optimizing sample acquisition through feedback to the clinicians obtaining diagnostic specimens, enabling pathologist-initiated reflex molecular testing, and enhancing interdisciplinary coordination at the local and provincial levels. The multidisciplinary network established by our initiative is well-positioned to facilitate future knowledge transfer and collaboration to improve the diagnosis and treatment of lung cancer in Ontario.

CONFLICT OF INTEREST DISCLOSURES
We have read and understood *Current Oncology*'s policy on disclosing conflicts of interest, and we declare the following interests: JCC reports personal fees and nonfinancial support from Pfizer, outside the submitted work. DMH reports personal fees from Pfizer and from Merck, outside the submitted work. SKR reports grants from AstraZeneca during the conduct of the study. REM reports personal fees from Boehringer Ingelheim, Eli Lilly, Pfizer, Novartis, AstraZeneca, and Roche, outside the submitted work. MST reports personal fees from Merck Canada, grants and personal fees from AstraZeneca, personal fees from Bristol–Myers Squibb, personal fees from Ventana Hoffmann–La Roche, and grants and personal fees from Pfizer Canada, outside the submitted work. NBL reports grants from the Ontario Institute of Cancer Research during the conduct of the study.

AUTHOR AFFILIATIONS
*Division of Medical Oncology, Princess Margaret Cancer Centre, University of Toronto, Toronto; †Department of Pathology and Laboratory Medicine, University of Ottawa, Ottawa; ‡Department of Pathology and Molecular Medicine, McMaster University, Hamilton; §Laboratory Medicine Program, University Health Network, University of Toronto, Toronto; ‖Molecular Diagnostics Laboratory, University Health Network, Toronto; #LifeLabs Genetics, Toronto; **Division of Thoracic Surgery, University of Toronto, Toronto; ††Division of Respirology, University of Toronto, Toronto; ‡‡Division of Respirology, Trillium Health Partners, Mississauga; §§Joint Department of Medical Imaging, University Health Network, Mount Sinai Hospital and Women's College Hospital, Toronto; ‖‖Department of Radiation Oncology, University of Toronto, Toronto; ##Simcoe Muskoka Regional Cancer Centre, Barrie; and ***Lung Cancer Canada, Toronto, ON.

REFERENCES
1. Canadian Cancer Society's Steering Committee on Cancer Statistics. *Canadian Cancer Statistics 2012.* Toronto, ON: Canadian Cancer Society; 2012.
2. Evans WK, Ung YC, Assouad N, Chyjek A, Sawka C. Improving the quality of lung cancer care in Ontario: the lung cancer disease pathway initiative. *J Thorac Oncol* 2013;8:876–82.
3. Sudhindra A, Ochoa R, Santos ES. Biomarkers, prediction, and prognosis in non-small-cell lung cancer: a platform for personalized treatment. *Clin Lung Cancer* 2011;12:360–8.
4. Inoue A, Kobayashi K, Usui K, *et al.* on behalf of the North East Japan Gefitinib Study Group. First-line gefitinib for patients with advanced non-small-cell lung cancer harboring epidermal growth factor receptor mutations without indication for chemotherapy. *J Clin Oncol* 2009;27:1394–400.
5. Shaw AT, Yeap BY, Solomon BJ, *et al.* Effect of crizotinib on overall survival in patients with advanced non-small-cell lung cancer harbouring *ALK* gene rearrangement: a retrospective analysis. *Lancet Oncol* 2011;12:1004–12.
6. Kwak EL, Bang YJ, Camidge DR, *et al.* Anaplastic lymphoma kinase inhibition in non-small-cell lung cancer. *N Engl J Med* 2010;363:1693–703.
7. Mok TS, Wu YL, Thongprasert S, *et al.* Gefitinib or carboplatin–paclitaxel in pulmonary adenocarcinoma. *N Engl J Med* 2009;361:947–57.
8. Rosell R, Carcereny E, Gervais R, *et al.* on behalf of the Spanish Lung Cancer Group in collaboration with Groupe Français de Pneumo-Cancérologie and the Associazione Italiana Oncologia Toracica. Erlotinib versus standard chemotherapy as first-line treatment for European patients with advanced *EGFR* mutation–positive non-small-cell lung cancer (eurtac): a multicentre, open-label, randomised phase 3 trial. *Lancet Oncol* 2012;13:239–46.
9. Fukuoka M, Wu YL, Thongprasert S, *et al.* Biomarker analyses and final overall survival results from a phase iii, randomized, open-label, first-line study of gefitinib versus carboplatin/paclitaxel in clinically selected patients with advanced non-small-cell lung cancer in Asia (ipass). *J Clin Oncol* 2011;29:2866–74.
10. Miller VA, Hirsh V, Cadranel J, *et al.* Afatinib versus placebo for patients with advanced, metastatic non-small-cell lung cancer after failure of erlotinib, gefitinib, or both, and one or two lines of chemotherapy (lux-Lung 1): a phase 2b/3 randomised trial. *Lancet Oncol* 2012;13:528–38.
11. Yu HA, Riely GJ. Second-generation epidermal growth factor receptor tyrosine kinase inhibitors in lung cancers. *J Natl Compr Canc Netw* 2013;11:161–9.
12. Yang JC, Sequist LV, Schuler MH, *et al.* Overall survival (os) in patients (pts) with advanced non–small cell lung cancer (nsclc) harboring common (Del19/L858R) epidermal growth factor receptor mutations (*EGFR* mut): pooled analysis of two large open-label phase iii studies (lux-Lung 3 [ll3] and lux-Lung 6 [ll6]) comparing afatinib with chemotherapy (ct) [abstract 8004^]. *J Clin Oncol* 2014;32:5s. [Available online at: http://meetinglibrary.asco.org/content/129081-144; cited 9 February 2017]
13. Sequist LV, Yang JC, Yamamoto N, *et al.* Phase iii study of afatinib or cisplatin plus pemetrexed in patients with metastatic lung adenocarcinoma with *EGFR* mutations. *J Clin Oncol* 2013;31:3327–34.
14. Shaw AT, Kim DW, Mehra R, *et al.* Ceritinib in *ALK*-rearranged non-small-cell lung cancer. *N Engl J Med* 2014;370:1189–97.
15. Lindeman NI, Cagle PT, Beasley MB, *et al.* Molecular testing guideline for selection of lung cancer patients for egfr and alk tyrosine kinase inhibitors: guideline from the College of American Pathologists, International Association for the Study of Lung Cancer, and Association for Molecular Pathology. *Arch Pathol Lab Med* 2013;137:828–60.
16. Cutz J, Craddock K, Torlakovic E, *et al.* Canadian anaplastic lymphoma kinase study: a model for multicenter standardization and optimization of *ALK* testing in lung cancer. *J Thorac Oncol* 2014;9:1255–63.
17. Leigh NB, Rekhtman N, Biermann WA, *et al.* Molecular testing for selection of patients with lung cancer for epidermal

growth factor receptor and anaplastic lymphoma kinase tyrosine kinase inhibitors: American Society of Clinical Oncology endorsement of the College of American Pathologists/International Association for the Study of Lung Cancer/Association for Molecular Pathology guideline. *J Clin Oncol* 2014;32:3673–9.

18. Ellis PM, Blais N, Soulieres D, *et al.* A systematic review and Canadian consensus recommendations on the use of biomarkers in the treatment of non-small cell lung cancer. *J Thorac Oncol* 2011;6:1379–91.

19. Jolly Graham A, Potti A. Translating genomics into clinical practice: applications in lung cancer. *Curr Oncol Rep* 2009;11:263–8.

20. Levy MA, Lovly CM, Pao W. Translating genomic information into clinical medicine: lung cancer as a paradigm. *Genome Res* 2012;22:2101–8.

21. Sawka C, Ross J, Srigley J, Irish J. The crucial role of clinician engagement in system-wide quality improvement: the Cancer Care Ontario experience. *Healthc Q* 2012;15:38–41.

22. Ionescu DN. Impact of the College of American Pathologists, the International Association for the Study of Lung Cancer, and the Association for Molecular Pathology clinical practice guidelines for *EGFR* and *ALK* testing in lung cancer in Canada. *Curr Oncol* 2013;20:220–6.

23. Zer A, Cutz JC, Sekhon HS, *et al.* A targeted intervention to improve awareness to molecular testing in nsclc [abstract 6547]. *J Clin Oncol* 2014;32:. [Available online at: http://meetinglibrary.asco.org/content/132268-144; cited 9 February 2017]

24. Murgu S, Colt H. Role of the pulmonologist in ordering post-procedure molecular markers in non-small-cell lung cancer: implications for personalized medicine. *Clin Lung Cancer* 2013;14:609–26.

25. Coghlin CL, Smith LJ, Bakar S, *et al.* Quantitative analysis of tumor in bronchial biopsy specimens. *J Thorac Oncol* 2010;5:448–52.

26. Reck M, Hermes A, Tan EH, Felip E, Klughammer B, Baselga J. Tissue sampling in lung cancer: a review in light of the merit experience. *Lung Cancer* 2011;74:1–6.

27. da Cunha Santos G, Saieg MA. Preanalytic parameters in epidermal growth factor receptor mutation testing for non–small cell lung carcinoma: a review of cytologic series. *Cancer Cytopathol* 2015;123:633–43.

28. Thunnissen E, Kerr KM, Herth FJ, *et al.* The challenge of nsclc diagnosis and predictive analysis on small samples. Practical approach of a working group. *Lung Cancer* 2012;76:1–18.

29. Karahalli E, Yilmaz A, Türker H, Ozvaran K. Usefulness of various diagnostic techniques during fiberoptic bronchoscopy for endoscopically visible lung cancer: should cytologic examinations be performed routinely? *Respiration* 2001;68:611–14.

30. Tanner NT, Pastis NJ, Sherman C, Simon GR, Lewin D, Silvestri GA. The role of molecular analyses in the era of personalized therapy for advanced nsclc. *Lung Cancer* 2012;76:131–7.

31. Eapen GA, Shah AM, Lei X, *et al.* on behalf of the American College of Chest Physicians Quality Improvement Registry, Education. Complications, consequences, and practice patterns of endobronchial ultrasound–guided transbronchial needle aspiration: results of the aquire registry. *Chest* 2013;143:1044–53.

32. Trisolini R, Cancellieri A, Tinelli C, *et al.* Rapid on-site evaluation of transbronchial aspirates in the diagnosis of hilar and mediastinal adenopathy: a randomized trial. *Chest* 2011;139:395–401.

33. Collins BT, Chen AC, Wang JF, Bernadt CT, Sanati S. Improved laboratory resource utilization and patient care with the use of rapid on-site evaluation for endobronchial

ultrasound fine-needle aspiration biopsy. *Cancer Cytopathol* 2013;121:544–51.

34. van der Heijden EH, Casal RF, Trisolini R, *et al.* on behalf of the World Association for Bronchology and Interventional Pulmonology, Task Force on Specimen Guidelines. Guideline for the acquisition and preparation of conventional and endobronchial ultrasound–guided transbronchial needle aspiration specimens for the diagnosis and molecular testing of patients with known or suspected lung cancer. *Respiration* 2014;88:500–17.

35. Lee HS, Lee GK, Lee HS, *et al.* Real-time endobronchial ultrasound–guided transbronchial needle aspiration in mediastinal staging of non–small cell lung cancer: how many aspirations per target lymph node station? *Chest* 2008;134:368–74.

36. Solomon SB, Zakowski MF, Pao W, *et al.* Core needle lung biopsy specimens: adequacy for *EGFR* and *KRAS* mutational analysis. *AJR Am J Roentgenol* 2010;194:266–9.

37. Tam AL, Kim ES, Lee JJ, *et al.* Feasibility of image-guided transthoracic core-needle biopsy in the battle lung trial. *J Thorac Oncol* 2013;8:436–42.

38. Moreira AL, Thornton RH. Personalized medicine for non-small-cell lung cancer: implications of recent advances in tissue acquisition for molecular and histologic testing. *Clin Lung Cancer* 2012;13:334–9.

39. Nasuti JF, Gupta PK, Baloch ZW. Diagnostic value and cost-effectiveness of on-site evaluation of fine-needle aspiration specimens: review of 5,688 cases. *Diagn Cytopathol* 2002;27:1–4.

40. Santambrogio L, Nosotti M, Bellaviti N, Pavoni G, Radice F, Caputo V. ct-guided fine-needle aspiration cytology of solitary pulmonary nodules: a prospective, randomized study of immediate cytologic evaluation. *Chest* 1997;112:423–5.

41. Garon EB, Rizvi NA, Hui R, *et al.* Pembrolizumab for the treatment of non-small-cell lung cancer. *N Engl J Med* 2015;372:2018–28.

42. Katsios C, Ziogas DE, Liakakos T, Zoras O, Roukos DH. Translating cancer genomes sequencing revolution into surgical oncology practice. *J Surg Res* 2012;173:365–9.

43. Evans WK, Ung YC, Assouad N, Chyjek A, Sawka C. Improving the quality of lung cancer care in Ontario: the lung cancer disease pathway initiative. *J Thorac Oncol* 2013;8:876–82.

44. Goss GD, O'Callaghan C, Lorimer I, *et al.* Gefitinib versus placebo in completely resected non-small-cell lung cancer: results of the ncic ctg br19 study. *J Clin Oncol* 2013;31:3320–6.

45. da Cunha Santos G, Lai SW, Saieg MA, *et al.* Cyto-histologic agreement in pathologic subtyping of non small cell lung carcinoma: review of 602 fine needle aspirates with follow-up surgical specimens over a nine year period and analysis of factors underlying failure to subtype. *Lung Cancer* 2012;77:501–6.

46. Li T, Kung HJ, Mack PC, Gandara DR. Genotyping and genomic profiling of non-small-cell lung cancer: implications for current and future therapies. *J Clin Oncol* 2013;31:1039–49.

47. Shiau CJ, Babwah JP, da Cunha Santos G, *et al.* Sample features associated with success rates in population-based *EGFR* mutation testing. *J Thorac Oncol* 2014;9:947–56.

48. Kim L, Tsao MS. Tumour tissue sampling for lung cancer management in the era of personalised therapy: what is good enough for molecular testing? *Eur Respir J* 2014;44:1011–22.

49. Sigel CS, Moreira AL, Travis WD, *et al.* Subtyping of non–small cell lung carcinoma: a comparison of small biopsy and cytology specimens. *J Thorac Oncol* 2011;6:1849–56.

50. Lindeman NI, Cagle PT, Beasley MB, *et al.* on behalf of the College of American Pathologists International Association for the Study of Lung Cancer and Association for Molecular Pathology. Molecular testing guideline for selection of lung cancer patients for egfr and alk tyrosine kinase inhibitors: guideline from the College of American Pathologists,

International Association for the Study of Lung Cancer, and Association for Molecular Pathology. *J Mol Diagn* 2013;15:415–53. [Erratum in: *J Mol Diagn* 2013;15:730]

51. Burlingame OO, Kessé KO, Silverman SG, Cibas ES. On-site adequacy evaluations performed by cytotechnologists: correlation with final interpretations of 5241 image-guided fine-needle aspiration biopsies. *Cancer Cytopathol* 2012;120:177–84.

52. Alsharif M, Carlo-Demovich J, Massey C, *et al.* Telecytopathology for immediate evaluation of fine-needle aspiration specimens. *Cancer Cytopathol* 2010;118:119–26.

53. Travis WD, Rekhtman N. Pathological diagnosis and classification of lung cancer in small biopsies and cytology: strategic management of tissue for molecular testing. *Semin Respir Crit Care Med* 2011;32:22–31.

54. Sterlacci W, Savic S, Schmid T, *et al.* Tissue-sparing application of the newly proposed IASLC/ATS/ERS classification of adenocarcinoma of the lung shows practical diagnostic and prognostic impact. *Am J Clin Pathol* 2012;137:946–56.

55. Ellis PM, Vandermeer R. Delays in the diagnosis of lung cancer. *J Thorac Dis* 2011;3:183–8.

56. Lim C, Tsao MS, Le LW, *et al.* Biomarker testing and time to treatment decision in patients with advanced nonsmall-cell lung cancer. *Ann Oncol* 2015;26:1415–21.

57. Wiesweg M, Ting S, Reis H, *et al.* Feasibility of preemptive biomarker profiling for personalised early clinical drug development at a comprehensive cancer center. *Eur J Cancer* 2013;49:3076–82.

58. Conron M, Phuah S, Steinfort D, Dabscheck E, Wright G, Hart D. Analysis of multidisciplinary lung cancer practice. *Intern Med J* 2007;37:18–25.

59. Sekhon HS, Souza CA, Gomes MM. Advances in cytopathology for lung cancer: the impact and challenges of new technologies. *Thorac Surg Clin* 2013;23:163–78.