Harmonization of PD-L1 testing in oncology: a Canadian pathology perspective

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ABSTRACT

Checkpoint inhibitors targeting the programmed cell death 1 protein (PD-1) and programmed cell death ligand 1 (PD-L1) are demonstrating promising efficacy and appear to be well tolerated in a number of tumour types. In non-small-cell lung cancer, head-and-neck squamous cell carcinoma, and urothelial carcinoma, outcomes appear particularly favourable in patients with high PD-L1 expression. However, assays for PD-L1 have been developed for individual agents, and they use different antibody clones, immunohistochemistry staining protocols, scoring algorithms, and cut-offs. Given that laboratories are unlikely to use multiple testing platforms, use of one PD-L1 assay in conjunction with a specific therapy will become impractical and could compromise treatment options. Methods to harmonize testing methods are therefore crucial to ensuring appropriate treatment selection. This paper focuses on lung, bladder, and head-and-neck cancer. It reviews and compares available PD-L1 testing methodologies, summarizes the literature about comparability studies to date, discusses future directions in personalized diagnostics, and provides a pathologist’s perspective on PD-L1 testing in the Canadian laboratory setting.

Key Words Immunotherapy, lung cancer, bladder cancer, PD-L1 assays, immunohistochemistry

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INTRODUCTION

The programmed cell death 1 protein (PD-1) is a checkpoint co-inhibitory receptor on the surface of immune cells that, under normal conditions, represents a negative feedback mechanism to switch T cell activation off, thereby protecting tissues from damage during immune system stimulation1. When activated through binding with one of its ligands, PD-L1 or PD-L2, PD-1 inhibits kinases involved in T cell activation through the phosphatase Shp2. Expression of PD-1 is high in CD4+Foxp3+ regulatory T cells, which are present in tumours in large numbers. In contrast, PD-L1 expression has been found in human tumour-associated antigen-presenting cells such as dendritic cells, macrophages, fibroblasts, and T cells2.

The induction of PD-L1 expression is controlled by cytokines such as interferon γ (IFNγ)2. Effector T cell IFNγ is thought to be responsible for high levels of PD-L1 expression in the tumour microenvironment, which occurs as a resistance mechanism after immune challenge. Other mechanisms of PD-L1 upregulation involve gene amplification and other cancer signalling pathways. Amplification of chromosome 9p24.1, which is responsible for PD-L1 and PD-L2, was observed in Hodgkin lymphoma and was subsequently detected in a subgroup of patients with gastric carcinoma, colon carcinomas, triple-negative breast cancers, and glioblastomas. In addition, signalling pathways thought to influence PD-L1 expression include PTEN deletions, PI3K or AKT mutations (or both), EGFR mutations, MYC overexpression, cyclin-dependent kinase 5 disruption, and an increase in PD-L1 transcripts3. A large proportion of tumour-infiltrating lymphocytes show PD-1 expression. By expressing PD-L1 to activate the PD-1 signalling cascade, cancer cells might evade immune-mediated destruction by tumour-infiltrating lymphocytes1. Novel checkpoint inhibitors that block the PD-1 pathway are therefore being developed to enhance antitumour immune functions in the treatment of cancer.

Several of the immune checkpoint inhibitors that have been developed have demonstrated remarkable clinical activity4. For the sake of brevity, the discussion here is limited to non-small-cell lung cancer (NSCLC), head-and-neck squamous cell carcinoma (HNSCC), and urothelial carcinoma (UC)—tumour types in which PD-L1 testing is currently necessary or provides important information to select patients who are most likely to benefit from checkpoint inhibitor therapies. Immune checkpoint therapy has, however, been used successfully in other
cancer types such as melanoma, Merkel cell carcinoma, Hodgkin lymphoma, and solid tumours with mismatch repair deficiency—uses that are not addressed further in this review.

To date, the PD-1 inhibitors nivolumab and pembrolizumab and the PD-L1 inhibitors atezolizumab, durvalumab, and avelumab have demonstrated promising efficacy and tolerability in the tumour types to be discussed here (supplementary Appendix A). Nivolumab and pembrolizumab have both been approved by Health Canada, conditional upon confirmatory results of further studies to verify clinical benefit in patients with incurable advanced-stage NSCLC who experience disease progression during or after platinum-containing chemotherapy. Nivolumab is also approved for the treatment of adult patients with recurrent or metastatic HNSCC. In addition, atezolizumab and durvalumab have been given marketing authorization, conditional on confirmatory results of further studies to verify clinical benefit for the treatment of locally advanced metastatic UC in patients with disease progression during or after platinum-containing chemotherapy or within 12 months of neoadjuvant or adjuvant treatment with platinum-containing chemotherapy. Of the foregoing agents, pembrolizumab for therapy of NSCLC requires that patients be PD-L1 positive, as determined using a validated test at a designated cut-off point for PD-L1 expression.

Given the likely future availability of additional PD-L1 inhibitors, PD-L1 assays will become increasingly important in identifying patients most likely to benefit from treatment. However, the PD-L1 assays for use with durvalumab, pembrolizumab, nivolumab, and atezolizumab have all been developed independently, and they use different antibody clones, immunohistochemistry (IHC) staining protocols, scoring algorithms, and cut-offs for determining PD-L1 status. Questions have therefore been raised about whether the tests can be used interchangeably to inform treatment decisions for the various PD-1 and PD-L1 inhibitors. Given the variability in PD-L1 testing paradigms and the lack of available resources to perform multiple tests within laboratories, harmonization of methods is crucial. In this paper, we review and compare available PD-L1 testing methods, summarize the literature about comparability studies to date, discuss future directions in personalized diagnostics, and provide a pathologist’s perspective on PD-L1 testing in the Canadian laboratory setting.

**COMPARING PD-L1 TESTING METHODS**

The PD-L1 assay is an IHC procedure in which the primary antibody recognizes and binds to a protein called the epitope, which is present in the tissue section. Several PD-L1 assays have been developed, each with a specific antibody that is designed to bind to a different epitope of PD-L1 and that was created to be used with a specific checkpoint inhibitor (Table 1). The methods differ in the choice of antibody clone, staining platform, and cut-off point for what is considered to be PD-L1-positive or -negative expression in NSCLC, HNSCC, and UC.

In addition to tests developed commercially (for example, PharmDx: Dako North America, Carpinteria, CA),...
U.S.A.), some diagnostic centres use laboratory-developed tests (LDTs) to reduce testing costs and to harmonize assays across disease sites. A LDT is any test that differs from the commercial assays, no matter how small the modification to one or more of its components or testing procedures. The LDTs use commercially available PD-L1 clones. Unfortunately, development of LDTs can be limited by an inability to standardize many of their components. As a result, they are likely to be less robust than the commercial tests and will introduce additional variability to results. Also, given that commercial tests have been examined within the context of clinical trials for specific checkpoint inhibitors, the clinical validity of LDT assays in regard to their ability to predict patient response is unclear. With the likely development of LDTs, external quality assessment (EQA) schemes for assurance of test quality in the clinical setting are an ongoing need.

Given variances between testing methods, PD-L1 assays are likely to differ in their classification of patients as PD-L1-positive or -negative. When different assays are used, patients might therefore be misclassified based on the method and scoring technique validated for a specific checkpoint inhibitor. As a consequence, patients might be inappropriately selected for or excluded from a specific treatment, potentially leading to unnecessary toxicities or exclusion from treatments that could improve outcomes. This misclassification might be further affected by the use of LDTs in which the variability of results is likely greater than that with commercial assays.

With the availability of checkpoint inhibitors that target the PD-1/PD-L1 pathway steadily increasing, robust methods that can identify the cohort of patients likely to respond to these agents is crucial. However, given that PD-L1 diagnostic assays have been developed independently for each agent, it is possible that several tests will become available in Canada. The use of multiple assays within individual laboratories is impractical given resource, platform (for example, Ventana (Ventana Medical Systems, Tucson, AZ, U.S.A.) vs. Dako (Dako North America, Carpinteria, CA, U.S.A.)), and tissue availability. To reduce the need for multiple tests and to provide testing options that are compatible with available platforms, an examination of the compatibility of testing assays is therefore important to determine whether those assays can be used interchangeably. That examination of compatibility is crucial to providing accurate and reliable tools that will optimize treatment decisions for patients.

**PD-L1 ASSAY COMPARABILITY STUDIES**

**Comparability Studies in NSCLC**

A number of studies have looked at the comparability of PD-L1 assays in NSCLC (Table 1). Of nine studies examining the 28-8, 22C3, and SP263 assays, all showed high levels of agreement in the ability of the assays to detect PD-L1 in tumour cells (tc). However, the level of agreement might depend on the cut-off points used. Of seven studies that included the SP142 assay, six showed lower agreement in comparisons with other antibodies such as 28-8, 22C3, and SP263. In addition, preliminary results from phase 2 of the Blueprint study, which also included the recently developed 73-10 assay, showed that more tc s were stained positive by the 73-10 assay than by the SP263, 28-8, and 22C3 assays, and confirmed that SP142 detected fewer positive tc s. The analytic similarity shown between the SP263, 28-8, and 22C3 assays suggests that those tests could be used interchangeably at appropriate cut-off points. However, the variability in both immune cell (ic) and tc staining with the SP142 assay suggests that it is not comparable to other assays.

Thus far, three studies have examined the interchangeability of LDTs with commercial assays. A study by Scheel et al. showed that staining in 6 of 11 LDT protocols was similar to that in the 22C3 and 28-8 assays. In addition, results of an ongoing retrospective study by Velcheti et al., which included a total of 1728 test results (323 from LDTs), demonstrated no difference in the measurement of PD-L1 expression using LDTs compared with other assays. Moreover, results of an ongoing study by Adam et al. showed that LDTs have various levels of agreement when compared with 3 commercial assays. Notably, LDTs using the SP263 clone had the greatest agreement across all platforms for both ic and tc staining, whereas some LDTs with clones 28-8, 22C3, and E1L3N, but not SP142, showed good correlation with the 3 commercial assays for tc s only. Interestingly, NordiQC, a leading European organization managing EQA for IHC, has used the 22C3, 28-8, and SP263 PharmDx assays, as well as LDTs, to assess 68 laboratories. Results of a pilot survey demonstrated a pass rate of 80% for the 3 PharmDx assays; however, the pass rate for LDTs was only 20%. Those results suggest that LDTs might be more variable than PharmDx assays and must be carefully validated, with adequate training and EQA schemes.

An important finding of studies into the interpretation of PD-L1 tests in NSCLC is that the variability of staining appears to be higher for ic s than for tc s. A study by Rehman et al. confirmed that finding, demonstrating 94% agreement between pathologists for tc staining, but only 27% agreement for stromal or ic assessment. However, the biologic implication of ic staining does not seem to be as important in NSCLC as in other tumours; it might depend on the assay and the specific checkpoint inhibitor in question. Therefore, it is not necessary that assays for NSCLC measure PD-L1 expression in ic s.

**Comparability Studies in HNSCC and UC**

In HNSCC, one comparability study is ongoing in 501 archival clinical HNSCC tumour samples, with interim results presented at the 2016 European Society for Medical Oncology meeting. As in NSCLC, preliminary results in uc showed good agreement between the 22C3, 28-8, and SP263 assays, with much lower agreement for SP142. Also, as happened for NSCLC, increased variability between pathologists was observed in the assessment of ic s. However, as in NSCLC, the biologic implications of ic staining do not seem to be as important in HNSCC as in other tumours. Analyses of uc tissue, using SP142 on the Ventana platform, have shown that PD-L1 is expressed more commonly on ic s than tc s, suggesting that ic PD-L1 expression could be more relevant in uc than in NSCLC. With the exception of the 28-8 assay, commercial assays tend to use ic s as components of their algorithms (Table 1).
However, the SP142 assay assesses only ICs, whereas the SP-263, 22C3, and 73-10 assays include both ICs and TCS. Another key difference with the SP142 assay is that it calculates PD-L1–expressing ICs in the tumour area only; other assays use the IC area as a whole. It is therefore likely that the SP142 and 28-8 assays could not be used interchangeably with the other assays in UC. In addition to the variability in testing compartments, commercial assays also differ in their scoring algorithms in UC. For example, the 22C3 assay uses a combined positive score,

### TABLE II: PD-L1 comparability studies in non-small-cell lung cancer

| Study                        | Assays          | Samples (n) | Findings                                                                 |
|------------------------------|-----------------|-------------|---------------------------------------------------------------------------|
| Gaule et al., 2016<sup>26</sup> | 28-8, 22C3, SP142, E1L3N, 9A11 | 30          | When using chromogenic staining in isogenic cell lines, high levels of agreement were observed for all antibodies ($R^2=0.76–0.99$). |
| Adam et al., 2017<sup>27</sup> (French study) | 28-8, 22C3, SP263, LDTs | 41          | Assays 22C3, 28-8, SP263 performed in several centres were highly concordant. Using a 4-category scale with 1%, 5%, and 10% thresholds, immune cell staining agreement was low. Laboratory-developed tests demonstrated variable levels of agreement, with SP263 being most concordant. |
| Brunnstrom et al., 2017<sup>28</sup> (Swedish study) | 28-8, 22C3, SP263, SP142 | 55          | The highest values for comparisons were seen between 22C3 and 28-8; the lowest were seen between SP142 and the other assays. Agreement was fairly good for 28-8, 22C3, and SP263, but not as good for SP142. |
| Hirsch et al., 2017<sup>29</sup> (Blueprint 1) | 28-8, 22C3, SP263, SP142 | 39          | The percentage of PD-L1–stained tumour cells was comparable with 22C3, 28-8, and SP263, but the SP142 assay showed fewer stained tumour cells. Variability of staining was higher for immune cells than for tumour cells. |
| Rimm et al., 2017<sup>30</sup> (Bristol–Myers Squibb, NCCN) | 28-8, 22C3, SP142, E1L3N | 90          | SP142 was an outlier, detecting significantly less PD-L1 expression in tumour cells and immune cells. Compared with either 28-8 or E1L3N, 22C3 showed slightly, yet statistically significantly, lower staining. The immune cell score showed poor agreement with any antibody. |
| Saito et al., 2017<sup>31</sup> | 22C3, 28-8 | 420         | Percentage agreement between assays was fair. Positive agreement was suboptimal when the cut-off was ≥25% and ≥50%. |
| Scheel et al., 2017<sup>32</sup> | 28-8, 22C3, SP263, SP142, LDTs | 21          | The SP142 staining pattern was distinct. Staining with 22C3, 28-8, SP263 was similar. Of 11 laboratory-developed tests, 6 showed staining similar to that with 22C3 and 28-8. |
| Scott et al., 2017<sup>33</sup>, Ratcliffe et al., 2017<sup>18</sup> (AstraZeneca study) | 28-8, 22C3, SP263, SP142, LDTs | 493         | SP263, 22C3, and 28-8 showed strong agreement in immune cell scoring. Immune cell agreement and PD-L1 staining or agreement were both less with SP142 than with SP263. |
| Tsao et al., 2017<sup>24</sup> (Blueprint 2, abstract) | 28-8, 22C3, SP263, SP142, 73-10 (from routine practice) | 81          | Results of Blueprint phase 1 were affirmed. The 22C3, 28-8, and SP263 assays are comparable; SP142 detects fewer cells, and 73-10 stains more PD-L1–positive tumour cells. Variability of staining was higher for immune cells than for tumour cells. |
| Velchetti et al., 2017<sup>14</sup> | 28-8, 22C3, SP142, LDTs | 1728        | No differences in PD-L1 expression using 22C3, 28-8, and laboratory-developed tests. SP142 was an outlier. |

NCCN = U.S. National Comprehensive Cancer Network.
which does not capture PD-L1 expression in tissue alone\textsuperscript{21}. Thus far, no published data from comparability studies in tissue are available. However, interim data from an ongoing study, sponsored by AstraZeneca and presented at the Society for Immunotherapy of Cancer 2017 annual meeting, showed a high level of agreement between the SP263, 22C3, and 28-8 assays for both tissue and cell staining\textsuperscript{49}. The SP142 assay showed good agreement with respect to cell staining, but significant differences for tissue staining. In addition, ongoing work by author MRD and colleagues has shown substantial inter-observer agreement in the interpretation of tissue staining in bladder using both SP263 and SP142 (data on file).

**FUTURE DIRECTIONS**

The use of PD-L1 expression as a biomarker of response is important in identifying patients who could obtain a clinically meaningful benefit from treatment with a checkpoint inhibitor. It is clear from clinical trials in non-small cell lung cancer, head and neck squamous cell cancer, and UC that responses to these inhibitors are improved in patients having PD-L1–positive tumours compared with patients having PD-L1–negative tumours\textsuperscript{40} (supplementary Appendix A). However, when checkpoint inhibitors are used in combination with agents involving non-redundant pathways, good responses have also been demonstrated in patients with low PD-L1 expression\textsuperscript{41,42}. Monotherapy might therefore be optimal for PD-L1–positive patients, with the use of immunotherapy combinations or alternative therapies being appropriate for those with low PD-L1 expression. The benefit of improved efficacy with combination therapies in PD-L1–negative patients will have to be weighed against an increased risk of toxicity. Those outcomes suggest that PD-L1 testing could aid in identifying patients who are most likely to respond to monotherapy with checkpoint inhibitors, thus aiding in the selection of the most appropriate therapy, optimizing resource utilization and fiscal expenditure, and minimizing treatment toxicities. In addition, the role of PD-L1 testing will likely extend beyond non-small cell lung cancer, head and neck squamous cell cancer, and UC, with the U.S. Food and Drug Administration having recently approved pembrolizumab for previously treated patients with recurrent locally advanced or metastatic gastric or gastroesophageal junction cancers whose tumours express PD-L1 as determined by the 22C3 PharmDx assay\textsuperscript{43}.

Currently, the process of validating and standardizing LDTs is difficult and laborious. However, standardization arrays using cell lines could allow laboratories to check assays to ensure that they are able to correctly quantify PD-L1 based on known limits of detection, quantification, and saturation. In addition, a framework being developed by the Canadian Partnership Against Cancer for molecular biomarker testing will recommend that testing laboratories undergo accreditation, participate in EQA schemes, and produce standard operating procedures for all tests. The Canadian Cancer Research Alliance is also developing a national network of molecular diagnosis and pathology groups that could be used to support translational research and personalized medicine.

In the future, combining PD-L1 IHC with T cell activation measures might boost the ability to predict patient response to checkpoint inhibitors. New testing paradigms that include multiplex gene expression markers, including PD-L1, are being developed by NanoString Technologies (Seattle, WA, U.S.A.) and might provide additional testing options\textsuperscript{44}. However, whether these newer paradigms will be superior to currently available IHC assays is unclear. Finally, microsatellite instability, mismatch repair deficiency, and tumour mutational burden are being targeted as genomic features in certain cancers that could be responsive to checkpoint inhibitors. Assays that identify patients with those genomic features might replace or complement PD-L1 immuno-expression. To date, PD-L1 testing is the most evaluated and accepted methodology used in selecting patients for checkpoint inhibitors in non-small cell lung cancer, head and neck squamous cell cancer, and UC.

**CANADIAN PERSPECTIVE ON PD-L1 TESTING**

The SP142 assay developed by Ventana, used according to its published protocol, differs significantly from others, given its distinct staining pattern showing both membranous and granular cytoplasmic staining in tissues\textsuperscript{35}. Studies to date in both non-small cell lung cancer and head and neck squamous cell cancer have demonstrated good comparability between the 22C3, 28-8, and SP263 assays, with the SP142 assay representing a clear outlier. In addition, preliminary results of the Blueprint 2 study demonstrated that the 73-10 assay shows greater staining of tissues in non-small cell lung cancer\textsuperscript{24}. Because the goal is to provide interchangeable PD-L1 assays, SP142 should not, given its low agreement, be used in place of other methods, with the exception of use for atezolizumab. Results of comparability studies, at least in non-small cell lung cancer and head and neck squamous cell cancer, suggest that the 22C3, 28-8, and SP263 assays can be used interchangeably based on their relevant cut-offs. The choice of assay should therefore be based on the platform and the availability of pathologists who are trained in interpreting the tests. The reliability of assay interpretation should also be considered when adopting a standard method.

As for any biomarker, a greater volume of samples allows laboratories to gain experience, becoming proficient in testing methods. Consequently, it is important that testing be conducted at sites that will receive a reasonable volume of samples. Given the high prevalence of lung cancer, it is anticipated that testing centres will receive a large volume of samples and that the use of PD-L1 testing will expand beyond its current indication. In addition, given differences in the testing methods by disease site, it is important that assays are interpreted by specialized pathologists with training in PD-L1 interpretation. At least initially, testing should be conducted centrally so that experience can be gained through the analysis of a large volume of samples. Centralized testing would also allow for standardization in recording PD-L1 results. One concern with non-small cell lung cancer samples is that, because of the heterogeneity of PD-L1 expression in a tumour and the minimal requirement for the presence of only 50 tumour cells (SP142) or 100 tumour cells (22C3 and 28-8) for the test\textsuperscript{19}, PD-L1 results from small biopsy or resection specimens might be discordant, with the concordance between them varying in the 52%–92% range\textsuperscript{3,45}. However, most non-small cell lung cancer patients are diagnosed at an advanced stage and are not treated with surgery; only biopsy or cytology specimens are available for ancillary testing, including PD-L1. Moreover, in clinical trials,
approximately 10% of nsclc patients reported as PD-L1–negative responded to immunotherapy19 (supplementary Appendix A). It is therefore likely that a fraction of patients are miscategorized as PD-L1–negative and might otherwise have responded to immunotherapy. To optimally treat such patients, testing methods that are more sensitive are therefore needed to identify PD-L1 expression from smaller samples.

Although oncologists might prefer to have access to the exact percentage of tumour cells expressing PD-L1, that result is likely less accurate than results reported within a broader category. As a compromise, pathologists might consider reporting the tumour proportion score within the comments section. It is also important for oncologists to specify the checkpoint inhibitor that they would like to choose for their patient, given that testing labs might not know which drug will be prescribed.

It is clear that even if testing is conducted centrally, testing sites will not have access to both the Dako and Ventana platforms. It is therefore crucial that there be a choice of assays within each tumour type, allowing for testing on either platform. In Europe, the Ventana platform is the most widely used, which poses a challenge for PD-L1 testing related to pembrolizumab. The Conformité Européenne designation has been awarded to SP263, which now allows the assay to be used in determining therapy with pembrolizumab and nivolumab in addition to durvalumab. To date, Health Canada has approved checkpoint inhibitors with the use of any validated assay. Consequently, there will be flexibility to use methods that are interchangeable across platforms. It is also clear that one specific assay and scoring algorithm will not be valid for multiple disease states. It will therefore be necessary to perform comparability studies in each tumour type to determine which assays can be used interchangeably. Ideally, the same antibodies could then be used across disease sites, thereby avoiding confusion for oncologists and pathologists.

Given the complexities of the testing methods and the need for EQA schemes to ensure the validity of LDRs, it is important to select commercial tests initially while centres gain experience. Over time, LDRs could be developed and validated, but further comparability studies are needed to ensure that they are interchangeable with the commercial tests. Academic laboratories with a large volume of samples might be able to research LDRs in their own populations treated with various checkpoint inhibitors to clinically validate those assays. In that context, it is important that LDRs be undertaken with caution, given their increased variability and lack of robustness. Ultimately, LDRs are useful only if they provide a more economical method of assessing PD-L1 status. It is possible that LDRs could prove cheaper than commercial assays, given their ability to test any volume of samples in one run. Larger runs might be of benefit to smaller laboratories; however, it is unclear whether smaller centres would receive sufficient samples to ensure the gain of sufficient technical and professional experience.

The reliability of PD-L1 scoring is also likely to be hindered by variability in interpretation between pathologists, which appears to be greater for ic than for tc staining. Increased variability is therefore probable for assays containing an ic component. For tumour types such as nsclc and hnscc, in which the biologic implications of the ic staining component is not as important, assays that focus on the ic component might produce less variability between pathologist readings. However, for uc, in which the ic component is more important, wider variability between pathologist readings is likely. Education and experience are therefore needed for improvement in the interpretation of ic staining and for ongoing assessment of intra- and inter-observer variability. Although PD-L1 tests are conducted using individual assays, sufficient comparability studies will have to be performed to permit the identification of a reasonable biomarker approach that can be implemented in a responsible and practical way across available platforms at the key testing sites.

**SUMMARY**

The number of PD-L1 inhibitors available for the treatment of nsclc, hnscc, and uc is growing, and indications are likely to expand to additional tumour types. However, PD-L1 assays have been developed independently for each agent and vary in their make-up. Use of a single PD-L1 assay in conjunction with a specific therapy will become impractical and could compromise treatment options for patients, driving the need for a reduction to the fewest possible tests. Test selection could then be based on the platform and the availability of pathologists trained in interpretation. Approaches to harmonizing testing methods are therefore crucial in ensuring appropriate treatment selection for Canadian patients.

Results of comparability studies, at least in nsclc and hnscc, suggest that the 22C3, 28-8, and SP263 assays can be used interchangeably at appropriate cut-offs. However, further studies are needed to determine the interchangeability of LDRs and to harmonize assays for other tumour types. A standardized plan has to be developed to allow for the rollout of new testing paradigms that considers the expanding need for PD-L1 testing. A reasonable approach could be to centralize testing using specialized pathologists. Commercial tests should be used until LDRs are validated with sufficient EQA. Eventually, the goal should be to develop a rational, cost-effective, and universal approach that can be applied in multiple indications to meet the needs of Canadian patients.

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**CONFLICT OF INTEREST DISCLOSURES**

We have read and understood *Current Oncology’s* policy on disclosing conflicts of interest, and we declare the following interests: DNI has received honoraria and served on advisory boards for Merck, Bristol–Myers Squibb, Pfizer, AstraZeneca, and Hoffmann–La Roche. MRD has received compensation from Hoffmann–La Roche and AstraZeneca for participating on advisory boards and has received honoraria from AstraZeneca. MST has received honoraria for advisory board meetings from Merck, AstraZeneca, and Bristol–Myers Squibb, and a research grant from Hoffmann–La Roche and Ventana.
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