SUMMARY OF SAFETY AND EFFECTIVENESS DATA (SSED)

I. GENERAL INFORMATION

Device Generic Name:	In vitro diagnostic immunohistochemistry (IHC) for detection of PD-L1 protein in formalin-fixed, paraffin-embedded (FFPE) human tissue sections
Device Trade Name:	PD-L1 IHC 28-8 pharmDx
Device Procode:	PLS
Applicant's Name and Address:	Dako North America, Inc. 6392 Via Real Carpinteria, CA 93013
Date(s) of Panel Recommendation:	None
Premarket Approval Application (PMA) Number:	P150025/S003

Date of FDA Notice of Approval: September 15, 2017

The original PMA (P150025) for PD-L1 IHC 28-8 pharmDx intended for use in detecting PD-L1 protein in non-squamous non-small cell lung cancer (NSCLC) was approved on October 9th 2015. A second PMA (P150027) for the same device was approved for use in detecting PD-L1 protein in melanoma on January 22, 2016. The SSED for each indication is available on the CDRH website and is incorporated by reference here. The current supplement (S003) was submitted to expand the intended use for the PD-L1 IHC 28-8 pharmDx to include detection of PD-L1 protein in squamous cell carcinoma of head and neck (SCCHN) and urothelial carcinoma (UC).

II. INDICATIONS FOR USE

For in vitro diagnostic use.

PD-L1 IHC 28-8 pharmDx is a qualitative immunohistochemical assay using Monoclonal Rabbit Anti-PD-L1, Clone 28-8 intended for use in the detection of PD-L1 protein in formalin-fixed, paraffin-embedded (FFPE) non-squamous non-small cell lung cancer (NSCLC), squamous cell carcinoma of the head and neck (SCCHN), urothelial carcinoma (UC), and melanoma tissues using EnVision FLEX visualization system on Autostainer Link 48. PD-L1 protein expression is defined as the percentage of evaluable tumor cells exhibiting partial or complete membrane staining at any intensity. Tumor PD-L1 status is defined by indication specific staining interpretation.

Tumor Indication*	Intended Use	PD-L1 Expression Clinical Cut off
nsNSCLC	PD-L1 expression as detected by PD-L1 IHC 28-8 pharmDx in non-squamous NSCLC and SCCHN may be associated with enhanced survival from OPDIVO® (nivolumab).	≥1%, ≥5%, ≥10%
SCCHN		≥1%
UC	PD-L1 expression as detected by PD-L1 IHC 28-8 pharmDx in UC may be associated with enhanced response rate from OPDIVO®.	≥1%
Melanoma	Positive PD-L1 status as determined by PD-L1 IHC 28-8 pharmDx in melanoma is correlated with the magnitude of the treatment effect on progression-free survival from OPDIVO®.	≥1%

*For details on staining interpretation, refer to section 13 of the package insert and indication specific PD-L1 IHC 28-8 pharmDx Interpretation Manuals.

III. CONTRAINDICATIONS

There are no known contraindications for the use of this test.

IV. WARNINGS AND PRECAUTIONS

The warnings and precautions can be found in the PD-L1 IHC 28-8 pharmDx product labeling.

V. DEVICE DESCRIPTION

PD-L1 IHC 28-8 pharmDx contains optimized reagents required to complete an immunohistochemical staining procedure for formalin-fixed and paraffin-embedded (FFPE) specimens using the Dako Autostainer Link 48 automated staining system and the EnVision FLEX visualization system. The principle component of the kit is the rabbit monoclonal anti PD-L1 clone 28-8 antibody that binds to PD-L1 protein expressed on FFPE tissue. The kit includes reagents required for pre-treatment of tissue, secondary antibodies, amplification and detection reagents that are all manufactured by Dako. PD-L1 IHC 28-8 pharmDx includes reagents sufficient to perform 50 tests in up to 15 individual runs and instructions for use (IFU). A total of 28 vials containing reagents including the EnVision FLEX visualization system, and 15 cell line control slides are provided in each kit. SeeTable 1, below.

Table 1: Overview of PD-L1 IHC 28-8 pharmDx Components

Component	Component Description
Peroxidase-Blocking Reagent	Buffered solution containing hydrogen peroxide, detergent and 0.015 mol/L sodium azide.
Primary Antibody: Monoclonal Rabbit anti-PD-L1, Clone 28-8	Monoclonal rabbit anti-PD-L1 in a buffered solution, containing stabilizing protein, and 0.015 mol/L sodium azide.
Negative Control Reagent (NCR)	Monoclonal rabbit control IgG antibody in a buffered solution, containing stabilizing protein, and 0.015 mol/L sodium azide.
Linker, anti-Rabbit	Mouse secondary antibody against rabbit immunoglobulins in a buffered solution containing stabilizing protein and 0.015 mol/L sodium azide.
Visualization Reagent- HRP	Dextran coupled with peroxidase molecules and goat secondary antibody molecules against rabbit and mouse immunoglobulins in a buffered solution containing stabilizing protein and an antimicrobial agent.
DAB+ Substrate Buffer	Buffered solution containing hydrogen peroxide and an antimicrobial agent.
DAB+ Chromogen	3,3'-diaminobenzidine tetrahydrochloride in an organic solvent.
DAB Enhancer	Cupric sulfate in water.
EnVision™ FLEX Target Retrieval Solution, Low pH, 50x	Buffered solution, pH 6.1, containing detergent and an antimicrobial agent (EnVision FLEX Target Retrieval Solution, Low pH, 50X).
PD-L1 IHC 28-8 pharmDx Control Slides	Each slide contains sections of two pelleted, formalin-fixed paraffin-embedded cell lines: NCI-H226 with moderate PD-L1 protein expression and MCF-7 with negative PD-L1 protein expression (PD-L1 IHC 28-8 pharmDx Control Slides).

Device Instrumentation and Software

PD-L1 IHC 28-8 pharmDx assay is performed on the Dako Autostainer Link 48 automated staining system using the DakoLink software (version 4.0.3). The Autostainer system is designed to mimic the staining steps performed manually by a lab technician. The PD-L1 IHC 28-8 pharmDx protocol is assay specific. The DakoLink software has been designed to recognize and group PD-L1 IHC 28-8 pharmDx reagents, requiring that all system reagents are used together. Deparaffinization, rehydration and target retrieval (3-in-1) procedures are performed in the PT Link Pre-treatment module.

Specimen Preparation

FFPE specimens must be processed appropriately to preserve the tissue for IHC staining. Handling and processing instructions recommended in the labeling are : < 30 minutes ischemia time prior to immersion in fixative, and 24-48 hours fixation time in neutral buffered formalin. Alternative fixative methods have not been validated and may give erroneous results. Tissue specimens should be cut into sections of 4-5 µm, mounted on charged slides and stored in the dark at 2-8 °C until staining, which should be performed within 4 months of sectioning.

Test Controls and Calibrators

Run controls are included in each staining run to establish the validity of the test results. The device labeling recommends the following controls to be run with the assay:

1) Control cell line slides provided as part of the kit should be used to verify the staining procedure. One Control Slide should be stained with the primary antibody to PD-L1 in each staining run. The evaluation of the Control Slide cell lines supplied in the kit indicates the validity of the staining run. The Control Slides should not be used as an aid in interpretation of patient results.

2) Positive run controls are to be provided by the end-user laboratory. It is recommended that tissue that stains weakly for PD-L1 is used to monitor for all aspects of pre-analytical variables such as fixation, processing and sectioning. Negative control tissue is required to detect unintended antibody cross reactivity to tissue and is expected to be negative for PD-L1 expression. Tissue features located in patient tissue or positive control tissue that is not expected to show PD-L1 expression may be used as negative control for staining.

3) The Kit includes a Negative Control Reagent that is used in parallel with the PD-L1 Clone 28-8 primary antibody on patient tissue. The matched negative control aids the reader in differentiating a true signal from tissue-specific background staining that occurs from reaction with detection chemistry and not the anti PD-L1 primary antibody.

Additional information about the use of controls is available in the product labeling.

Principle of Operation

Patient FFPE tissue specimens are subject to deparaffinization, rehydration, and target retrieval in the Target Retrieval Solution (3-in-1) for 20 minutes at 97 °C in the PT Link instrument to expose the PD-L1 antigen if present on the tissue. The slides are then loaded onto the Autostainer Link 48 automated stainer and incubated with the primary monoclonal antibody to PD-L1 (Clone 28-8) or the Negative Control Reagent. This step enables binding of the primary antibody to tumor tissue section when PD-L1 antigen is expressed. The slides are then incubated with an anti-Rabbit linker antibody, specific to Fc region of the primary antibody. Following this, the slides are incubated with a ready-to-use Visualization Reagent consisting of secondary antibody molecules and horseradish peroxidase molecules coupled

to a dextran polymer backbone. The enzymatic conversion of the subsequently added DAB+ Chromogen results in precipitation of a visible reaction product at the antigen sites. The color of the chromogenic reaction is modified by a chromogen enhancement reagent, DAB Enhancer. The specimens are then counterstained with hematoxylin and cover-slipped and observed under a microscope to visually determine if the PD-L1 protein is expressed in patient tumor tissue.

Staining Protocol:
The staining protocol for PD-L1 IHC 28-8 pharmDx on the Dako Autostainer Link 48 is as follows:
Rinse in buffer
Peroxidase-Blocking Reagent (2 x 150 µL): 5 minutes
Rinse in buffer
Monoclonal Rabbit anti-PD-L1 (or Negative Control Reagent) (2 x 150 µL): 30 minutes
Rinse in buffer
Linker Reagent (2 x 150 µL): 30 minutes
Rinse in buffer
Visualization Reagent-HRP (2 x 150 µL): 30 minutes
Rinse in buffer
Rinse in buffer: 5 minutes
DAB+ solution (2 x 150 µL): 2 x 5 minutes
Rinse in buffer
DAB Enhancer (2 x 150 µL): 5 minutes
Rinse in buffer
Hematoxylin (2 x 150 µL): 7 minutes
Rinse in deionized water
Rinse in buffer: 5 minutes
Rinse in deionized water

Interpretation of PD-L1 Staining in SCCHN and UC

The device labeling states that interpretation of specimens should be performed by a pathologist using a light microscope. For evaluation of the PD-L1 immunohistochemical staining and scoring, 4x objective magnification can be used for initial assessment of the entire specimen, followed by the 10-20x objectives for scoring (40x can be utilized for confirmation if needed). PD-L1 staining is indicated with a brown (3, 3'-diaminobenzidine, DAB) reaction product. All viable tumor cells on the entire PD-L1 stained patient slide must be evaluated and included in the PD-L1 scoring assessment. A minimum of 100 viable tumor cells should be present in the PD-L1 stained patient slide to determine the percentage of stained cells. The specimen is considered PD-L1 positive if ≥ 1% of cancer cells exhibit circumferential and/or partial linear plasma membrane PD-L1 staining of tumor cells at any intensity. Specimen is considered PD-L1 negative if < 1% of cancer cells exhibit circumferential and/or partial linear plasma membrane PD-L1 staining of tumor cells at any intensity.

Cytoplasmic staining, if present, is not considered for scoring purposes. Non-malignant cells and immune cells (e.g., infiltrating lymphocytes or macrophages) may also stain with PD-L1; however, these should not be included in the scoring for the determination of PD-L1

positivity.

VI. ALTERNATIVE PRACTICES AND PROCEDURES

There are no other FDA-cleared or approved alternatives for the testing of PD-L1 expression in FFPE SCCHN or UC human specimens to assess association to improved benefit for treatment with nivolumab.

VII. MARKETING HISTORY

PD-L1 IHC 28-8 pharmDx has been marketed in the United States following FDA approval of the device on October 9, 2015, for determining PD-L1 expression in FFPE tumor tissue from patients with advanced non-squamous NSCLC for whom OPDIVO ® treatment is considered. Subsequent to approval on January 23, 2016 the device has been marketed for melanoma indication in addition to the NSCLC indication. The device has not been withdrawn from marketing for any reason related to its safety or effectiveness

VIII. POTENTIAL ADVERSE EFFECTS OF THE DEVICE ON HEALTH

Failure of the device to perform as expected or failure to correctly interpret test results may lead to incorrect PD-L1 test results and subsequently improper interpretation of the benefit/risks for patients who are considering treatment with OPDIVO®.

For the specific adverse events that occurred in the clinical studies, please see Section X below.

IX. SUMMARY OF NONCLINICAL STUDIES

A. Laboratory Studies

Preclinical studies were performed using the PD-L1 IHC 28-8 pharmDx kit to establish analytical performance of device. These studies were conducted to characterize the assay, demonstrate the impact of pre-analytical variables on assay performance, verify precision and robustness of the assay, and establish assay stability. The study results detailed below establish sensitivity, specificity, precision and reproducibility of the device.

The scoring algorithm used in these studies included a clinical score (i.e., PD-L1 positive or negative) and/or an analytical quality score (0-3 scale for staining signal intensity). Clinical scores were recorded for all studies with the scoring algorithm developed for clinical interpretation of the PD-L1 28-8 IHC Assay. Analytical scores were assessed for some studies to evaluate assay performance across the range of staining including borderline cases. The analytical score is not part of the interpretation of PD-L1 staining status in the device labeling. Analytical validation of UC and SCCHN was performed in independent studies. Assay precision for SCCHN indication leveraged data from analytical validation submitted under P150025 and P150027 and results from abbreviated precision and reproducibility studies is summarized in the sections below.

1. Analytical Specificity

Assessment of analytical specificity of the primary antibody for PD-L1 IHC 28-8 pharmDx- the monoclonal rabbit anti -human PD-L1 clone 28-8 antibody- was reviewed under P150025 and P150027. Refer to the SSED associated with P150025 or P150027 for study design and results.

2. Analytical Sensitivity

SCCHN

Analytical sensitivity of PD-L1 IHC 28-8 pharmDx was tested on 236 unique SCCHN FFPE cancer tissues using one lot of the device. The specimens were chosen at random and represented the full range of PD-L1 expression and tumor stage. PD-L1 expression in tumor specimens ranged from 0-100% and 0-3 staining intensity. The number of PD-L1 positive cases at the ≥1% expression level was 104 (44%).

UC

Analytical sensitivity of PD-L1 IHC 28-8 pharmDx was tested on 138 unique UC FFPE cancer tissues using one lot of the device. The specimens were chosen at random and represented the full range of PD-L1 expression and tumor stage. PD-L1 expression in tumor specimens ranged from 0-100% and 0-3 staining intensity. The number of PD-L1 positive cases at the ≥1% expression level was 56 (41%).

3. Repeatability

The objective of this study was to demonstrate that PD-L1 IHC 28-8 pharmDx would produce consistent staining in normal day-to-day testing with multiple lots of test kit.

SCCHN

The study was performed with 39 SCCHN specimens spanning the range of PD-L1 expression, and 4 of these specimens represented specimens around cut off. The specimens along with control slides were stained, then blinded and randomized prior to evaluation of PD-L1 expression status. Specimens were determined to be positive if PD-L1 expression was ≥1 % or negative if PD-L1 expression was <1 %. Statistical analysis was conducted on the dichotomized data.

Results for average negative percent agreement (ANA), average positive percent agreement (APA), and overall percent agreement (OA) were calculated for all non-redundant pair-wise comparisons across all the conditions in a test. The respective 95% confidence intervals were calculated based on the two sided percentile Bootstrap method.

The study met acceptance criteria when evaluating SCCHN specimens. Results are presented in Table 2.

Table 2: **Lot to Lot Repeatability in SCCHN**

Repeatability	Method	% Agreement (95% CI) at ≥1% Expression Level	
Inter-Lot	Each of 39 specimens with a range of PD-L1 IHC expression was tested with each of three kit lots on the Autostainer Link 48 instrument. A total of 115 pair-wise comparisons were performed.	ANA APA OA	100 (96.9, 100) 100 (96.6, 100) 100 (98.4, 100)

Inter-instrument, inter-day and intra-run precision study data were leveraged from the two prior FFPE tissue approvals (non-squamous NSCLC and melanoma).

UC

Precision was assessed in 4 separate studies: intra-day /run, combined (operator/day/instrument) precision, Lot to Lot precision and reader precision. Intra-day/run, combined precision and Lot to Lot studies were performed with 30 UC specimens spanning the range of PD-L1 expression, and at least 25% of these represented specimens around the 1% cut off. Reader precision studies included 72 unique UC specimens with 42 around the 1% cut off. Study specimens along with control slides were stained, then blinded and randomized prior to evaluation of PD-L1 expression status. Specimens were determined to be positive if PD-L1 expression was ≥1 % or negative if PD-L1 expression was <1 %. Statistical analysis was conducted on the dichotomized data and 95% confidence intervals (CI) calculated by pairwise bootstrap method.

Results for average negative percent agreement (ANA), average positive percent agreement (APA), and overall percent agreement (OA) were calculated for all non-redundant pair-wise comparisons across all the conditions in a test. The study demonstrated acceptable precision when evaluating UC specimens. The respective 95% confidence intervals were calculated based on two sided percentile bootstrap method. Study design and results are summarized in Table 3 below:

Table 3: Summary of Repeatability in UC

Repeatability	Method	% Agreement (95% CI) ≥1% Expression Level
Combined Precision	Each of 29* urothelial carcinoma specimens with a range of PD-L1 IHC expression was tested in the following conditions: 5 operators/instruments/days using one assay lot. A total of 286 independent pair-wise comparisons were performed.	ANA 96.6 (91.7, 100.0) APA 96.4 (90.3, 100.0) OA 96.5 (91.0, 100.0)
Inter-lot	Each of 29* urothelial carcinoma specimens with a range of PD-L1 IHC expression was tested with 3 different assay build lots on the Autostainer Link 48 instrument. A total of 87 independent pair-wise comparisons were performed.	ANA 97.8 (93.8, 100.0) APA 97.6 (92.3, 100.0) OA 97.7 (93.1, 100.0)
Intra-run	Each of 29* urothelial carcinoma specimens with a range of PD-L1 IHC expression was tested with five replicates within the same run by the same operator on the same Autostainer Link 48 instrument. A total of 290 independent pair-wise comparisons were performed.	ANA 96.3 (90.3, 100.0) APA 96.8 (92.7, 100.0) OA 96.6 (91.7, 100.0)
Inter-observer	Each of 72 urothelial carcinoma specimens with a range of PD-L1 IHC expression was read 3 times by 3 different pathologists with a 2-week washout period in between reads. A total of 1944 independent pair-wise comparisons were performed.	ANA 90.3 (85.2, 94.1) APA 93.1 (89.2, 96.4) OA 91.9 (87.6, 95.7)
Intra-observer	Each of 72 urothelial carcinoma specimens with a range of PD-L1 IHC expression was read 3 times by the same pathologist with a 2-week washout period in between reads. A total of 3 pathologists were used and 648 independent pair-wise comparisons were performed.	ANA 95.2 (92.6, 97.4) APA 96.6 (94.6, 96.2) OA 96.0 (93.8, 97.8)

***** A total of 30 urothelial carcinoma specimens were tested in combined precision, inter-lot, and intra-run studies, one specimen was found to be not evaluable.

4. External Reproducibility

Reproducibility studies for SCCHN and UC were designed to evaluate the performance of PD-L1 IHC 28-8 pharmDx for PD-L1 detection across laboratories on the Dako Autostainer Link 48. The study included specimens that were to represent full PD-L1 expression range and a minimum of 25% of the specimens had staining expected to be around the cut off (1%). This specimen set was randomized and blinded prior to testing

at 3 external reproducibility sites and assessed for performance with regard to site-to-site and day-to-day reproducibility, or intra-site reproducibility.

Reproducibility studies also included assessment of observer-to-observer variability with specimens that spanned the expression range of PD-L1. These specimens were stained at the Dako facility and shipped to the 3 external reproducibility sites. All IHC tests were interpreted by certified clinical pathologists to determine the positive/negative results based on the ≥1% expression levels at three external sites.

SCCHN:
Reproducibility in SCCHN was assessed in two separate studies, an abbreviated study with 14 SCCHN specimens tested at 3 sites over 3 non consecutive days and the reader reproducibility evaluated with 30 pre-stained specimens read two times by three pathologists at one site (results in Table 5). Due to the limited number of specimens, a second full scale study was conducted with 32 specimens tested over 5 non consecutive days and reader reproducibility with 38 pre stained specimens read by three pathologists at 3 sites with a 14 day washout between reads. The results of the reproducibility studies are included in Table 4 (abbreviated study) and Table 5 (Full scale). The acceptance criteria were met for all sources of variability that were examined.

Table 4: Abbreviated Reproducibility Study

Reproducibility	Method	% Agreement (95% CI) ≥1% Expression Level
Inter-site (three sites)	A set of 14 SCCHN specimens, with a range of PD-L1 IHC expression, was tested on three non-consecutive days at each of three sites. Inter-site analysis was performed between three sites on a total of 378 pair-wise comparisons.	ANA 100 (99.0, 100) APA 100 (99.0, 100) OA 100 (99.5, 100)
Intra-site	A set of 14 SCCHN specimens, with a range of PD-L1 IHC expression, was tested on three non-consecutive days at each of three sites. Intra-site analysis was performed for three sites on a total of 126 pair-wise comparisons.	ANA 100 (97.0, 100) APA 100 (97.0, 100) OA 100 (98.5, 100)
Inter-observer (three observers)	Two separate scoring evaluations of a set of 30 SCCHN specimens, demonstrating a range of PD-L1 IHC expression, stained with PD-L1 IHC 28-8 pharmDx, were performed by three pathologists, with a minimum of a 5-day washout period between reads. Inter-observer analysis was performed between three pathologists on a total of 360 pair-wise comparisons.	ANA 95.5 (89.8, 100) APA 95.6 (90.3, 100) OA 95.6 (90.0, 100)
Intra-observer	Two separate scoring evaluations of a set of 30 SCCHN specimens, demonstrating a range of PD-L1 IHC expression, stained with PD-L1 IHC 28-8 pharmDx, were performed by three pathologists, with a minimum of a 5-day washout period between reads. Intra-observer analysis was performed for three pathologists on a total of 90 pair-wise comparisons.	ANA 94.4 (87.4, 100) APA 94.5 (87.6, 100) OA 94.4 (87.8, 100)

Table 5: Three site Reproducibility of the PD-L1 IHC 28-8 pharmDx - SCCHN,

Reproducibility	Method	% Agreement (95% CI) ≥1% Expression Level
Inter-site (three sites)	A set of 32 SCCHN specimens, with a range of PD-L1 IHC expression, was tested on five non-consecutive days at each of three sites. Inter-site analysis was performed between three sites on a total of 2400 pair-wise comparisons.	ANA 96.0 (91.5, 99.2) APA 96.3 (92.9, 99.2) OA 96.2 (92.2, 99.2)
Intra-site	A set of 32 SCCHN specimens, with a range of PD-L1 IHC expression, was tested on five non-consecutive days at each of three sites. Intra-site analysis was performed for three sites on a total of 960 pair-wise comparisons.	ANA 97.2 (94.7, 99.1) APA 97.4 (95.3, 99.2) OA 97.3 (95.0, 99.2)
Inter-observer (three observers)	Three separate scoring evaluations of a set of 38 SCCHN specimens, demonstrating a range of PD-L1 IHC expression, stained with PD-L1 IHC 28-8 pharmDx, were performed by three pathologists, with a minimum of a 14-day washout period between reads. Inter-observer analysis was performed between three pathologists on a total of 1026 pair-wise comparisons.	ANA 97.1 (94.6, 99.4) APA 97.1 (94.7, 99.4) OA 97.1 (94.7, 99.4)
Intra-observer	Three separate scoring evaluations of a set of 38 SCCHN specimens, demonstrating a range of PD-L1 IHC expression, stained with PD-L1 IHC 28-8 pharmDx, were performed by three pathologists, with a minimum of a 14-day washout period between reads. Intra-observer analysis was performed for three pathologists on a total of 342 pair-wise comparisons.	ANA 97.1 (94.2, 99.4) APA 97.1 (94.3, 99.4) OA 97.1 (94.2, 99.4)

UC

Reproducibility was assessed with 46 specimens being tested across 3 sites over 5 non consecutive days and 78 pre stained specimens being assessed 3 times by 3 readers with a two week washout between reads. The acceptance criteria were met for all sources of variability that were examined. The results of the reproducibility studies are included in Table 6. Lower bound of 95% Confidence intervals for ANA, APA and OA for the inter site study were below 85% and did not meet acceptance criteria. The study had equal representation of positive and negative specimens (24 negative and 22 positive), however 30 of the 46 specimens in the study represented challenging samples around the clinical cut off of ≥1%.

Table 6: Three Site Reproducibility in UC

Reproducibility	Method	% Agreement (95% CI) ≥1% Expression Level
Inter-site (three sites)	46 urothelial carcinoma specimens with a range of PD-L1 IHC expression were tested on five non-consecutive days. Inter-site analysis was performed between three sites on a total of 3440 pair-wise comparisons.	ANA 88.1 (82.2, 93.7) APA 85.8 (77.7, 92.7) OA 87.0 (80.5, 93.2)
Intra-site	46 urothelial carcinoma specimens with a range of PD-L1 IHC expression were tested on five non-consecutive days. Intra-site analysis was performed for three sites on a total of 1376 pair-wise comparisons.	ANA 93.2 (89.5, 96.5) APA 91.9 (87.2, 96.0) OA 92.6 (88.6, 96.2)
Inter-observer (one observer at each of three sites)	Scoring of 78 urothelial carcinoma specimens with a range of PD-L1 IHC expression was performed by three pathologists, one at each of three study sites, on three non-consecutive days. Inter-observer analysis was performed between three sites on a total of 2106 pair-wise comparisons.	ANA 91.9 (87.8, 95.7) APA 92.5 (88.5, 96.1) OA 92.2 (88.1, 95.9)
Intra-observer	Scoring of 78 urothelial carcinoma specimens with a range of PD-L1 IHC expression was performed by three pathologists, one at each of three study sites, on three non-consecutive days. Intra-observer analysis was performed for three sites on a total of 702 pair-wise comparisons.	ANA 96.1 (94.0, 98.0) APA 96.4 (94.4, 98.2) OA 96.3 (94.3, 98.0)

5. Robustness Studies:

Robustness of staining performance for the PD-L1 IHC 28-8 pharmDx was evaluated by evaluating variability in assay output when varying the different assay conditions with one lot of reagent.

SCCHN

Robustness of PD-l1 IHC 28-8 performance in SCCHN was evaluated by varying the following conditions:

a) Target retrieval solution temperature (97^0C, 95^0C, and 99^0C)
b) Target retrieval time (20, 18 and 22 minutes)
c) Target retrieval solution pH (pH 6.1, 5.8 and 6.4)
d) Tissue Section Thickness at 3, 4 and 5 μM

The target retrieval study included 55 and tissue thickness study 39 SCCHN specimens spanning the range of PD-L1 expression and included specimens around the cut off. Staining performance was evaluated for both percent (%) tumor staining and intensity of staining. A change in the PD-L1 status or drop in intensity by 0.5 on a scale of 0-3 was considered a test condition failure. No significant difference in results was observed for any of the recommended experimental conditions above.

UC

Robustness of the staining performance of PD-L1 IHC 28-8 pharmDx in UC was evaluated by testing the performance of the assay when varying the following conditions as described below. On one lot of reagents was assessed.

a) Tissue section thickness (4 and 5 μM)
b) Microscope slide type (Fisher Brand SuperFrost Plus, Dako Flex IHC microscope slides)
c) Target retrieval solution temperature (97^0C, 95^0C, and 99^0C)
d) Target retrieval time (20, 18 and 22 minutes)
e) Target retrieval solution pH (pH 6.1, 5.9 and 6.3)
f) Target retrieval solution reuse (3 re-uses)

The study included 30 UC specimens spanning the range of PD-L1 expression and included specimens around the cut off. Staining performance was evaluated for both percent (%) tumor staining and intensity of staining. A change in the PD-L1 status or drop in intensity by 0.5 on a scale of 0-3 was considered a test condition failure. No significant difference in results was observed for any of the recommended experimental conditions above.

6. Impact of Intra-Case Heterogeneity

The objective of this study was to investigate whether tumor heterogeneity affects PD-L1 IHC staining results with PD-L1 IHC 28-8 pharmDx.

SCCHN

a. Primary vs. Metastatic Tumor Tissues

Matched primary versus metastatic FFPE tumor blocks were obtained from 35 subjects and evaluated by PD-L1 IHC 28-8 pharmDx. Three (3) of the specimens were unevaluable due to invalid test results or insufficient tissue for evaluation of PD-L1 status and omitted from final analysis. In 22 of 32 matched SCCHN pairs the diagnostic outcome was identical for primary and metastatic specimens. Ten (10) primary/metastatic pairs showed discordant results at the ≥1% expression level such that the in 6 cases the primary tumor was positive for PD-L1 and metastatic tumor negative and in 4 cases the primary tumor was PD-L1 negative and metastatic tumor was positive.

b. Multiple FFPE Blocks from the Same Subjects (Variability in PD-L1 expression between anatomic sites within patients)

Multiple blocks (2 or more) from each of 53 SCCHN subjects obtained from the same tumor were evaluated. In 46 of 53 sets of SCCHN intra-subject specimens the diagnostic outcomes were identical for all specimens. Seven (7) cases showed discordant results at the ≥1% expression level.

UC

a. Primary vs. Metastatic Tumor Tissues

Matched primary versus metastatic blocks were obtained from 30 subjects and evaluated by PD-L1 IHC 28-8 pharmDx. Two (2) of the specimens were unevaluable and omitted from final analysis. In 23 of 28 matched UC pairs the diagnostic outcome was identical for primary and metastatic specimens. Five (5) primary/metastatic pairs showed discordant results at the ≥1% expression level such that the in 3 cases the primary tumor was positive for PD-L1 and metastatic tumor negative and in 2 cases the primary tumor was PD-L1 negative and metastatic tumor was positive.

b. Multiple FFPE Blocks from the Same Subjects (Variability in PD-L1 expression between anatomic sites within patients)

Multiple blocks (2 or more) from each of 55 UC subjects obtained from the same tumor were evaluated. In 52 of 55 sets of UC intra-subject specimens the diagnostic outcomes were identical for all specimens. Three (3) cases showed discordant results at the ≥1% expression level.

7. Stability testing

a. PD-L1 IHC 28-8 pharmDx Stability

The real-time shelf life/stability of the PD-L1 IHC 28-8 pharmDx was assessed under original PMA applications P150025 and P150027. A stability testing protocol has been approved for extending the stability claim for the kit which is currently at 7 months and no change is proposed at the time of this review. See P150025 and P150027 SSED for details on stability testing. The reagent stability claims are as follows:

Total Shelf Life
7 months at 2-8°C

Finished Good Shelf Life
7 months at 2-8°C

In-Use/On-Board Stability Testing
Six cycles to room temperature

Working/Reconstituted Stability Testing
DAB Substrate-Chromogen Solution: 5 Days at 2-8°C, Protected from Light.
Target Retrieval Solution: 5 days at Room Temperature in a PT-Link with up to 3 uses for 3-in-1 Pretreatment.

b. FFPE Cut Section Stability

A real-time stability study was designed to evaluate the shelf life of cut tissue sections of SCCHN and UC FFPE blocks using PD-L1 IHC 28-8 pharmDx when stored in the dark at 2-8 °C or 25 °C. The study included 4 SCCHN and 4 UC specimens. Based on these studies, cut-slide stability dating for SCCHN is 4 months and UC is 6 months.

B. Animal Studies

None

C. Additional Studies

1. *PT Link PT100/101 and PT200 Concordance Evaluation*

The objective of this study was to demonstrate that the performance of PD-L1 IHC 28-8 pharmDx (SK005) assay is concordant when the 3-in-1 pretreatment procedure for deparaffinization, rehydration, and heat-induced epitope-retrieval (HIER) of formalin fixed, paraffin-embedded (FFPE) tissue sections is conducted using the PT Link model PT200 and PT Link model PT100/101. The study was conducted with 120 specimens from 3 tumor indications: UC, SCCHN and squamous NSCLC (sq. NSCLC). Concordance in PD-L1 expression status (positive or negative agreement according to 1% clinical cut-off) was assessed when pretreatment of FFPE tumor specimens was conducted in PT Link model PT200 or PT Link model PT100/101. Results are presented in Table 7. Acceptance criteria were met.

Table 7: Concordance results of PT Link (PT100/PT101) and PT Link (PT200)

Concordance	Method	% Agreement (95% CI)
		≥ 1% Expression Level
Concordance at 1% Cutoff (UC, sq. NSCLC, and SCCHN)	40 UC, 40 sq. NSCLC and 40 SCCHN specimens were evaluated for staining after being pretreated with PT100 and compared to replicate slides pretreated using the PT200 instrument. A total of 120 comparisons were made.	NPA 98.4% (91.5, 99.7%) PPA 98.2% (90.7, 99.7%) OA 98.3% (94.1, 99.5%)

X. SUMMARY OF PRIMARY CLINICAL STUDY(IES)

SCCHN

A. Study Design

The clinical performance of PD-L1 IHC 28-8 pharmDx was evaluated in study CheckMate141 (CA209141/ NCT02105636): An Open Label, Randomized Phase 3 Clinical Trial of Nivolumab vs. Therapy of Investigator's Choice in Recurrent or

Metastatic Platinum-refractory Squamous Cell Carcinoma of the Head and Neck (SCCHN). A total of 361 subjects were randomized at 55 sites in 15 countries (Argentina, Brazil, Canada, France, Germany, Hong Kong, Italy, Japan, Korea, Netherlands, Spain, Switzerland, Taiwan, United Kingdom, and United States of America). Of these randomized subjects, 171 (47.4%) were in Europe, 145 (40.2%) were in North America, and 45 (12.5%) were from the rest of the world. The enrollment period lasted approximately 14 months (May-2014 to Jul-2015). The first patient first visit date was 29-May-2014 and the last patient first treatment date was 31-Aug-2015. The clinical database lock for this CSR occurred on 18-Dec-2015 and the PD-L1 biomarker database lock occurred on 03-Feb-2016.

Patients were randomized to receive nivolumab administered intravenously (IV) at 3 mg/kg every 2 weeks or investigators choice of

- cetuximab 400 mg/m2 loading dose IV followed by 250 mg/m2 weekly,
- methotrexate 40 to 60 mg/m2 IV weekly, or
- Docetaxel 30 to 40 mg/m2 IV weekly.

Subjects were enrolled and randomized regardless of PD-L1 expression status; however, pre-study (baseline) FFPE tumor tissue specimens were systematically collected prior to randomization and prior to first treatment, in order to conduct pre-planned analyses of efficacy according to PD-L1 expression status. Tumor tissue was obtained from an unresectable site of disease or from a site of metastatic disease, and Assessment of FFPE tumor sample sections for PD-L1 expression was performed centrally

1. Clinical Inclusion and Exclusion Criteria

   Enrollment in the CheckMate141 study was limited to patients who met the following inclusion criteria:

   - Males and Females ≥ 18 years of age with an Eastern Cooperative Oncology Group (ECOG) performance status ≥ 1.
   - Histologically confirmed recurrent or metastatic SCCHN (oral cavity, pharynx, larynx), stage III/IV and not amenable to local therapy with curative intent (surgery or radiation therapy with or without chemotherapy).
   - Tumor progression or recurrence within 6 months of last dose of platinum therapy in the adjuvant (i.e., with radiation after surgery), primary (i.e., with radiation), recurrent, or metastatic setting.
   - Measurable disease by CT or MRI per RECIST 1.1 criteria.
   - A FFPE tumor tissue block or a minimum of 10 unstained slides of tumor sample (archival or recent) obtained in the metastatic setting or from unresectable site of disease must be available for biomarker evaluation
   - Documentation of p16-positive or p16-negative disease to determine human papillomavirus (HPV) status of tumor for Squamous Cell Carcinoma of the oropharynx.
   - Prior palliative radiotherapy must have been completed at least 2 weeks before study drug administration.

Patients were not permitted to enroll in the CheckMate141 study if they met any of the following exclusion criteria:

- Active brain metastases or leptomeningeal metastases.
- Histologically confirmed recurrent or metastatic carcinoma of the nasopharynx, squamous cell carcinoma of unknown primary, and salivary gland or non-squamous histologies (e.g., mucosal melanoma).
- Prior treatment with an anti-PD-1, anti-PD-L1, anti-PD-L2, anti-CTLA-4 antibody, or any other antibody or drug specifically targeting T-cell co-stimulation or immune checkpoint pathways.
- Prior malignancy active within the previous 3 years except for locally curable cancers that have been apparently cured.
- Subjects with active, known or suspected autoimmune disease.
- Subjects with a condition requiring systemic treatment with either corticosteroids (> 10 mg daily prednisone equivalents) or other immunosuppressive medications within 14 days of study drug administration. Inhaled or topical steroids and adrenal replacement doses > 10 mg daily prednisone equivalents are permitted in the absence of active autoimmune disease.

2. Follow-up Schedule

   The first tumor assessments were conducted 9 weeks after randomization and continued every 6 weeks thereafter until disease progression or treatment was discontinued
   Study subjects assessed for safety in two additional follow ups within 100 days from last dose of study therapy.

3. Clinical Endpoints
   Primary efficacy end point for the CheckMate141 study was overall survival (OS) in nivolumab vs. Investigators choice arms. Overall survival was defined as the time from randomization to the date of death from any cause. For subjects that are alive at the time of analysis, their survival time was censored at the date of last contact ("last known alive date"). Secondary endpoint was investigator assessed Progression Free Survival (PFS) and investigator assessed Overall Response Rate (ORR). Association between tumor PD-L1 status and safety and efficacy outcomes was an exploratory endpoint for the study.

   Endpoints for the safety assessments were frequency of deaths, serious adverse events (SAEs), adverse events (AEs) leading to discontinuation or dose modification, select AEs, clinical laboratory assessments (hematology, serum chemistry, and liver and thyroid function tests), and vital sign measurements. Endpoints for the assessment of Immunogenicity were serum antidrug antibodies (ADA) and neutralizing ADA.

B. Accountability of PMA Cohort

At the time of database lock, of 506 patients enrolled in the PMA study, 361 patients were randomized into the study and of these 347 were treated. Data from all randomized was used for primary efficacy analysis and all treated for primary safety analyses. At database lock 329 (90.6%) of randomized subjects had tumor tissue collected at baseline. Among these 260 (72%) had tumor samples with quantifiable PD-L1 expression at baseline and 101 (28%) did not have quantifiable PD-L1 expression at baseline. Of the 101 subjects without quantifiable PD-L1 expression at baseline, 67 (18.6%) had a baseline tumor sample with non-evaluable PDL1 expression and 32 (8.9%) did not have a tumor tissue sample submitted at any time, and 2 (0.6%) had a tumor tissue sample collected after baseline). No subjects had indeterminate PD-L1 expression at baseline. The distribution of specimens is shown in Table 8.

Table 8: Accountability of PMA Cohort

	Nivolumab (n)	**Investigators Choice (n)**	**Total (n)**
Subjects enrolled and randomized	240	121	361
Subjects with tumor sample collected at baseline	213	114	327
Subjects with quantifiable PD-L1 expression	161	99	260
Subjects without PD-L1 expression at baseline	79	22	101
Reason for not testing:			
Subjects with INDETERMINATE [(a)] PD-L1 expression at baseline	0	0	0
Subjects with NOT EVALUABLE[(b)] PD-L1 expression at baseline	52	15	67
Subjects without tumor tissue at baseline	25	7	32
Subjects with tumor tissue collected after baseline	2	0	2

(a) Indeterminate = tumor cell membrane staining hampered for reasons attributed to the biology of the tumor

(b) Not evaluable = tumor biopsy specimen was not optimally collected or prepared

C. Study Population Demographics and Baseline Parameters

In CheckMate-141, total of 361 patients were randomized; 240 patients to OPDIVO and 121 patients to investigator's choice (45% received docetaxel, 43% received methotrexate, and 12% received cetuximab). The median age was 60 years (range: 28 to 83) with 31%, 65 years of age, 83% were White, 12% Asian, and 4% were Black, and

83% male. Baseline ECOG performance status was 0 (20%) or 1 (78%), 76% were former/current smokers, 90% had Stage IV disease, 45% of patients received only one prior line of systemic therapy, the remaining 55% received two or more prior lines of systemic therapy, and 25% had HPVp16-positive tumors, 24% had HPV p16-negative tumors, and 51% had unknown status.

D. Safety and Effectiveness Results

1. Safety Results

   As an in vitro diagnostic test, the PD-L1 IHC 28-8 pharmDx Assay involves testing on FFPE non-squamousSCCHN . These tissues are routinely removed as part of the practice of medicine for the diagnosis of SCCHN by pathologists. Removal of these tissues, therefore, presents no additional safety hazard to the patient being tested. As compared to the overall study population, no meaningful differences in safety were observed based on PD-L1 expression level within each treatment arm.

2. Effectiveness Results

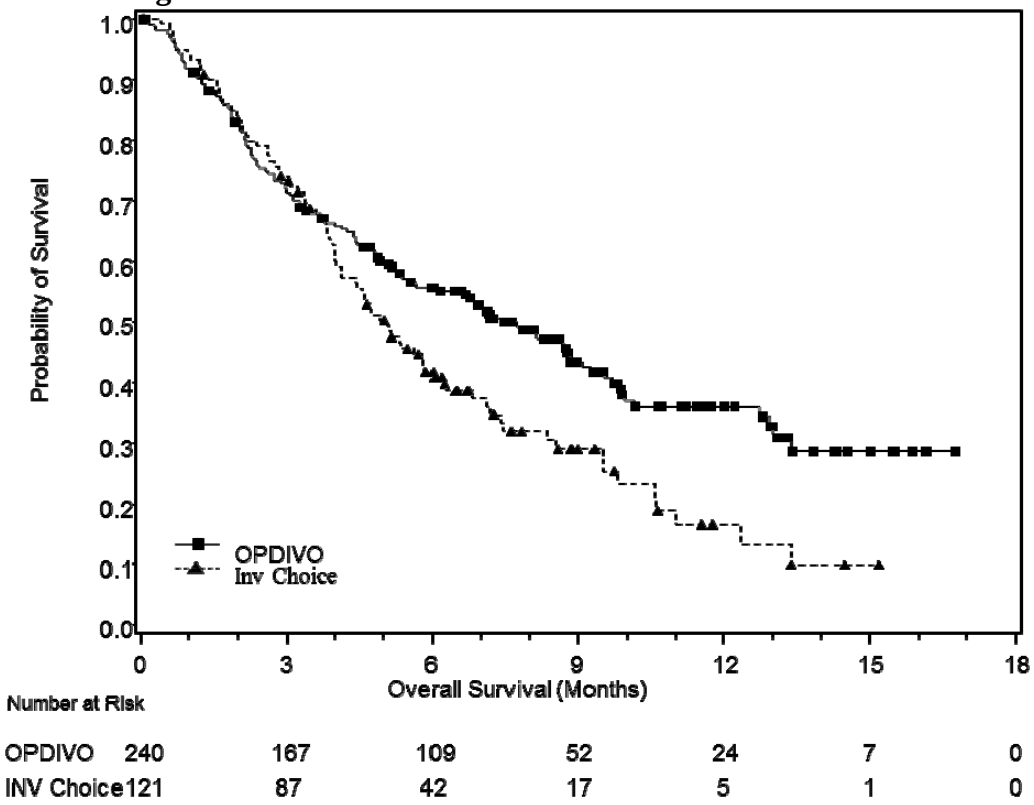
   The analysis of effectiveness for nivolumab in SCCHN was based on overall survival (OS) analysis in 361 evaluable patients enrolled in CheckMate141 trial, irrespective of tumor PD-L1 expression status was based on the at pre-specified interim analysis (78% of planned number of events for final analysis). The trial demonstrated a statistically significant improvement in OS for patients randomized to nivolumab as compared with investigator's choice therapy (IC) (Hazard Ratio (HR) = 0.70 [97.73% CI: 0.51, 0.96]; stratified log-rank test p-value = 0.0101). Median OS in nivolumab treated group was 7.5 (95% CI of 5.5, 9.1 months) vs. 5.1 (95% CI of 4.0, 6.0) in IC arm of the trail. The survival results are displayed in Table 9 and Figure 1. There were no statistically significant differences between the two arms for PFS (HR=0.89; 95% CI: 0.70, 1.13) or ORR (13.3% [95% CI: 9.3, 18.3] vs. 5.8% [95% CI: 2.4, 11.6] for nivolumab and investigator's choice, respectively).

Table 9: Overall Survival in CheckMate-141

	Nivolumab (n=240)	**Investigators Choice (n=121)**
Overall Survival		
Deaths (%)	133 (55%)	85 (70%)
Median (months) (95% CI)	7.5 (5.5, 9.1)	5.1 (4.0, 6.0)
Hazard Ratio (95% CI) [a]	0.70 (0.53, 0.92)	
p-value [b, c]	0.0101	

[a] Based on stratified proportional hazards model.
[b] Based on stratified log-rank test.
[c] p-value is compared with 0.0227 of the allocated alpha for this interim analysis.

Figure 01: Overall Survival - CheckMate-141



3. **Efficacy Results based on pre-study PD-L1 expression status**

Clinical performance of PD-l1 IHC 28-8 pharmDx was assessed in CheckMate-141 trial. Of 361 subjects randomized into the trial 327 had tumor specimens tested with the PD-L1 IHC 28-8 pharmDx assay. PD-L1 status was quantifiable for 161(67.1%) subjects in the nivolumab arm and 99 (81.8%) subjects in the investigators choice arm. Data for 101 study subjects was not available due to missing specimens (32), or not evaluable due to inadequate quality of tumor biopsy specimen (67). The distribution of patients with evaluable specimens that are PD-L1 positive or negative across the study arms is shown in Table 10.

Table 10: Tumor PD-L1 Status for CheckMate-141 Study Subjects

	Number of Subjects, n (%)		
	Nivolumab	**Investigators Choice**	**Total**
Number of subjects with quantifiable PD-L1 expression	161	99	260
Subjects with baseline PD-L1 Expression ≥ 1%	88 (54.7)	61 (61.6)	149(57.3)
Subjects with baseline PD-L1 Expression < 1%	73(45.3)	38(38.4)	111(42.7)

In pre-specified exploratory subgroup analysis using PD-L1 IHC 28-8 pharmDx response, response in PD-L1 subgroups (<1% and ≥1%), the hazard ratio (HR) was 0.89 (95% CI: 0.54, 1.45) in the PD-L1 negative (<1%) subgroup and 0.55 (95% CI: 0.36, 083) in the PD-L1 positive (≥1%) subgroup for nivolumab vs. investigators choice treatment arms. Median overall survival in PD-L1 positive subjects was 8.7 months in the nivolumab treated arm vs. 4.6 months in investigators choice treatment arm. In the PD-L1 negative subgroups median overall survival was 5.7 months in the nivolumab treated arm vs. 5.8 months in investigators choice arms of the study. The OS trends in PD-L1 subgroups are captured in Table 11.

Table 11: Overall Survival in PD-L1 Subgroups in CHECKMATE-141

	Median Overall Survival in months (95% CI)	
PD-L1 Expression Level	**Nivolumab**	**Investigators choice therapy**
<1%	5.7	5.8
≥1%	8.7	4.6
	Hazard Ratios (95% CI)	
	Nivolumab vs. Investigators Choice	
<1%	0.89 (0.54, 1.45)	
≥1%	0.55 (0.36,0.83)	

The efficacy analysis of outcomes in the PD-L1 subgroups did not consider missing PD-L1 results for 101 specimens. In order to evaluate the potential impact of the missing specimens on the efficacy data, a sensitivity analysis by imputing missing PD-L1 status flags of <1% vs. ≥1% was performed. PD-L1status was imputed based on 'with replacement' sampling from the 260 randomized subjects in the study with non-missing PD-L1 status data. The hazard ratio analysis was run using model-based imputed PD-L1 status and adjusting for the prognostic covariates. The imputation analysis yielded HRs and associated 95% CIs of 0.598 (0.401-0.891) and 1.044 (0.641-1.702) for PD-L1 expression ≥ 1% and PD-L1 expression level <1%, respectively. The imputed hazard ratios are summarized in Table 12. These results are consistent with data obtained for samples with PD-L1 results.

Table 12: Overall Survival by PD-L1 Subset, After Imputing Missing PD-L1 Expression, All Randomized Subjects, Study CA209141

	PD-L1 ≥ 1 %	PD-L1 < 1%
Hazard Ratio (95% CI)[a]	0.598 (0.401 - 0.891)	1.044 (0.641 - 1.702)
Median HR from 20 imputations	0.603	1.036
Minimum HR - Maximum HR	0.535 - 0.715	0.861 - 1.235

[a] Association between PD-L1 status (<1%, ≥1%) and baseline covariates was examined through univariate logistic regressions. To impute missing PD-L1 status, covariates with p-value<0.1 were included in final model. The final model contained the following covariates: sex (Male/Female), race (white/non-white), and primary site of disease (oral cavity/not oral cavity). The multiple imputations were based on 20 simulations. Estimated hazard ratio of nivolumab to IC and its associated 95% CI were obtained from combining 20 individual HRs and standard errors via Rubin's method. Hazard ratio is from stratified Cox Model with treatment effect adjusted for prognostic covariates of gender, ECOG PS, primary site of tumor, time from prior platinum to progression, time from initial diagnosis to randomization and prior radiotherapy.

4. Pediatric Extrapolation
   In this premarket application, existing clinical data was not leveraged to support approval of a pediatric patient population.

E. Financial Disclosure

The Financial Disclosure by Clinical Investigators regulation (21 CFR 54) requires applicants who submit a marketing application to include certain information concerning the compensation to, and financial interests and arrangement of, any clinical investigator conducting clinical studies covered by the regulation. The pivotal clinical study included 3 investigators. None of the clinical investigators had disclosable financial interests/arrangements as defined in sections 54.2(a), (b), (c), and (f). The information provided does not raise any questions about the reliability of the data.

Clinical Studies in Urothelial Bladder Cancer

A2. Study Design

The clinical performance of PD-L1 IHC 28-8 pharmDx was evaluated in study CheckMate-275 (CA209275/ NCT02387996): A phase II single arm clinical trial of nivolumab (BMS-936558) in subjects with metastatic or unresectable urothelial cancer who have progressed or recurred following treatment with a platinum agent. Three hundred eighty-six (386) subjects were enrolled at sixty-three (63) sites in eleven (11) countries (Australia, Belgium, Czech Republic, Finland, Germany, Italy, Japan, Poland, Spain, Sweden, and United States of America). Enrollment of first 70 subjects' in the study was based on confirmed PD-L1 tumor expression of ≥5%. The first patient first

visit date (FPFV) was 09-Mar-2015. Last patient final visit was on 12 November 2015. The clinical database lock for this CSR occurred on 30 May 2016 with a clinical cut off 15 April, 2016. An update to ORR, DoR, PFS and OS was based on database lock of September 02, 2016 based on clinical cut off of July 21, 2016.

Patients receive nivolumab administered intravenously (IV) over 60 minutes at 3 mg/kg every 2 weeks until progression or unacceptable toxicity. Evaluable tissue for prospective assessment of PD-L1 expression was submitted prior to treatment. Subjects were assigned to a cohort according to PD-L1 status (PD-L1 ≥ 1%, PDL1 < 5%, or indeterminate) and treated with nivolumab. Archived tissue was obtained from prior biopsy of unresectable or metastatic disease or from prior surgical resection. Assessment of FFPE tumor sample sections for PD-L1 expression was performed centrally using the PD-L1 IHC 28-8 pharmDx test.

1. Clinical Inclusion and Exclusion Criteria

   Enrollment in the CheckMate-275 study was limited to patients who met the following inclusion criteria:

   - Histological or cytological evidence of metastatic or surgically unresectable (cT4b or any N+ (N1-3) or may be M1) transitional cell carcinoma of the urothelium involving the bladder, urethra, ureter, or renal pelvis.
   - Tumor progression or recurrence within 6 months of last dose of platinum therapy in the adjuvant (i.e., with radiation after surgery), primary (i.e., with radiation), recurrent, or metastatic setting.
   - Measurable disease by CT or MRI per RECIST 1.1 criteria.
   - Subjects must have progression or recurrence after treatment with at least 1 platinum containing chemotherapy regimen for metastatic/surgically unresectable UC or within 12 months of peri-operative treatment with platinum agent in cystectomy setting for muscle invasive UC.
   - Evaluable tumor tissue (archival or recent biopsy) must be provided for biomarker analysis as FFPE tumor block or minimum of 10 slides.
   - Eastern Cooperative Oncology Group (ECOG) performance status (PS) 0 or 1.

   Patients were not permitted to enroll in the CheckMate141 study if they met any of the following exclusion criteria:

   - Active brain metastases or leptomeningeal metastases.
   - Any serious or uncontrolled medical disorder
   - Prior treatment with an anti-PD-1, anti-PD-L1, anti-PD-L2, anti-CTLA-4 antibody, or any other antibody or drug specifically targeting T-cell co-stimulation or immune checkpoint pathways.
   - Prior malignancy active within the previous 3 years except for locally curable cancers that have been apparently cured.
   - Subjects with active, known or suspected autoimmune disease.
   - Subjects with a condition requiring systemic treatment with either corticosteroids (> 10 mg daily prednisone equivalents) or other immunosuppressive medications within 14 days of study drug administration.

Inhaled or topical steroids and adrenal replacement doses > 10 mg daily prednisone equivalents are permitted in the absence of active autoimmune disease.

2. Follow-up Schedule
   On-study tumor assessments began 8 weeks (+/- 1 week) from first dose and continued every 8 weeks (+/- 1 week) thereafter up to 48 weeks, then every 12 weeks (+/- 1 week) until disease progression or treatment discontinuation. Treatment beyond initial investigator-assessed RECIST 1.1-defined progression was permitted if the subject had investigator-assessed clinical benefit and was tolerating study drug.

3. Clinical Endpoints
   The primary endpoint of the study was ORR based on assessments by BIRC in subjects with tumor expressing PD-L1 (membranous staining in ≥ 5% and ≥ 1% tumor cells) and overall treated subjects. The final analysis of the primary endpoint ORR (based on BIRC assessments) was performed six months after approximately 70 subjects with PD-L1 expression of ≥ 5% have been treated (i.e., six months after last patient first treatment).

   Endpoints for the safety assessments were frequency of deaths, serious adverse events (SAEs), adverse events (AEs) leading to discontinuation or dose modification, select AEs, clinical laboratory assessments (hematology, serum chemistry, and liver and thyroid function tests), and vital sign measurements. Endpoints for the assessment of Immunogenicity were serum antidrug antibodies (ADA) and neutralizing ADA.

B2. Accountability of PMA Cohort

At the time of database lock, specimens from 270 subjects had been tested prospectively prior to treatment. A total of 386 patients enrolled in the clinical study CheckMate-275; of the 386 patients 116 were not treated [14 (3.6%) withdrew consent, 80 (20.7%) failed to meet study criteria, 6 (1.6%) due to adverse events, and 2 (0.5%) due to death or 14 (3.6%) due to poor compliance or other reasons. 270 (70%) patients were treated after tumor PD-L1 expression status was prospectively determined. Data from all treated subjects was used for primary efficacy analysis and all treated for primary safety analyses. As of database lock, 100% of treated subjects had a tumor tissue sample collected at baseline. Among these subjects, all of them had quantifiable PD-L1 expression at baseline. Refer to Table 13.

Table 13: Accountability of PMA Cohort in CheckMate-275

	Number of subjects
Subjects Enrolled into the study	**386**
Enrolled subjects randomized and Treated	**270**
Subjects with Tumor sample collected at baseline	270
Treated subjects with quantifiable PD-L1 expression	270
Treated subject Without PD-L1 expression at baseline	0
Site of Collection:	
Primary Site	212 (78.5%)
Metastatic Site	54 (20%)
Other site	1 (0.4)
Site Not reported	3 (1.1)

C2. Study Population Demographics and Baseline Parameters

In CheckMate-275, total of 386 patients were enrolled. Of these 270 (69.9%) were treated with nivolumab. The median age was 66 years (range: 38 to 90), 78% were male; 86% of patients were white. 27% had non-bladder urothelial carcinoma and 84% had visceral metastases. 34% of patients had disease progression following prior platinum-containing neoadjuvant or adjuvant therapy. 29% of patients had received ≥2 prior systemic regimens in the metastatic setting. 36% of patients received prior cisplatin only, 23% received prior carboplatin only, and 7% were treated with both cisplatin and carboplatin in the metastatic setting. 46% of patients had an ECOG performance status of 1. 18% of patients had hemoglobin <10 g/dL and 28% of patients had liver metastases at baseline. Patients were included regardless of their PD-L1 status.

D2. Safety and Effectiveness Results

1. Safety Results

   As an in vitro diagnostic test, the PD-L1 IHC 28-8 pharmDx Assay involves testing on FFPE UC sections. These tissues are routinely removed as part of the practice of medicine for the diagnosis of UC by pathologists. Removal of these tissues, therefore, presents no additional safety hazard to the patient being tested.

As compared to the overall study population, no meaningful differences in safety were observed based on PD-L1 expression level within each treatment arm.

2. Effectiveness Results

Clinical performance of PD-L1 IHC 28-8 pharmDx was assessed in CheckMate-275 trial. Of 270 subjects treated in the trial 270 had tumor specimens tested with the PD-L1 IHC 28-8 pharmDx assay. PD-L1 status was quantifiable for all 270 (100%) tumor specimens. 54.7% of study subjects had PD-L1 <1% and 45.9% had PD-L1 ≥1%. Refer to Table 14.

Table 14: Tumor PD-L1 Status for CheckMate-141 Study Subjects

	Number of Subjects, n (%)
Number of subjects with quantifiable PD-L1 expression	270
Subjects with baseline PD-L1 Expression <1%	146 (54.1)
Subjects with baseline PD-L1 Expression ≥ 1%	124(45.9)

Nivolumab monotherapy demonstrated clinically meaningful ORR that compared favorably to single agent chemotherapy historical control. In PD-L1 subgroups ≥1% and all treated subjects, the lower bound of 95% CI for ORR was greater than 10%, the pre specified threshold below which ORR is not considered an improvement over single-agent chemotherapy historical control. Treatment with nivolumab led to an ORR of 19.6% (95% CI: 15.1, 24.9) in all treated subjects, 25.0% (95% CI: 17.7, 33.6) in PD-L1 ≥ 1% cohort and 15.1% (95% CI: 9.7, 21.9)
in PD-L1 <1% cohort. Seven subjects achieved a CR.

Table 15: Overall Survival in CheckMate-275

	All Patients (N=270)	**PD-L1 <1% (N=146)**	**PD-L1 ≥1% (N=124)**
Confirmed Objective Response Rate, n (%), 95% CI	53 (19.6%) 15.1-24.9	22 (15.1%) 9.7-21.9	31 (25%) 17.7-33.6
Complete Response Rate	7 (2.6%)	1 (0.7%)	6 (4.8%)
Partial Response Rate	46 (17.0%)	21 (14.4%)	25 (20.2%)
Median Duration of Response * (months) (Range)	10.3 (1.9+, 12.0+)	7.6 (3.7+,12.0+)	NE^ (1.9+, 12.0+)

* Estimated from the Kaplan-Meier Curve; ^ Not Estimable

3. Pediatric Extrapolation
   In this premarket application, existing clinical data was not leveraged to support approval of a pediatric patient population.

E2. Financial Disclosure

The Financial Disclosure by Clinical Investigators regulation (21 CFR 54) requires applicants who submit a marketing application to include certain information concerning the compensation to, and financial interests and arrangement of, any clinical investigator conducting clinical studies covered by the regulation. The pivotal clinical study included 3 investigators. None of the clinical investigators had disclosable financial interests/arrangements as defined in sections 54.2(a), (b), (c), and (f). The information provided does not raise any questions about the reliability of the data.

XI. PANEL MEETING RECOMMENDATION AND FDA'S POST-PANEL ACTION

In accordance with the provisions of section 515(c) (2) of the act as amended by the Safe Medical Devices Act of 1990, this PMA was not referred to the Hematology and Pathology Devices Panel, an FDA advisory committee, for review and recommendation because the information in the PMA substantially duplicates information previously reviewed by this panel.

XII. CONCLUSIONS DRAWN FROM PRECLINICAL AND CLINICAL STUDIES

A. Effectiveness Conclusions

SCCHN:

The clinical benefit of PD-L1 IHC 28-8 pharmDx was evaluated in CheckMate-141 (CA209141) study, a phase 3 study in which study subjects received whether nivolumab or investigators choice of treatment. Efficacy analysis in overall population demonstrated that nivolumab showed a statistically significant, clinically meaningful improvement in overall survival benefit over investigators choice arms with an hazard ratio of 0.70 (95% CI: 0.53, 0.92). The median overall survival of 7.5 months was observed in nivolumab subjects when compared to 5.1 months in investigators choice arms.

Baseline tumor specimens were retrospectively assessed for PD-L1 status and defined PD-L1 positive tumors as those that express >1% tumor cells expressed the PD-L1 protein. Tumor specimens were retrospectively evaluated for PD-L1 expression using the PD-L1 IHC 28-8 pharmDx assay. In pre-specified exploratory subgroup analyses, the hazard ratio for survival was 0.89 (95% CI: 0.54, 1.45) with median survivals of 5.7

and 5.8 months for the nivolumab and chemotherapy arms, respectively, in patients with <1% PD-L1 expression. The HR for survival was 0.55 (95% CI: 0.36, 0.83) with median survivals of 8.7 and 4.6 months for the nivolumab and chemotherapy arms, respectively, in the PD-L1 positive SCCHN subgroup. Thus the overall survival benefit for treatment with nivolumab when compared to investigators choice may be higher in PD-L1 >1% sub group than in PD-L1 <1% subgroup.

UC:
Clinical benefit of PD-L1 IHC 28-8 pharmDx was assessed in CheckMate-275 (CA2029275) study: a phase II single arm clinical trial of nivolumab in subjects with metastatic or unresectable urothelial cancer who had progressed or recurred following treatment with platinum agent. A total of 270 patients were enrolled regardless of tumor PD-L1 status; patient tumor specimens were collected prospectively and tested with the PD-L1 IHC 28-8 pharmDx to determine tumor PD-L1 status prior to treatment. Treatment with nivolumab led to an ORR of 19.6% (95% CI: 15.1, 24.9) in all treated subjects with 7 (2.6 %) subjects achieving a CR. In the PD-L1 ≥ 1% cohort, the ORR was 25.0% (95% CI: 17.7, 33.6) and 15.1% (95% CI: 9.7, 21.9) in PD-L1 <1% cohort. These results suggest that ORR in PD-L1 ≥ 1% may be higher than in PD-L1 <1%.

The performance of PD-L1 IHC 28-8 pharmDx was also supported by the analytical validation studies.

B. Safety Conclusions

The PD-L1 IHC 28-8 pharmDx is an *in vitro* diagnostic device, which tests tumor FFPE specimens collected from patients with SCCHN and UC. The risks of the device are based on data collected in the clinical study. Risks of the PD-L1 IHC 28-8 pharmDx are associated with failure of the device to perform as expected or failure to correctly interpret test results. The process of testing on FFPE tumor specimens does not present additional significant safety concerns, as these samples are routinely removed for SCCHN and UC diagnosis.

C. Benefit-Risk Determination

In clinical study CHECKMATE-141 for subjects with SCCHN, tumor PD-L1 expression at ≥ 1 % based on the PD- L1 IHC 28-8 pharmDx assay, was associated with enhanced survival from OPDIVO® (nivolumab) with HR of 0.55 (95% CI: 0.36, 0.83) when compared to chemotherapy. Whereas in subjects with PD-L1 expression <1% the hazard ratio was 0.89 (95% CI: 0.54, 1.45), suggesting that patients with squamous cell carcinoma of the head and neck cancer may potentially benefit from the use of the device. Erroneous device results (false negative or false positive results) could adversely influence expectation of benefit from OPDIVO® (nivolumab) treatment of squamous cell carcinoma of head and neck cancer patients. . Based on the data collected in the clinical study which were used to support PMA approval as described above, the probable benefits outweigh the probable risks.

In Clinical study CHECKMATE-275, Urothelial carcinoma patients whose tumors expressed PD-L1 at ≥ 1 % based on the PD-L1 IHC 28-8 pharmDx assay, showed higher ORR (25.0% (95% CI: 17.7, 33.6) when compared to subjects whose tumors expressed PD-L1 at <1 % (ORR : 15.1% (95% CI: 9.7, 21.9)) on treatment with OPDIVO® (nivolumab) suggesting potentially benefit from the use of the device. Erroneous device results (false negative or false positive results) could adversely influence expectation of benefit from OPDIVO® (nivolumab) treatment of urothelial cancer patients. Based on the data collected in the clinical study which were used to support PMA approval as described above, the probable benefits outweigh the probable risks.

1. Patient Perspectives
   This submission did not include specific information on patient perspectives for this device.

D. Overall Conclusions

The data in this application support the reasonable assurance of safety and effectiveness of this device when used in accordance with the indications for use and product labeling. The provided studies support use of PD-L1 IHC 28-8 pharmDx in squamous cell carcinoma of the head and neck (SCCHN) and urothelial cancer patients who may be considered for treatment with OPDIVO® (nivolumab).

XIII. CDRH DECISION

CDRH issued an approval order on September 15, 2017.

The applicant's manufacturing facilities have been inspected and found to be in compliance with the device Quality System (QS) regulation (21 CFR 820).

XIV. APPROVAL SPECIFICATIONS

Directions for use: See device labeling.

Hazards to Health from Use of the Device: See Indications, Contraindications, Warnings, Precautions, and Adverse Events in the device labeling.

Post-approval Requirements and Restrictions: See approval order.