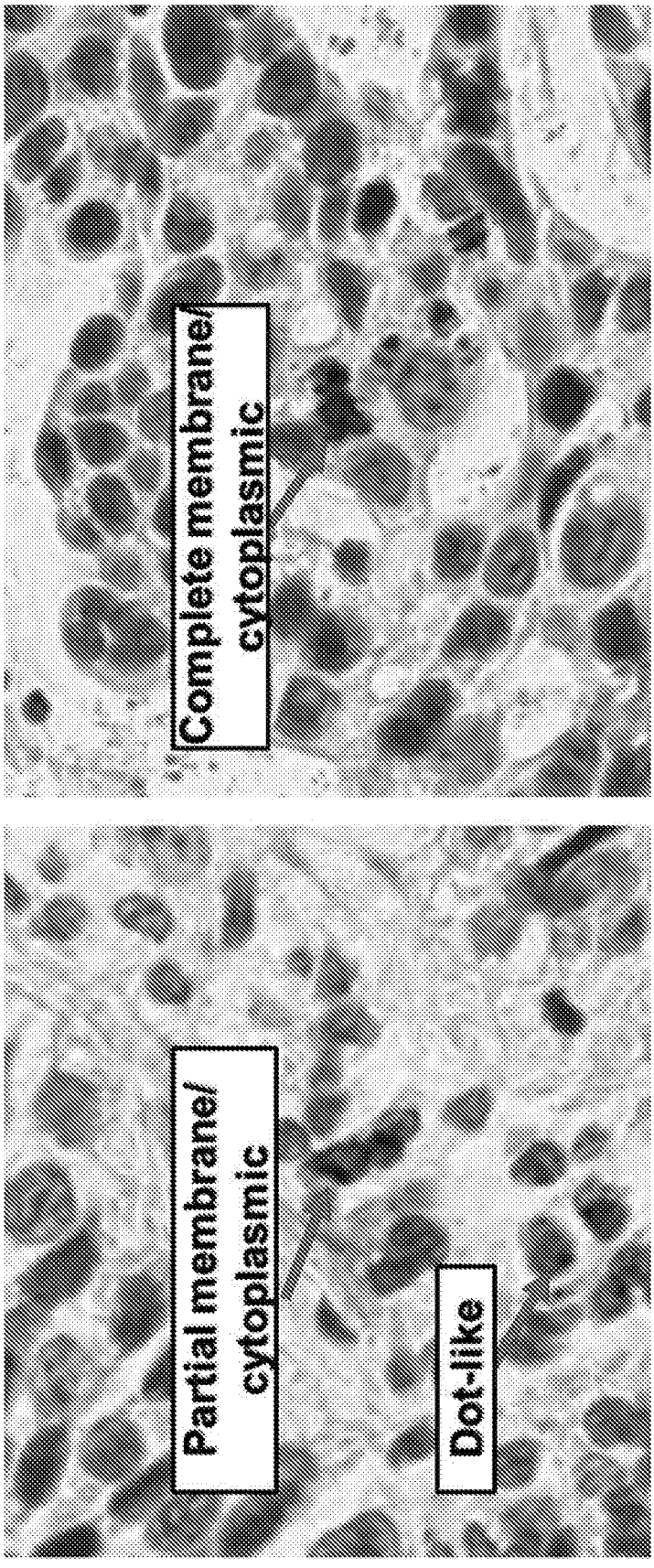
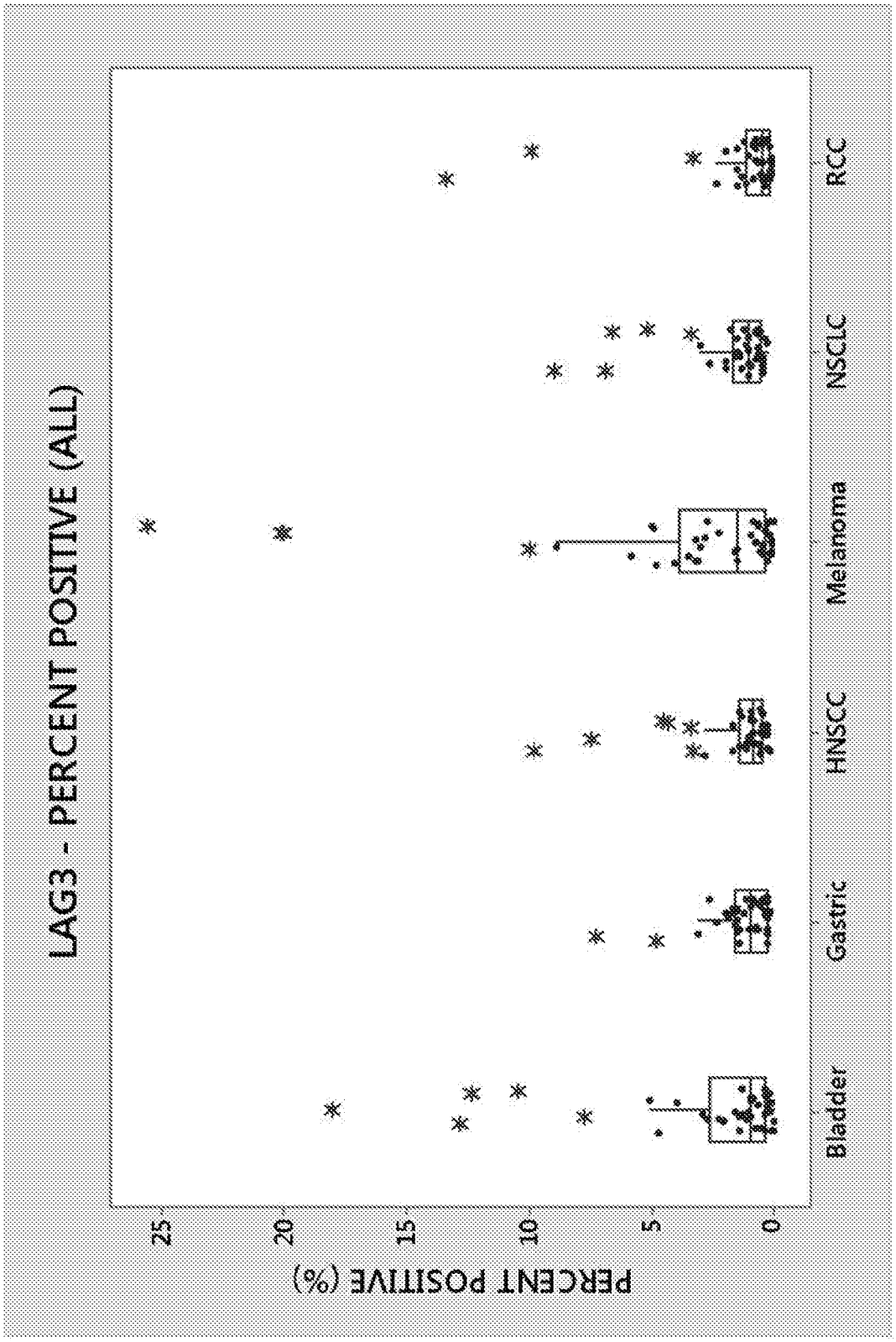
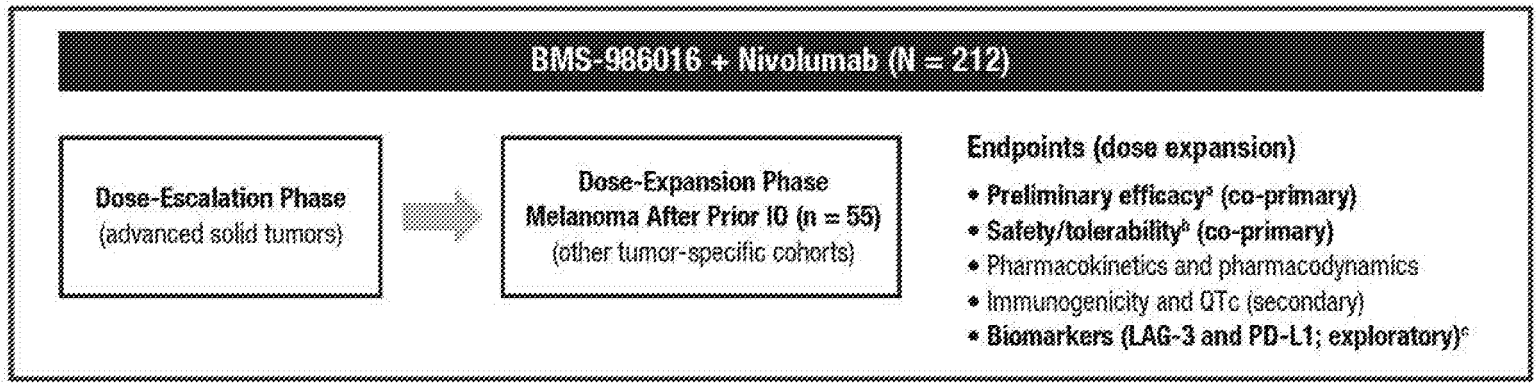

Complete membrane/
cytoplasmic
Partial membrane/
cytoplasmic
Dot-like

Figure 1

Figure 2


LAG3 - PERCENT POSITIVE (ALL)
PERCENT POSITIVE (%)
0
5
10
15
20
25
Bladder
Gastric
HNSCC
Melanoma
NSCLC
RCC

## Figure 3A




[a]Tumor response evaluated per Response Evaluation Criteria in Solid Tumors (RECIST) v1.1[19] (investigator assessment). [b]Safety evaluated per Common Terminology Criteria for Adverse Events v4.0[41] during treatment and up to 135 days after discontinuation. [c]LAG-3 and PD-L1 expression (percent of positive cells within invasive margin, tumor, and stroma) evaluated using immunohistochemistry (IHC) assays on formalin-fixed, paraffin-embedded tumor sections. Immune cell LAG-3 expression (≥ 1% or < 1%) determined using mouse antibody clone 17B4; tumor cell PD-L1 expression (≥ 1% or < 1%) determined using Dako 28-8 PD-L1 IHC 28-8 kit.

## Figure 3B

### Key Eligibility Criteria for Patients in the Melanoma Prior-IO Expansion Cohort

| Inclusion | Exclusion |
|---|---|
| At least 18 years of age and ECOG PS ≤ 1 | Prior exposure to IOs other than anti–CTLA-4 and/or anti–PD-1/PD-L1 antibody therapy (eg, anti–PD-L2, anti–KIR, anti–CD137, and anti–OX40) |
| Advanced (metastatic and/or unresectable) melanoma | Uveal melanoma |
| Progression on/intolerance of ≥ 1 and ≤ 3 prior standard regimens | Uncontrolled brain metastases |
| Prior treatment with anti–PD-1/PD-L1 (± anti–CTLA-4 ± BRAF ± MEK inhibitors) required | Active autoimmune disease |
| Prior adjuvant or neoadjuvant therapy with cytokines (IL-2 or IFN) or anti–CTLA-4 antibodies allowed | |
| At least 1 measurable lesion per RECIST v1.1[19] | |

CD137, cluster of differentiation 137; CTLA-4, cytotoxic T lymphocyte antigen-4; ECOG PS, Eastern Cooperative Oncology Group performance status; IFN, interferon; IL-2, interleukin-2; KIR, anti–killer-cell immunoglobulin-like receptor.

## Figure 4

Table 1. **Baseline Demographics and Disease Characteristics**

| | Mel Prior IO (n = 55) |
|---|---|
| **Median age (range), years** | 59 (25–80) |
| < 65 years, n (%) | 36 (65) |
| **Male, n (%)** | 39 (71) |
| **Race, n (%)** | |
| White | 52 (95) |
| Black | 1 (1.8) |
| Asian | 1 (1.8) |
| Other | 1 (1.8) |
| **ECOG PS, n (%)** | |
| 0 | 38 (69) |
| 1 | 17 (31) |
| **M stage at study entry, n (%)** | |
| M0 | 7 (13) |
| M1A | 6 (11) |
| M1B | 4 (7.3) |
| M1C with brain metastasis | 1 (1.8) |
| M1C without brain metastasis | 37 (67) |
| **LDH, n (%)** | |
| Normal | 25 (45) |
| Normal to < 2 × ULN | 13 (24) |
| ≥ 2 × ULN | 8 (15) |
| Unknown | 9 (16) |
| **Liver metastases, n (%)** | |
| Yes | 11 (20) |
| No | 44 (80) |
| **BRAF status, n (%)** | |
| Mutation | 21 (38) |
| No mutation | 31 (56) |
| Unknown | 3 (5.5) |

M, metastasis.

Figure 5

Table 2. **Prior Therapy**

| Patients, n (%) | Mel Prior IO (n = 55) |
|---|---|
| Prior radiotherapy | 16 (29) |
| Prior systemic therapy | 54 (98) |
| Immunotherapy | 54 (98) |
| Anti–CTLA-4[a] | 32 (58) |
| Anti–PD-1/PD-L1[b] | 52 (95) |
| Best response to prior anti–PD-1/PD-L1[c] | |
| CR | 1 (1.8) |
| PR | 12 (22) |
| SD | 16 (29) |
| PD | 22 (40) |
| BRAF inhibitors | 16 (29) |
| MEK inhibitors | 11 (20) |
| Number of systemic regimens | |
| 1 | 12 (22) |
| 2 | 17 (31) |
| ≥ 3 | 25 (45) |
| Median (range) | 2 (1–5) |

CR, complete response; PR, partial response; SD, stable disease. [a]Two patients received anti–PD-1/PD-L1 + anti–CTLA-4, 5 patients received anti–CTLA-4 after anti–PD-1/PD-L1, and 29 patients received anti–CTLA-4 before anti–PD-1/PD-L1; prior anti–CTLA-4 therapy was not reported in 4 patients. [b]Twenty-five patients received nivolumab, 25 patients received pembrolizumab, and 2 patients received other therapies; 2 patients did not receive prior anti–PD-1/PD-L1; prior anti–PD-1/PD-L1 was unknown in 1 patient. [c]Response in 1 patient was reported as not applicable.

Figure 6

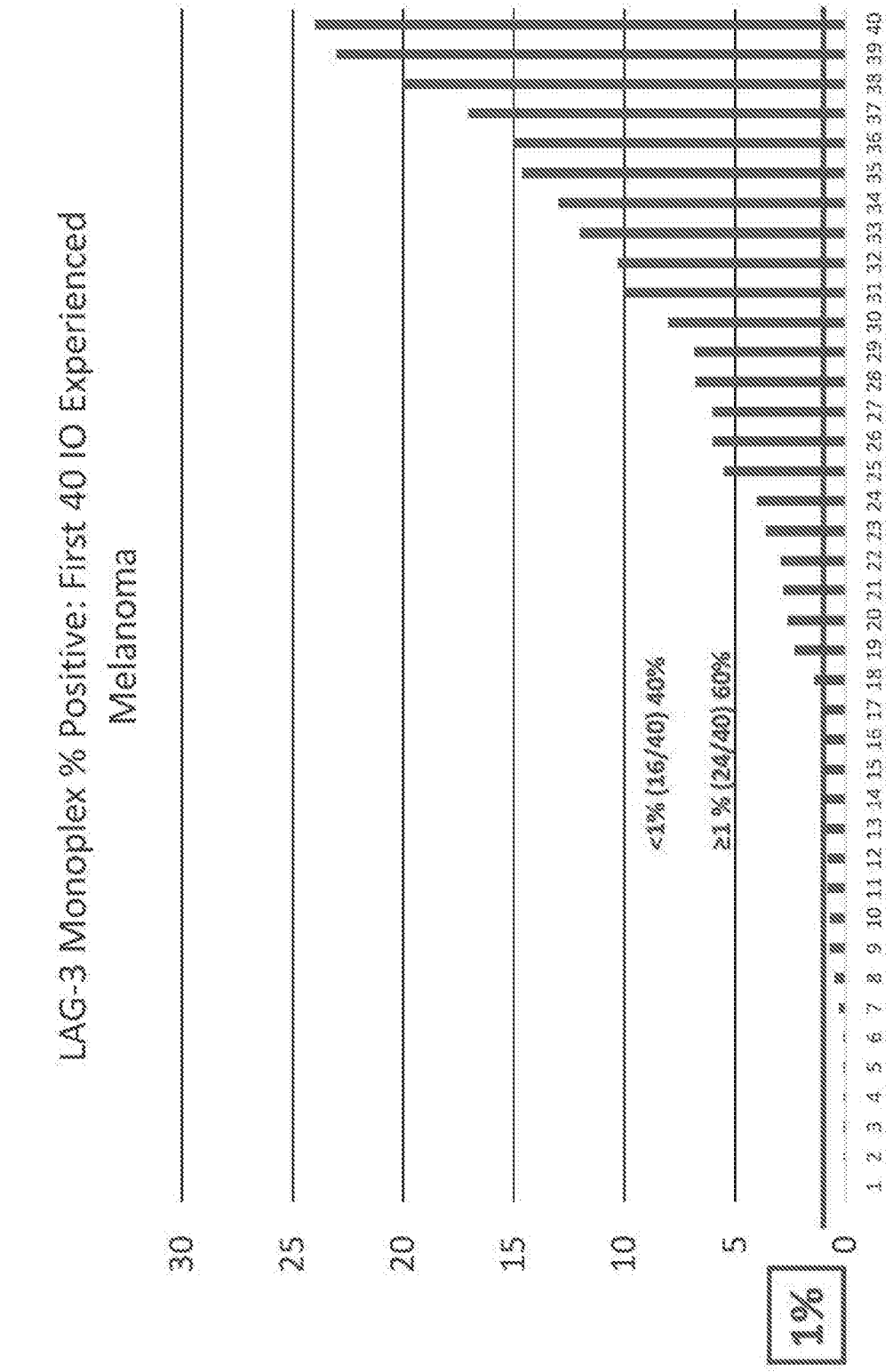

LAG-3 Monoplex % Positive: First 40 IO Experienced
Melanoma
30
25
20
15
10
5
0
1%
<1% (16/40) 40%
≥1 % (24/40) 60%
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40

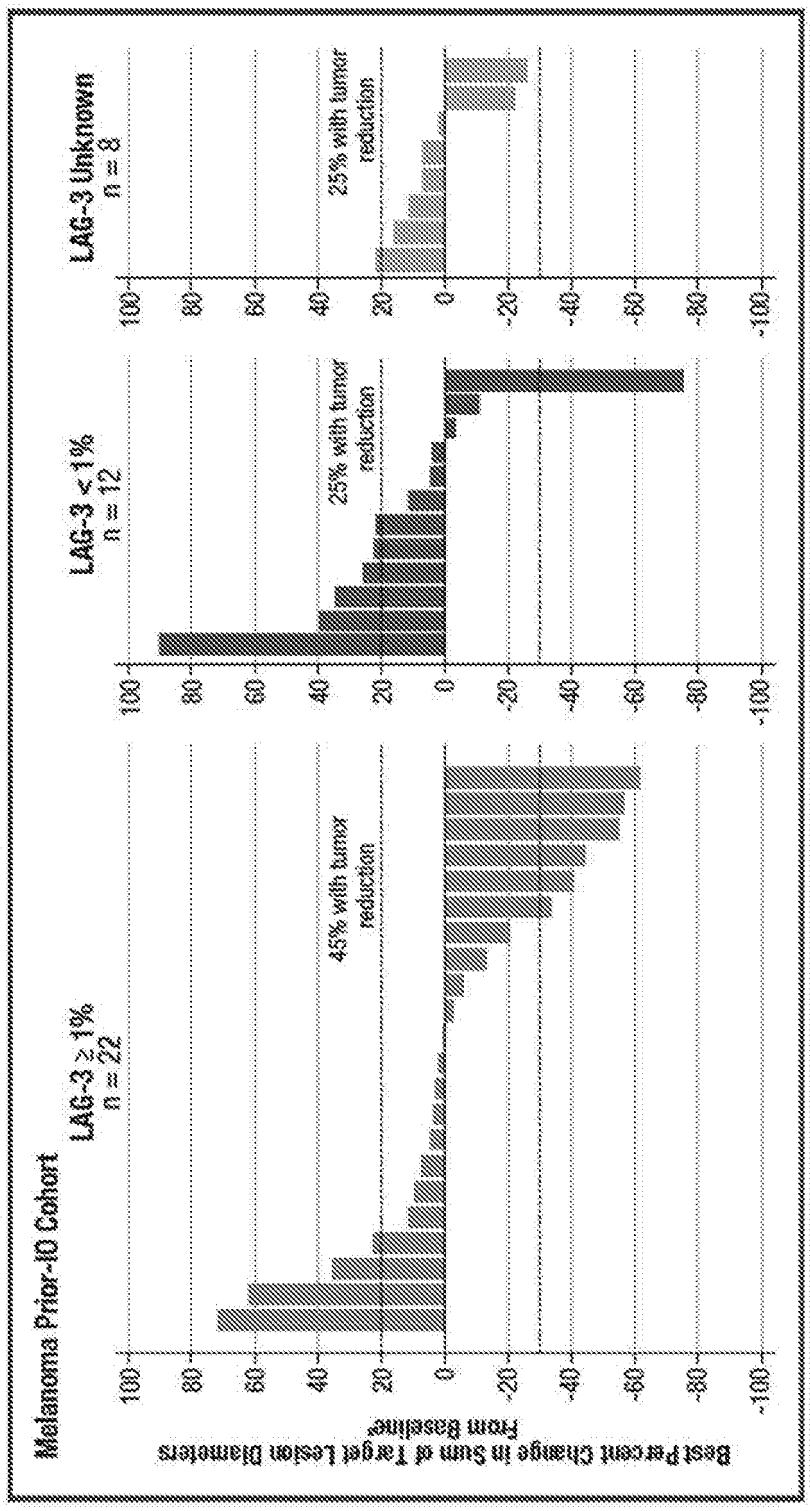
Figure 7

Table 3. **Preliminary Evidence of Antitumor Activity**

| Patients, n (%) | Mel Prior IO (n = 48[a]) |
|---|---|
| **BOR** | |
| CR | 0 |
| PR[b] | 6 (13) |
| SD | 20 (42) |
| PD | 16 (33) |
| Clinical progressions[c] | 6 (13) |
| **ORR**, 95% CI[b] | 6 (13), 4.7, 25 |
| LAG-3 ≥ 1% (n = 25) | 5 (20), 6.8, 41 |
| LAG-3 < 1% (n = 14) | 1 (7.1), 0.2, 34 |
| **DCR** (CR + PR + SD)[b] | 26 (54) |
| LAG-3 ≥ 1% (n = 25) | 16 (64) |
| LAG-3 < 1% (n = 14) | 5 (36) |

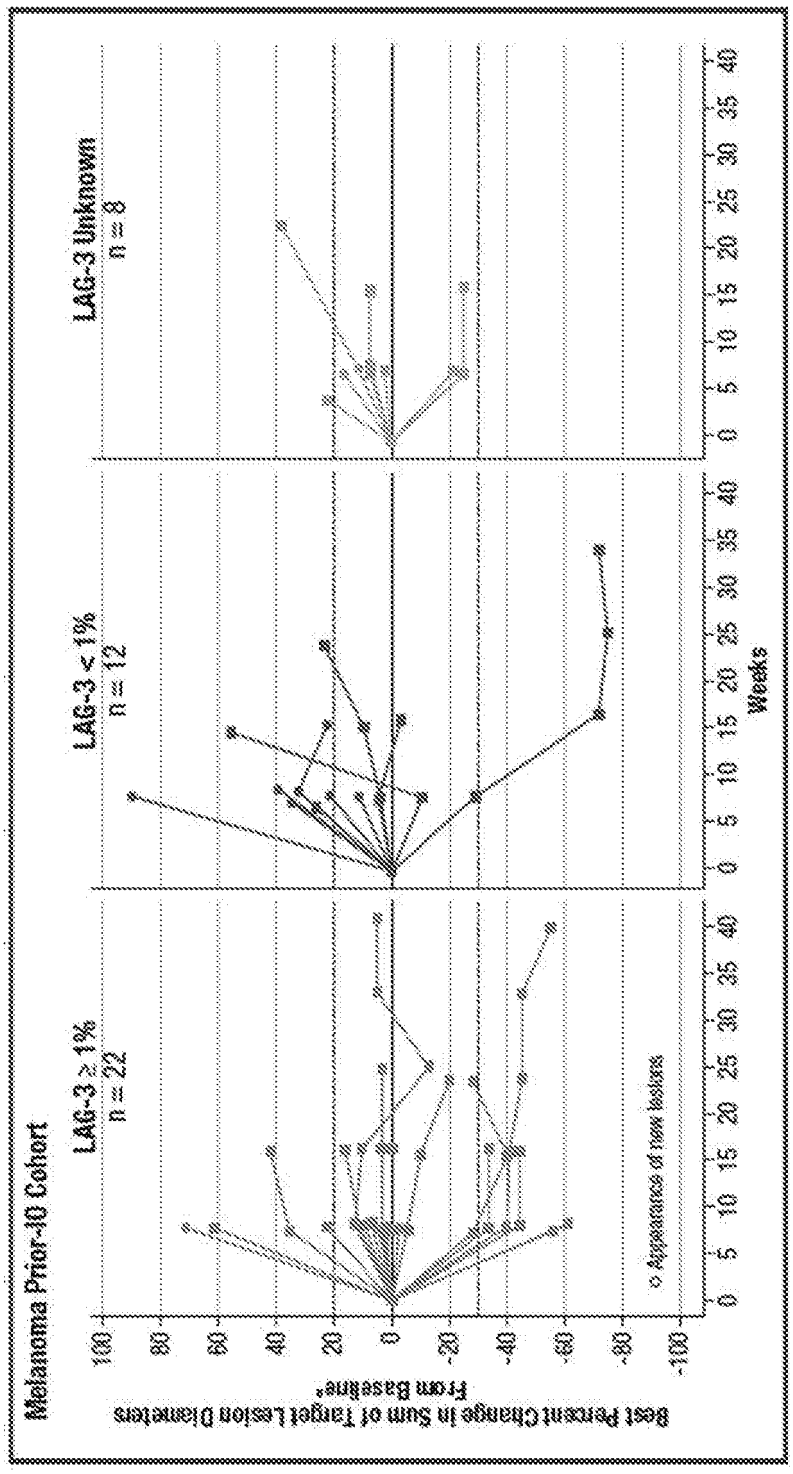
BOR, best overall response; DCR, disease control rate. [a]All response-evaluable patients; all progressed on prior anti-PD-1/PD-L1 therapy. [b]Two responses were unconfirmed. [c]Occurred prior to first radiographic scan.

Figure 8



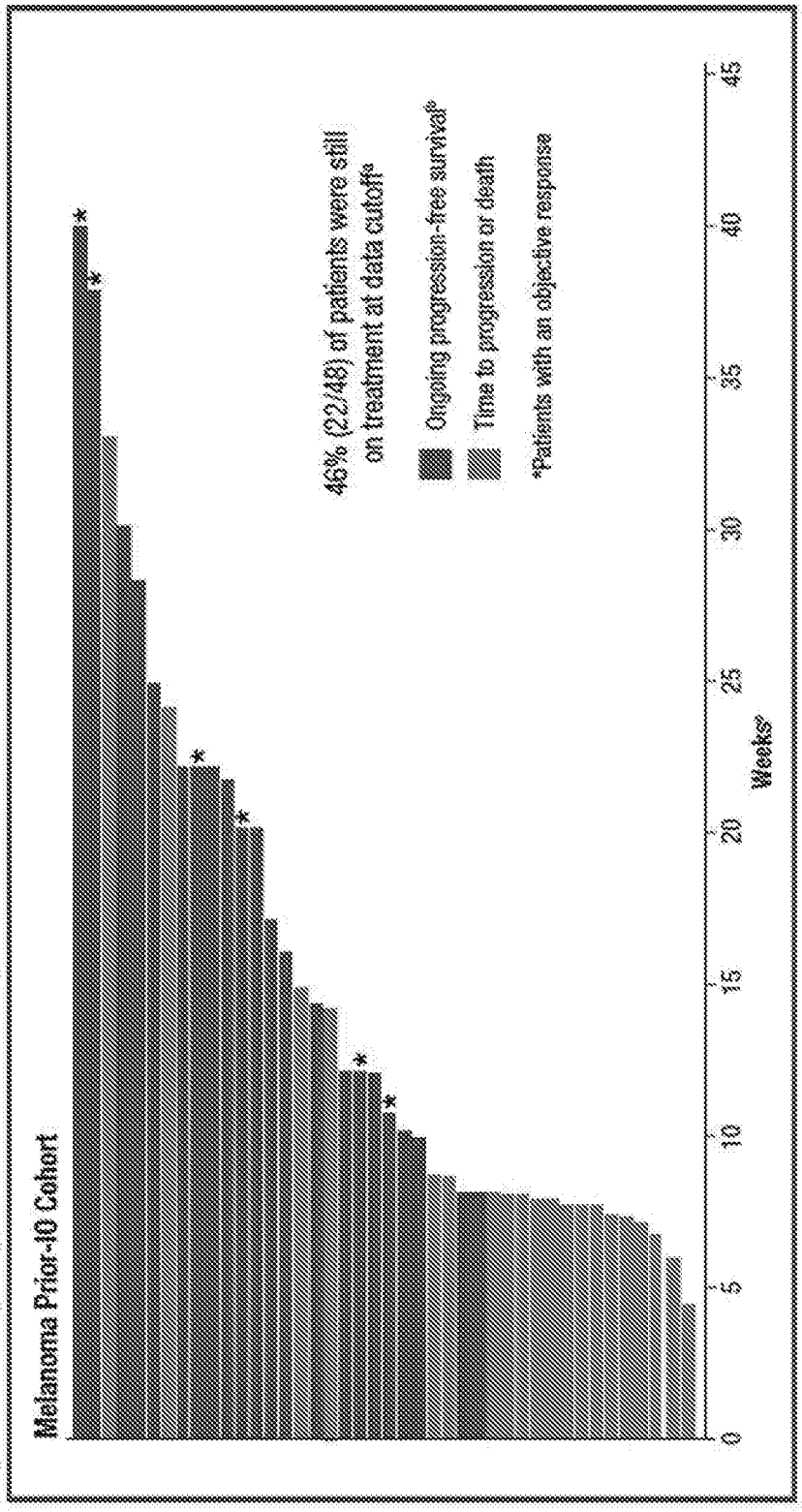

*Six patients had clinical progression prior to their first scan and are not included in the plot. One patient with best change from baseline > 30% had an unconfirmed best response of SD.

Figure 9




*Six patients had clinical progression prior to their first scan and are not included in the plot.

Figure 10




*Six patients had clinical progression prior to their first scan and are not included in the plot. *Censored on last visit. *Evaluations are planned for every 8 weeks.

Figure 11

Table 4. Response by Baseline Characteristics (investigator assessed)

| | Mel Prior IO (n = 40*) | | |
|---|---|---|---|
| | | ORR | |
| | n | n (%) | 95% CI |
| **LAG-3 expression** | | | |
| ≥ 1% | 25 | 5 (20) | 6.8, 41 |
| < 1% | 14 | 1 (1.7) | 0.2, 34 |
| **PD-L1 expression** | | | |
| ≥ 1% | 16 | 2 (13) | 1.6, 38 |
| < 1% | 19 | 4 (21) | 6.1, 46 |
| **Response based upon prior BOR to anti–PD-1/PD-L1 only** | | | |
| CR | 1 | 0 | – |
| PR | 12 | 2 (17) | – |
| SD | 14 | 1 (7.1) | – |
| CR + PR + SD | 27 | 3 (11) | 2.4, 29 |
| PD | 20 | 2 (10) | 1.2, 32 |
| **M stage at study entry** | | | |
| M1A | 6 | 1 (17) | 0.4, 64 |
| M1B | 3 | 0 | 0, 71 |
| M1C with brain metastasis | 1 | 0 | 0, 97.5 |
| M1C without brain metastasis | 32 | 4 (13) | 3.5, 29 |
| **Liver metastases** | | | |
| Yes | 11 | 1 (9.1) | 0.2, 41 |
| No | 37 | 5 (14) | 4.5, 29 |
| **BRAF status** | | | |
| Mutation | 19 | 0 | 0, 18 |
| No mutation | 28 | 6 (21) | 8.3, 41 |

*All response-evaluable patients; all progressed on prior anti–PD-1/PD-L1 therapy.

**Figure 12**

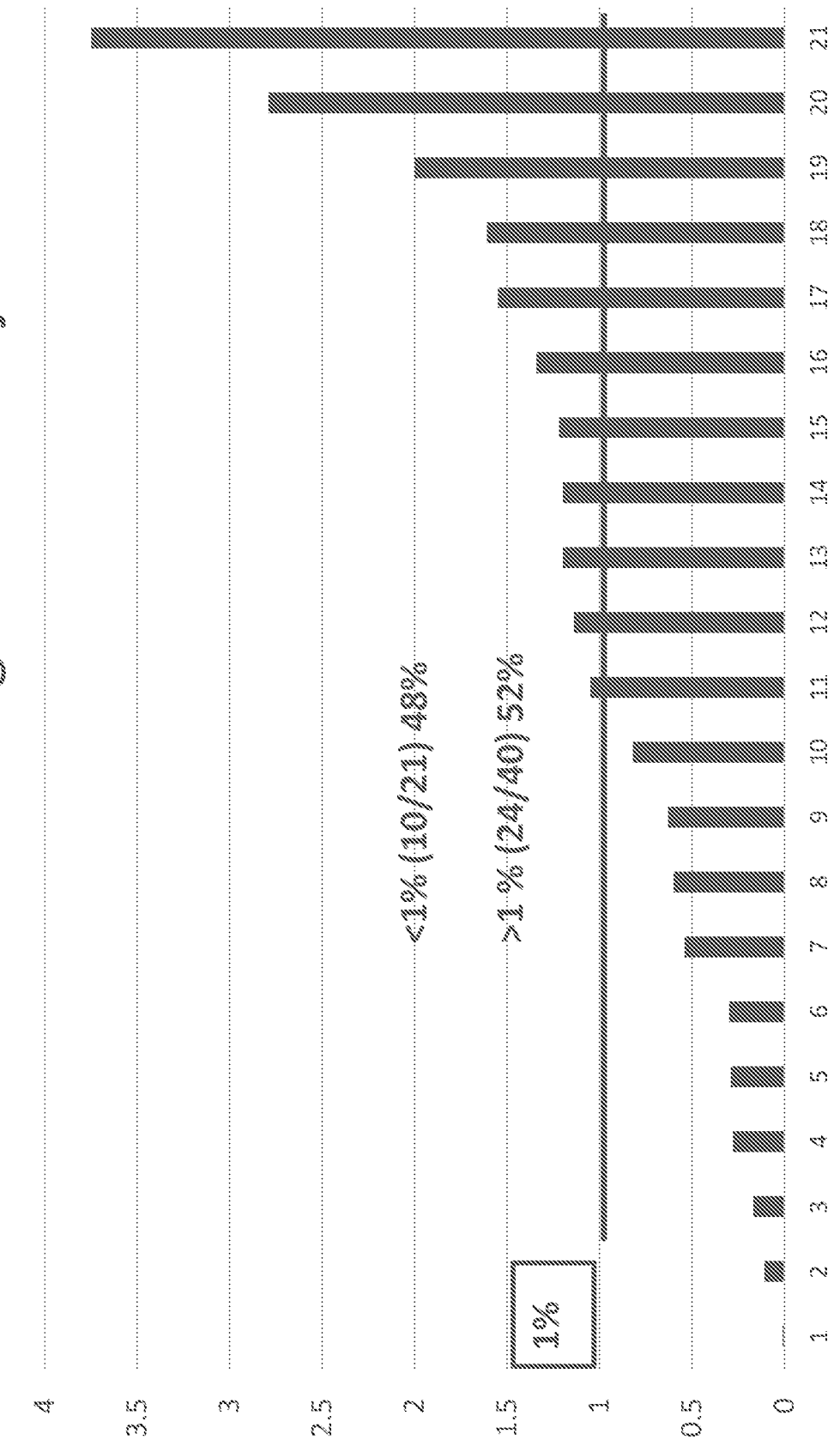

Gastric LAG3 Staining First 21 Subjects
4
3.5
3
2.5
2
1.5
1
0.5
0
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21
1%
<1% (10/21) 48%
>1 % (24/40) 52%

Figure 13

**Preliminary Clinical Activity of LAG-3 + nivo in IO Naïve Gastric Patients Enriched in LAG+ Patients**

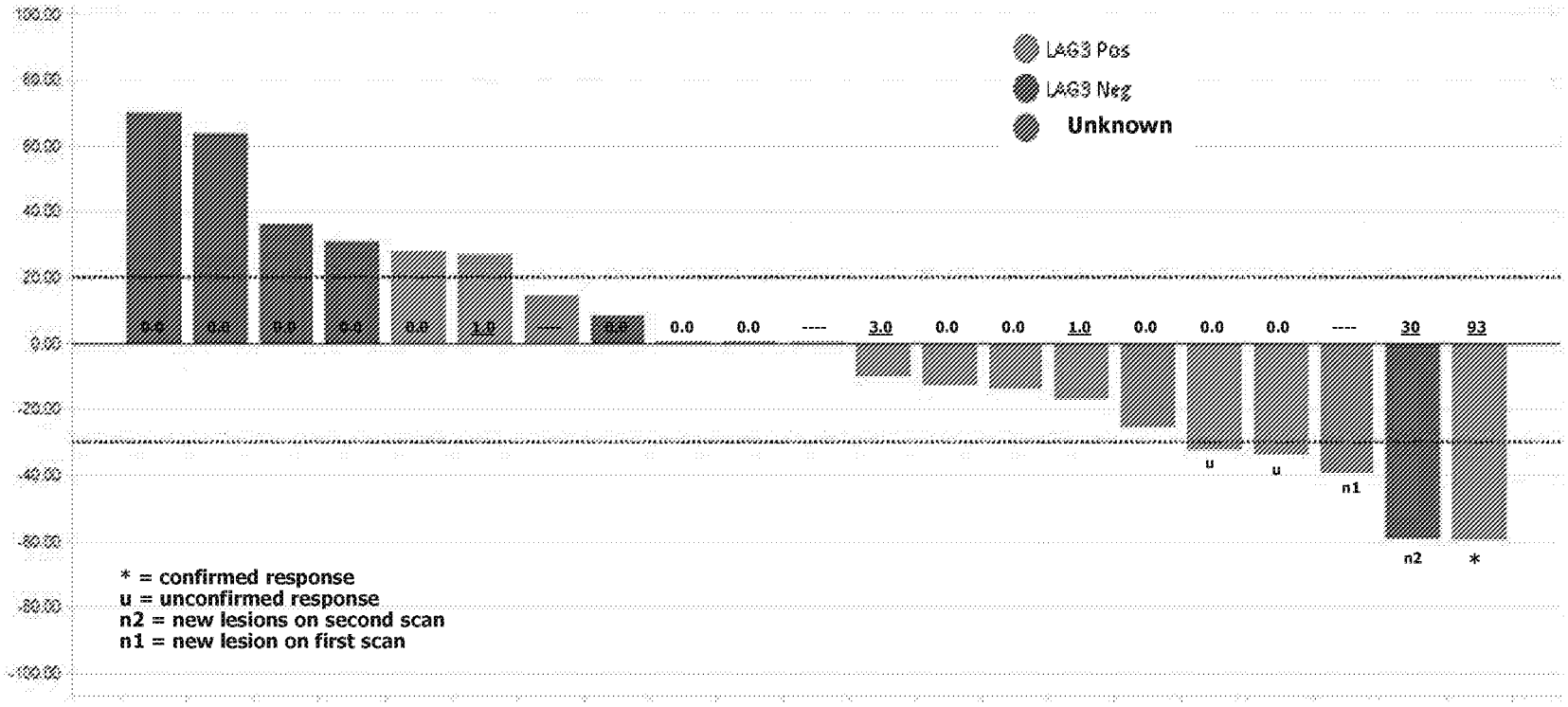


**Figure 14**

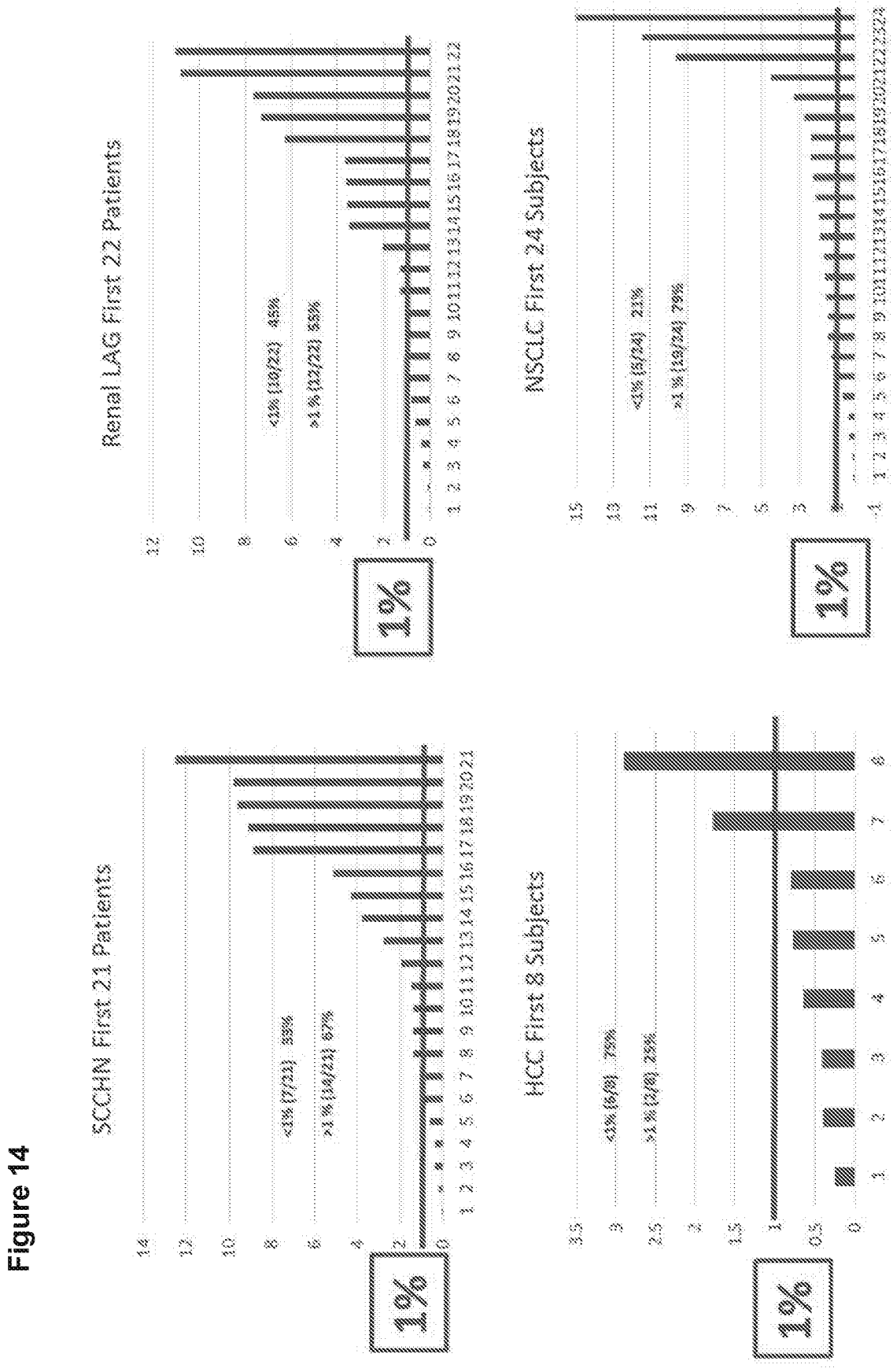
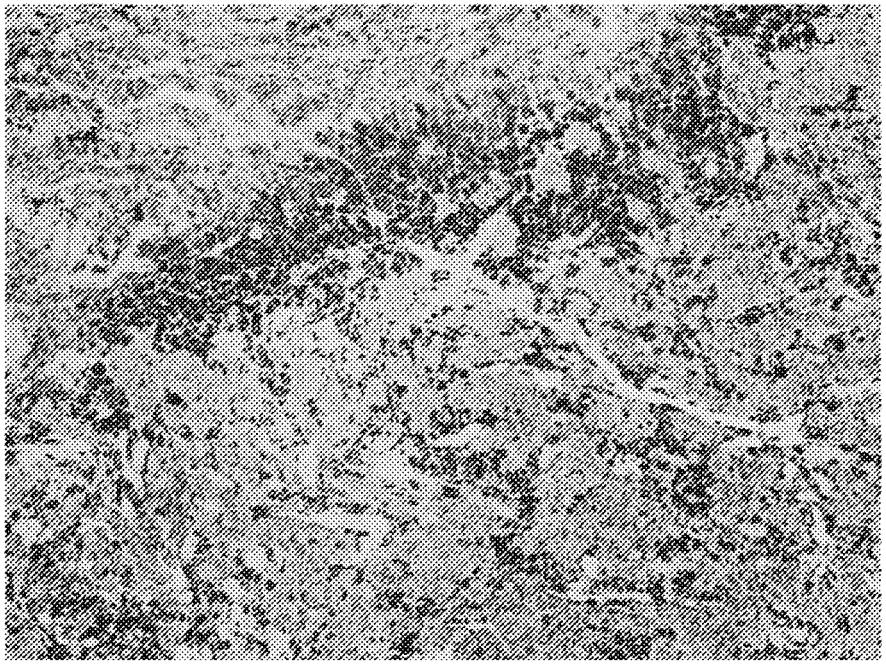
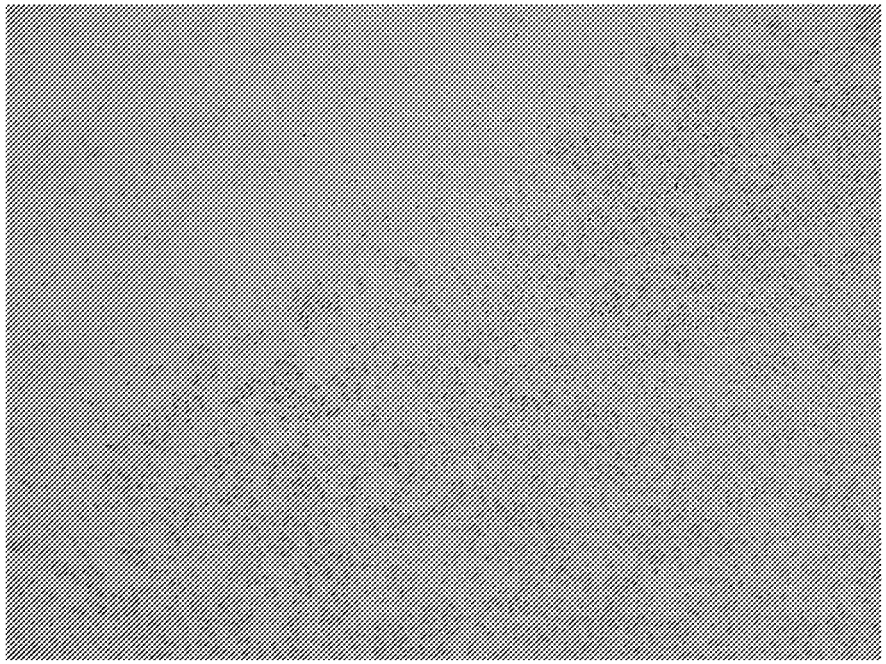

SCCHN First 21 Patients
<1% (7/21) 33%
>1 % (14/21) 67%
1%
Renal LAG First 22 Patients
<1% (10/22) 45%
>1 % (12/22) 55%
1%
HCC First 8 Subjects
<1% (6/8) 75%
>1 % (2/8) 25%
1%
NSCLC First 24 Subjects
<1% (5/24) 21%
>1 % (19/24) 79%
1%

**Figure 15A**

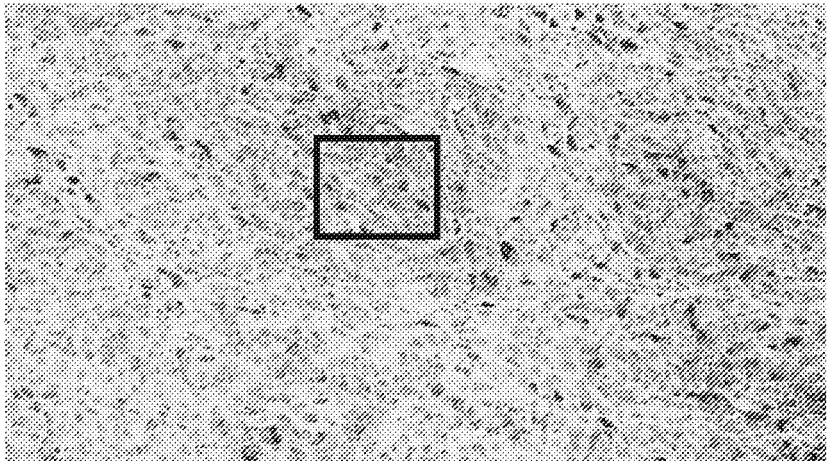


Pigmented melanoma without bleaching
(nuclei counterstained with hematoxylin)

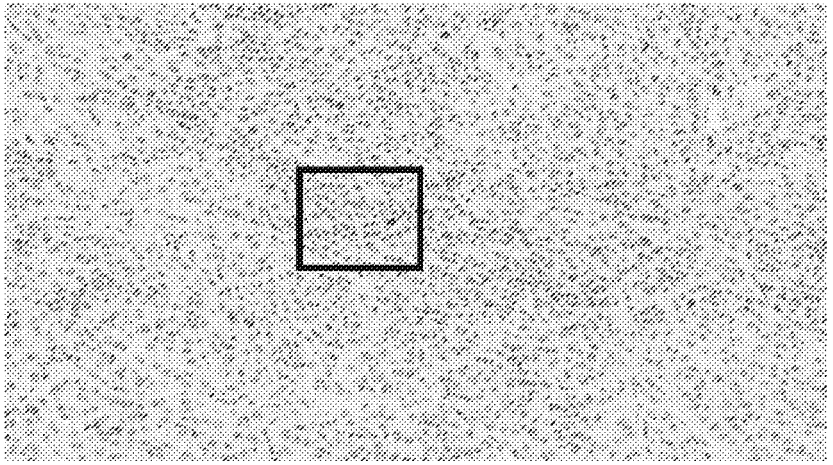


Pigmented melanoma following bleaching
(nuclei counterstained with hematoxylin)

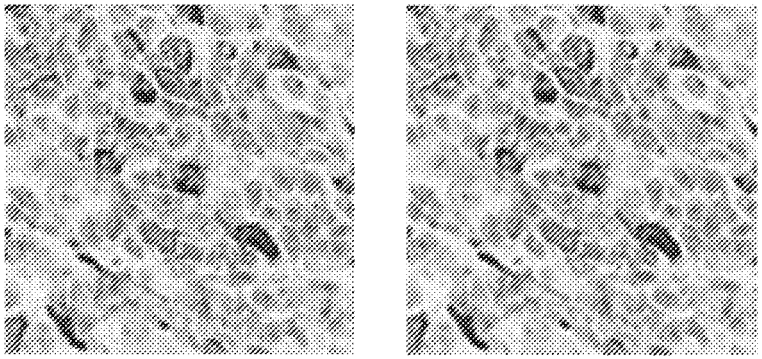
**Figure 15B**



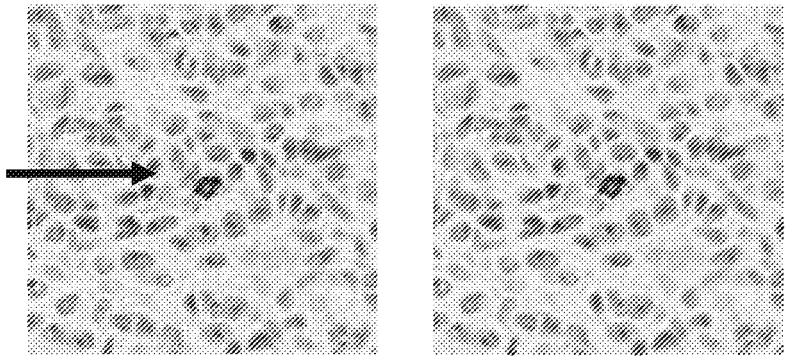
Pigmented melanoma - LAG-3 by IHC
without bleaching
(nuclei counterstained with hematoxylin)



Pigmented melanoma - LAG3 by IHC following
bleaching
(nuclei counterstained with hematoxylin)



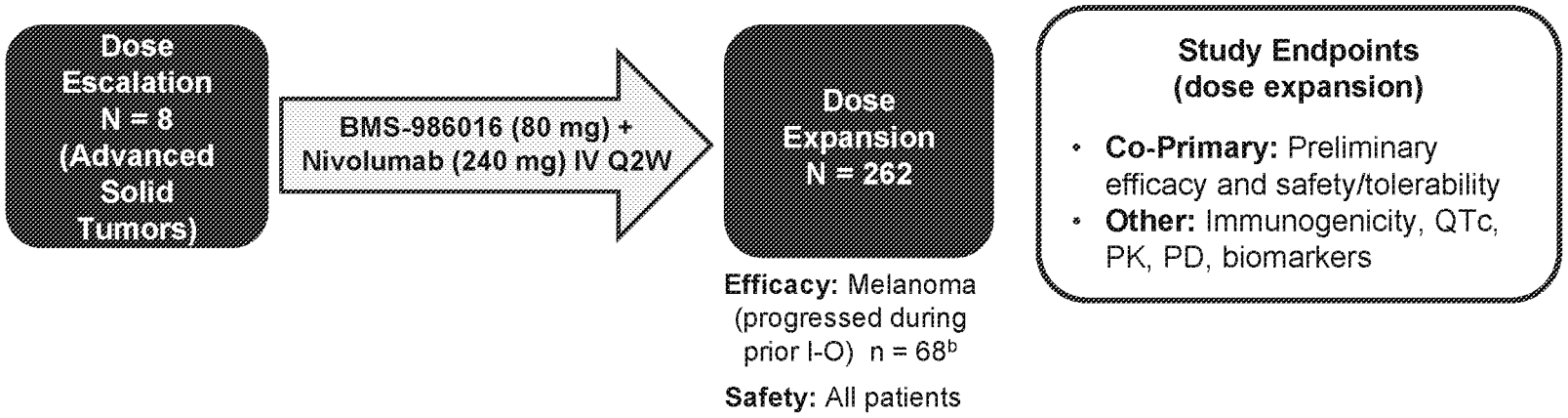
LAG-3 IHC indistinguishable from melanin



LAG-3 IHC following bleaching

**Figure 16**

## Study Rationale and Design




[b]Sixty-one patients were response-evaluable.

Figure 17

## Baseline Demographics and Disease Characteristics

| | Mel Prior PD-(L)1 n = 68 |
|---|---|
| Median age (range), years | 60 (25–81) |
| < 65 years, n (%) | 44 (65) |
| Male, n (%) | 46 (68) |
| Race, n (%) | |
| White | 65 (96) |
| Other | 3 (4.4) |
| ECOG PS, n (%) | |
| 0 | 44 (65) |
| 1 | 24 (35) |
| M stage at study entry, n (%) | |
| M0 | 6 (8.8) |
| M1A | 7 (10) |
| M1B | 8 (12) |
| M1C with brain metastasis | 1 (1.5) |
| M1C without brain metastasis | 46 (68) |

| | Mel Prior PD-(L)1 n = 68 |
|---|---|
| LDH, n (%) | |
| Normal | 35 (51) |
| Normal to < 2 × ULN | 15 (22) |
| ≥ 2 × ULN | 9 (13) |
| Unknown | 9 (13) |
| Liver metastases, n (%) | |
| No | 51 (75) |
| Yes | 17 (25) |
| *BRAF* status, n (%) | |
| No mutation | 43 (63) |
| Mutation | 23 (34) |
| Unknown | 2 (2.9) |

ECOG PS, Eastern Cooperative Oncology Group performance status; LDH, lactate dehydrogenase; M, metastasis; Mel Prior PD-(L)1, patients with melanoma who progressed during prior anti–PD-1/PD-L1 therapy; ULN, upper limit of normal.

**Figure 18**

## Prior Therapies

| | Anti-PD-1/PD-L1 n = 68 |
|---|---|
| Prior radiotherapy, n (%) | 19 (28) |
| Prior systemic therapy, n (%) | 68 (100) |
| Immunotherapy | 68 (100) |
| Anti–CTLA-4[a] | 39 (57) |
| Anti–PD-1/PD-L1[b] | 68 (100) |
| Best response to prior anti–PD-1/PD-L1[c] | |
| CR | 1 (1.5) |
| PR | 12 (18) |
| SD | 20 (29) |
| PD | 31 (46) |
| BRAF inhibitors | 19 (28) |
| MEK inhibitors | 13 (19) |
| Number of systemic regimens | |
| 1 | 16 (24) |
| 2 | 21 (31) |
| ≥ 3 | 31 (46) |
| Median (range) | 2 (1–5) |

CR, complete response; CTLA-4, cytotoxic T lymphocyte antigen-4;
PD, progressive disease; PR, partial response; SD, stable disease.
[a]Four patients received anti–PD-1/PD-L1 + anti–CTLA-4, 8 patients received anti–CTLA-4 after anti–PD-1/PD-L1, and 33 patients received anti–CTLA-4 before anti–PD-1/PD-L1; prior anti–CTLA-4 therapy was not reported in 2 patients [b]Thirty-three patients received nivolumab, 33 patients received pembrolizumab, and 2 patients received other therapies. [c]Response in 1 patient was reported as not applicable.

Figure 19

## Antitumor Activity of BMS-986016 + Nivolumab

| | Mel Prior PD-(L)1[a] | |
|---|---|---|
| | All n = 61 | LAG-3 ≥ 1%[b] n = 33 |
| ORR,[c] n (%)<br>95% CI | 7 (11.5)[d]<br>4.7, 22 | 6 (18)[d]<br>7, 36 |
| BOR,[c] n (%) | | |
| CR | 1 (1.6) | 1 (3.0) |
| PR | 6 (9.8)[d] | 5 (15)[d] |
| SD | 23 (38) | 15 (45) |
| PD | 25 (41) | 8 (24) |
| Clinical progression[e] | 6 (9.8) | 4 (12) |
| DCR (CR + PR + SD),[c] n (%)<br>95% CI | 30 (49)<br>36, 62 | 21 (64)<br>45, 80 |

BOR, best overall response.

[a]Response-evaluable patients; all progressed during prior anti–PD-1/PD-L1 therapy. [b]Immune-cell LAG-3 expression (percent of positive cells within invasive margin, tumor, and stroma) evaluated by IHC in tumor sections with antibody clone 17B4. [c]Tumor response evaluated by investigator per Response Evaluation Criteria in Solid Tumors v1.1. [d]One response was unconfirmed. [e]Occurred prior to first radiographic scan.

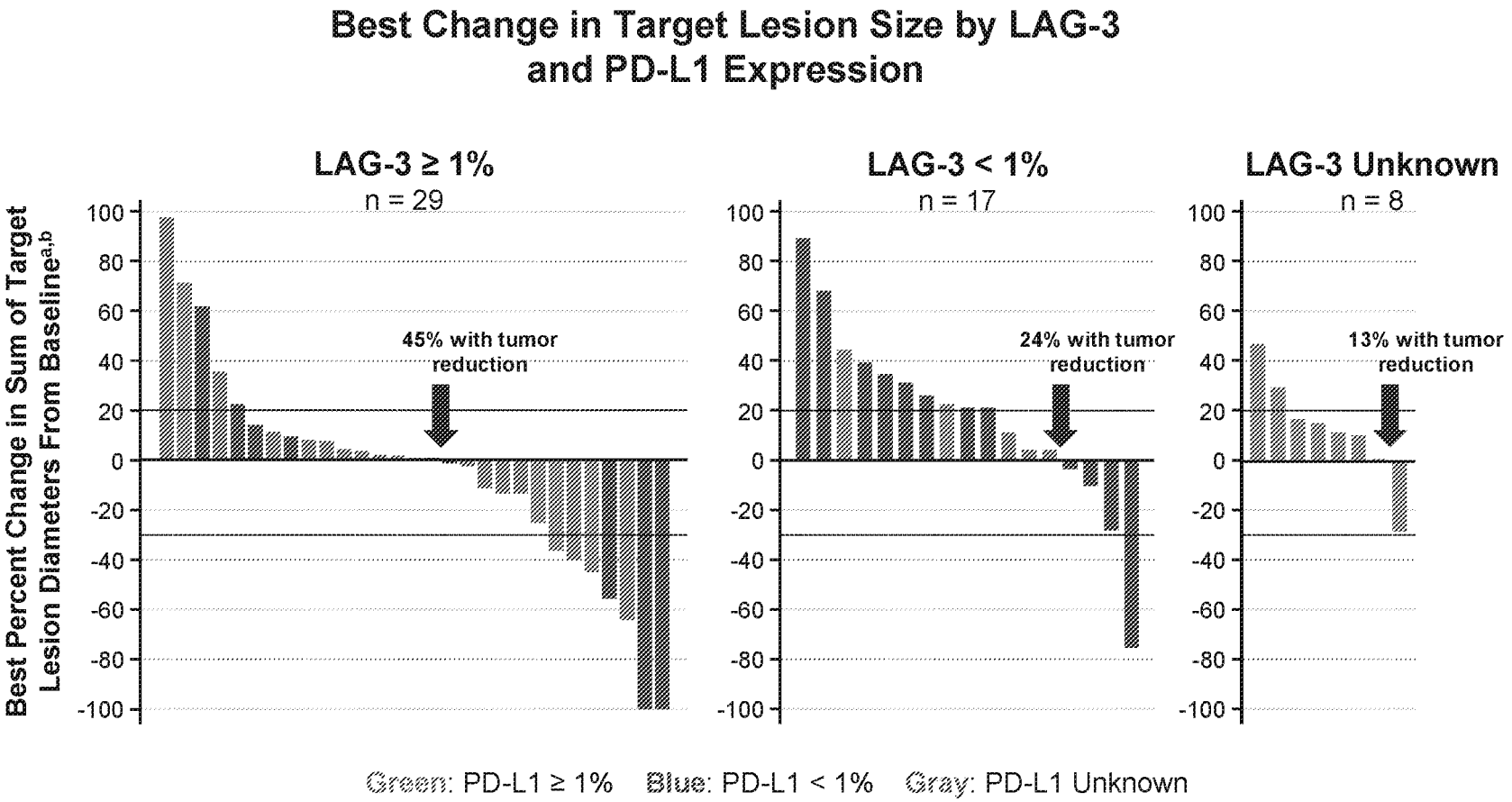
Figure 20

## Response by Baseline Characteristics and LAG-3 Expression

| | Met Prior PD (L)1[a] | | | |
|---|---|---|---|---|
| | LAG-3 Expression (N = 53) | | | |
| | LAG-3 ≥ 1% | | LAG-3 < 1% | |
| | N | ORR, n (%) | N | ORR, n (%) |
| PD-L1 expression[b] (N = 44) | | | | |
| ≥ 1% | 16 | 1 (6.3) | 4 | 0 |
| < 1% | 11 | 3 (27) | 13 | 1 (7.7) |
| *BRAF* status (N = 52) | | | | |
| No mutation | 21 | 5 (24) | 11 | 1 (9.1) |
| Mutation | 11 | 1 (9.1) | 9 | 0 |
| Prior anti–CTLA-4 (N = 53) | | | | |
| No | 12 | 1 (8.3) | 7 | 1 (14) |
| Yes | 21 | 5 (24) | 13 | 0 |

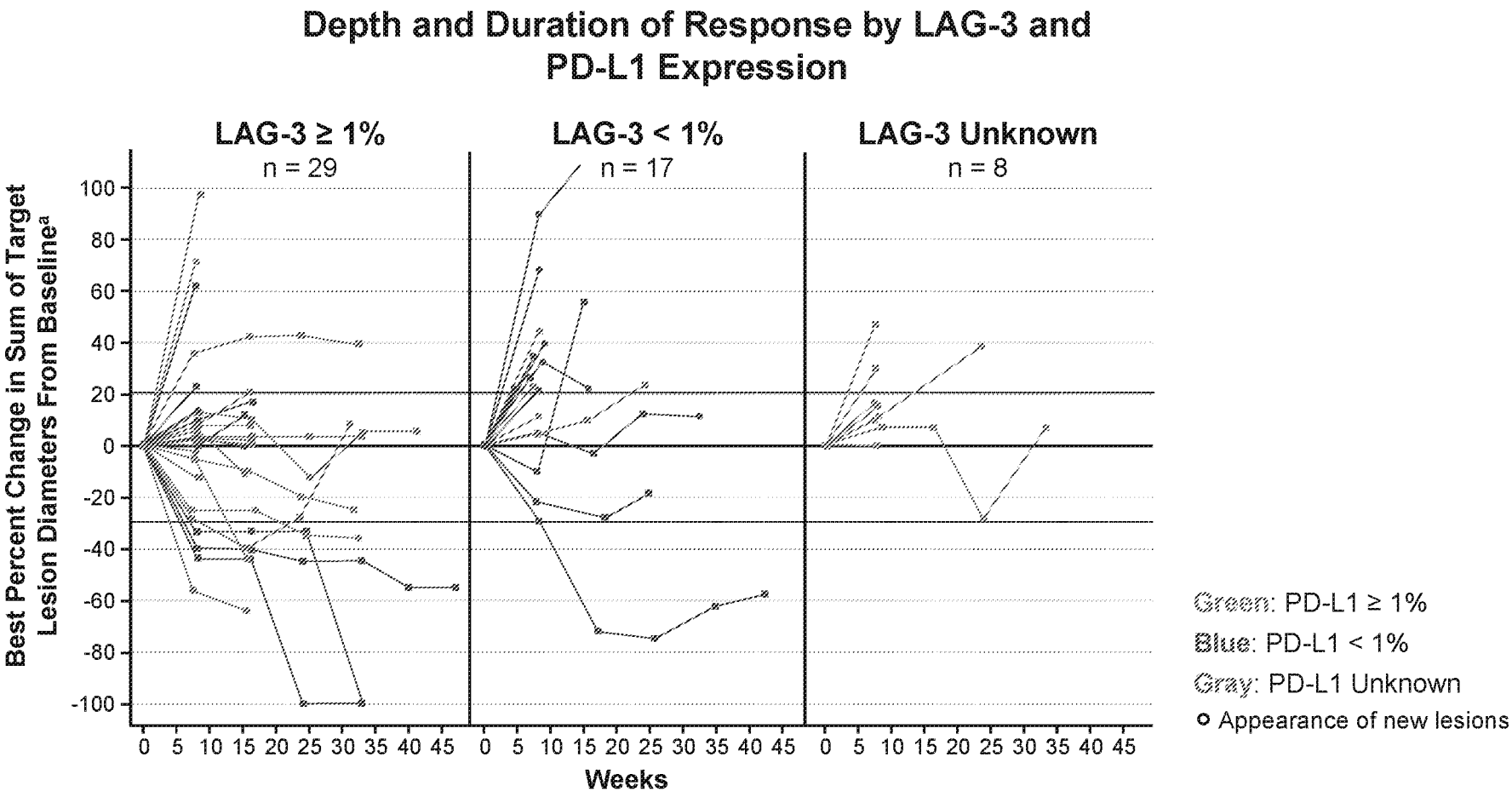
[a]Response-evaluable patients; all progressed during prior anti–PD-1/PD-L1 therapy. [b]Tumor-cell PD-L1 expression (percent of positive cells within invasive margin, tumor, and stroma) evaluated by IHC in tumor sections with the Dako PD-L1 IHC 28-8 kit.

Figure 21



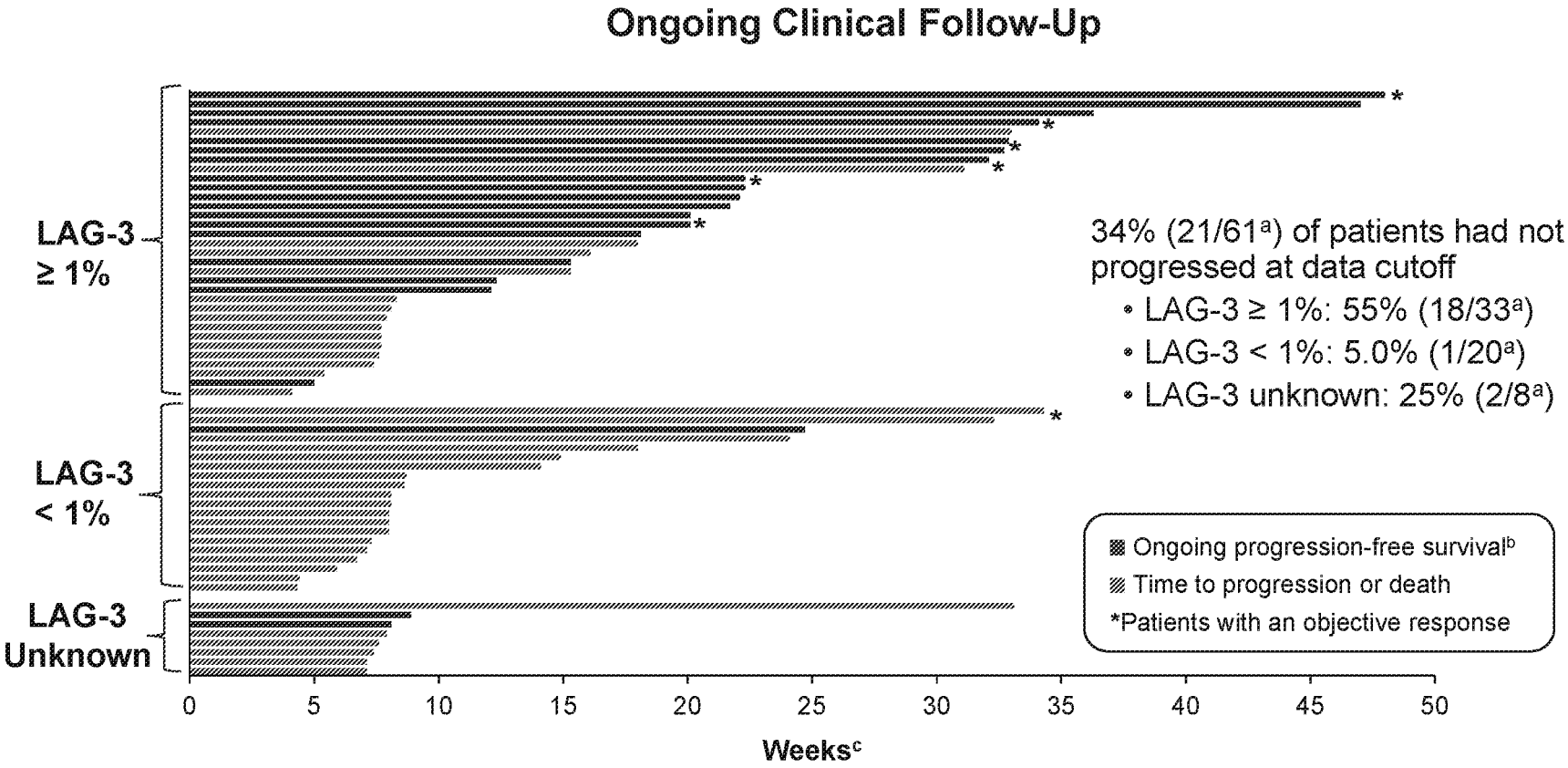

[a]Six patients with clinical progression prior to their first scan and one with PD due to an imaged new symptomatic brain metastasis prior to getting full scans were not included.
[b]One patient with best change from baseline > 30% had a best response of SD.

**Figure 22**




[a]Six patients with clinical progression prior to their first scan and one patient with PD due to a new symptomatic brain metastasis prior to getting full scans were not included.

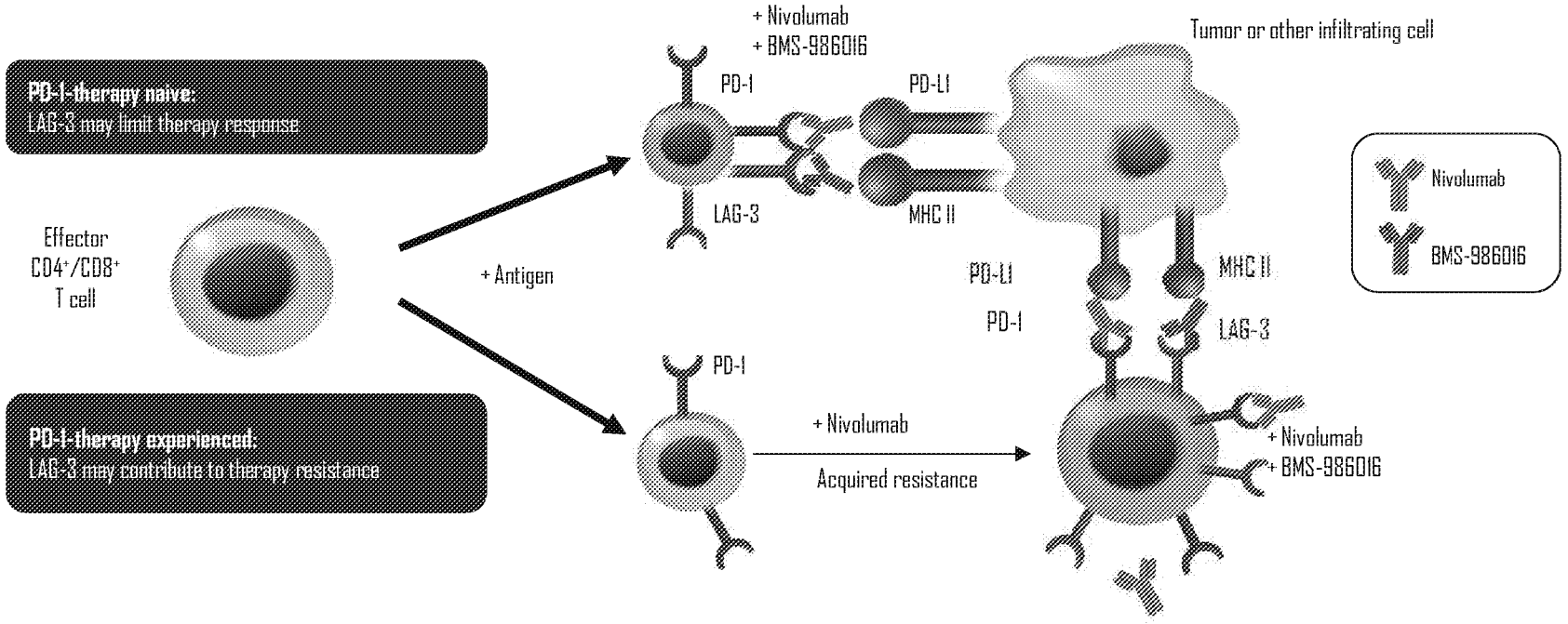
**Figure 23**




[a]Response-evaluable patients; all progressed during prior anti–PD-1/PD-L1 therapy. [b]Censored on last visit.
[c]Evaluations are planned for every 8 weeks.

**Figure 24**

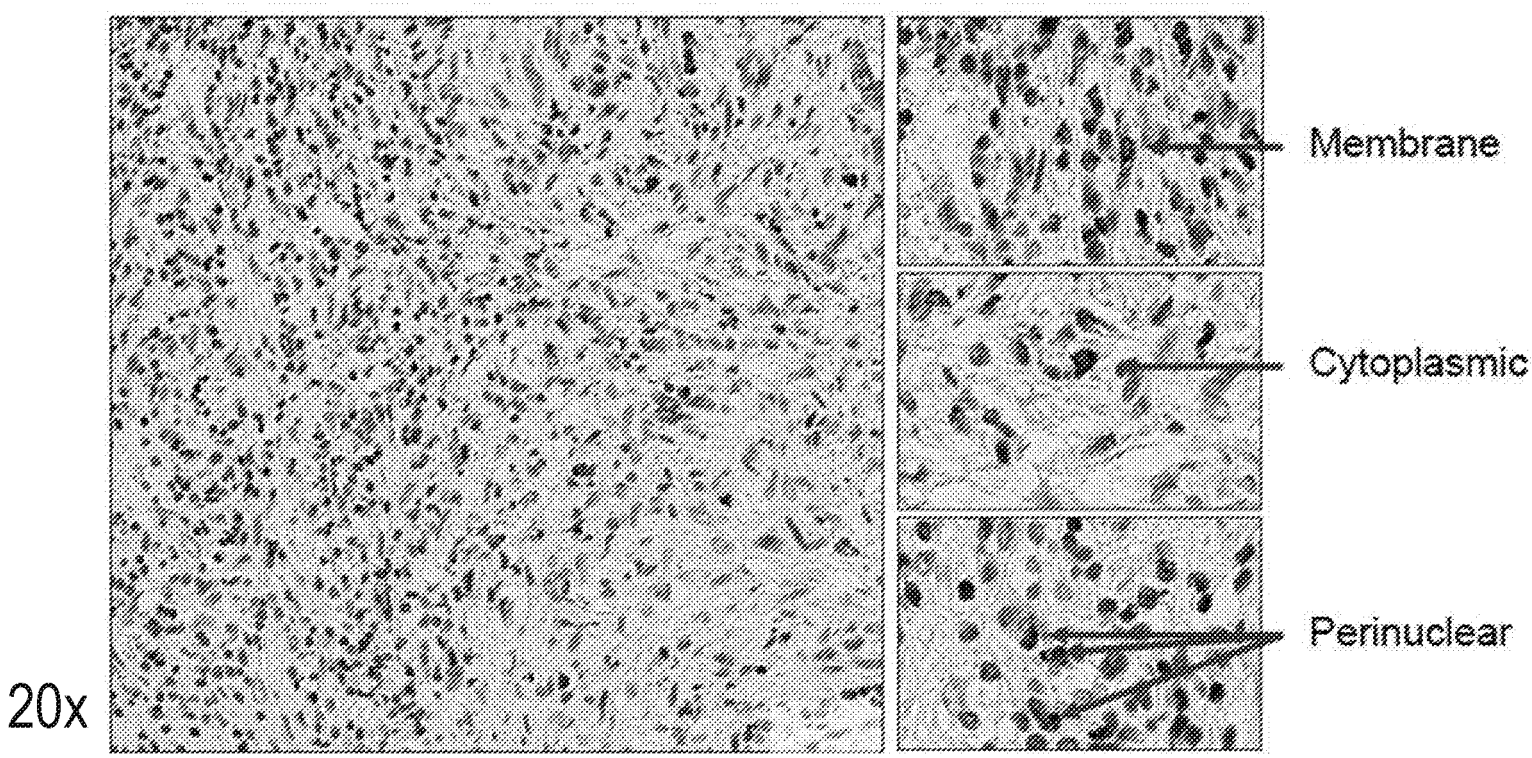
# Role of LAG-3 and PD-1 in T-cell exhaustion and proposed clinical utility of BMS-986016 combined with nivolumab




CD = cluster of differentiation; LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; PD-1 = programmed death-1; PD-L1 = programmed death-1 ligand 1.

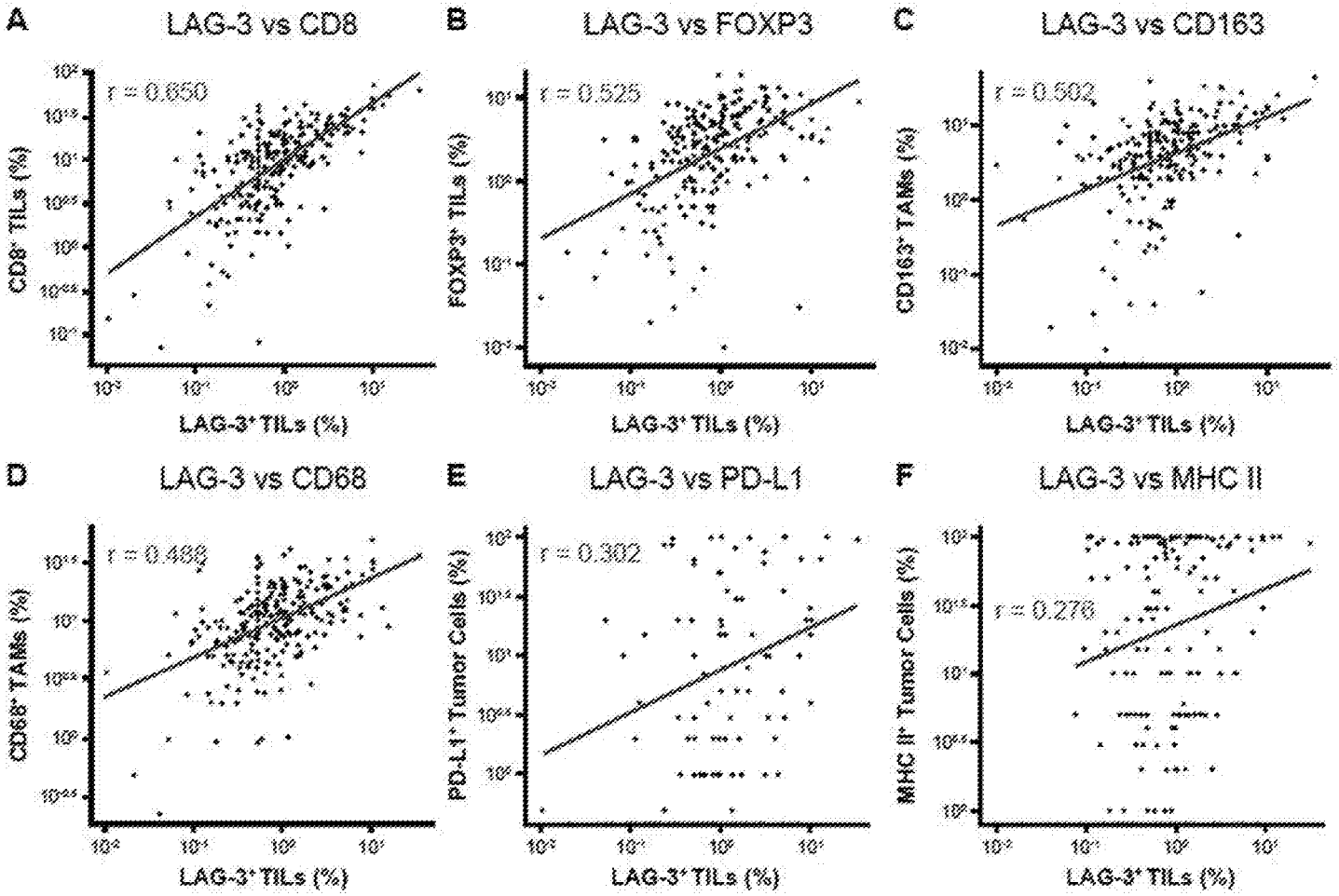
Figure 25

## LAG-3 patterns of expression by IHC staining of total nucleated cells in a melanoma tumor specimen



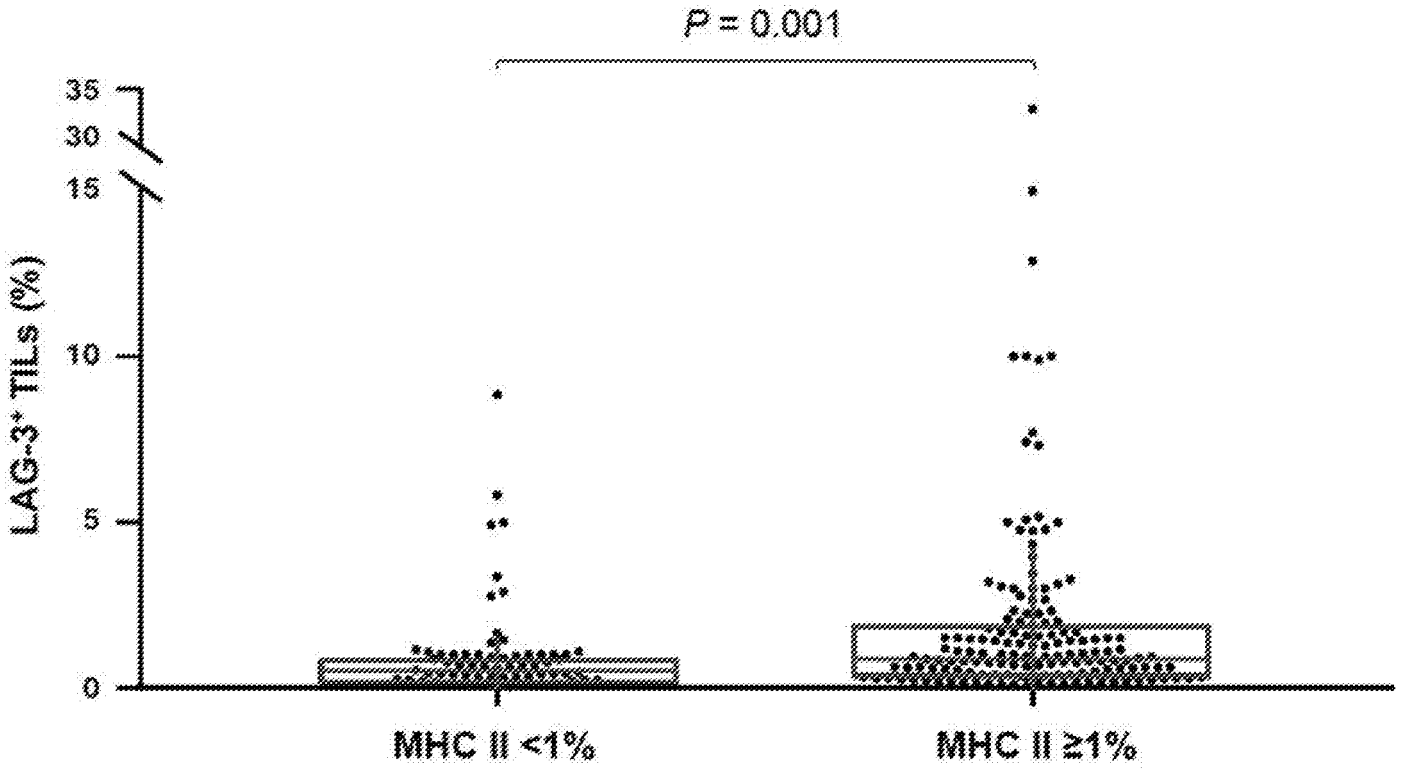

IHC = immunohistochemistry; LAG-3 = lymphocyte-activation gene 3.

Figures 26A-F. **Association of LAG-3 with immune and inflammatory biomarkers**




n = 237. LAG-3, CD8, FOXP3, CD163, CD68 quantified on immune cells; PD-L1 and MHC II quantified on tumor cells. CD = cluster of differentiation; FOXP3 = forkhead box P3; LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; PD-L1 = programmed death-1 ligand 1; TAMs = tumor-associated macrophages; TILs = tumor-infiltrating lymphocytes.

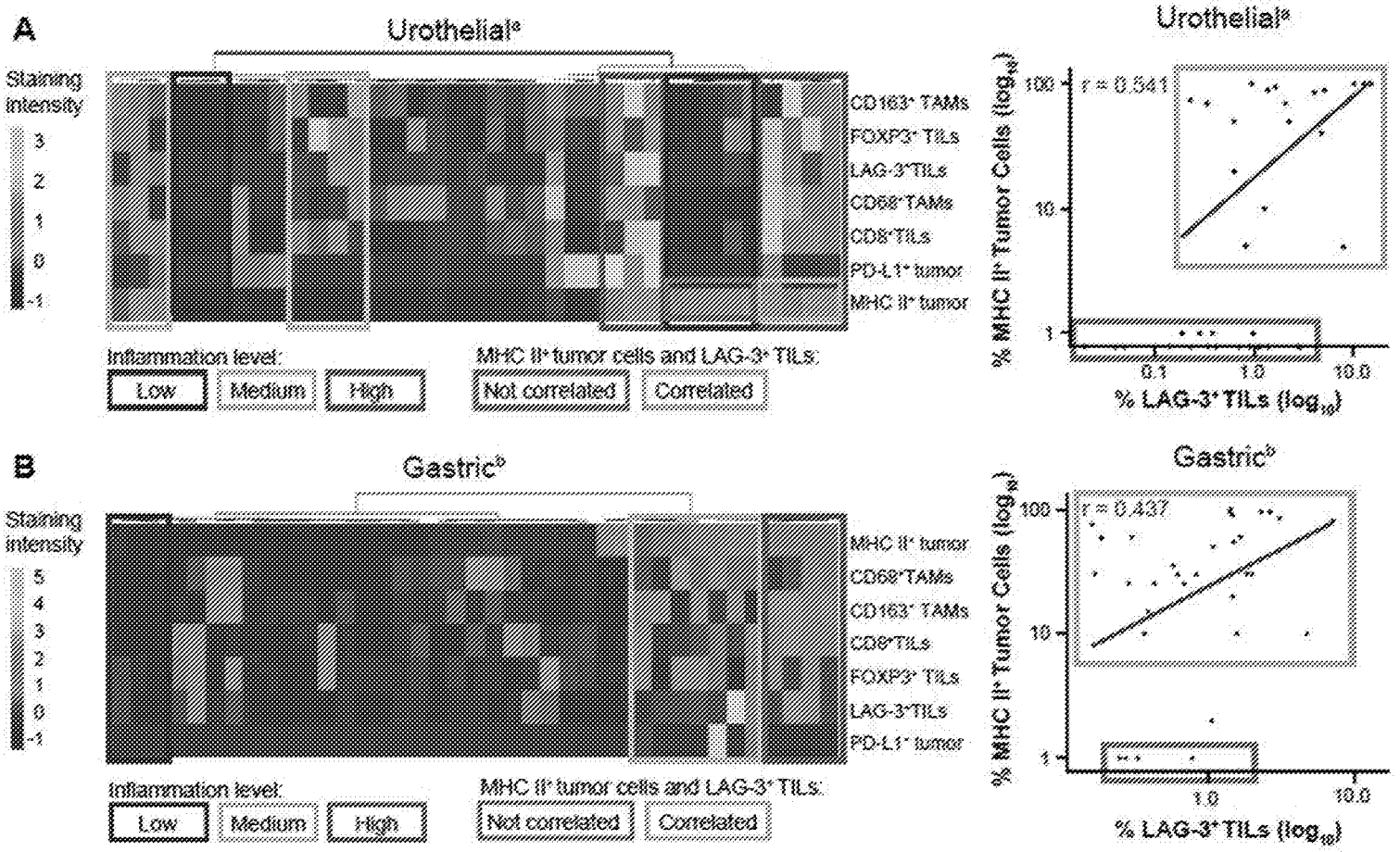
Figure 27. LAG-3+ TILs by MHC II tumor cell expression




n = 241. Vertical bars represent the interquartile range. LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; TILs = tumor-infiltrating lymphocytes.

Figures 28A-B.

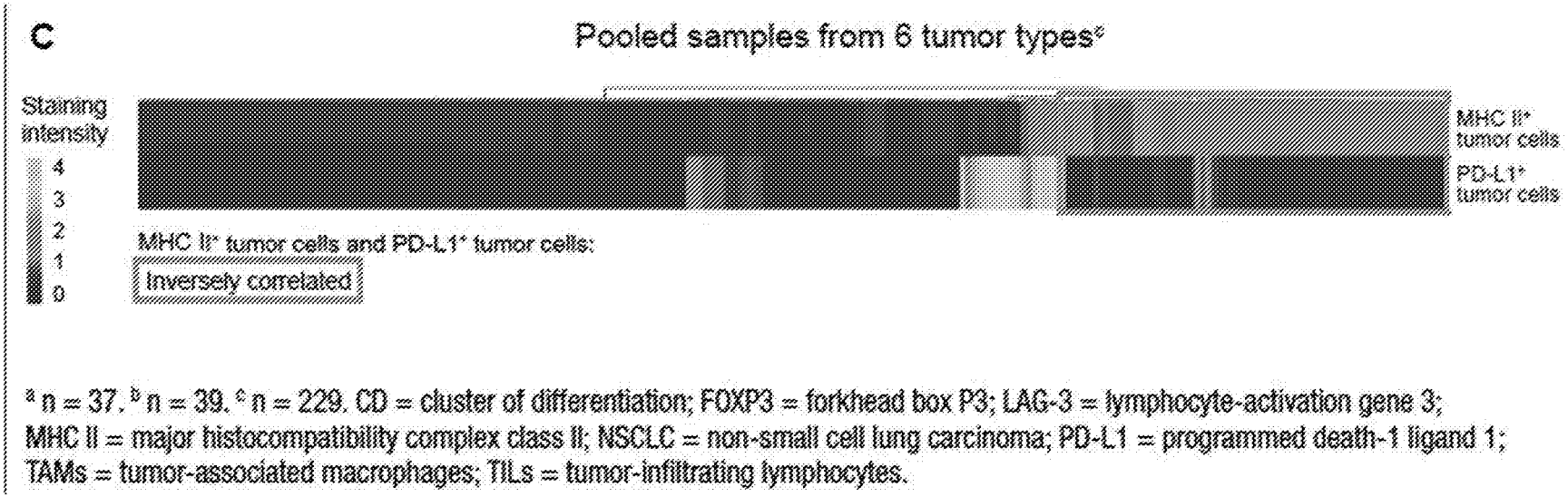
# Relationship between inflammation clusters and biomarker expression




[a] n = 37. [b] n = 39. [c] n = 229. CD = cluster of differentiation; FOXP3 = forkhead box P3; LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; NSCLC = non-small cell lung carcinoma; PD-L1 = programmed death-1 ligand 1; TAMs = tumor-associated macrophages; TILs = tumor-infiltrating lymphocytes.

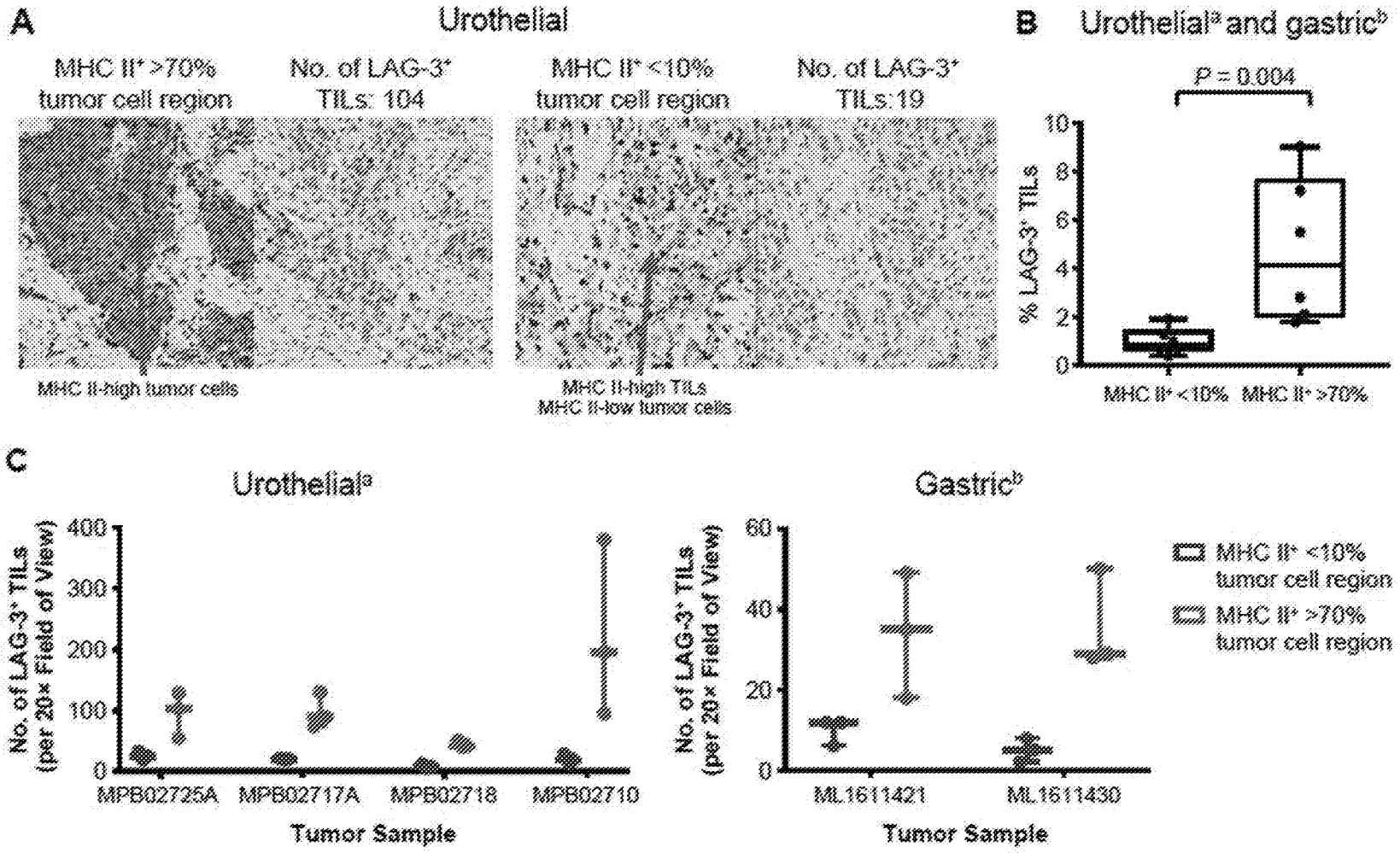
**Figures 28C.**

## Relationship between inflammation clusters and biomarker expression



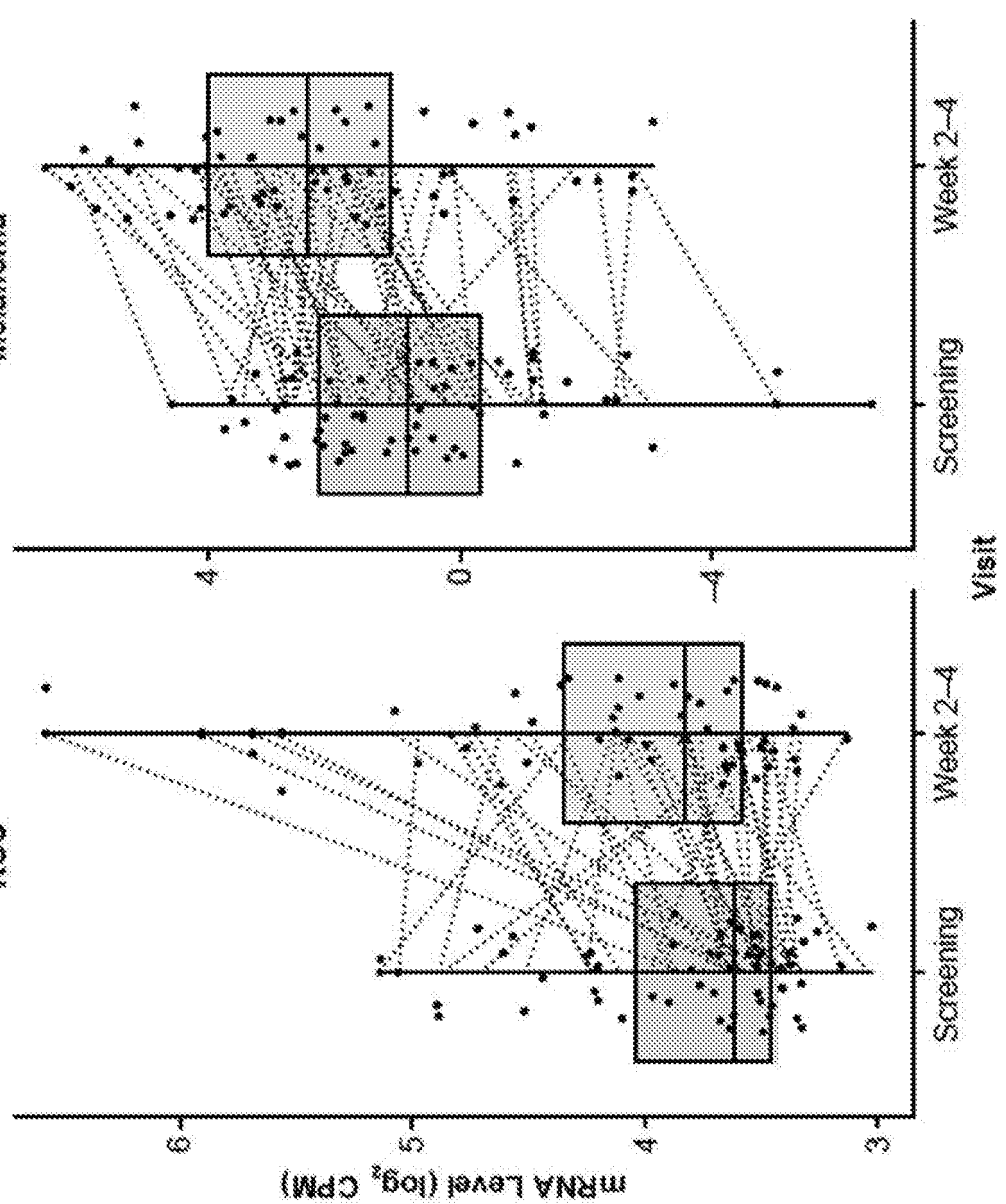

[a] n = 37. [b] n = 39. [c] n = 229. CD = cluster of differentiation; FOXP3 = forkhead box P3; LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; NSCLC = non-small cell lung carcinoma; PD-L1 = programmed death-1 ligand 1; TAMs = tumor-associated macrophages; TILs = tumor-infiltrating lymphocytes.

Figures 29A-C. Heterogeneous MHC II tumor cell expression and LAG-3+ TILs




[a] n = 4. [b] n = 2. LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; TILs = tumor-infiltrating lymphocytes.

Figures 30A.


LAG-3 mRNA levels at screening and at week 2–4 of nivolumab monotherapy
RCC
Melanoma
mRNA Level (log2 CPM)
6
5
4
3
4
0
−4
Screening
Week 2–4
Visit

Figures 30B.

## LAG-3 mRNA levels at screening and at week 2–4 of nivolumab monotherapy

| Changes in LAG-3 mRNA levels | RCC | Melanoma |
|---|---|---|
| n: pretreatment, posttreatment | 59, 55 | 61, 62 |
| *P* value | 0.0479 | 0.00002 |
| Mean fold increase from pretreatment baseline | 1.2 | 3.1 |

Dashed lines represent samples from the same patient. CPM = counts per million; LAG-3 = lymphocyte-activation gene 3; RCC = renal cell carcinoma.