Review

Clinical Cancer Research

Reimagining IDO Pathway Inhibition in Cancer Immunotherapy via Downstream Focus on the Tryptophan–Kynurenine–Aryl Hydrocarbon Axis

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Abstract

Significant progress has been made in cancer immunotherapy with checkpoint inhibitors targeting programmed cell death protein 1 (PD-1)–programmed death-ligand 1 signaling pathways. Tumors from patients showing sustained treatment response predominately demonstrate a T cell–inflamed tumor microenvironment prior to, or early on, treatment. Not all tumors with this phenotype respond, however, and one mediator of immunosuppression in T cell–inflamed tumors is the tryptophan–kynurenine–aryl hydrocarbon receptor (Trp–Kyn–AhR) pathway. Multiple mechanisms of immunosuppression may be mediated by this pathway including depletion of tryptophan, direct immunosuppression of Kyn, and activity of Kyn-bound AhR. Indoleamine 2,3-dioxygenase 1 (IDO1), a principle enzyme in Trp catabolism, is the target of small-molecule inhibitors in clinical development in combination with PD-1 checkpoint inhibitors. Despite promising results in early-phase clinical trials in a range of tumor types, a phase III study of the IDO1-selective inhibitor epacadostat in combination with pembrolizumab showed no difference between the epacadostat-treated group versus placebo in patients with metastatic melanoma. This has led to a diminution of interest in IDO1 inhibitors; however, other approaches to inhibit this pathway continue to be considered. Novel Trp–Kyn–AhR pathway inhibitors, such as Kyn-degrading enzymes, direct AhR antagonists, and tryptophan mimetics are advancing in early-stage or preclinical development. Despite uncertainty surrounding IDO1 inhibition, ample preclinical evidence supports continued development of Trp–Kyn–AhR pathway inhibitors to augment immune-checkpoint and other cancer therapies.

Introduction

It has long been understood that cancer cells express antigens that are recognized and prompt elimination by the immune system. Despite clinical trial evidence that cancer vaccines led to efficient antigen presentation with subsequent priming and infiltration of cytotoxic T cells into tumors, regression of tumors only occurred in a small subset of patients (1). This finding led to the prediction that important barriers downstream of initial T-cell priming must exist that limit meaningful tumor elimination (2). It is now appreciated that evasion of immune-mediated elimination occurs through multiple mechanisms, including immunoediting, decreased antigen presentation, and importantly, local immunosuppression in the tumor microenvironment (TME; refs. 3, 4).

Tumor analysis from patients with metastatic melanoma receiving vaccine and cytokine therapies suggested a paradigm of two broad phenotypes characterized by the presence or absence a T cell–inflamed tumor. T cell–inflamed tumors are characterized by tumor-infiltrating lymphocytes, a type I/II IFN transcriptional profile, and high degree of expression of immunosuppressive mechanisms. In contrast, a non-T cell–inflamed tumor is observed to have a low inflammatory signature and the absence of tumor-infiltrating lymphocytes (5–7).

Immunotherapy in the treatment of solid malignancies has evolved significantly over the past decade with the emergence of mAbs against programmed cell death protein 1 (PD-1)–PD-L1 and cytotoxic T-lymphocyte associated antigen 4 (CTLA-4), delivering meaningful clinical benefit across multiple solid tumors. Patients more likely to benefit from checkpoint immunotherapy include those with tumors demonstrating a high density of somatic mutations, elevated PD-L1 expression, and/or are enriched with IFNγ transcriptional profiles (8–13). However, a subset of tumors identified as having a T cell–inflamed tumor phenotype do not respond to checkpoint immunotherapy, suggesting other immunosuppressive mechanisms contribute to limiting immune-mediated tumor regression besides the PD-1/L1 axis. Preclinical studies have identified multiple immunosuppressive mechanisms that are present in the T-cell–inflamed tumors, including, but not limited to, extrinsic inhibition by regulatory cell populations such as forkhead box P3 (FoxP3)–positive regulatory T cells (Treg) and metabolic mechanisms of immunosuppression such as the tryptophan–kynurenine–aryl hydrocarbon receptor (Trp–Kyn–AhR) pathway (14, 15).

Specific clinical focus has increasingly centered on the immunosuppressive actions of tryptophan catabolism as regulated by indoleamine 2,3-dioxygenase 1 and 2 (IDO1/IDO2), tryptophan

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2–3-dioxygenase (TDO) and kynureninase as well as downstream signaling of tryptophan catabolites as agonists of AhR. In preclinical models, heightened activity of the Trp–Kyn–AhR pathway has been linked to impairment of antitumor immunity and tumor growth (16, 17). More recent characterization AhR has demonstrated multiple mechanisms by which it facilitates a tolerogenic immune environment (18). Inhibition of the Trp–Kyn–AhR pathway has become an attractive therapeutic target and the focus of substantial biotechnology and pharmaceutical effort. Currently, several IDO1, combination IDO1/TDO inhibitors, AhR inhibitors as well as a recombinant kynureninase are in clinical development or late preclinical testing.

Immunomodulatory Role of the Trp–Kyn–AhR Pathway

The Trp–Kyn–AhR pathway, in which the essential amino acid tryptophan is converted to Kyn and other secondary metabolites, is the primary route of tryptophan catabolism (19). Three enzymes catalyze the rate-limiting step of tryptophan catabolism to kynurenines: IDO1, IDO2, and TDO. The enzyme kynureninase hydrolyzes 3-hydroxykynurenine to 3-hydroxyanthranilic acid in the production of nicotinamide adenine dinucleotide. Regulation of these enzymes is notably divergent, with IDO1 being influenced by the interplay of IFNγ and IL6 as compared with TDO being regulated by tryptophan, cholesterols, and prostaglandin E2 (20, 21). The regulation and role of IDO2 is uncertain. Immunosuppression associated with Kyn was first described in experiments that demonstrated increased tryptophan catabolism limits allogeneic fetal rejection in mice (22).

In tumors, IDO1 is expressed by stromal cells of the TME and is induced by IFNγ as a result of $CD8^+$ T-cell infiltration and activation of other immunosuppressive pathways (7, 23, 24). TDO is ectopically expressed by tumor cells in certain malignancies (25). Kynurenines act as potent agonists of AhR, a ligand-gated transcription factor that is expressed in many immune cells and mediates a wide range of immunomodulatory effects (26, 27). Elevated IDO1 and TDO activity and Kyn levels are associated with increased tumor grade and poor prognosis in many cancers (28).

Several mechanisms have been proposed to explain the role of the Trp–Kyn–AhR pathway in tumor-associated immunosuppression. T cells are exquisitely sensitive to local depletion of tryptophan in which low tryptophan levels suppress mTORC pathways and activate general control nondepressible 2 (GCN2) kinase leading to cell-cycle arrest and anergy of infiltrating T cells via eIF-2–dependent pathways (Fig. 1; refs. 29–31). However, recent studies have questioned the significance of this mechanism (32, 33). Accumulation of kynurenines induce effector T-cell arrest and lead to binding of AhR. This results in nuclear translocation and promotion of FoxP3 transcripts and IL10, eventually producing Treg populations (18, 34–37). *In vitro* studies of AhR-deficient lung dendritic cells demonstrate failure to promote Treg development and an increase in Th2 cell differentiation and proinflammatory responses to allergen exposure (38). AhR suppresses innate immunogenicity of antigen-presenting cells and promotes IL10 production by natural killer cells (Fig. 2; refs. 39–41). In addition, the Kyn–AhR interaction has been shown to upregulate PD-1 expression by $CD8^+$

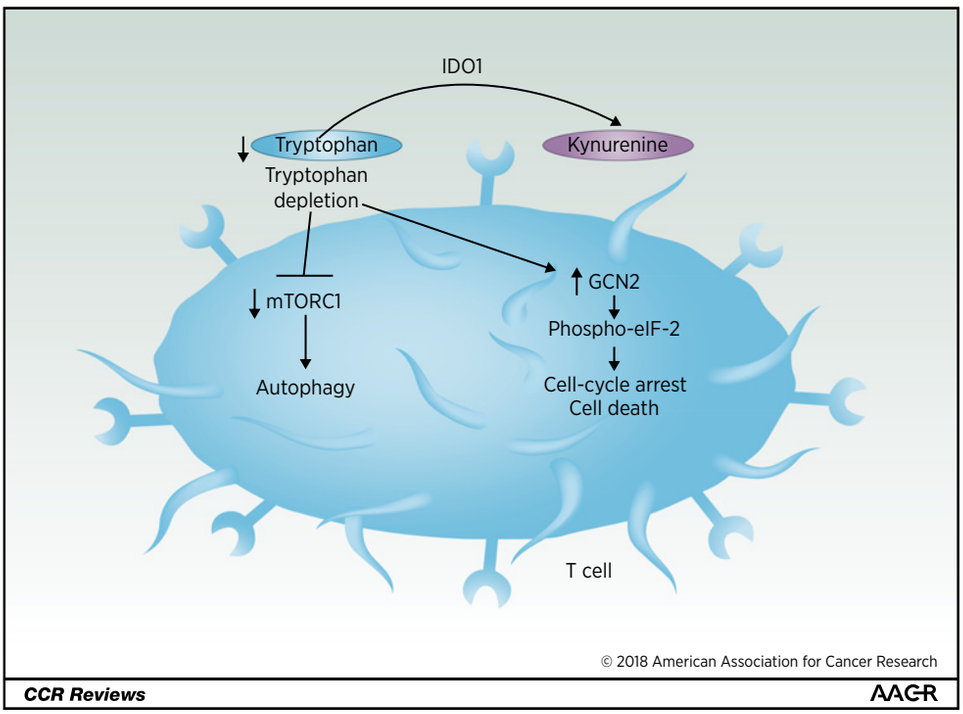


Figure 1.
Tryptophan depletion–dependent signaling. Depletion of tryptophan suppresses activity in the mTORC1 signaling pathway, leading to autophagy in T cells, and releases GCN2-mediated phosphorylation of eIF-2, inducing cell-cycle arrest and death in T cells.

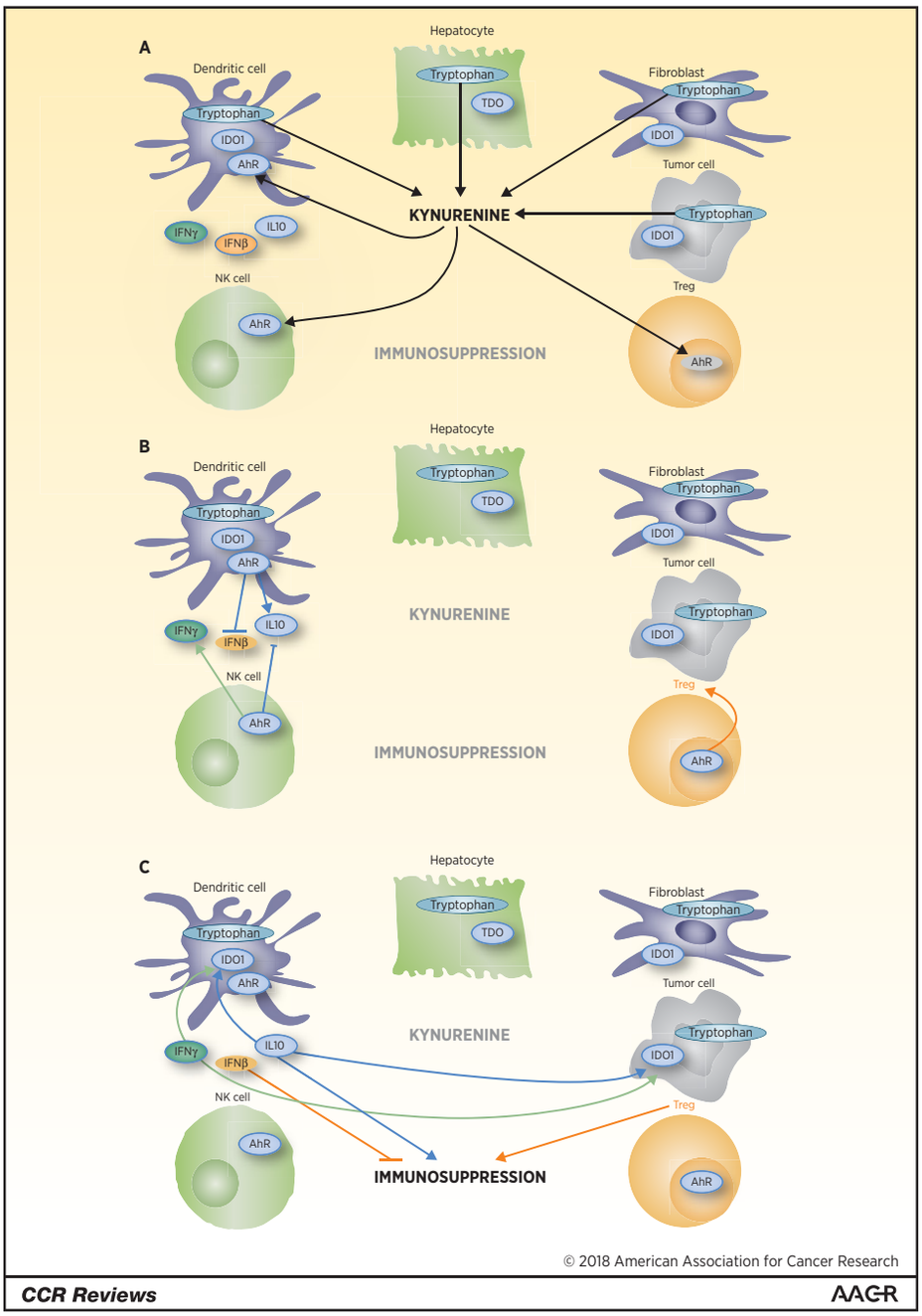


Figure 2.
IDO1–kynurenine–AhR signaling in TME immunosuppression. **A,** IDO1 in tumor cells, dendritic cells, and fibroblasts. TDO in hepatocytes are the rate-limiting enzymes in the conversion of tryptophan to kynurenine and kynurenine derivatives. Kynurenine binds to and activates the AhR, a ligand-activated transcription factor, in Tregs, natural killer (NK) cells, and dendritic cells. **B,** Activation and nuclear translocation of the AhR (1) in dendritic cells induces synthesis and release of IL10 and inhibits IFNβ signaling, (2) in NK cells induces synthesis and release of IL10 and IFNγ, and (3) in Tregs promotes Treg development. **C,** Tregs and IL10 promote immunosuppression within the TME, whereas inhibition of IFNβ by AhR releases regulation of immunosuppression from inhibitory IFNβ signaling. In addition, both IL10 and IFNγ promote IDO1 activity, establishing a positive feedback loop for IDO1–kynurenine–AhR signaling.

T cells via transcellular signaling mechanism in the tumor microenvironment (42).

Prominent IDO1/TDO Inhibitors and Trp–Kyn Pathway Inhibitors in Clinical Development

Several biochemical strategies exist to inhibit the Trp–Kyn–AhR pathway. IDO1 knockout mice demonstrate no clinical phenotype, in contrast to the inflammatory phenotype observed for knockouts of the immune checkpoints CTLA-4 and PD-1, and thus, IDO1 inhibitors have predominantly been used in combination with other treatment modalities (43, 44). Selective IDO1 enzyme inhibitors such as epacadostat, NLG-919, and BMS-986205 either compete with tryptophan for the catalytic site of IDO1 or bind the enzyme with very high affinity (44–47). In contrast, the tryptophan mimetic indoximod appears to have pleiotropic effects on downstream Kyn–AhR pathway signaling and has been shown to relieve immunosuppressive signaling normally induced by tryptophan depletion (48, 49). AhR inhibitors and recombinant kynureninase have more recently entered clinical development and will be discussed below.

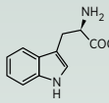
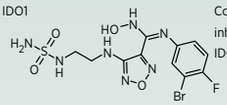
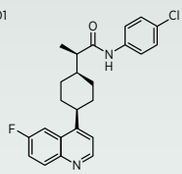
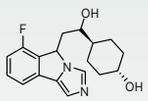
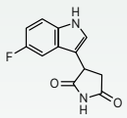
Drug	Company	Target	Structure	Mechanism	Dosing	Human IDO1 enzymatic assay (IC_{50})	Human IDO1 cell-based assay (IC_{50})	Human TDO enzymatic activity	Phase of development
Indoximod	NewLink	Tryptophan mimetic		Stimulates mTOR kinase to reduce T-cell autophagy (49)	1200 mg BID (77)		>2.5 mM (HeLa cells) (46) ~30 uM (Human DCs)	Nonselective	III
Epacadostat	Incyte	IDO1		Competitive inhibition of IDO1 (46)	100 mg BID (59)	72 nM (46)	7–23 nM (46)	>100-fold (46)	III
BMS986205	Bristol-Meyers Squib	IDO1		Irreversible inhibition of IDO1 (78)	150 mg QD (79)		1 nM (HEK293 cells) 2 nM (IFNγ-stimulated HeLa cells) (44)	>100-fold (44) >2 uM in HEK293 cells	III
Navoximod	NewLink	IDO1		Noncompetitive inhibition of IDO1	50–800 mg BID (80)	28 nM (82)	75 nM (81)	10–20-fold (78)	Ib
PF-06840003	iTeos	IDO1		Noncompetitive inhibition of IDO1 (82)	250–500 mg BID (83)	120 nM (82)	1100 nM (82)	>100-fold (82)	I
KHK2455	Kyowa Hakko Kirin	IDO1	Not available	Competitive inhibition, apo-conformation (84)	1 mg QD (85)		14 nM (84)	>100-fold (84)	I
RG70099	Roche	IDO1/TDO	Not available	Competitive inhibition	Unspecified	16 nM (69)	12 nM (69)	6-fold (69)	Preclinical
IOM-E	Merck	IDO1	Not available	Unknown	Unspecified		100 nM (70)	>100-fold (70)	Preclinical
IOM-D	Merck	IDO1/TDO	Not available	Unknown	Unspecified		365 (70)	10 nM (70)	Preclinical

Figure 3.
Trp–Kyn pathway inhibitors in current or prior clinical development. BID, twice a day; DC, dendritic cell; QD, every day.

A primary pharmacodynamic measure reported for selective IDO1 inhibitors in clinical trials was reduction in peripheral blood Kyn levels. Initial peripheral blood Kyn suppression data demonstrated approximately 50% reduction, suggesting other enzymes contribute to the production of systemic kynurenine, such as TDO. To date, assessment of intratumoral Kyn has not been consistently collected or reported in clinical trials (50, 51). Figure 3 describes the prominent IDO, TDO inhibitors, and Trp–Kyn pathway inhibitors currently in clinical development.

IDO1, TDO, and Trp–Kyn–AhR Inhibition in Combination Treatment

Association between the Trp–Kyn–AhR pathway and PD-1/L1 was suggested by the observation that both pathways are induced by IFNγ signaling in the TME (7, 14). Indeed, across 30 human solid tumors from The Cancer Genome Atlas (TCGA) database, we have observed that the gene expression of *IDO1* was strongly correlated with the expression of PD-1 across increasing level of IFNγ-responsive gene expression from non-T cell–inflamed to highly T cell–inflamed tumors (Fig. 4A). In contrast, expression of *IDO2*, *TDO2*, *KYNU*, *AHR*, and *GCN2* (alias *EIF2AK4*) does not appear to correlate with *PD-1* expression or demonstrate IFNγ responsiveness on a transcriptional level as strongly as *IDO1* (Fig. 4B).

Despite early observations for lack of monotherapy activity of selective IDO1 inhibitors (52), combination strategies utilizing IDO1 inhibitors were quickly advanced. Indeed, IDO1 and PD-1/L1 inhibitor combinations appeared to show great promise in early-phase clinical trials across multiple tumor types (Supplementary Tables S1 and S2).

A substantial literature also supports the potential utility of inhibition of the IDO pathway in conjunction with other anticancer modalities. Studies of IDO pathway blockade with radiation, chemotherapy, and tumor vaccines suggest an improvement relative to those treatments alone (53, 54). Several clinical trials evaluating combinations across these modalities are ongoing (Supplementary Table S1).

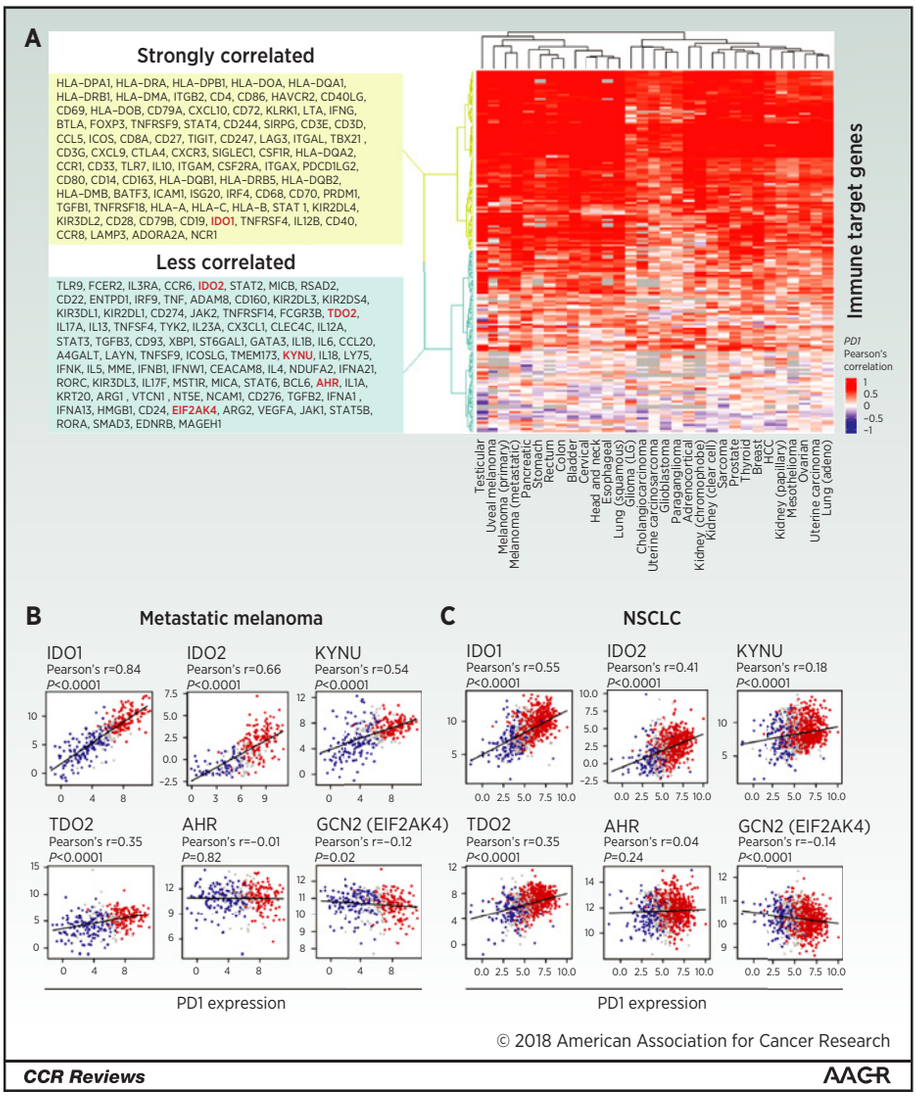


Figure 4.

Expression of *PD1* is positively correlated with immunotherapy-relevant target genes across solid tumors from TCGA. **A,** Heatmap of Pearson product–moment correlation coefficient *r* between *PD1* and immune target genes by tumor type. Immune target genes were separated into those strongly or less correlated with *PD1* expression. Methods: level 3 RNA-seq data (release date February 4, 2015) were downloaded for 30 solid tumor types from TCGA and processed as described previously (86). Acute myeloid leukemia, diffuse large B-cell lymphoma, and thymoma were excluded because of high tumor-intrinsic immune cell transcripts. Skin cutaneous melanoma had both primary and metastatic samples available, whereas the other 29 cancers had only primary tumors available. A noncomprehensive list of 171 immune molecules representative of the interactions between tumor cells and immune cells in the TME were selected and correlated with *PD1* (alias *PDCD1*) gene expression. For each tumor type, Pearson *r* was computed between each immune molecule and *PD1* and used for clustering the genes by hierarchical unsupervised clustering with Euclidean distance. Two distinct groups are shown, consisting of (1) strongly correlated genes and (2) less correlated genes. Glioma (LG), low-grade glioma; HCC, hepatocellular carcinoma; Lung (adeno), lung adenocarcinoma. **B** and **C,** Correlation plots of *PD1* versus *IDO1, IDO2, KYNU, TDO2, AHR*, and *GCN2* (alias *EIF2AK4*; highlighted in red in **A**) in metastatic melanoma (**B**) and NSCLC (**C**). Patients were categorized into T cell–inflamed (red), non-T cell–inflamed (blue), and intermediate groups using a defined T cell–inflamed gene signature (86). Each data point represents one patient. NSCLC, non–small cell lung carcinoma.

Epacadostat

Epacadostat, a competitive, selective inhibitor of IDO1, reached the most advanced stage of development, with the recent early termination of ECHO-301/Keynote-252; a phase III clinical trial in combination with pembrolizumab in metastatic melanoma. At median follow-up of 14 months, patients treated with epacadostat plus pembrolizumab demonstrated a progression-free survival of 4.7 months versus 4.9 months in those treated with pembrolizumab plus placebo (HR = 1.00; CI, 0.83–1.21; P = 0.517.) The overall response rate was 34.2% versus 31.5% in the epacadostat plus pembrolizumab and placebo plus pembrolizumab groups, respectively. Treatment-related adverse events occurred in 79.3% of patients receiving epacadostat plus pembrolizumab versus 81.0% receiving placebo plus pembrolizumab and grade >3 treatment-related adverse events occurred in 21.8% versus 17.0%, respectively (55). These negative results were unexpected as preclinical studies and early-phase clinical trials of this combination in as many as 14 different solid tumors showed encouraging results (Supplementary Table S1). Multiple hypotheses have been advanced to explain the seeming discrepancy between early-phase clinical trial success of epacadostat and the failure of the late-phase ECHO-301 trial. A nonexhaustive list of possible explanations to differentiate early- versus late-phase results could include differences between the treatment populations, inappropriately low dosing of epacadostat, and incomplete suppression of intratumoral Kyn.

Regarding the patient populations for these studies, the patient characteristics appeared to be relatively similar from the early- (ECHO-202) to late-phase (ECHO-301) melanoma studies across multiple variables including but not limited to performance status of zero (77% vs. 76%), M1c staging (55% vs. 61%), elevated lactate dehydrogenase (37% vs. 32%), and no prior therapy (71 vs. 89%), respectively (55, 56). As IDO1 expression is intimately linked to IFNγ gene expression, the baseline quality of the T cell–inflamed TME may be of relevance to the results. To date, this is not well characterized in either study. In ECHO-301, IDO1 expression was not an

inclusion criterion (although PD-L1 expression was a randomization stratification factor) with only PD-L1 status as a surrogate from the early-phase studies. Given the seeming complete lack of activity between epacadostat and placebo in ECHO-301, some have wondered whether further study should be given to the patients treated in the early-phase study. Despite the lack of obvious clinical differentiators, perhaps the tumors from these patients were disproportionately T cell–inflamed and thus much more likely to respond to pembrolizumab and possibly epacadostat.

Other less commonly controlled for differences of potential clinical significance could exist between the early- to late-phase trial populations also however. One of these would be to note that the early-phase trial was conducted at a select number of sites in the United States, whereas the late-phase study was predominately international (Australia, Europe, Asia, South America) with a lesser accrual in the United States. It has been observed in previous trials of melanoma that outcomes appeared to be substantially different between these populations in the phase III setting (57). Consideration might be given to whether dietary or environmental exposures could be variable in these different localities especially given the canonical role of AhR as a xenobiotic sensor that is responsive to signals from the gut microbiome (18). With an evolving literature supporting the microbiome as a potential influencer of PD-1 antibody response (58), focus on diet, medication use, and microbiome contents may be important variables to track in clinical trials moving forward.

Regarding dosing of epacadostat, 100 mg was taken forward out of the phase I study despite higher dose levels being tolerable and no MTD being established (59). This dose was chosen by Incyte as it was deemed that maximal inhibition of IDO1 activity was observed at doses of $\geq$100 mg with a relative plateau in decrease of kynurenine level in peripheral blood at higher doses. This point of the most appropriate dose is debated however particularly given that published modeling of IDO inhibition only reaches approximately 50% to 70% at the 100 mg dose (60). It is somewhat notable that an early-phase study of epacadostat with nivolumab was simultaneously pursued using 300 mg of epacadostat (61) and published modeling implies a higher likelihood of IDO1 inhibition at this dose (60). The optimal dose of epacadostat continues to be explored in ongoing clinical trials.

Perhaps of most relevance to the failure of ECHO-301 however is the open question of intratumoral pharmacodynamics. Although peripheral blood monitoring of kynurenine was reported for epacadostat in phase I, to date, no intratumoral data have been released from any clinical trial. Given the lack of consensus surrounding whether limiting tryptophan depletion versus suppression of Kyn acts as a primary mechanism of immunosuppression, these data are essential to inform the field surrounding next steps. Even very low levels of canonical AhR ligands, such as Kyn and dioxin, can activate AhR-associated gene expression and recent preclinical studies have suggested kynureninase and direct AhR inhibitors have higher potency relative to IDO1-selective inhibitors (62–64). If suppression of Kyn is indeed the dominant mechanism, it is very possible that IDO1 inhibition alone may be inadequate to drive intratumoral levels consistently low enough to alleviate the immunosuppressive effects of Kyn-activated AhR, including production of IL10 and suppression of type I IFN (41).

Other Selective IDO1 Inhibitors

Pivotal studies of BMS-986205, an irreversible IDO1 inhibitor developed by Flexus Biosciences and Bristol-Myers Squibb, have been predominately scaled back in the wake of epacadostat's late-stage trial failure, although a randomized study in bladder cancer is still planned. Relative to epacadostat, BMS-986205 demonstrates higher potency based on IC_{50} in IDO1-expressing cell lines. In contrast to the lack of such data for epacadostat, analysis of 39 paired pre- versus on-treatment tumor samples across various tumor types from the phase I trial of BMS-986205 plus nivolumab demonstrated decreased Kyn levels (and mostly near zero levels on-treatment) and increased the percentage of proliferating $CD8^+$ T cells (65).

Genentech has recently terminated rights to NewLink's NLG-919 (navoximod/GDC-0919), another selective inhibitor of IDO1.

Tryptophan Mimetics

Indoximod, the D-enantiomer of 1-methyl-tryptophan, has demonstrated inhibition of the IDO pathway as a tryptophan mimetic. Indoximod limits IDO-mediated immunosuppression by at least two mechanisms including (i) serving as an artificial Trp-sufficiency signal that prevents activation of GCN2 and inhibition of mTORC1 and (ii) modulation of AhR-dependent transcriptional activity (66). Indoximod increased activity and proliferation of $CD8^+$ T cells by limiting tryptophan depletion-mediated mTORC1 suppression (66). mTORC1 activation has been associated with ICOS expression, a T-cell coregulatory receptor seen on tumor-infiltrating T cells that has been associated with clinical response (67). In an AhR-dependent manner, indoximod was shown to stimulate $CD4^+$ T-cell differentiation to $Th17^+$ helper T cells, inhibit FoxP3 Tregs, and downregulate expression of IDO in dendritic cells (66).

Data from a single-arm phase II trial of indoximod plus anti–PD-1 in advanced melanoma achieved an ORR of 56% and CR in 19% with low rates of high-grade immune-related adverse events (68). Multiple phase II and III trials combining indoximod with other current modalities of treatment, including chemotherapy, cancer vaccines, and checkpoint immunotherapy are ongoing (Supplementary Tables S1 and S2).

Dual IDO1/TDO Inhibitors

Analysis of tumor and immune cells by IHC revealed differences in the expression of IDO1 and TDO among tumor types, suggesting the potential for a possible advantage with dual IDO and TDO inhibitors in certain tumors (69). Dual IDO/TDO inhibitors such as RG70099 decrease serum Kyn levels by approximately 90%. IOM-E and IOM-D are selective IDO1 and dual IDO1 and TDO inhibitors, respectively. Preclinical studies have revealed a promising pharmacokinetic profile. Significant *in vivo* efficacy was observed in mouse pancreatic adenocarcinoma cells treated with IOM-E, the selective IDO1 inhibitor, in combination with gemcitabine and Abraxane. Particular efficacy in preclinical lung cancer models has also been seen (70). Several companies have disclosed preclinical programs surrounding the development of dual IDO/TDO inhibitors, although the current status of these programs is in flux in the wake of ECHO-301. It is worth pointing out however that in murine models, complete IDO/TDO inhibition results in significant alteration of Trp metabolism,

which has raised concern over potential neurologic toxicity (seizures) from dual IDO1/TDO inhibition (71).

AhR Inhibitors

Inhibition of AhR signaling with small-molecule antagonists interferes with the downstream immunomodulatory effects irrespective of the source of Kyn production. Early inhibitors in this class have been shown to block nuclear translocation of AhR and enhanced production of IFNγ, TNFα, IL2, and reduction in tumor-associated M2-like macrophages. In mouse models, AhR inhibitors have demonstrated activity as monotherapy, a notable contrast to IDO1 inhibitors. Furthermore, enhanced activity is seen with AhRi combination with anti–PD-1 (63). Several biotechnology companies including Hercules Pharmaceuticals, Ideaya Biosciences, and KYN Therapeutics have disclosed the development of AhR inhibitors.

Kynurenine-Degrading Enzymes

Recombinant Kyn-degrading enzymes, kynureninase or KYNase, have been shown to reduce Kyn levels in IDO1, TDO, and IDO1/TDO dual positive cancer cells without impact on systemic tryptophan levels. Preclinical studies of a recombinant PEG-KYNase in established tumor models have demonstrated inhibition of tumor growth and increase in tumor-infiltrating effector T cells as monotherapy. Synergistic activity with anti–PD-1 has also been demonstrated (72, 73). An ongoing development program of recombinant kynureninase has been disclosed by KYN Therapeutics. Intratumoral injection of engineered *E. coli* strains that metabolize kynurenine have demonstrated reduction in *in vivo* kynurenine levels and generation of antitumor response (74).

Conclusions and Future Directions

The degree of tumor inflammation, as assessed by the presence of type I/II IFN signaling and infiltrating effector T cells, is associated with an improved response to checkpoint immunotherapy (8–11, 13). However, despite robust T-cell inflammation, a considerable percentage of tumors progress by virtue of multiple immunosuppressive mechanisms (14, 15). Upregulation of the Trp–Kyn–AhR pathway has been identified as one such mechanism. The immunosuppressive effect of this pathway is believed to be mediated by Trp depletion, T-cell cycle arrest mediated by Kyn cytotoxicity and activation of immune-tolerogenic AhR.

Recent studies have cast doubt on the Trp-depletion mechanisms (32, 33) and highlighted the potent immunosuppressive activity of intratumoral Kyn and AhR signaling. To this end, in the evaluation of Trp–Kyn–AhR inhibitors, reduction of extracellular Kyn within the TME or downstream AhR transcriptional programs should be emphasized as major pharmacodynamic endpoints. Indeed, a major concern pertaining to the clinical evaluation of selective IDO1 inhibitor epacadostat was the absence of intratumoral Kyn biomarker analysis. Prior studies have demonstrated serum Kyn:Trp correlate with response to anti–PD-1 (75, 76); however, it is unclear whether serum kynurenine is a surrogate for intratumoral Kyn.

Despite the failed experience of epacadostat in unselected melanoma patients, a strong translational rationale still exists for targeting of the Trp–Kyn–AhR pathway in conjunction with immunotherapy. Alternative mechanisms to achieve intratumoral Kyn reduction are currently being investigated. Comprehensive inhibition of kynurenine production by dual IDO1/TDO inhibitors and/or degradation of Kyn molecules by recombinant kynurenine-degrading enzymes may provide more robust intratumoral Kyn reduction. Alternatively, inhibition of AhR may serve to alleviate the immunosuppressive TME regardless of the source of Kyn production. Novel agents in each of these classes are approaching phase I studies, and preclinical experiments have shown promising results (63, 72–74).

A contrarian view to acknowledge surrounding this pathway would be that in a T cell–inflamed tumor, inhibition of Trp–Kyn–AhR may be insufficient to elicit further antitumor immune response due to the presence of further escape mechanisms (6, 7, 15). In these settings, this pathway may fail to make an inflamed environment even more inflamed. However, it may be that targeting downstream in the pathway could mediate the induction of the T cell–inflamed TME in previously noninflamed tumors given studies suggesting regulation of type I IFN response by AhR (41). This concept awaits further investigation and biospecimens from phase I studies of AhR antagonists will be especially interesting in this regard.

Cancer immunotherapy has advanced significantly with the development of CTLA-4 and PD-1–PD-L1 inhibitors. Progress in understanding the biology underlying the T cell–inflamed tumor microenvironment suggests that the Trp–Kyn–AhR pathway and PD-1–PD-L1 signaling are both associated with IFNγ response, but mediate independent mechanisms of immunosuppression. Combinatorial therapies may thus benefit a subset of patients. Despite uncertainty surrounding selective IDO1 inhibition, ample preclinical evidence supports continued development of Trp–Kyn–AhR pathway inhibitors to augment immune-checkpoint and other cancer therapies. Novel Trp–Kyn–AhR inhibitors have demonstrated promising preclinical activity and as new candidates undergo lead optimization and early evaluation, lessons learned from recent IDO1 inhibitor failure must guide the field moving forward.

Disclosure of Potential Conflicts of Interest

J.J. Luke reports receiving commercial research grants from Array, CheckMate, Evelo, and Palleon, holds ownership interest (including patents) in Actym and Alphamab Oncology, and is a consultant/advisory board member for Actym, Aduro, Alphamab Oncology, Array, Astellas, AstraZeneca, BeneVir, Bristol-Myers Squibb, Castle, CheckMate, Compugen, EMD Serono, Ideaya, Janssen, Merck, NewLink, Novartis, RefleXion, 7 Hills, Spring Bank, Syndax, Tempest, TTC Oncology, Vividion, and WntRx. No potential conflicts of interest were disclosed by the other authors.

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References

1. Rosenberg SA, Sherry RM, Morton KE, Scharfman WJ, Yang JC, Topalian SL, et al. Tumor progression can occur despite the induction of very high levels of self/tumor antigen-specific CD8+ T cells in patients with melanoma. J Immunol 2005;175:6169–76.
2. Gajewski TF. Monitoring specific T-cell responses to melanoma vaccines: ELISPOT, tetramers, and beyond. Clin Diagn Lab Immunol 2000;7:141–4.
3. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell 2011;144:646–74.
4. Becker JC, Andersen MH, Schrama D, thor Straten P. Immune-suppressive properties of the tumor microenvironment. Cancer Immunol Immunother 2013;62:1137–48.
5. Gajewski TF, Louahed J, Brichard VG. Gene signature in melanoma associated with clinical activity: a potential clue to unlock cancer immunotherapy. Cancer J 2010;16:399–403.
6. Harlin H, Meng Y, Peterson AC, Zha Y, Tretiakova M, Slingluff C, et al. Chemokine expression in melanoma metastases associated with CD8+ T-cell recruitment. Cancer Res 2009;69:3077–85.
7. Spranger S, Spaapen RM, Zha Y, Williams J, Meng Y, Ha TT, et al. Up-regulation of PD-L1, IDO, and T(regs) in the melanoma tumor microenvironment is driven by CD8(+) T cells. Sci Transl Med 2013;5:200ra116.
8. Ayers M, Lunceford J, Nebozhyn M, Murphy E, Loboda A, Kaufman DR, et al. IFN-gamma-related mRNA profile predicts clinical response to PD-1 blockade. J Clin Invest 2017;127:2930–40.
9. Ji R-R, Chasalow SD, Wang L, Hamid O, Schmidt H, Cogswell J, et al. An immune-active tumor microenvironment favors clinical response to ipilimumab. Cancer Immunology, Immunotherapy 2012;61:1019–31.
10. Le DT, Durham JN, Smith KN, Wang H, Bartlett BR, Aulakh LK, et al. Mismatch repair deficiency predicts response of solid tumors to PD-1 blockade. Science 2017;357:409–13.
11. Ribas A, Hamid O, Daud A, Hodi FS, Wolchok JD, Kefford R, et al. Association of pembrolizumab with tumor response and survival among patients with advanced melanoma. JAMA 2016;315:1600–9.
12. Tang H, Liang Y, Anders RA, Taube JM, Qiu X, Mulgaonkar A, et al. PD-L1 on host cells is essential for PD-L1 blockade-mediated tumor regression. J Clin Invest 2018;128:580–8.
13. Garon EB, Rizvi NA, Hui R, Leighl N, Balmanoukian AS, Eder JP, et al. Pembrolizumab for the treatment of non–small-cell lung cancer. N Engl J Med 2015;372:2018–28.
14. Lanitis E, Dangaj D, Irving M, Coukos G. Mechanisms regulating T-cell infiltration and activity in solid tumors. Ann Oncol 2017;28(suppl_12): xii18–xii32.
15. Gajewski TF, Meng Y, Blank C, Brown I, Kacha A, Kline J, et al. Immune resistance orchestrated by the tumor microenvironment. Immunol Rev 2006;213:131–45.
16. Munn DH, Sharma MD, Hou D, Baban B, Lee JR, Antonia SJ, et al. Expression of indoleamine 2,3-dioxygenase by plasmacytoid dendritic cells in tumor-draining lymph nodes. J Clin Invest 2004;114:280–90.
17. Brandacher G, Perathoner A, Ladurner R, Schneeberger S, Obrist P, Winkler C, et al. Prognostic value of indoleamine 2,3-dioxygenase expression in colorectal cancer: effect on tumor-infiltrating T cells. Clin Cancer Res 2006;12:1144–51.
18. Gutierrez-Vazquez C, Quintana FJ. Regulation of the immune response by the aryl hydrocarbon receptor. Immunity 2018;48:19–33.
19. Badawy AA-B. Kynurenine pathway of tryptophan metabolism: regulatory and functional aspects. Int J Tryptophan Res 2017;10: 1178646917691938.
20. Ochs K, Ott M, Rauschenbach KJ, Deumelandt K, Sahm F, Opitz CA, et al. Tryptophan-2,3-dioxygenase is regulated by prostaglandin E2 in malignant glioma via a positive signaling loop involving prostaglandin E receptor-4. J Neurochem 2016;136:1142–54.
21. Pallotta MT, Orabona C, Volpi C, Vacca C, Belladonna ML, Bianchi R, et al. Indoleamine 2,3-dioxygenase is a signaling protein in long-term tolerance by dendritic cells. Nat Immunol 2011;12:870–8.
22. Munn DH, Zhou M, Attwood JT, Bondarev I, Conway SJ, Marshall B, et al. Prevention of allogeneic fetal rejection by tryptophan catabolism. Science 1998;281:1191–3.
23. Ryan JM, Barry F, Murphy JM, Mahon BP. Interferon-γ does not break, but promotes the immunosuppressive capacity of adult human mesenchymal stem cells. Clin Exp Immunol 2007;149:353–63.
24. Vigneron N, van Baren N, Van den Eynde BJ. Expression profile of the human IDO1 protein, a cancer drug target involved in tumoral immune resistance. Oncoimmunology 2015;4:e1003012.
25. Opitz CA, Litzenburger UM, Sahm F, Ott M, Tritschler I, Trump S, et al. An endogenous tumour-promoting ligand of the human aryl hydrocarbon receptor. Nature 2011;478:197–203.
26. Kewley RJ, Whitelaw ML, Chapman-Smith A. The mammalian basic helix-loop-helix/PAS family of transcriptional regulators. Int J Biochem Cell Biol 2004;36:189–204.
27. Cheong JE, Sun L. Targeting the IDO1/TDO2-KYN-AhR pathway for cancer immunotherapy - challenges and opportunities. Trends Pharmacol Sci 2018;39:307–25.
28. Reznik E, Luna A, Aksoy BA, Liu EM, La K, Ostrovnaya I, et al. A landscape of metabolic variation across tumor types. Cell Syst 2018;6:301–13.
29. Munn DH, Shafizadeh E, Attwood JT, Bondarev I, Pashine A, Mellor AL. Inhibition of T cell proliferation by macrophage tryptophan catabolism. J Exp Med 1999;189:1363–72.
30. Munn DH, Sharma MD, Baban B, Harding HP, Zhang Y, Ron D, et al. GCN2 kinase in T cells mediates proliferative arrest and anergy induction in response to indoleamine 2,3-dioxygenase. Immunity 2005;22:633–42.
31. Eleftheriadis T, Pissas G, Antoniadi G, Liakopoulos V, Stefanidis I. Indoleamine 2,3-dioxygenase depletes tryptophan, activates general control non-derepressible 2 kinase and down-regulates key enzymes involved in fatty acid synthesis in primary human CD4+ T cells. Immunology 2015;146:292–300.
32. Sonner JK, Deumelandt K, Ott M, Thome CM, Rauschenbach KJ, Schulz S, et al. The stress kinase GCN2 does not mediate suppression of antitumor T cell responses by tryptophan catabolism in experimental melanomas. Oncoimmunology 2016;5:e1240858.
33. Badawy AA, Namboodiri AM, Moffett JR. The end of the road for the tryptophan depletion concept in pregnancy and infection. Clin Sci 2016;130:1327–33.
34. Mellor AL, Chandler P, Baban B, Hansen AM, Marshall B, Pihkala J, et al. Specific subsets of murine dendritic cells acquire potent T cell regulatory functions following CTLA4-mediated induction of indoleamine 2,3 dioxygenase. Int Immunol 2004;16:1391–401.
35. Veldhoen M, Hirota K, Christensen J, O'Garra A, Stockinger B. Natural agonists for aryl hydrocarbon receptor in culture medium are essential for optimal differentiation of Th17 T cells. J Exp Med 2009;206:43–9.
36. Mezrich JD, Fechner JH, Zhang X, Johnson BP, Burlingham WJ, Bradfield CA. An interaction between kynurenine and the aryl hydrocarbon receptor can generate regulatory T cells. J Immunol 2010;185:3190–8.
37. Gandhi R, Kumar D, Burns EJ, Nadeau M, Dake B, Laroni A, et al. Activation of the aryl hydrocarbon receptor induces human type 1 regulatory T cell-like and Foxp3(+) regulatory T cells. Nat Immunol 2010;11:846–53.
38. Thatcher TH, Williams MA, Pollock SJ, McCarthy CE, Lacy SH, Phipps RP, et al. Endogenous ligands of the aryl hydrocarbon receptor regulate lung dendritic cell function. Immunology 2016;147:41–54.
39. Nguyen NT, Kimura A, Nakahama T, Chinen I, Masuda K, Nohara K, et al. Aryl hydrocarbon receptor negatively regulates dendritic cell immunogenicity via a kynurenine-dependent mechanism. Proc Natl Acad Sci U S A 2010;107:19961–6.
40. Wagage S, John B, Krock BL, Hall AO, Randall LM, Karp CL, et al. The aryl hydrocarbon receptor promotes IL-10 production by NK cells. J Immunol 2014;192:1661–70.
41. Yamada T, Horimoto H, Kameyama T, Hayakawa S, Yamato H, Dazai M, et al. Constitutive aryl hydrocarbon receptor signaling constrains type I interferon-mediated antiviral innate defense. Nat Immunol 2016;17: 687–94.
42. Liu Y, Liang X, Dong W, Fang Y, Lv J, Zhang T, et al. Tumor-repopulating cells induce PD-1 expression in CD8(+) T cells by transferring kynurenine and AhR activation. Cancer Cell 2018;33:480–94.
43. Vacchelli E, Aranda F, Eggermont A, Sautes-Fridman C, Tartour E, Kennedy EP, et al. Trial watch: IDO inhibitors in cancer therapy. Oncoimmunology 2014;3:e957994.
44. Hunt JT, Balog A, Huang C, Lin T-A, Lin T-A, Maley D, et al. Structure, in vitro biology and in vivo pharmacodynamic characterization of a novel clinical IDO1 inhibitor [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2017; 2017 Apr 1–5;

Washington, DC. Philadelphia (PA): AACR; Cancer Res 2017;77(13 Suppl):Abstract nr 4964. doi:10.1158/1538-7445.AM2017-4964.
45. Yue EW, Douty B, Wayland B, Bower M, Liu X, Leffet L, et al. Discovery of potent competitive inhibitors of indoleamine 2,3-dioxygenase with in vivo pharmacodynamic activity and efficacy in a mouse melanoma model. J Med Chem 2009;52:7364–7.
46. Liu X, Shin N, Koblish HK, Yang G, Wang Q, Wang K, et al. Selective inhibition of IDO1 effectively regulates mediators of antitumor immunity. Blood 2010;115:3520–30.
47. Mautino MR, Jaipuri FA, Waldo J, Kumar S, Adams J, Van Allen C, et al. NLG919, a novel indoleamine-2,3-dioxygenase (IDO)-pathway inhibitor drug candidate for cancer therapy [abstract]. In: Proceedings of the 104th Annual Meeting of the American Association for Cancer Research; 2013 Apr 6–10; Washington, DC. Philadelphia (PA): AACR; Cancer Res 2013;73(8 Suppl):Abstract nr 491. doi:10.1158/1538-7445.AM2013-491.
48. Hou DY, Muller AJ, Sharma MD, DuHadaway J, Banerjee T, Johnson M, et al. Inhibition of indoleamine 2,3-dioxygenase in dendritic cells by stereoisomers of 1-methyl-tryptophan correlates with antitumor responses. Cancer Res 2007;67:792–801.
49. Metz R, Rust S, Duhadaway JB, Mautino MR, Munn DH, Vahanian NN, et al. IDO inhibits a tryptophan sufficiency signal that stimulates mTOR: A novel IDO effector pathway targeted by D-1-methyl-tryptophan. Oncoimmunol 2012;1:1460–8.
50. Platten M, Wick W, Van den Eynde BJ. Tryptophan catabolism in cancer: beyond IDO and tryptophan depletion. Cancer Res 2012;72:5435–40.
51. Platten M, von Knebel Doeberitz N, Oezen I, Wick W, Ochs K. Cancer immunotherapy by targeting IDO1/TDO and their downstream effectors. Front Immunol 2015;5:673.
52. Kristeleit R, Davidenko I, Shirinkin V, El-Khouly F, Bondarenko I, Goodheart MJ, et al. A randomised, open-label, phase 2 study of the IDO1 inhibitor epacadostat (INCB024360) versus tamoxifen as therapy for biochemically recurrent (CA-125 relapse)-only epithelial ovarian cancer, primary peritoneal carcinoma, or fallopian tube cancer. Gynecol Oncol 2017;146:484–90.
53. Muller AJ, DuHadaway JB, Donover PS, Sutanto-Ward E, Prendergast GC. Inhibition of indoleamine 2,3-dioxygenase, an immunoregulatory target of the cancer suppression gene Bin1, potentiates cancer chemotherapy. Nat Med 2005;11:312–9.
54. Bahary N, Wang-Gillam A, Haraldsdottir S, Somer BG, Lee JS, O'Rourke MA, et al. Phase 2 trial of the IDO pathway inhibitor indoximod plus gemcitabine / nab-paclitaxel for the treatment of patients with metastatic pancreas cancer. J Clin Oncol 36:15s, 2018 (suppl; abstr 4015).
55. Long GV, Dummer R, Hamid O, Gajewski T, Caglevic C, Dalle S, et al. Epacadostat (E) plus pembrolizumab (P) versus pembrolizumab alone in patients (pts) with unresectable or metastatic melanoma: results of the phase 3 ECHO-301/KEYNOTE-252 study. J Clin Oncol 36, 2018 (suppl; abstr 108).
56. Hamid O, Gajewski TF, Frankel AE, Bauer TM, Olszanski AJ, Luke JJ, et al. Epacadostat plus pembrolizumab in patients with advanced melanoma: phase 1 and 2 efficacy and safety results from ECHO-202/KEYNOTE-037. Ann Oncol 2017;28(suppl_5):v428–v48.
57. Grob J, Schadendorf D, Wagstaff J, Márquez-Rodas I, Lebbé C, Ascierto PA, et al. 1222PD - Regional differences in overall survival (OS) in patients with advanced melanoma (MEL) who received nivolumab (NIVO) combined with ipilimumab (IPI). Ann Oncol 2017;28:428–48.
58. Matson V, Fessler J, Bao R, Chongsuwat T, Zha Y, Alegre ML, et al. The commensal microbiome is associated with anti-PD-1 efficacy in metastatic melanoma patients. Science 2018;359:104–8.
59. Beatty GL, O'Dwyer PJ, Clark J, Shi JG, Bowman KJ, Scherle PA, et al. First-in-human phase I study of the oral inhibitor of indoleamine 2,3-dioxygenase-1 epacadostat (INCB024360) in patients with advanced solid malignancies. Clin Cancer Res 2017;23:3269–76.
60. Mitchell TC, Hamid O, Smith DC, Bauer TM, Wasser JS, Olszanski AJ, et al. Epacadostat plus pembrolizumab in patients with advanced solid tumors: phase I results from a multicenter, open-label phase I/II trial (ECHO-202/KEYNOTE-037). J Clin Oncol 2018;36:3223–30.
61. Daud A, Saleh MN, Hu J, Bleeker JS, Riese MJ, Meier R, et al. Epacadostat plus nivolumab for advanced melanoma: updated phase 2 results of the ECHO-204 study. J Clin Oncol 36:15s, 2018 (suppl; abstr 9511).
62. Sherr D, Kenison-Whte J, Wang Z. The aryl hydrocarbon receptor as driver of cancer immunity [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2018; 2018 Apr 14–18; Chicago, IL. Philadelphia (PA): AACR; Cancer Res 2018;78(13 Suppl):Abstract nr LB-128.
63. Joseph J, Gonzalez-Lopez M, Galang C, Garcia C, Lemar H, Lu J, et al. Small-molecule antagonists of the aryl hydrocarbon receptor (AhR) promote activation of human PBMCs *in vitro* and demonstrate significant impact on tumor growth and immune modulation *in vivo* [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2018; 2018 Apr 14–18; Chicago, IL. Philadelphia (PA): AACR; Cancer Res 2018;78(13 Suppl):Abstract nr 4719.
64. Tchaicha J, McGovern K, Campesato LF, Coma S, Zhang XM, Walsh M, et al. Targeting the IDO and TDO pathway through inhibition of the aryl hydrocarbon receptor [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2018; 2018 Apr 14–18; Chicago, IL. Philadelphia (PA): AACR; Cancer Res 2018;78(13 Suppl):Abstract nr 4723.
65. Luke JJ, Gelmon K, Pachynski R, Desai J, Siu LL. Preliminary antitumor and immunomodulatory activity of BMS-986205, an optimized indoleamine 2,3-dioxygenase 1 (IDO1) inhibitor, in combination with nivolumab in patients with advanced cancers. J Immunother Cancer 2017;5 (Suppl3):89.
66. Brincks EL, Adams J, Essmann M, Turner BA, Wang L, Ke J, et al. Indoximod modulates AhR-driven transcription of genes that control immune function [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2018; 2018 Apr 14–18; Chicago, IL. Philadelphia (PA): AACR; Cancer Res 2018;78(13 Suppl):Abstract nr 3753.
67. Fu T, He Q, Sharma P. The ICOS/ICOSL pathway is required for optimal antitumor responses mediated by anti-CTLA-4 therapy. Cancer Res 2011; 71:5445–54.
68. Zakharia Y, Rixe O, Ward JH, Drabick JJ, Shaheen MF, Milhem MM, et al. Phase 2 trial of the IDO pathway inhibitor indoximod plus checkpoint inhibition for the treatment of patients with advanced melanoma. J Clin Oncol 36:15s, 2018 (suppl; abstr 9512).
69. Gyulveszi G, Fischer C, Mirolo M, Stern M, Green L, Ceppi M, et al. RG70099: a novel, highly potent dual IDO1/TDO inhibitor to reverse metabolic suppression of immune cells in the tumor micro-environment [abstract]. In: Proceedings of the 107th Annual Meeting of the American Association for Cancer Research; 2016 Apr 16–20; New Orleans, LA. Philadelphia (PA): AACR; Cancer Res 2016;76(14 Suppl):Abstract nr LB-085.
70. Wise A, McGuinness BE, Trewick SC, Brown TJ, Cowley PM. Pre-clinical development of next generation inhibitors of the enzymes indoleamine 2,3-dioxygenase 1 and tryptophan 2,3-dioxygenase as cancer immunotherapies [abstract]. In: Proceedings of the 107th Annual Meeting of the American Association for Cancer Research; 2016 Apr 16–20; New Orleans, LA. Philadelphia (PA): AACR; Cancer Res 2016;76(14 Suppl): Abstract nr 5115.
71. Kanai M, Funakoshi H, Takahashi H, Hayakawa T, Mizuno S, Matsumoto K, et al. Tryptophan 2,3-dioxygenase is a key modulator of physiological neurogenesis and anxiety-related behavior in mice. Mol Brain 2009;2:8.
72. Zhang M, Stone E, Triplett TA, Triplett K, Lamb C, Karamitros CS, et al. A novel approach to targeting the IDO/TDO pathway through degradation of the immunosuppressive metabolite kynurenine [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2017; 2017 Apr 1–5; Washington, DC. Philadelphia (PA): AACR; Cancer Res 2017;77(13 Suppl):Abstract nr 5570. doi:10.1158/1538-7445.AM2017-5570.
73. Triplett TA, Garrison KC, Marshall N, Donkor M, Blazeck J, Lamb C, et al. Reversal of indoleamine 2,3-dioxygenase-mediated cancer immune suppression by systemic kynurenine depletion with a therapeutic enzyme. Nat Biotechnol 2018;36:758–64.
74. West KA, Fisher A, Leventhal D, Sokolovska A, Li N, Plescia C, et al. Metabolic modulation of the tumor microenvironment using Synthetic Biotic™ Medicines [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2018; 2018 Apr 14–18; Chicago, IL. Philadelphia (PA): AACR; Cancer Res 2018;78(13 Suppl): Abstract nr 2920.
75. Giannakis M, Li H, Jin C, Gopal S, Desai K, Horak C, et al. Metabolomic correlates of response in nivolumab-treated renal cell carcinoma and melanoma patients. J Clin Oncol 35, 2017 (suppl; abstr 3036).

76. Botticelli A, Cerbelli B, Lionetto L, Zizzari I, Pisano A, Roberto M, et al. The key role of kynurenine in anti-PD-1 failure [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2018; 2018 Apr 14–18; Chicago, IL. Philadelphia (PA): AACR; Cancer Res 2018;78(13 Suppl):Abstract nr 5705.
77. Soliman HH, Jackson E, Neuger T, Dees EC, Harvey RD, Han H, et al. A first in man phase I trial of the oral immunomodulator, indoximod, combined with docetaxel in patients with metastatic solid tumors. Oncotarget 2014;5:8136–46.
78. Prendergast GC, Malachowski WP, DuHadaway JB, Muller AJ. Discovery of IDO1 inhibitors: from bench to bedside. Cancer Res 2017;77:6795–811.
79. Siu LL, Gelmon K, Chu Q, Pachynski R, Alese O, Basciano P, et al. BMS-986205, an optimized indoleamine 2,3-dioxygenase 1 (IDO1) inhibitor, is well tolerated with potent pharmacodynamic (PD) activity, alone and in combination with nivolumab (nivo) in advanced cancers in a phase 1/2a trial [abstract]. In: Proceedings of the American Association for Cancer Research Annual Meeting 2017; 2017 Apr 1–5; Washington, DC. Philadelphia (PA): AACR; Cancer Res 2017;77(13 Suppl):Abstract nr CT116. doi:10.1158/1538-7445.AM2017-CT116.
80. Nayak A, Hao Z, Sadek R, Vahanian N, Ramsey WJ, Kennedy E, et al. A phase I study of NLG919 for adult patients with recurrent advanced solid tumors. J Immunother Cancer 2014;2(Suppl 3):P250.
81. Spahn J, Peng J, Lorenzana E, Kan D, Hunsaker T, Segal E, et al. Improved anti-tumor immunity and efficacy upon combination of the IDO1 inhibitor GDC-0919 with anti-PD-l1 blockade versus anti-PD-l1 alone in preclinical tumor models. J Immunother Cancer 2015;3(Suppl 2):P303.
82. Crosignani S, Bingham P, Bottemanne P, Cannelle H, Cauwenberghs S, Cordonnier M, et al. Discovery of a novel and selective indoleamine 2,3-dioxygenase (IDO-1) inhibitor 3-(5-Fluoro-1H-indol-3-yl)pyrrolidine-2,5-dione (EOS200271/PF-06840003) and its characterization as a potential clinical candidate. J Med Chem 2017;60:9617–29.
83. Reardon D, Desjardins A, Rixe O, Cloughesy T, Alekar S, Gamelin E, et al. A phase 1 study of PF-06840003, an oral indole 2,3-dioxygenase 1 (IDO1) inhibitor in patients with malignant gliomas. Neurooncol 2017;19 (Suppl 6):vi32.
84. Mie M, Kunieda K, Koshiba S, Murakami T, Horita S, Fukuda Y, et al. In vitro and in vivo characterization of KHK2455, a highly potent and selective indoleamine 2,3-dioxygenase 1 (IDO1) inhibitor with a novel mechanism of action. J Immunother Cancer 2017;5:P149.
85. Sahebjam S, Yap T, Hong D, Rao A, Adams S, Efuni S, et al. KHK2455, a long-acting selective IDO-1 inhibitor, in combination with mogamulizumab, an anti-CCR4 monoclonal antibody, in patients with advanced solid tumors: preliminary safety report and pharmacodynamic activity from a first-in-human study. J Immunother Cancer 2017;5(Suppl 2):P148.
86. Spranger S, Luke JJ, Bao R, Zha Y, Hernandez KM, Li Y, et al. Density of immunogenic antigens does not explain the presence or absence of the T-cell-inflamed tumor microenvironment in melanoma. Proc Natl Acad Sci U S A 2016;113:E7759–E68.