A Mouse Model of Melanoma Driven by Oncogenic KRAS

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Abstract

The small G-protein NRAS is mutated in 22% of human melanomas, whereas the related proteins KRAS and HRAS are mutated in only 2% and 1% of melanomas, respectively. We have developed a mouse model of melanoma in which Cre recombinase/LoxP technology is used to drive inducible expression of G12V KRAS in the melanocytic lineage. The mice develop skin hyperpigmentation, nevi, and tumors that bear many of the cardinal histopathology features and molecular characteristics of human melanoma. These tumors invade and destroy the underlying muscles and cells derived from them can grow as subcutaneous tumors and colonize the lungs of nude mice. These data establish that oncogenic KRAS can be a founder event in melanomagenesis.

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Introduction

The RAS proteins are components of a conserved signal transduction cascade that regulates cell responses to the environment (1, 2). These small guanine nucleotide binding proteins are activated downstream of growth factor, cytokine, and hormone receptors. Specifically, they are activated by guanine-nucleotide exchange factors, which replace GDP for GTP, and inactivated by GTPase-activating proteins, which stimulate GTP hydrolysis and returning RAS to the GDP-bound state. RAS activates several effector proteins, including phosphoinositide 3-kinase (PI3K), the RAF protein kinases, and the Ral guanine-nucleotide exchange factor (2), to regulate the activity of signaling pathways that control proliferation, senescence, survival, and differentiation. There are three RAS genes in mammals, HRAS, KRAS, and NRAS, and the proteins they encode couple overexpression, but not oncogenic transformation if a mutation occurs, or it may possess distinct biological properties. Accordingly, G12V NRAS and G12V KRAS both activate RAF and induce anchorage-independent growth in melanocytes in vitro, but NRAS-transformed melanocytes are more tumorigenic than KRAS-transformed melanocytes in mice (7). Thus, KRAS may be a weaker oncogene than NRAS in melanocytes.

Several transgenic mouse melanoma models driven by RAS have been developed, but in the absence of additional events, the induction of melanoma in these models is generally inefficient. Thus, constitutive or inducible expression of G12V HRAS only induces tumor formation efficiently when Cdkn2a (p16INK4a) or Tp53 is deleted (8–10) or when the mice are exposed to UV light (11). The tumors are dermal, display variable pigmentation, and do not metastasize. Similarly, conditional expression of G12V HRAS in melanocytes using Cre-recombinase/LoxP technology only induces melanoma efficiently when p16INK4a is deleted (12). The tumors are heavily pigmented and also nonmetastatic. Further, constitutive state and promoting tumorigenesis (2). RAS mutations are found in about 25% of melanomas, a form of skin cancer that arises from melanocytes, which are specialized pigment cells that provide skin and hair tone and protection from UV radiation. In most western societies, melanoma incidence is doubling every decade, and due to a lack of effective therapies, metastatic malignant melanoma has a particularly poor outcome (6).

NRAS is mutated in 22% of human melanomas, whereas KRAS and HRAS are mutated in 2% and 1% of cases, respectively. More than 80% of the NRAS and HRAS mutations involve Q61, whereas ~77% of the KRAS mutants involve G12 (http://www.sanger.ac.uk/genetics/CGP/cosmic/). It is unclear why NRAS mutations predominate in melanoma, whereas in cancers such as pancreatic carcinoma and colorectal cancer, KRAS mutations predominate. It is possibly that codon Q61 of NRAS is a mutation hotspot in melanoma, but this is difficult to consolidate based on DNA sequence conservation. Alternatively, NRAS may be expressed at levels that are optimal for oncogenic transformation if a mutation occurs, or it may possess distinct biological properties. Accordingly, G12V NRAS and G12V KRAS both activate RAF and induce anchorage-independent growth in melanocytes in vitro, but NRAS-transformed melanocytes are more tumorigenic than KRAS-transformed melanocytes in mice (7). Thus, KRAS may be a weaker oncogene than NRAS in melanocytes.

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expression of G12V KRAS in melanocytes using a melanocyte-specific tyrosinase promoter only induces tumors efficiently when p16INK4a is deleted (13). The tumors in these mice are pigmented and frequently ulcerated, and tumor cells frequently invade and colonize the epidermis and epidermal/dermal junctions of the skin. They invade the reticular dermis, the subcutis, and the underlying muscles and, in some mice, spread to the lymph nodes, liver, and lungs.

These data show that HRAS and NRAS can induce melanomagenesis in mice, albeit inefficiently. Because no such studies are reported for KRAS, in this study we used Cre-recombinase/LoxP technology–induced expression of G12V KRAS in melanocytes to determine whether it can also induce melanomagenesis in mice.

Materials and Methods

Transgenic mouse procedures

Transgenic mouse procedures were done under Home Office license authority with local Ethics Committee approval. Transgenic β-actin::LSL::G12V KRAS and Tyr::CreERT2 mice, and the primers required for their genotyping have been described (14, 15). The mice were treated with tamoxifen (TM) and samples were prepared for DNA genotyping and reverse transcription-PCR (RT-PCR) analysis as described (16). Cell lines for use in intravitral imaging were prepared and analyzed as described (16). For allograft tumor growth, 1 × 10⁶ melanoma cells were injected s.c. into the flanks of nude mice in 100 µl of PBS. Tumor size was measured using calipers and volume was calculated using the formula length × width × depth × 0.5. For Western blotting, protein extracts were prepared using NP40 buffer (17) and the following antibodies: mouse anti-c-myc and rabbit anti-extracellular signal–regulated kinase 2 (Erk2; Santa Cruz Biotechnology) and rabbit anti-TP53 (Novacorda).

To sequence Tp53 mRNA, representative 8-µm frozen sections from four tumors were prepared and microdissected as described (18) to provide samples with >90% tumor cells from which cDNA was prepared as described (19). The entire Tp53 cDNA was amplified using eight paired primers (Supplementary Table S1) for sequencing using Big Dye (Applied Biosystems) as described (20). Mutation analysis was done using Mutation surveyor software (SoftGenetics LLC). All samples were sequenced in duplicate from separately microdissected samples.

Immunohistochemistry and immunofluorescence

Immunohistochemistry analysis was as described (16) using rabbit polyclonal antibody ZO331 (1/800) for S100 (Dako), rat monoclonal antibody M7249 (1/25) for Ki67 (Dako), and a rabbit polyclonal antibody for p16INK4a (1/25; Santa Cruz Biotechnology). S100 antigen retrieval was done using Labvision pretreatment module citrate (pH 6) for 30 minutes (Labvision) or by microwave (p16INK4a, Ki67) in Dako (pH 6) buffer (Dako) for 18 minutes. Antibody binding was detected using the Rabbit enVision Peroxidase kit, the AEC substrate chromogen (S100), and the Vectastain Elite ABC kit (p16INK4a, Ki67), or with AK-5001 rabbit IgG Vectastain ABC-AP kit (S100) and rat IgG Vectastain ABC-AP (Ki67), with SK-5300 Vector Blue substrate or 3,3′-diaminobenzidine substrate (all from Vector). Negative controls for each run included appropriate nonimmune sera (Dako; Jackson Immunoresearch).

Immunofluorescence was done with mouse antibody MP-606-PG1 for S100 (Menarini) and rabbit antibody M-157 for p16INK4a (Santa Cruz Biotechnology). Antigens were retrieved using Menarini Antigen Access Unit with Super buffer (pH 9.5). Primary antibody incubation was overnight and antibody binding was detected with AlexaFluor488 goat anti-rabbit IgG (Invitrogen) and AlexaFluor555 goat anti-mouse IgG (Invitrogen) for p16INK4a and S100, respectively. Slides were counterstained with 4′,6-diamidino-2-phenylindole (DAPI). Negative controls were included for each immunohistochemical run using an appropriate nonimmune serum IgG (Dako).

Results

To develop a KRAS-driven model of melanoma, we crossed β-actin::LSL::G12V KRAS and Tyr::CreERT2 mice. In β-actin::LSL::G12V KRAS mice, G12V KRAS is downstream of a β-actin promoter, but its expression is blocked by an intervening LoxP-STOP-LoxP (LSL) cassette that can be removed by Cre recombinase (Supplementary Fig S1; ref. 14). In Tyr::CreERT2 mice, a TM-activated version of Cre recombinase (CreERT2) is expressed using a melanocyte-specific tyrosinase promoter (15). The double transgenic mice thus allow exquisite temporal and spatial control of G12V KRAS expression because CreERT2 is restricted to the melanocytes and only activated upon TM treatment, allowing the human acquisition of somatic mutations in KRAS to be mimicked in the mice.

Mice at 1 to 2 months of age were treated with topical application of TM to their shaved backs. Two to three months later, the β-actin::LSL::G12V KRAS;Tyr::CreERT2 mice (henceforth G12V KRAS;CreERT2 mice) developed diffuse skin hyperpigmentation on their backs, tails, and ears (Fig. 1A). Importantly, pigmentation was never seen in ethanol-treated G12V KRAS;CreERT2 mice or in TM-treated β-actin::LSL::G12V KRAS or Tyr::CreERT2 mice (Fig. 1A and data not shown). The TM-treated G12V KRAS;CreERT2 mice also developed several types of melanocytic lesion. Most of the animals developed multiple small lesions bearing a striking similarity to human blue nevi, predominantly within the TM-treated areas but also in the nontreated regions of the skin. In approximately 30% of the animals, these lesions were visible as dome-shaped lesions on the surface of the skin (Fig. 1A, arrow) and were located in the superficial dermal layers (Fig. 1B). More commonly (~70% of animals), the lesions were centered on the deeper regions of the dermo-hypodermal interface (Fig. 1C) and were not obvious until postmortem examination of the skin. These lesions were composed of epithelioid and dendritic melanocytes immersed in a collageinous stroma, occasionally forming vague nests and displaying low levels of pigmentation (Fig. 1B and C). They did not invade the epidermis and no junctional components were observed (Fig. 1B and C). Nuclear pleomorphism and atypia were not found and mitotic figures were absent. Ki67 staining
was negligible (<1%; Fig. 1D). These lesions therefore showed low levels of proliferation and, overall, did not exhibit any features of malignancy. Importantly, however, no such lesions were observed in control animals either macroscopically or on histologic examination (Fig. 1A; data not shown).

Slightly different but still relatively benign melanocytic lesions were observed in the periorbital areas of the TM-treated G12VKRAS;CreERT2 mice (Fig. 1A, arrowheads; Fig. 2A), but again not in the controls (Fig. 1A). These darkly pigmented lesions were also first evident 2 to 3 months after TM treatment and were characterized by an accumulation of heavily pigmented cells within the dermis, but again without epidermal or epidermal-dermal junction involvement (Fig. 2B). These lesions were significantly larger than the blue nevi and possessed occasional extensions into the subcutaneous tissue, but without displaying overt infiltrating or destructive growth patterns. They were also composed of admixed heavily melanin-laden epithelioid (Fig. 2C, arrows) and less pigmented spindle/dendritic cells (Fig. 2C, arrowheads). The nuclei of these cells were slightly enlarged, but were still uniform and possessed small discrete nucleoli. Overt nuclear atypia and nuclear pleomorphism were not observed. Mitotic figures were scant at <1 per 10 high-power fields (HPF) and restricted to the superficial areas of the lesions. These histologic features are consistent with a diagnosis of pigmented epithelioid melanocytoma, a melanocytic lesion characterized by...
by local aggressiveness and borderline metastatic potential in humans (21).

Importantly, all of the mice eventually developed large, rapidly growing tumors, either within the TM-treated areas of skin (seen in 85% of the mice; Fig. 3A) or in the perianal region (seen in 79% of the mice; Fig. 3B). These tumors arose between 1 and 10 months of TM treatment with a median time of 4 months (Fig. 3C). Ultimately, one or other of these lesions necessitated sacrifice of the animals. The TM-treated skin lesions were hypopigmented, asymmetrical, and dome-shaped (Fig. 3A). The perianal lesions were ostensibly black, but also dome-shaped, asymmetrical (Fig. 3B), and often ulcerated. Histologically, the superficial aspects of the perianal lesions were similar in appearance to the periorbital lesions. They lacked epithelial or junctional components and were composed of heavily pigmented epithelioid cells mixed with less pigmented spindle/dendritic cells (Fig. 3D). However, in their deeper aspects, the perianal lesions were composed largely of hypo/amelanotic spindle cells that exhibited conspicuous nuclear pleomorphism with large nucleoli (Fig. 4A) and pseudo-nuclear inclusions (Fig. 4B). The lesions displayed diffuse positivity for Ki67 (Fig. 4C; Supplementary Fig. S2), which correlated with high mitotic activity and an average of 6 mitoses/10 HPFs in both the superficial and deep aspects of the lesions (Fig. 4D, arrows).

The TM-treated skin lesions lacked the superficial pigmented cells seen in the perianal lesions, but were otherwise very similar to their deeper aspects (summarized in Supplementary Table S2). They were asymmetrical, grossly infiltrative and destructive, and often ulcerated. Generally, they were composed largely of invasive hypo/amelanotic spindle cells with atypical and markedly pleomorphic nuclei, large nucleoli, pseudo-nuclear inclusions, and diffuse Ki67 positivity (Supplementary Table S2). Compared with the perianal lesions, they seemed to be more aggressive, with higher mitotic activity (9 mitoses/10 HPFs; Supplementary Table S2) and mitotic figures promptly found in their deep aspects. Consistent with their higher proliferative fraction, they reached tumor burden limits within 3 weeks of first presentation, whereas the perianal lesions took up to 10 weeks to reach tumor burden limits.

Using PCR, we provide clear evidence that the β-actin::LSL::G12VKRAS locus was rearranged both in the TM-treated skin and in perianal tumors (Fig. 5A, tumors A and B, respectively). Unlike previously described RAS-driven models of melanomagenesis, in our studies tumor induction was efficient without the need for further manipulation of the mice, and thus we examined p16INK4a and Tp53 status in these tumors. Using double-staining immunofluorescence with the melanocytic marker S100, we show that p16INK4a is expressed in the cancer cells in the tumors in our mice (Fig. 5B). We also performed complete sequencing of the Tp53 cDNA from these tumors and did not detect any mutations (data not shown). Further, by Western blot we showed that Tp53
protein was not detectable in cell lines derived from the tumors, but that it responded normally and was stabilized when the cells were treated with the DNA-damaging agent doxorubicin (Fig. 5C). Together, these data show that p16$\text{INK4a}$ and Tp53 are intact in the G12VKRAS-driven tumors.

The histopathologic features of the deeper aspects of the TM-treated skin and perianal lesions were remarkably similar and both stained positive for S100 (Supplementary Table S2). However, our attempts to stain these tumors with other melanocytic markers, including HMB45/gp100, Melan A, and Mitf, were unsuccessful because the antibodies were raised against human antigens and do not seem to have sufficient sensitivity and specificity for mouse tissues. Furthermore, using immunohistochemistry, it was recently shown that human spindle cell melanomas frequently lack expression of melanocytic markers other than S100 (22). We therefore used RT-PCR to investigate the expression of melanocytic genes in the tumors because it is more sensitive. Tyrosinase, Dct, Pax3, and silver (which encodes HMB45/gp100) were all strongly expressed in the tumors from both the TM-treated skin and the perianal regions (Fig. 5D, tumors A and B, respectively). As a control, we show that these genes were not expressed in brain or kidney from TM-treated animals. We also examined these markers in the benign lesions in the mice. We show expression of Dct and, albeit more weakly, of tyrosinase, Pax3, and silver in the periorbital lesions from TM-treated G12V KRAS;CreERT2 mice, but not in the eyelids of wild-type animals (Fig. 5D). We also observed expression of tyrosinase, Dct, Pax3, and silver in the nevus-rich skin from TM-treated G12V KRAS;CreERT2 mice but not, or at considerably lower levels, in the skin from wild-type mice (Fig. 5D).

Together, these data confirm that the benign lesions derive from melanocytic cells and provide evidence to support a diagnosis of melanoma in the TM-treated skin and perianal tumors. Although these tumors display many features that are used to differentiate malignant from benign melanocytic lesions in human pathology (Supplementary Table S2), we did not observe metastatic deposits in our mice on postmortem examination (data not shown). However, the cells from these lesions did invade the subcutaneous tissue and the skeletal muscle fibers in a destructive growth fashion (Fig. 6A). Therefore, to confirm the tumorigenicity of the cancer cells, we show that cell lines developed from these lesions grew as subcutaneous tumors in nude mice (Fig. 6B). We also engineered these cells to express firefly luciferase and injected them into the tail veins of nude mice. One month after injection, intravital imaging indicated that melanoma cells had colonized the lungs of all (5 of 5) of the recipient animals (Fig. 6C), and multiple tumor foci were observed in the lungs of these mice on postmortem examination (Fig. 6D, arrows). Importantly, immunohistochemistry revealed that the subcutaneous lesions and the lung lesions both contained metastatic deposits harboring cells with morphologic features similar to those in the original tumors (Supplementary Fig. S3). Thus, despite the lack of metastatic deposits in situ, these data confirm the malignant potential of the cancer cells in these tumors.

Discussion

Here we describe a mouse model of melanoma driven by oncogenic KRAS. We show that G12V KRAS expression in the melanocytes induces rapid-onset skin hyperpigmentation, followed by induction of two types of benign melanocytic lesions that are characterized by an accumulation of pigmented epithelioid and spindle cells. Despite the obvious expansion of pigmented cells, these lesions were benign and possessed low levels of proliferation. They were largely (but not exclusively) restricted to the TM-treated areas of skin where they displayed the characteristic histologic features of human blue nevi, or they occurred on the eyelids, where they resembled pigmented epithelioid melanocytomas. They were restricted to the dermal layers of the skin and did not possess epidermal or junctional components, presumably reflecting the fact that mouse melanocytes, unlike human melanocytes, are largely restricted to the dermis. We note that although TM was topically applied to the skin on backs of the mice, they mounted a systemic response, developing pigmented lesions at distant sites. This is similar to the responses we observed when oncogenic Braf was expressed in mouse melanocytes (16) and presumably occurs because TM is absorbed through the dermis or ingested during posttreatment grooming.

All of the mice developed aggressive tumors on the TM-treated skin or in their perianal regions. Blue nevi were...
common at the periphery of these tumors, but as with melanoma induced by oncogenic Braf (16), it is not possible to prove that the tumors emerged directly from these nevi or if they developed de novo and their juxtaposition was circumstantial. We confirmed the tumorigenicity of the lesions by showing that cells derived from them grew as subcutaneous tumors in nude mice, and their metastatic potential was confirmed by their locally invasive and destructive growth patterns, their high proliferation indices, and their ability to colonize the lungs of recipient mice following tail vein injections. The median latency for tumor induction was 4 months after \(G^{12V}\)KRAS expression, but tumors occurred as early as 1 month, and within 10 months all mice had tumors on at least one site; 64% of the mice developed tumors on both sites. Generally, the tumors on the two sites were similar and displayed the hallmark features of malignant melanocytic tumors in humans, including asymmetry, infiltrative and destructive growth patterns, and areas of ulceration. In agreement with studies showing that oncogenic RAS suppresses tyrosinase expression (23), the tumors were generally hypo/amelanotic. The cells were generally spindle-shaped with atypical, pleomorphic nuclei, large nucleoli, and pseudonuclear inclusions. They expressed S100, tyrosinase Dct, Pax3, and silver, markers of the melanocytic lineage, and importantly, the \(\beta\text{-actin::LSL::G}^{12V}\)KRAS locus had been recombined in these tumors. This provides strong circumstantial evidence to support a diagnosis of metastatic melanoma driven by oncogenic KRAS for both types of tumor.

Despite the general overall similarity of the tumors on the two sites, some subtle differences were seen. There were pigmented cells in the superficial aspects of the perianal tumors that were not seen in the TM-treated skin tumors.

**Figure 5.** Molecular characterization of melanocytic lesions. A, PCR-mediated genotyping for recombined \(G^{12V}\)KRAS (lox-G\(^{12V}\)KRAS; 240bp) from a dorsally located tumor (tumor A) and a perianal tumor (tumor B). Kidney DNA is used as a negative control and Tyr::Cre\(^{ERT2}\) is present in all three samples. B, double immunofluorescence staining for p16\(^{INK4a}\) (p16; green) and S100 (red) in a \(G^{12V}\)KRAS-driven tumor. DAPI nuclear staining (blue) reveals the nuclei and a merged image is shown. C, Western blot for Tp53 in three \(G^{12V}\)KRAS-driven tumor cell lines (#8352, #5954, #4755) treated with DMSO (control) or doxorubicin (DOX; 1 \(\mu\)mol/L; 8 h). Erk2 is used as a loading control. D, RT-PCR analysis for expression of Tyrosinase (Tyr.), Dct, Pax3, Silver (Si/gp100), and Gapdh (loading control). Tumors were taken from a section of TM-treated skin (tumor A) or from the perianal region (tumor B). The indicated tissues were also analyzed from a TM-treated \(G^{12V}\)KRAS;Cre\(^{ERT2}\) (G\(^{12V}\)KRAS) or control (WT) mouse.
perianal tumors were also generally less aggressive. They had fewer proliferating cells and it took them three times as long as the TM-treated skin tumors to reach tumor burden limits. It is unclear what governs these differences or why distinct types of lesions develop in various sites. It may stem from differences in melanocyte distribution or melanocyte type in the perianal and periorbital regions due to the transition from skin to nonsquamous epithelium, or it may be due to the presence of distinct microenvironments caused by differences in growth factor production by the epithelial cells lining the mucosa and skin or by differences in the types of fibroblasts or inflammatory cells found locally. Thus cell-intrinsic or cell-extrinsic factors could mediate these differences, but the mechanism is currently unclear.

We note several differences between our model and the RAS-driven models previously reported. In particular, our mice seem to succumb to a progressive phenotype and a broader spectrum of melanocytic lesions. Skin hyperpigmentation was seen when G12VHRAS or Q61LNRAS was constitutively expressed using a tyrosinase enhancer/promoter (13, 24) but not when G12VHRAS was expressed using a doxycyclin- or TM-inducible system (10, 12). We also report induction of nevi—only previously reported in a tyrosinase-driven G12VHRAS model (24)—and, unique for Ras-driven models, pigmented epithelioid melanocytomas, although oncogenic Braf induces both types of lesion (16). The most significant difference in our model is its penetrance. All of our mice developed melanoma within 10 months without the loss of p16INK4a or Tp53 or the exposure to UV light required by previous models (8, 9, 12, 13). Intriguingly, p16INK4a remains intact in the tumors induced by G12VHRAS combined with UV light (11), and we show that p16INK4a and Tp53 also remain intact in the tumors in our mice. We therefore conclude that loss of these tumor suppressors is not required for G12VHRAS-induced melanomagenesis in this model.

The differences between the various mouse models presumably reflect differences in the design of the transgenes used in the various studies. Many of the previous models used the tyrosinase promoter (8, 10, 12, 13, 24), which is activated early and thus would drive constitutive oncogene expression throughout melanocyte development. This approach does not therefore mimic the acquisition of somatic mutations that occurs in humans and may allow melanocytes to adapt to the oncogene during their development. Furthermore, different tyrosinase enhancer elements could regulate subtle differences in expression or, as has even been previously suggested (12), expression in distinct subpopulations of melanocytes. This could explain why Powell and colleagues reported a constitutive Tyr::HRAS model that gives hyperpigmentation and nevi (24), whereas Chin and colleagues reported another Tyr::HRAS model that does not develop these lesions (9). To overcome some of these concerns, we used a ubiquitous promoter with exquisite control over timing of expression and observed a broader spectrum of skin lesions than previously reported. Surprisingly, however, we recently reported that expression of G12DKras using the...
endogenous mouse gene to ensure physiologic expression levels induced some skin darkening but no nevi, and tumors only when a second genetic manipulation (expression of kinase-dead BRAF) was also engineered (25).

The data above suggest that the timing and levels of expression, and possibly also the precise mutation and RAS isoform involved, are important factors in determining whether tumors are induced in RAS-driven models. Importantly, we present a model of melanomagenesis driven by oncogenic KRAS that is 100% penetrant without the need for additional genetic manipulation or exposure to UV light. Despite this high level of penetrance, we argue that G12V KRAS alone is not sufficient to induce melanoma even in this model. The progressive nature of the response and the long latency required for tumor induction suggest that additional genetic events are likely to be required and we are currently attempting to identify those events. KRAS is reported to be less oncogenic than NRAS in melanocytes in vitro (7) and KRAS mutations are less common than NRAS or HRAS mutations in human melanoma. It has been suggested that this is because KRAS is less efficient at blocking Myc phosphorylation by GSK3 (26), and although we show that Myc is expressed in the tumors from our mice (Supplementary Fig. S4), we have yet to investigate its phosphorylation status. Alternatively, it has been suggested that, unlike HRAS and NRAS, which are in lipid rafts, KRAS is disordered in the plasma membrane and thus less able to activate PI3K/AKT (26). However, our data now establish that G12V KRAS can transform melanocytes in a nonsensitized background in vivo, suggesting that its oncogenicity may be context specific.

Thus, we have developed a mouse model of melanoma driven by oncogenic KRAS that is initiated by a single, inducible genetic event. We establish that oncogenic KRAS can induce nevi and be a founder event in melanomagenesis. The lesions we observe resemble those of amelanotic/oligomelanotic malignant metastatic melanoma in humans, and the model is relevant because although KRAS mutations are rare in human melanoma, when present, they usually (>75%) involve a G12 substitution (http://www.sanger.ac.uk/genetics/CGP/cosmic/). This tractable model is therefore a powerful tool in the study of KRAS-driven melanoma.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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