

## Molecular Mechanisms and Clinical Implications in Melanoma Brain Metastasis: New Insights

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### ABSTRACT

**AIM:** Metastatic spread of melanoma to the brain is a common and devastating manifestation of disease progression. Understanding the various molecular events is critical to develop new therapies for melanoma brain metastasis.

**METHODS:** The literature-based review was conducted by searching for keywords in Pubmed using the search terms: 'melanoma', 'metastatic melanoma', 'brain metastasis', 'molecular basis', 'immunotherapy' and 'targeted therapy'.

**RESULTS:** The molecular mechanisms implicated in melanoma brain metastasis include signaling pathways, proteases and surface markers involved in the formation of brain metastases. Dabrafenib (specific inhibitor of mutated BRAFV600E protein) may be more effective than vemurafenib (mutant BRAF inhibitor) for patients with advanced melanoma with brain metastases.

**CONCLUSIONS:** Since the central nervous system (CNS) lacks a lymphatic system, tumor cells can only reach the brain parenchyma by hematogenous metastasis formation. During this process metastatic cells need to traverse brain endothelial cells which in turn form the morphological basis of the blood-brain barrier (BBB). The most important cellular elements of the BBB are endothelial cells, astrocytes and pericytes. Targeted therapies are highly recommended to be incorporated into the multimodality management of melanoma brain metastasis. Nanoparticles have been studied extensively for drug delivery, to increase anticancer treatment efficacy. Drugs delivered by nanoparticles may have a longer biological life, due to packaging protection, and may be concentrated in the site of cancer due to enhanced permeability and retention (EPR) at cancer sites. Nanoparticles produce a constant release of delivered drugs. Nanoencapsulation of the recent FDA-approved drugs might be beneficial.

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**Key words:** Melanoma; Brain metastasis; Metastatic melanoma; Targeted therapy; Immunotherapy

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### INTRODUCTION

Brain metastasis is a major cause of morbidity and mortality in advanced melanoma<sup>[1]</sup>. Malignant melanoma is the third most common cancer leading to brain metastases after carcinomas of the lung and breast, having the highest propensity to metastasize to the brain of all primary neoplasms in adults. Autopsy data indicate a prevalence of 55-75% of brain metastasis in melanoma<sup>[2]</sup>. Brain metastasis contributes to death in nearly 95% of patients<sup>[2]</sup>. In general, the median survival from the time of diagnosis of cerebral metastases is less than 6 months<sup>[1]</sup>. A key event in brain metastasis is the migration of cancer cells through the blood brain barrier (BBB)<sup>[3]</sup>. Since the central nervous system (CNS) lacks a lymphatic system,

tumor cells can only reach the brain parenchyma by hematogenous metastasis formation. During this process metastatic cells need to traverse brain endothelial cells which in turn form the morphological basis of the blood-brain barrier. Different cell surface and adhesion molecules, proteolytic enzymes and signaling pathways have been shown to facilitate invasive and migratory capacities of melanoma cells and their transfer through the blood-brain barrier<sup>[2]</sup>. The MAP-kinase signaling pathways (including the mutations in BRAF) are critical in the development and progression of primary and brain metastatic melanoma<sup>[4]</sup>. BRAFV600E has been implicated in different mechanisms of melanoma progression, and principally the activation of the downstream MEK/ERK pathway, evasion of senescence and apoptosis, unchecked replicative potential, angiogenesis (through MEK-dependent activation of hypoxia-inducible factor (HIF)-1 $\alpha$  and vascular endothelial growth factor), tissue invasion and metastasis (via upregulation of several proteins involved in migration, integrin signaling, cell contractility, tumor- and microenvironment-derived interleukin-8), as well as the evasion of immune response<sup>[5]</sup>. It is important that understanding the molecular mechanisms in melanoma brain metastasis could make substantial contributions to reducing mortality from melanoma.

Treatments for melanoma brain metastases have included surgical resection for patients with a single or limited number of lesions, stereotactic radiosurgery, or whole brain radiation therapy (with or without surgery/stereotactic radiosurgery)<sup>[1]</sup>. It is becoming clearer that the genetic background of a certain patient (i.e., germline mutations) or a tumor should dictate its treatment regimen, and that targeted therapy against these tumor-specific alterations may be more efficacious<sup>[6]</sup>. Melanoma is a multifactorial disease whose risk depends on genetic susceptibility as well as on external factors<sup>[3]</sup>. Further understanding of the various molecular events that occur in melanoma brain metastasis is critical to develop new targeted therapies<sup>[7]</sup>.

## MATERIALS AND METHODS

### Search strategies and selection criteria

The literature-based review was conducted by searching for keywords in Pubmed using the search terms: ‘melanoma’, ‘metastatic melanoma’, ‘brain metastasis’, ‘molecular basis’, ‘immunotherapy’ and ‘targeted therapy’. Only papers published in English and focusing on the association between the molecular mechanisms and melanoma brain metastasis were included.

## RESULTS

This paper will outline the new insights of molecular mechanisms and clinical implications in melanoma brain metastasis. Molecular mechanisms are classified into 5 groups.

### Group 1. Signaling pathways (PI3K/AKT and BRAF/MAPK) involved in the formation of brain metastases

#### 1. Mitogen activated protein kinase (MAPK) signaling pathway:

The MAP-kinase signaling pathways (including the mutations in BRAF) are critical in the development and progression of primary and brain metastatic melanoma<sup>[8]</sup>. Among the BRAF mutations observed in melanoma, over 90 % are at codon 600, and among these, over 90 % are a single nucleotide mutation resulting in substitution of glutamic acid for valine (BRAFV600E)<sup>[9]</sup>.

Clinical aspect: Vemurafenib, a selective BRAF inhibitor (BRAFi), has been approved by the US Food and Drug Administration for the

treatment of unresectable or metastatic melanoma in patients with BRAFV600E mutations<sup>[10]</sup>.

Mechanisms of BRAFi resistance:

It is noteworthy that resistance to vemurafenib occurred rather quickly in melanoma patients and the median progression-free survival was only 5.3 months<sup>[10]</sup>. The majority of BRAF mutant melanomas responds to BRAF inhibitors (BRAFi) rapidly but acquires drug resistance within a median time of 6-7 months. The specific mechanisms of acquired BRAFi resistance are variegated but fall under two core pathways: (1) reactivation of RAF-MEK-ERK MAPK signaling, and (2) activation of MAPK-redundant signaling via the receptor tyrosine kinase (RTK)-PI3K-AKT pathway, which is parallel but interconnected to the MAPK pathway<sup>[11]</sup>.

Dabrafenib (GSK2118436) is a stronger and specific inhibitor of mutated BRAFV600E protein<sup>[12]</sup>. A dedicated phase 2 study was conducted to further examine the effect of dabrafenib in those with untreated, or previously treated but relapsed, brain metastases. In BRAFV600E melanoma patients, the overall intracranial response rates (OIRRs) in untreated patients and in previously treated patients were 39% and 31%, respectively. The intracranial disease control rate (defined as complete+partial response+stable disease) was 80%–90%. In BRAFV600E patients in both cohorts, the median PFS was 16 weeks and the median OS was 31-33 weeks<sup>[13]</sup>.

Clinical results: Dabrafenib has activity and an acceptable safety profile in patients with BRAFV600E-mutant melanoma and brain metastases irrespective of whether they are untreated or have been previously treated but have progressed<sup>[13]</sup>.

In order to address the problem of resistance to therapy with BRAF kinase inhibitors associated with reactivation of the mitogen-activated protein kinase (MAPK) pathway, Flaherty and colleagues conducted a phase 1 and 2 trial of combined treatment with dabrafenib, a selective BRAF inhibitor, and trametinib, a selective MAPK kinase (MEK) inhibitor<sup>[14]</sup>.

Clinical results: These results revealed that dabrafenib and trametinib were safely combined at full monotherapy doses with significant improvement of response rate and progression-free survival. The rate of complete or partial response with combination therapy (150 mg of dabrafenib and 2 mg of trametinib) was 76%, as compared with 54% with monotherapy ( $p = 0.03$ ). Median progression-free survival in the combination group (150 mg of dabrafenib and 2 mg of trametinib) was 9.4 months, as compared with 5.8 months in the monotherapy group (hazard ratio for progression or death, 0.39; 95% confidence interval, 0.25 to 0.62;  $p < 0.001$ )<sup>[14]</sup>.

Several common nanoparticles, including liposome, polymersomes, dendrimers, carbon-based nanoparticles, and human albumin, have been used to deliver chemotherapeutic agents, and small interfering ribonucleic acids (siRNAs) against signaling molecules have also been tested for the treatment of melanoma. Nanoparticles have been studied extensively for drug delivery, to increase anticancer treatment efficacy. Drugs delivered by nanoparticles may have a longer biological life, due to packaging protection, and may be concentrated in the site of cancer due to enhanced permeability and retention (EPR) at cancer sites. Nanoparticles produce a constant release of delivered drugs<sup>[15]</sup>. Nanoencapsulation of the recent FDA-approved drugs (e.g., vemurafenib, ipilimumab), using proper nanocarriers, or rational combination of these drugs with available adjuvant nanotherapeutics might be beneficial as they might enhance the overall pharmacological features (e.g., bioavailability and targeting)<sup>[16]</sup>.

#### 2. The phosphoinositide 3-kinase (PI3K)-AKT pathway:

Davies and colleagues reported that BRAF-mutant tumors had

higher levels of phosphorylated AKT-Ser473, phosphorylated AKT-Thr308, and phosphorylated GSK3 $\alpha$ /beta than NRAS-mutant tumors. Brain metastases had significantly higher phosphorylated AKT and lower PTEN than lung or liver metastases<sup>[17]</sup>.

Clinical implication: These findings have implications for the rational development of targeted therapy for melanoma brain metastasis<sup>[17]</sup>. PI3K, PDK1 and AKT are targets for therapeutic purposes in melanoma. PI3K inhibitor, LY294002, works through ATP competitive inhibition, preventing the phosphorylation of PI3K. Continuing down the AKT signaling pathway, the next attractive target is PDK1. UCN-01 is a nonselective inhibitor of PDK1. AKT inhibitor, Perifosine, functionally inhibits the activation of AKT by preventing phosphorylation at both sites within the activation loop, as well as impeding AKT membrane translocation<sup>[18]</sup>.

### Group 2. Proteases involved in the formation of brain metastases

The role of serine proteases in the formation of brain metastases:

Melanoma cells coming in contact with cerebral endothelial cells (CECs) disrupted the tight and adherens junctions of CECs. During this process melanoma cells produced and released large amounts of proteolytic enzymes, mainly serine proteases<sup>[2]</sup>.

Clinical implication: The serine protease inhibitor Pefabloc<sup>®</sup> was able to decrease to 44-55% the number of melanoma cells migrating through CECs<sup>[2]</sup>.

### Group 3. The surface biomarkers involved the formation of brain metastases

#### 1. Melanotransferrin and its clinical implication:

Administration of a single dose of a monoclonal antibody (L235) directed against human melanotransferrin (MTf) significantly reduced the development of human melanoma brain metastases in nude mice<sup>[19]</sup>.

Clinical implication: The data suggested MTf as an attractive target and demonstrated the therapeutic potential of an anti-MTf monoclonal antibody for preventing metastatic melanoma<sup>[19]</sup>.

#### 2. Integrins:

Vogetseder and colleagues investigated the expression of  $\alpha\beta3$ ,  $\alpha\beta5$ ,  $\alpha\beta6$  and  $\alpha\beta8$  integrins in primary malignancies and metastases to brain of breast, lung and renal carcinomas and in malignant melanoma. Integrin  $\alpha\beta3$  expression was increased in brain metastases compared to primary cutaneous melanoma ( $p < 0.01$ )<sup>[20]</sup>.

Clinical implication:  $\alpha$ v-integrins may be therapeutic targets for patients with melanoma brain metastasis<sup>[20]</sup>. Cilengitide is a cyclized pentapeptide peptidomimetic designed to compete for the arginine-glycine-aspartic acid (RGD) peptide sequence that regulates integrin ligand binding. Specifically, cilengitide selectively and potently blocks the ligation of the  $\alpha\beta3$  and  $\alpha\beta5$  integrins to provisional matrix proteins such as vitronectin, fibronectin, fibrinogen, von Willebrand factor, osteopontin, and others. These integrins play a significant role in tissue remodeling and angiogenesis and contribute to the growth and malignancy of a wide range of cancers<sup>[21]</sup>.  $\alpha$ v-integrin antagonists not only disrupt tumor angiogenesis but in some cases have a direct impact on the growth and malignant properties of tumor cells themselves<sup>[21]</sup>. Cilengitide is currently in clinical phase II for treatment of metastatic melanoma<sup>[22]</sup>.

### Group 4. Other biomarker involved in the formation of brain metastasis

#### Endothelin receptor B:

Cruz-Munoz and colleagues identified alterations in expression of endothelin receptor B (EDNRB) as a potential factor that influenced

brain metastasis. Induced overexpression of this gene mediated enhanced overall metastatic disease, and resulted in an increased incidence of spontaneous CNS metastases<sup>[23]</sup>.

Clinical implication: The identification of an influential role of EDNRB in CNS melanoma spontaneous metastasis may provide both a target for therapeutic intervention as well as a potential prognostic marker for patients having an increased predisposition for incidence of CNS melanoma metastases<sup>[23]</sup>.

### Group 5. Monoclonal antibody: Ipilimumab

Ipilimumab, a fully human, IgG1 monoclonal antibody, blocks cytotoxic T-lymphocyte-associated antigen 4 (CTLA-4), a negative regulator of T cells, and thereby augments T-cell activation and proliferation<sup>[24]</sup>.

Clinical results: Ipilimumab in combination with dacarbazine, as compared with dacarbazine plus placebo, improved overall survival in patients with previously untreated metastatic melanoma<sup>[24]</sup>. Margolin and colleagues conducted an open-label, phase II trial in patients with melanoma brain metastases. Fifty-one patients in cohort A were neurologically asymptomatic and were not receiving corticosteroid treatment at study entry; 21 patients in cohort B were symptomatic and on a stable dose of corticosteroids. When the brain alone was assessed, 12 patients in cohort A (24%) and two in cohort B (10%) achieved disease control<sup>[25]</sup>. These preliminary data point to the encouraging efficacy of ipilimumab against active melanoma brain metastases.

The completed clinical trials of melanoma brain metastasis are summarized in table 1, and the clinical trials of melanoma brain metastasis currently in progress are summarized in table 2.

## CONCLUSIONS

Melanoma brain metastasis, a common and devastating manifestation of disease progression, remains not well understood.

Vemurafenib, a selective BRAF inhibitor, has been approved by the US Food and Drug Administration for the treatment of unresectable or metastatic melanoma in patients with BRAFV600E mutations. Compared with dacarbazine chemotherapy, vemurafenib significantly improved the 6-month overall survival of patients. The median progression-free survival for vemurafenib is only 5.3 months. Dabrafenib has activity and an acceptable safety profile in patients with BRAFV600E-mutant melanoma and brain metastases irrespective of whether they are untreated or have been previously treated but have progressed<sup>[13]</sup>. The duration of benefit is usually brief because of the development of acquired resistance. Dabrafenib and trametinib were safely combined with significant improvement of response rate and progression-free survival<sup>[14]</sup>. Preliminary data demonstrated the encouraging efficacy of immunotherapy using monoclonal antibody, ipilimumab, against active melanoma brain metastases<sup>[25]</sup>. Ipilimumab in combination with dacarbazine, as compared with dacarbazine plus placebo, improved overall survival in patients with previously untreated metastatic melanoma<sup>[24]</sup>. In the near future, there will most likely be additional targeted therapies and immunotherapies for metastatic melanoma. The critical issues will include the design of small molecule inhibitors that have a high penetrance across the blood-brain barrier as well as immunotherapies that can drive more anti-melanoma cytotoxic T cells into the CNS<sup>[12]</sup>. Nanoencapsulation of the recent FDA-approved drugs (e.g., vemurafenib, ipilimumab), using proper nanocarriers, or rational combination of these drugs with available adjuvant nanotherapeutics might be beneficial as they might enhance the overall pharmacological features (e.g., bioavailability and targeting)<sup>[16]</sup>.

**Table 1** Completed Clinical Trials of Melanoma Brain Metastasis.

Author	Phase	No. patients	Intervention	Main results	Ref.
Dummer2013	II	24	Drug: Vemurafenib	Overall PR: 10/24 (42%) Median PFS: 3.9 months Median OS:5.3 months	[26]
Long 2012	II	172	Drug: Dabrafenib	BRAFV600E patients: OIRR in untreated patients: 39% OIRR in previously treated patients: 31% median PFS in both cohorts: 16 weeks OS in both cohorts:31-33 weeks	[11]
Falchook 2012	I	184	Drug: Dabrafenib	No deaths or discontinuations resulted from adverse events. Response rate (Val600Glu BRAF-mutant): 21/27 (78%)	[27]
Margolin 2012	II	72 cohort A:51 cohort B:21	Drug: Ipilimumab	Disease control rate in the brain: Cohort A: 12/51 (24%) Cohort B:2/21 (10%) Ipilimumab has activity in some patients with advanced melanoma and brain metastases.	[18]
Chiariion-Sileni 2011	III	150	Drug: CTI: cisplatin, temozolomide- and dacarbazine CDI: cisplatin, dacarbazine and interleukin-2	Development of CNS metastasis: CTI: 24/74 (32%) CDI: 34/75 (45%) Median OS: CTI: 8.4 months CDI: 8.7 months No difference in toxicity was observed between the two arms.	[28]
Papadopoulos 2011	I	16	Drug: temozolomide, thalidimide, and lomustine	PR: 2 SD: 2 The combination therapy is safe and well tolerated .	[29]

PR: partial response; SD: stable disease; PFS: progression-free survival; OS: overall survival; OIRR: overall intracranial response rate; Cohort A: No corticosteroid; Cohort B: Corticosteroid required.

**Table 2** Clinical Trials of Melanoma Brain Metastasis Currently in Progress.

Clinical Trial	Estima. No. Patients	Condition	Intervention	Study Start Date	Estimated Study Completion Date	Estimated Primary Completion Date	Ref.
A Phase 2 Prospective Trial of Dabrafenib With Stereotactic Radiosurgery in BRAFV600E Melanoma Brain Metastases	39	BRAFV600E Melanoma Patients	Drug: Dabrafenib; Procedure: Gamma Knife Radiosurgery	April 2013	October 2015	January 2015	[30]
Phase 2 Study of Neoadjuvant Vemurafenib in Melanoma Patients With Untreated Brain Metastases	34	Melanoma	Drug: Vemurafenib	April 2013	March 2016	March 2015	[30]
A Phase 2 Study of Vemurafenib in Metastatic Melanoma Patients With Brain Metastases	146	Melanoma	Drug: Vemurafenib	July 2011	December 2013	December 2013	[30]
Melanoma Gamma Knife (Phase 3)	80	Melanoma brain Metastases	Radiation: SRS; Radiation: WBRT; Behavioral:Cognitive Function Tests; Behavioral: Questionnaires8436	August 2012	Not available	August 2016	[30]
Ipilimumab and Whole-Brain Radiation Therapy or Stereotactic Radiosurgery in Treating Patients With Melanoma With Brain Metastases (Phase 1)	24	Recurrent Melanoma; Stage IV Melanoma; Tumors Metastatic to Brain	Drug: Ipilimumab; Radiation: Whole-Brain Radiation Therapy; Radiation: Stereotactic Radiosurgery	Nov. 2012	November 2019	November 2017	[30]
Whole Brain Radiotherapy Following Local Treatment of Intracranial Metastases of Melanoma (Phase 3)	200	Intracranial Metastases of Melanoma	Radiation: Whole-Brain Radiation Therapy	Oct. 2007	June 2016	June 2015	[30]

## CONFLICT OF INTERESTS

There are no conflicts of interest with regard to the present study.

## REFERENCES

- Rochet NM, Dronca RS, Kottschade LA, Chavan RN, Gorman B, Gilbertson JR, Markovic SN. Melanoma brain metastases and vemurafenib: need for further investigation. *Mayo Clin Proc* 2012; **87**: 976-981
- Fazakas C, Wilhelm I, Nagyoszi P, Farkas AE, Haskó J, Molnár J, Bauer H, Bauer HC, Ayaydin F, Dung NT, Siklós L, Krizbai IA. Transmigration of melanoma cells through the blood-brain barrier: role of endothelial tight junctions and melanoma-released serine proteases. *PLoS One* 2011; **6**: e20758
- Yashin AI, Wu D, Arbeev KG, Kulminski AM, Stallard E, Ukraintseva SV. Why does melanoma metastasize into the brain? Genes with pleiotropic effects might be the key. *Front Genet* 2013; **4**: 75
- Wilhelm I, Molnár J, Fazakas C, Haskó J and Krizbai IA. Role of the blood-brain barrier in the formation of brain me-

- tastases. *Int J Mol Sci* 2013; **14**: 1383-411
- 5 Ascierto PA, Kirkwood JM, Grob JJ, Simeone E, Grimaldi AM, Maio M, Palmieri G, Testori A, Marincola FM, Mozzillo N: The role of BRAF V600 mutation in melanoma. *J Transl Med* 2012; **10**: 85
  - 6 Gaziel-Sovran A, Osman I, Hernando E. *In vivo* modeling and molecular characterization: a path toward targeted therapy of melanoma brain metastasis. *Front Oncol* 2013; **3**: 127
  - 7 Rahmathulla G, Toms SA, Weil RJ. The molecular biology of brain metastasis. *J Oncol* 2012; **2012**: 723541
  - 8 Wilhelm I, Molnár J, Fazakas C, Haskó J, Krizbai IA. Role of the blood-brain barrier in the formation of brain metastases. *Int J Mol Sci* 2013; **14**: 1383-1411
  - 9 Ascierto PA, Kirkwood JM, Grob JJ, Simeone E, Grimaldi AM, Maio M, Palmieri G, Testori A, Marincola FM, Mozzillo N. The role of BRAF V600 mutation in melanoma. *J Transl Med* 2012; **10**: 85
  - 10 Chapman PB, Hauschild A, Robert C, Haanen JB, Ascierto P, Larkin J, Dummer R, Garbe C, Testori A, Maio M, Hogg D, Lorigan P, Lebbe C, Jouary T, Schadendorf D, Ribas A, O'Day SJ, Sosman JA, Kirkwood JM, Eggermont AM, Dreno B, Nolop K, Li J, Nelson B, Hou J, Lee RJ, Flaherty KT, McArthur GA, BRIM-3 Study Group. Improved survival with vemurafenib in melanoma with BRAF V600E mutation. *N Engl J Med* 2011; **364**: 2507-2516
  - 11 Lo RS. Receptor tyrosine kinases in cancer escape from BRAF inhibitors. *Cell Res* 2012; **22**:945-947, 2012.
  - 12 Fonkem E, Uhlmann EJ, Floyd SR, Mahadevan A, Kasper E, Eton O and Wong ET: Melanoma brain metastasis: overview of current management and emerging targeted therapies. *Expert Rev Neurother* 2012; **12**:1207-1215
  - 13 Long GV, Trefzer U, Davies MA, Kefford RF, Ascierto PA, Chapman PB, Puzanov I, Hauschild A, Robert C, Algazi A, Mortier L, Tawbi H, Wilhelm T, Zimmer L, Switzky J, Swann S, Martin AM, Guckert M, Goodman V, Streit M, Kirkwood JM and Schadendorf D: Dabrafenib in patients with Val600Glu or Val600Lys BRAF-mutant melanoma metastatic to the brain (BREAK-MB): a multicentre, open-label, phase 2 study. *Lancet Oncol* 2012; **13**:1087-1095
  - 14 Flaherty KT, Infante JR, Daud A, Gonzalez R, Kefford RF, Sosman J, Hamid O, Schuchter L, Cebon J, Ibrahim N, Kudchadkar R, Burris HA 3rd, Falchook G, Algazi A, Lewis K, Long GV, Puzanov I, Lebowitz P, Singh A, Little S, Sun P, Allred A, Ouellet D, Kim KB, Patel K and Weber J: Combined BRAF and MEK inhibition in melanoma with BRAF V600 mutations. *N Engl J Med* 2012; **367**:1694-1703
  - 15 Chen J, Shao R, Zhang XD, Chen C. Applications of nanotechnology for melanoma treatment, diagnosis, and theranostics. *Int J Nanomedicine* 2013; **8**:2677-2688
  - 16 Mena F: Latest approved therapies for metastatic melanoma: what comes next? *J Skin Cancer* 2013; **2013**:735282
  - 17 Davies MA, Stemke-Hale K, Lin E, Tellez C, Deng W, Gopal YN, Woodman SE, Calderone TC, Ju Z, Lazar AJ, Prieto VG, Aldape K, Mills GB and Gershenwald JE: Integrated molecular, clinical analysis of AKT activation in metastatic melanoma. *Clin Cancer Res* 2009; **15**: 7538-7546
  - 18 Jazirehi AR, Wenn PB, Damavand M: Therapeutic implications of targeting the PI3Kinase/AKT/mTOR signaling module in melanoma therapy. *Am J Cancer Res* 2012; **2**: 178-191
  - 19 Rolland Y, Demeule M, Fenart L, Beliveau R. Inhibition of melanoma brain metastasis by targeting melanotransferrin at the cell surface. *Pigment Cell Melanoma Res* 2009; **22**: 86-98
  - 20 Vogetseder A, Thies S, Ingold B, Roth P, Weller M, Schraml P, Goodman SL, Moch H.  $\alpha$ -Integrin isoform expression in primary human tumors and brain metastases. *Int J Cancer* 2013; **133**: 2362-2371
  - 21 Reardon DA and Cheresch D: Cilengitide: A Prototypic Integrin Inhibitor for the Treatment of Glioblastoma and Other Malignancies. *Genes Cancer* 2011; **2**:1159-1165
  - 22 Kim KB, Prieto V, Joseph RW, Diwan AH, Gallick GE, Papadopoulos NE, Bedikian AY, Camacho LH, Hwu P, Ng CS, Wei W, Johnson MM, Wittemer SM, Vardeleon A, Reckeweg A and Colevas AD: A randomized phase II study of cilengitide (EMD 121974) in patients with metastatic melanoma. *Melanoma Res* 2012; **22**:294-301
  - 23 Cruz-Munoz W, Jaramillo ML, Man S, Xu P, Banville M, Collins C, Nantel A, Francia G, Morgan SS, Cranmer LD, O'Connor-McCourt MD and Kerbel RS: Roles for endothelin receptor B and BCL2A1 in spontaneous CNS metastasis of melanoma. *Cancer Res* 2012; **72**: 4909-4919
  - 24 Robert C, Thomas L, Bondarenko I, O'Day S, M D JW, Garbe C, Lebbe C, Baurain JF, Testori A, Grob JJ, Davidson N, Richards J, Maio M, Hauschild A, Miller WH Jr, Gascon P, Lotem M, Harmankaya K, Ibrahim R, Francis S, Chen TT, Humphrey R, Hoos A, Wolchok JD. Ipilimumab plus dacarbazine for previously untreated metastatic melanoma. *N Engl J Med* 2011; **364**: 2517-2526
  - 25 Margolin K, Ernstoff MS, Hamid O, Lawrence D, McDermott D, Puzanov I, Wolchok JD, Clark JI, Sznol M, Logan TF, Richards J, Michener T, Balogh A, Heller KN and Hodi FS. Ipilimumab in patients with melanoma and brain metastases: an open-label, phase 2 trial. *Lancet Oncol* 2013; **13**: 459-465
  - 26 Dummer R, Goldinger SM, Turttschi CP, Eggmann NB, Michielin O, Mitchell L, Veronese L, Hilfiker PR, Felderer L, Rinderknecht JD. Vemurafenib in patients with BRAFV600 mutation-positive melanoma with symptomatic brain metastases: Final results of an open-label pilot study. *Eur J Cancer* 2013 Nov 29. pii: S0959-8049(13)00979-9. doi: 10.1016/j.ejca.2013.11.002. [Epub ahead of print]
  - 27 Falchook GS, Long GV, Kurzrock R, Kim KB, Arkenau TH, Brown MP, Hamid O, Infante JR, Millward M, Pavlick AC, O'Day SJ, Blackman SC, Curtis CM, Lebowitz P, Ma B, Ouellet D, Kefford RF. Dabrafenib in patients with melanoma, untreated brain metastases, and other solid tumours: a phase 1 dose-escalation trial. *Lancet* 379:1893-1901, 2012.
  - 28 Chiarion-Sileni V, Guida M, Ridolfi L, Romanini A, Del Bianco P, Pigozzo J, Brugnara S, Colucci G, Ridolfi R, De Salvo GL and Italian Melanoma Intergroup (IMI): Central nervous system failure in melanoma patients: results of a randomised, multicentre phase 3 study of temozolomide and dacarbazine-based regimens. *Br J Cancer* 2011; **104**: 1816-1821
  - 29 Papadopoulos NE, Hwu W, Cain S, Posada L, Kim KB, Homsy J, Bedikian AY, Davies MA and Hwu P: Phase I trial of temozolomide, thalidomide, and lomustine in patients with metastatic melanoma in the brain. *J Clin Oncol* 2011; **29**: suppl, 8571
  - 30 ClinicalTrials.gov <http://www.clinicaltrials.gov>

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