

Answer 1:

### Bibliographic Information

**Knocking down galectin 1 in human Hs683 glioblastoma cells impairs both angiogenesis and endoplasmic reticulum stress responses.** Le Mercier, Marie; Mathieu, Veronique; Haibe-Kains, Benjamin; Bontempi, Gianluca; Mijatovic, Tatjana; Decaestecker, Christine; Kiss, Robert; Lefranc, Florence. Laboratory of Toxicology, Institute of Pharmacy, Free University of Brussels (ULB), Brussels, Belg. *Journal of Neuropathology & Experimental Neurology* (2008), 67(5), 456-469. Publisher: Lippincott Williams & Wilkins, CODEN: JNENAD ISSN: 0022-3069. Journal written in English. AN 2008:653285 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

### Abstract

Galectin (Gal) 1 is a hypoxia-regulated proangiogenic factor that also directly participates in glioblastoma cell migration. To det. how Gal-1 exerts its proangiogenic effects, we investigated Gal-1 signaling in the human Hs683 glioblastoma cell line. Galectin 1 signals through the endoplasmic reticulum transmembrane kinase/RNase inositol-requiring  $1\alpha$ , which regulates the expression of oxygen-regulated protein 150. Oxygen-regulated protein 150 controls vascular endothelial growth factor maturation. Galectin 1 also modulates the expression of 7 other hypoxia-related genes (i.e. CTGF, ATF3, PPP1R15A, HSPA5, TRA1, and CYR61) that are implicated in angiogenesis. Decreasing Gal-1 expression in Hs683 orthotopic xenografts in mouse brains by siRNA administration impaired endoplasmic reticulum stress and enhanced the therapeutic benefits of the proautophagic drug temozolomide. These results suggest that decreasing Gal-1 expression (e.g. through brain delivery of nonviral infusions of anti-Gal-1 siRNA in patients) can represent an addnl. therapeutic strategy for glioblastoma.

Answer 2:

### Bibliographic Information

**Noninvasive Imaging of Apoptosis and Its Application in Cancer Therapeutics.** Coppola, Julia M.; Ross, Brian D.; Rehemtulla, Alnawaz. Departments of Biological Chemistry and Radiation Oncology, University of Michigan Medical School, Ann Arbor, MI, USA. *Clinical Cancer Research* (2008), 14(8), 2492-2501. Publisher: American Association for Cancer Research, CODEN: CCRE4 ISSN: 1078-0432. Journal written in English. AN 2008:495584 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

### Abstract

**PURPOSE:** Activation of the apoptotic cascade plays an important role in the response of tumors to therapy. Noninvasive imaging of apoptosis facilitates optimization of therapeutic protocols regarding dosing and schedule and enables identification of efficacious combination therapies. **Exptl. Design:** We describe a hybrid polypeptide that reports on caspase-3 activity in living cells and animals in a noninvasive manner. This reporter, ANLucBCLuc, constitutes a fusion of small interacting peptides, peptide A and peptide B, with the NLuc and CLuc fragments of luciferase with a caspase-3 cleavage site (DEVD) between pepANLuc (ANLuc) and pepBCLuc (BCLuc). During apoptosis, caspase-3 cleaves the reporter, enabling sepn. of ANLuc from BCLuc. A high-affinity interaction between peptide A and peptide B restores luciferase activity by NLuc and CLuc complementation. Using a D54 glioma model, we show the utility of the reporter in imaging of apoptosis in living subjects in response to various chemotherapy and radiotherapy regimens. **RESULTS:** Treatment of live cells and mice carrying D54 tumor xenografts with chemotherapeutic agents such as temozolomide and perifosine resulted in induction of bioluminescence activity, which correlated with activation of caspase-3. Treatment of mice with combination therapy of temozolomide and radiation resulted in increased bioluminescence activity over individual treatments and increased therapeutic response due to enhanced apoptosis. **CONCLUSION:** The data provided show the utility of the ANLucBCLuc reporter in dynamic, noninvasive imaging of apoptosis and provides a rationale for use of this technol. to optimize dose and schedule of novel therapies or to develop novel combination therapies using existing drugs.

Answer 3:

### Bibliographic Information

**Impact of Angiogenesis Inhibition by Sunitinib on Tumor Distribution of Temozolomide.** Zhou, Qingyu; Guo, Ping; Gallo, James M. Department of Pharmaceutical Sciences, School of Pharmacy, Temple University, Philadelphia, PA, USA. *Clinical Cancer Research* (2008), 14(5), 1540-1549. Publisher: American Association for Cancer Research, CODEN: CCREF4 ISSN: 1078-0432. Journal written in English. AN 2008:270837 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

#### Abstract

**PURPOSE:** As combination chemotherapy of antiangiogenic agents with conventional chemotherapeutic drugs continues to evolve, an understanding of the pharmacokinetic and pharmacodynamic variables assocd. with optimal treatment is needed. Thus, the effect of the multitargeted tyrosine kinase inhibitor sunitinib on tumor distribution of temozolomide was investigated to evaluate conditions for optimal combination chemotherapy. **Exptl. Design:** In mice bearing SF188V+ human glioma xenografts, measurements of temozolomide pharmacokinetic properties and sunitinib pharmacodynamic activities were evaluated, the latter including determinants for vascular normalization, including CD31, collagen IV, and  $\alpha$ -SMA. **RESULTS:** Sunitinib given in a daily dose of either 10 or 40 mg/kg orally over 14 days increased temozolomide tumor distribution, as indicated by the tumor-to-plasma AUC ratio compared with control; however, only the 10 mg/kg group reached statistical significance ( $P < 0.05$ ). From the pharmacodynamic anal., a "vascular normalization index" incorporating the microvessel d. (MVD) and protein expression of  $\alpha$ -SMA and collagen IV was proposed as an indication of the no. of tumor vessels with relatively good quality, which was found to be significantly correlated with the unbound temozolomide AUC in tumor interstitial fluid ( $P = 0.05$ ). Furthermore, both sunitinib-treated groups maintained the mol. balance between angiopoietins Ang-1 and Ang-2, suggesting a crit. role of angiopoietins in vascular normalization. **CONCLUSIONS:** Several important factors relevant to the antiangiogenic agent-induced tumor vascular normalization have been identified and incorporated into a vascular normalization index that may serve to correlate the angiogenic phenotype to the distribution of cytotoxic drugs in solid tumors.

Answer 4:

#### Bibliographic Information

**In vitro and In vivo Radiosensitization Induced by the DNA Methylating Agent Temozolomide.** Kil, Whoon Jong; Cerna, David; Burgan, William E.; Beam, Katie; Carter, Donna; Steeg, Patricia S.; Tofilon, Philip J.; Camphausen, Kevin. Radiation Oncology Branch, National Cancer Institute, Bethesda, MD, USA. *Clinical Cancer Research* (2008), 14(3), 931-938. Publisher: American Association for Cancer Research, CODEN: CCREF4 ISSN: 1078-0432. Journal written in English. CAN 149:194214 AN 2008:158770 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

#### Abstract

**PURPOSE:** Temozolomide, a DNA methylating agent, is currently undergoing clin. evaluation for cancer therapy. Because temozolomide has been shown to increase survival rates of patients with malignant gliomas when given combined with radiation, and there is conflicting preclin. data concerning the radiosensitizing effects of temozolomide, we further investigated the possible temozolomide-induced enhancement of radiosensitivity. **Exptl. Design:** The effects of temozolomide on the in vitro radiosensitivity of U251 (a human glioma) and MDA-MB231BR (a brain-seeking variant of a human breast tumor) cell lines was evaluated using clonogenic assay. DNA damage and repair were evaluated using phosphorylated histone H2AX ( $\gamma$ H2AX), and mitotic catastrophe was measured using nuclear fragmentation. Growth delay was used to evaluate the effects of temozolomide on in vivo (U251) tumor radiosensitivity. **RESULTS:** Exposure of each cell line to temozolomide for 1 h before irradiation resulted in an increase in radiosensitivity with dose enhancement factors at a surviving fraction of 0.1 ranging from 1.30 to 1.32. Temozolomide had no effect on radiation-induced apoptosis or on the activation of the G2 cell cycle checkpoint. As a measure of DNA double strand breaks,  $\gamma$ H2AX foci were detd. as a function of time after the temozolomide + irradiation combination. The no. of  $\gamma$ H2AX foci per cell was significantly greater at 24 h after the combined modality compared with the individual treatments. Mitotic catastrophe, measured at 72 h, was also significantly increased in cells receiving the temozolomide + irradiation combination compared with the single treatments. In vivo studies revealed that temozolomide administration to mice bearing U251 tumor xenografts resulted in a greater than additive increase in radiation-induced tumor growth delay with a dose enhancement factor of 2.8.

**CONCLUSIONS:** These results indicate that temozolomide can enhance tumor cell radiosensitivity in vitro and in vivo and suggest that this effect involves an inhibition of DNA repair leading to an increase in mitotic catastrophe.

Answer 5:

**Bibliographic Information**

**Antiangiogenic compounds interfere with chemotherapy of brain tumors due to vessel normalization.** Claes, An; Wesseling, Pieter; Jeuken, Judith; Maass, Cathy; Heerschap, Arend; Leenders, William P. J. Departments of Pathology and Radiology, Nijmegen Medical Centre, Radboud University, Nijmegen, Neth. *Molecular Cancer Therapeutics* (2008), 7(1), 71-78. Publisher: American Association for Cancer Research, CODEN: MCTOCF ISSN: 1535-7163. Journal written in English. CAN 148:322147 AN 2008:64840 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

Glioblastomas are highly aggressive primary brain tumors. Curative treatment by surgery and radiotherapy is generally impossible due to the presence of diffusely infiltrating tumor cells. Furthermore, the blood-brain barrier (BBB) in infiltrative tumor areas is largely intact, and this hampers chemotherapy as well. The occurrence of angiogenesis in these tumors makes these tumors attractive candidates for antiangiogenic therapies. Because antiangiogenic compounds have been shown to synergize with chemotherapeutic compounds in other tumor types, based on vessel normalization, there is a tendency toward such combination therapies for primary brain tumors also. However, vessel normalization in brain may result in restoration of the BBB with consequences for the efficacy of chemotherapeutic agents. In this study, we investigated this hypothesis. BALB/c nude mice with intracerebral xenografts of the human glioblastoma lines E98 or U87 were subjected to therapy with different dosages of vandetanib (an angiogenesis inhibitor), temozolomide (a DNA alkylating agent), or a combination ( $n > 8$  in each group). Vandetanib selectively inhibited angiogenic growth aspects of glioma and restored the BBB. It did not notably affect diffuse infiltrative growth and survival. Furthermore, vandetanib antagonized the effects of temozolomide presumably by restoration of the BBB and obstruction of chemodistribution to tumor cells. The tumor microenvironment is an extremely important determinant for the response to antiangiogenic therapy. Particularly in brain, antiangiogenic compounds may have adverse effects when combined with chemotherapy. Thus, use of such compounds in neuro-oncol. should be reconsidered. [*Mol Cancer Ther* 2008;7(1):71-8].

Answer 6:

**Bibliographic Information**

**Contributing factors of temozolomide resistance in MCF-7 tumor xenograft models.** Kato, Yoshinori; Okollie, Baasil; Raman, Venu; Vesuna, Farhad; Zhao, Ming; Baker, Sharyn D.; Bhujwala, Zaver M.; Artemov, Dmitri. Department of Radiology, Division of MR Research, The Johns Hopkins University School of Medicine, Baltimore, MD, USA. *Cancer Biology & Therapy* (2007), 6(6), 891-897. Publisher: Landes Bioscience, CODEN: CBTAAO ISSN: 1538-4047. Journal written in English. CAN 148:135281 AN 2007:1287475 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

Vasculature mediated drug resistance in tumors was studied in female SCID mice bearing wild type MCF-7 and adriamycin resistant MCF-7/ADR xenograft using temozolomide (TMZ). A strong tumor growth inhibitory effect of TMZ treatment was observed in MCF-7 tumors during the initial treatment phase with subsequent relapse, but not in MCF-7/ADR tumors. Non-invasive MRI measurements of tumor vascular volume and vascular permeability-surface area product (PS) demonstrated significant reduction of PS in long-term treated MCF-7, but not in MCF-7/ADR tumors. O6-Methylguanine-DNA methyltransferase (MGMT) mRNA, and VEGF expression was analyzed using real-time RT-PCR and ELISA, respectively. No significant changes in MGMT mRNA and VEGF expression were observed in either MCF-7 or MCF-7/ADR tumors. However, in vitro incubation of MCF-7 cells with TMZ did induce the expression of MGMT mRNA. In addition, p53 and p21 levels were scored by immunoblotting. Exposure of cells to TMZ did not affect either the p21 or the p53 expression in both MCF-7 and MCF-7/ADR cells. The absence of these molecular responses to TMZ treatment in MCF-7 tumors in vivo supports the possibility that the onset of cancer drug resistance is associated with reduced PS, which can decrease delivery of the drug to cancer cells.

Answer 7:

**Bibliographic Information**

**Adenovirally Delivered Tumor Necrosis Factor- $\alpha$  Improves the Antiglioma Efficacy of Concomitant Radiation and Temozolomide Therapy.** Yamini, Bakhtiar; Yu, Xiaohong; Pytel, Peter; Galanopoulos, Nicholas; Rawlani, Vinay; Veerapong, Jula; Bickenbach, Kai; Weichselbaum, Ralph R. Section of Neurosurgery, Department of Surgery, Pritzker School of Medicine, The University of Chicago, Chicago, IL, USA. *Clinical Cancer Research* (2007), 13(20), 6217-6223. Publisher: American Association for Cancer Research, CODEN: CCREF4 ISSN: 1078-0432. Journal written in English. CAN 148:369366 AN 2007:1183225 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

**PURPOSE:** Treatment of malignant glioma involves concomitant temozolomide and ionizing radiation (IR). Nevertheless, overall patient survival remains poor. This study was designed to evaluate if addn. of Ad.Egr-tumor necrosis factor (TNF), a replication defective adenovector encoding a cDNA for TNF- $\alpha$ , to temozolomide and IR can improve overall anti-glioma effect. **Exptl. Design:** The efficacy of combination treatment with Ad.Egr-TNF, IR, and temozolomide was assessed in two glioma xenograft models. Animal toxicity and brain histopathol. after treatment were also examd. In addn., in an attempt to explain the antitumor interaction between these treatments, the activation status of the transcription factor nuclear factor- $\kappa$ B was examd. **RESULTS:** Triple therapy (Ad.Egr-TNF, IR, and temozolomide) leads to significantly increased survival in mice bearing glioma xenografts compared with dual treatment. Fifty percent of animals treated with the triple regimen survive for >130 days. Pathol. examn. shows that triple therapy leads to a complete response with formation of a collagenous scar. No significant change in myelination pattern is noted after triple therapy, compared with any double treatment. Treatment of intracranial glioma bearing mice with Ad.Egr-TNF and IR leads to cachexia and poor feeding that does not improve, whereas triple therapy results in less toxicity, which improves over 21 days. Both Ad.Egr-TNF and IR activate nuclear factor- $\kappa$ B, and temozolomide inhibits this activity in an inhibitor of  $\kappa$ B $\alpha$  (I $\kappa$ B $\alpha$ )-independent manner. **CONCLUSION:** This work shows that the addn. of adenoviral TNF- $\alpha$  gene delivery to temozolomide and IR significantly improves anti-glioma efficacy and illustrates a potential new treatment regimen for use in patients with malignant glioma.

Answer 8:

**Bibliographic Information**

**Targeting Methylguanine-DNA Methyltransferase in the Treatment of Neuroblastoma.** Wagner, Lars M.; McLendon, Roger E.; Yoon, K. Jin; Weiss, Brian D.; Billups, Catherine A.; Danks, Mary K. Division of Pediatric Hematology/Oncology, Cincinnati Children's Hospital Medical Center, University of Cincinnati College of Medicine, Cincinnati, OH, USA. *Clinical Cancer Research* (2007), 13(18, Pt. 1), 5418-5425. Publisher: American Association for Cancer Research, CODEN: CCREF4 ISSN: 1078-0432. Journal written in English. CAN 148:135231 AN 2007:1041626 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

**PURPOSE:** The combination of temozolomide and irinotecan has preclin. schedule-dependent synergy against neuroblastoma but is not curative for relapsed high-risk patients. We hypothesized that the DNA repair protein methylguanine-DNA methyltransferase (MGMT) is an important resistance factor, and that inactivation of MGMT would sensitize neuroblastoma cells to these agents. **Exptl. Design:** MGMT protein expression was assessed in 74 primary neuroblastoma tumors. Growth inhibition assays were done to det. the IC50 and the extent of synergy obsd. with various concns. of temozolomide, irinotecan, and the MGMT-inactivating agent O6-benzylguanine, using cultured syngeneic neuroblastoma cells with either low or high levels of MGMT expression. We then assessed efficacy in a mouse xenograft model of metastatic neuroblastoma. **RESULTS:** MGMT was expressed by all 74 tumors evaluated. Pretreatment of neuroblastoma cells with O6-benzylguanine reduced the IC50 of temozolomide by 10-fold regardless of level of MGMT expression, and pretreatment with BG followed by temozolomide + irinotecan further reduced the IC50 in cells with high MGMT expression another 10-fold, to well below clin. achievable concns. The combination index was 0.27 to 0.30 for all three drugs in both cell lines, indicating strong synergy. Survival at 100 days for mice with metastatic neuroblastoma was 56% with three-drug treatment, compared with untreated controls (0%,  $P < 0.001$ ) or temozolomide + irinotecan (10%,  $P = 0.081$ ). **CONCLUSIONS:** MGMT is widely expressed in primary neuroblastoma tumors, and is a relevant therapeutic target. Both in vitro and in vivo studies suggest inactivation of MGMT with O6-benzylguanine may increase the activity of temozolomide and irinotecan against neuroblastoma.

Answer 9:

**Bibliographic Information**

**Bioluminescence monitoring of intracranial glioblastoma xenograft: response to primary and salvage temozolomide therapy.** Dinca, Eduard B.; Sarkaria, Jann N.; Schroeder, Mark A.; Carlson, Brett L.; Voicu, Ramona; Gupta, Nalin; Berger, Mitchel S.; James, C. David. Neuroscience Graduate Program, Mayo Clinic, Rochester, MN, USA. *Journal of Neurosurgery* (2007), 107(3), 610-616. Publisher: American Association of Neurological Surgeons, CODEN: JONSAC ISSN: 0022-3085. Journal written in English. CAN 147:479960 AN 2007:1039856 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

Object: Bioluminescence imaging (BLI) offers a rapid and accurate means for longitudinal study of tumor cell growth and response to therapy in rodent models. Because this technol. has only recently come into use in the field of small animal imaging, applications in this area have been limited. In the current study we have applied BLI to the anal. of clin. relevant issues involving use of the DNA methylating agent temozolomide (TMZ) in a mouse model. Methods: An invasive glioblastoma multiforme xenograft was modified for BLI via transduction with a luciferase-encoding lentivirus. Supratentorial tumors were established in athymic nude mice that were subsequently assigned randomly to control and TMZ treatment groups, and the extent of intracranial tumor was monitored using BLI. Results: In an expt. designed to compare the extent of antitumor effect between a single high-dose TMZ treatment and a protracted low-dose TMZ regimen, BLI revealed the protracted regimen as having superior antitumor effect, and this interpretation was consistent with results from a survival comparison between the two TMZ treatment groups. In a second expt. designed to assess the utility of BLI for testing therapies against recurrent glioblastoma multiforme, mice with intracranial tumors were retreated with TMZ at a time when BLI monitoring revealed tumor regrowth following initial TMZ treatment, and retreatment was successful in providing addnl. survival benefit. Conclusion: The results of these expts. indicate that BLI monitoring can be used as a surrogate for predicting survival benefit from TMZ treatment, permits early detn. of relative survival benefit assocd. with distinct TMZ therapeutic regimens, and offers a means of investigating secondary/salvage therapy efficacy following tumor regrowth from initial therapy.

Answer 10:

**Bibliographic Information**

**Preclinical selection of a novel poly(ADP-ribose) polymerase inhibitor for clinical trial.** Thomas, Huw D.; Calabrese, Christopher R.; Batey, Michael A.; Canan, Stacie; Hostomsky, Zdenek; Kyle, Suzanne; Maegley, Karen A.; Newell, David R.; Skalitzky, Donald; Wang, Lan-Zhen; Webber, Stephen E.; Curtin, Nicola J. Northern Institute for Cancer Research, Medical School, Newcastle University, Newcastle upon Tyne, UK. *Molecular Cancer Therapeutics* (2007), 6(3), 945-956. Publisher: American Association for Cancer Research, CODEN: MCTOCF ISSN: 1535-7163. Journal written in English. CAN 146:513969 AN 2007:289482 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

Poly(ADP-ribose) polymerase (PARP)-1 (EC 2.4.2.30) is a nuclear enzyme that promotes the base excision repair of DNA breaks. Inhibition of PARP-1 enhances the efficacy of DNA alkylating agents, topoisomerase I poisons, and ionizing radiation. Our aim was to identify a PARP inhibitor for clin. trial from a panel of 42 potent PARP inhibitors (Ki, 1.4-15.1 nmol/L) based on the quinazolinone, benzimidazole, tricyclic benzimidazole, tricyclic indole, and tricyclic indole-1-one core structures. We evaluated chemosensitization of temozolomide and topotecan using LoVo and SW620 human colorectal cells; in vitro radiosensitization was measured using LoVo cells, and the enhancement of antitumor activity of temozolomide was evaluated in mice bearing SW620 xenografts. Excellent chemopotential and radiopotential were obsd. in vitro, with 17 of the compds. causing a greater temozolomide and topotecan sensitization than the benchmark inhibitor AG14361 and 10 compds. were more potent radiosensitizers than AG14361. In tumor-bearing mice, none of the compds. were toxic when given alone, and the antitumor activity of the PARP inhibitor-temozolomide combinations was unrelated to toxicity. Compds. that were more potent chemosensitizers in vivo than AG14361 were also more potent in vitro, validating in vitro assays as a prescreen. These studies have identified a compd., AG14447, as a PARP inhibitor with outstanding in vivo chemosensitization potency at tolerable doses, which is at least 10 times more potent than the initial lead, AG14361. The phosphate salt of AG14447 (AG014699), which has improved aq. soly., has been selected for clin. trial.

Answer 11:

**Bibliographic Information**

**The effects of the oral, pan-VEGF-R kinase inhibitor CEP-7055 and chemotherapy in orthotopic models of glioblastoma and colon carcinoma in mice.** Jones-Bolin, Susan; Zhao, Hugh; Hunter, Kathryn; Klein-Szanto, Andres; Ruggeri, Bruce. *Oncology Research*, Cephalon, Inc., West Chester, PA, USA. *Molecular Cancer Therapeutics* (2006), 5(7), 1744-1753. Publisher: American Association for Cancer Research, CODEN: MCTOCF ISSN: 1535-7163. Journal written in English. CAN 145:241183 AN 2006:771727 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

CEP-7055, a fully synthetic, orally active N,N-dimethylglycine ester of CEP-5214, a C3-(isopropylmethoxy)-fused pyrrolocarbazole with potent pan-vascular endothelial growth factor receptor (VEGFR) kinase inhibitory activity, has recently completed phase I clinical trials in cancer patients. These studies evaluated the antitumor efficacy of CEP-7055 using orthotopic models of glioblastoma and colon carcinoma in combination with temozolomide, and irinotecan and oxaliplatin, resp., for their effects on primary and metastatic tumor burden and median survival. Chronic administration of CEP-7055 (23.8 mg/kg/dose) and temozolomide resulted in improvement of median survival of nude mice bearing orthotopic human glioblastoma xenografts compared with temozolomide alone (261 vs. 192 days, resp.;  $P \leq 0.02$ ). Redns. in neurop. dysfunction, brain edema, hemorrhage, and intratumoral microvessel d. (CD34 staining) were obsd. in glioma-bearing mice receiving CEP-7055 alone, temozolomide alone, and the combination of CEP-7055 and temozolomide relative to vehicle and to temozolomide monotherapy. The administration of CEP-7055 in combination with irinotecan (20 mg/kg/dose i.p. x 5 days), and to a lesser degree with oxaliplatin (10 mg/kg/dose i.v.), showed redns. on primary colon carcinoma and hepatic metastatic burden in the CT-26 tumor model relative to that achieved by irinotecan and oxaliplatin monotherapy. These data show the significant efficacy and tolerability of optimal efficacious doses of CEP-7055 when given in combination with temozolomide and irinotecan relative to monotherapy with these cytotoxic agents in preclin. orthotopic glioma and colon carcinoma models and lend support for the use of these treatment regimens in a clin. setting in patients with glioblastoma and colon carcinoma.

Answer 12:

**Bibliographic Information**

**Cimetidine, an unexpected anti-tumor agent, and its potential for the treatment of glioblastoma (review).** Lefranc, Florence; Yeaton, Paul; Brotchi, Jacques; Kiss, Robert. Department of Neurosurgery, Erasmus University Hospital, Belg. *International Journal of Oncology* (2006), 28(5), 1021-1030. Publisher: International Journal of Oncology, CODEN: IJONES ISSN: 1019-6439. Journal; General Review written in English. CAN 145:327416 AN 2006:464075 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

A review. Cimetidine (CIM), the prototypical histamine H2 receptor antagonist (H2RA), was brought to market based on its ability to accelerate healing of gastrointestinal ulcers through the inhibition of gastric acid secretion. Cimetidine, the most studied H2RA, has been demonstrated to possess anti-tumor activity against colon, gastric and kidney cancers, and melanomas. This activity involves a no. of different mechanisms of action: (a) CIM antagonizes tumor cell-mediated interleukin-1-induced activation of selectins in liver sinusoids, inhibiting tumor cell binding on liver sinusoids, thereby reducing the development of liver metastasis; (b) histamine acts as a growth factor in various tumor cell types via the activation of H2 receptors; CIM therefore may antagonize this effect; (c) CIM acts as an immunomodulator by enhancing the host's immune response to tumor cells. With respect to malignant gliomas, CIM added to temozolomide was superior in vivo when compared to temozolomide alone in extending survival of nude mice with human glioblastoma cells orthotopically xenografted into their brain. We review the various mechanisms of action potentially assocd. with the therapeutic effects of CIM in the case of exptl. glioblastomas, observations we hope will encourage clin. investigation of CIM in the management of highly malignant gliomas.

Answer 13:

**Bibliographic Information**

**Effect of chemotherapy-induced DNA repair on oncolytic Herpes Simplex viral replication.** Aghi, Manish; Rabkin, Samuel; Martuza, Robert L. Department of Neurosurgery, Harvard Medical School, Massachusetts General Hospital, Boston, MA, USA. *Journal of the National Cancer Institute* (2006), 98(1), 38-50. Publisher: Oxford University Press, CODEN: JNCIEQ ISSN: 0027-8874. Journal written in English. CAN 145:39887 AN 2006:25878 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

Gliomas treated with the alkylating agent temozolomide have incomplete responses in part because of tumoral repair of chemotherapy-induced DNA damage. Data from phase I trials suggest that G207, an oncolytic herpes simplex virus (HSV) with mutated ribonucleotide reductase (RR) and  $\gamma$ 34.5 genes, is safe but needs greater viral oncolysis to be effective. We hypothesized that temozolomide and G207 treatment limitations could be jointly addressed using temozolomide-induced tumor-protective DNA repair pathways to enhance viral replication. Human glioblastoma cells (U87, T98, and U373) and U87 cells transfected with the gene for the DNA repair enzyme O6-methylguanine DNA methyltransferase (MGMT) were treated with G207 and/or temozolomide. Drug interactions, expression of the growth arrest DNA damage 34 (GADD34) and RR transcripts before and after their knockdown with short interfering RNAs, DNA strand breaks, and apoptosis were measured using Chou-Talalay anal., real-time reverse transcription-polymerase chain reaction, the comet assay, and flow cytometry, resp. Survival of mice (groups of ten) with intracranial U87 xenograft tumors treated with temozolomide and/or G207 was analyzed using Kaplan-Meier anal. Temozolomide exhibited strong synergy with G207 in both MGMT-neg. and the MGMT inhibitor O6-benzylguanine-treated MGMT-expressing gliomas (Chou-Talalay combination indexes = 0.005 to 0.39) and induced GADD34 expression primarily in nonapoptotic MGMT-neg. U87 glioma cells (fold difference = 16, 95% confidence interval [CI] = 12.6 to 20.4, compared with untreated cells). MGMT-expressing T98 and U87/MGMT cells treated with temozolomide plus O6-benzylguanine had higher RR expression than untreated cells (fold difference = 14.9, 95% CI = 10.1 to 22.0 [T98]; 9.9, 95% CI = 7.0 to 13.8 [U87/MGMT]). GADD34 and RR knockdown increased temozolomide-induced DNA damage and inhibited the synergy of G207 and temozolomide in U87 and O6-benzylguanine-treated U87/MGMT cells.

Mice bearing intracranial U87 tumors survived longer after combination therapy (100% survival at 90 days) than after single-agent therapy (median survival = 46 and 48 days with G207 and temozolomide treatment, resp.). Temozolomide-induced DNA repair pathways vary with MGMT expression and enhance HSV-mediated oncolysis in glioma cells. These findings unveil the potential of HSV to target cells surviving temozolomide treatment.

Answer 14:

**Bibliographic Information**

**O6-(4-bromothienyl)guanine reverses temozolomide resistance in human breast tumour MCF-7 cells and xenografts.** Clemons, M.; Kelly, J.; Watson, A. J.; Howell, A.; McElhinney, R. S.; McMurry, T. B. H.; Margison, G. P. Cancer Research UK Carcinogenesis Group, Paterson Institute for Cancer Research, Manchester, UK. *British Journal of Cancer* (2005), 93(10), 1152-1156. Publisher: Nature Publishing Group, CODEN: BJCAAI ISSN: 0007-0920. Journal written in English. CAN 144:246644 AN 2005:1188736 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

Tumor resistance to chemotherapy involving methylating agents such as DTIC (dacarbazine) and temozolomide is linked to expression of the DNA repair protein O6-alkylguanine-DNA alkyltransferase (MGMT). There is considerable interest in improving the efficacy of such O6-alkylating chemotherapy by the prior inactivation of MGMT. We have examd. the effect of the modified guanine base, O6-(4-bromothienyl)guanine (PaTrin-2, Patrin, Lomeguatrib) on MGMT activity and cell or xenograft tumor growth inhibition by temozolomide in the human breast carcinosarcoma cell line, MCF-7. PaTrin-2 effectively inactivated MGMT in MCF-7 cells (IC<sub>50</sub> .apprx.6 nM) and in xenografts there was complete inactivation of MGMT within 2 h of dosing (20 mg kg<sup>-1</sup> i.p.) and only slight recovery by 24 h. MGMT inactivation in a range of murine host tissues varied between complete and .apprx.60%, with extensive recovery by 24 h. PaTrin-2 (10  $\mu$ M) substantially increased the growth inhibitory effects of temozolomide in MCF-7 cells (D<sub>60</sub>=10  $\mu$ M with PaTrin-2 vs 400  $\mu$ M without). In MCF-7 xenografts, neither temozolomide (100 mg kg<sup>-1</sup> day<sup>-1</sup> for 5 days) nor PaTrin-2 (20 mg kg<sup>-1</sup> day<sup>-1</sup> for 5 days) had any significant effect on tumor growth. In contrast, the PaTrin-2-temozolomide combination produced a substantial tumor growth delay: median tumor quintupling time was increase by 22 days (P<0.005) without any significant increase in toxicity as

assessed from animal wt. A PaTrin-2-temozolomide combination may therefore be beneficial in the treatment of human breast cancers.

Answer 15:

#### Bibliographic Information

**Poly(ADP-ribose) polymerase-1 inhibition reverses temozolomide resistance in a DNA mismatch repair-deficient malignant glioma xenograft.** Cheng, C. Lynn; Johnson, Stewart P.; Keir, Stephen T.; Quinn, Jennifer A.; Ali-Osman, Francis; Szabo, Csaba; Li, Hongshan; Salzman, Andrew L.; Dolan, M. Eileen; Modrich, Paul; Bigner, Darell D.; Friedman, Henry S. Department of Surgery, Duke University Medical Center, Durham, NC, USA. *Molecular Cancer Therapeutics* (2005), 4(9), 1364-1368. Publisher: American Association for Cancer Research, CODEN: MCTOCF ISSN: 1535-7163. Journal written in English. CAN 143:318623 AN 2005:1064573 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

#### Abstract

Temozolomide is a DNA-methylating agent used in the treatment of malignant gliomas. In this study, we have examd. if inhibition of poly(ADP-ribose) polymerase (PARP) could increase the cytotoxicity of temozolomide, particularly in cells deficient in DNA mismatch repair. Athymic mice, transplanted with mismatch repair - proficient [D-245 MG] or deficient [D-245 MG (PR)] xenografts, were treated with a combination of temozolomide and the PARP inhibitor, INO-1001. For the tumors deficient in mismatch repair, the most ED of INO-1001 was found to be 150 mg/kg, given i.p. thrice at 4-h intervals with the first injection in combination with 262.5 mg/kg temozolomide (0.75 LD10). This dose of temozolomide by itself induced no partial regressions and a 4-day growth delay. In two sep. expts., the combination therapy increased the growth delay by 21.6 and 9.7 days with partial regressions obsd. in four of eight and three of nine mice, resp. The addn. of INO-1001 had a more modest, yet statistically significant, increase in tumor growth delay in the mismatch repair - proficient xenografts. In these expts., mice were treated with a lower amt. of temozolomide (88 mg/kg), which resulted in growth delays of 43.1 and 39.2 days. When the temozolomide treatment was in combination with 200 mg/kg INO-1001, there was an increase in growth delay to 48.9 and 45.7 days, resp. These results suggest that inhibition of PARP may increase the efficacy of temozolomide in the treatment of malignant gliomas, particularly in tumors deficient in DNA mismatch repair.

Answer 16:

#### Bibliographic Information

**Combined cimetidine and temozolomide, compared with temozolomide alone: significant increases in survival in nude mice bearing U373 human glioblastoma multiforme orthotopic xenografts.** Lefranc, Florence; James, Cyril; Camby, Isabelle; Gaussin, Jean-Francois; Darro, Francis; Brotchi, Jacques; Gabius, Joachim; Kiss, Robert. Department of Neurosurgery, Erasmus University Hospital, Brussels, Belg. *Journal of Neurosurgery* (2005), 102(4), 706-714. Publisher: American Association of Neurological Surgeons, CODEN: JONSAC ISSN: 0022-3085. Journal written in English. CAN 143:19449 AN 2005:388490 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

#### Abstract

Objective: Malignant gliomas consist of both heterogeneous proliferating and migrating cell subpopulations, with migrating glioma cells exhibiting less sensitivity to antiproliferative or proapoptotic drugs than proliferative cells. Therefore, the authors combined cimetidine, an antiinflammatory agent already proven to act against migrating epithelial cancer cells, with temozolomide to det. whether the combination induces antitumor activities in exptl. orthotopic human gliomas compared with the effects of temozolomide alone. Methods: Cimetidine added to temozolomide compared with temozolomide alone induced survival benefits in nude mice with U373 human glioblastoma multiforme (GBM) cells orthotopically xenografted in the brain. Computer-assisted phase-contrast microscopy analyses of 9L rat and U373 human GBM cells showed that cimetidine significantly decreased the migration levels of these tumor cells in vitro at concns. at which tumor growth levels were not modified (as revealed on monotetrazolium calorimetric assay). Computer-assisted microscope analyses of neoglycoconjugate-based glycohistochem. staining profiles of 9L gliosarcomas grown in vivo revealed that cimetidine significantly decreased expression levels of endogenous receptors for fucose and, to a lesser extent, for

N-acetyl-lactosamine moieties. Endogenous receptors of this specificity are known to play important roles in adhesion and migration processes of brain tumor cells. Conclusions: Cimetidine, acting as an antiadhesive and therefore an antimigratory agent for glioma cells, could be added in complement to the cytotoxic temozolomide compd. to combat both migrating and proliferating cells in GBM.

Answer 17:

#### Bibliographic Information

##### **Distinct Responses of Xenografted Gliomas to Different Alkylating Agents Are Related to Histology and Genetic Alterations.**

Leuraud, Pascal; Taillandier, Luc; Medioni, Jacques; Aguirre-Cruz, Lucinda; Criniere, Emmanuelle; Marie, Yannick; Kujas, Michele; Golmard, Jean-Louis; Duprez, Adrien; Delattre, Jean-Yves; Sanson, Marc; Poupon, Marie-France. Institut National de la Sante et de la Recherche Medicale, Laboratoire de Biologie des Interactions Neurons-Glie, Groupe Hospitalier Pitie-Salpetriere, Paris, Fr. Cancer Research (2004), 64(13), 4648-4653. Publisher: American Association for Cancer Research, CODEN: CNREA8 ISSN: 0008-5472. Journal written in English. CAN 141:116709 AN 2004:537842 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

#### Abstract

A series of 12 human gliomas was established as xenografts in nude mice and used to evaluate the relationship between histol., genetic parameters, and response to alkylating agents. Eight were high-grade oligodendroglial tumors, and four were glioblastoma. They were characterized for their genetic alterations, including those considered as "early" alterations, namely loss of chromosome 1 ± loss of chromosome 19q, TP53 mutation, and those considered as "late" alterations, namely loss of chromosome 10, loss of chromosome 9p, EGFR genomic amplification, PTEN mutation, CDKN2A homozygous deletion, and telomerase reactivation. Chemosensitivity of xenografts to four alkylating agents, temozolomide (42 mg/kg, days 1-5, p.o.), 1,3-bis(2-chloroethyl)-1-nitrosourea (5 mg/kg, day 1, i.p.), Ifosfamide (90 mg/kg, days 1-3, i.p.), and carboplatin (66 mg/kg, day 1, i.p.) was tested by administration of drugs to tumor-bearing mice. Although each tumor presented an individual response pattern, glioblastoma had a lower chemosensitivity than oligodendrogliomas, and temozolomide was the most effective drug. Deletion of 1p ± 19q was assocd. with higher chemosensitivity, whereas late mol. alterations, particularly EGFR amplification, were assocd. with chemoresistance. These results suggest that the combined use of histol. and mol. markers should eventually be helpful selecting the most appropriate agents for treatment of malignant oligodendrogliomas and astrocytomas.

Answer 18:

#### Bibliographic Information

##### **Anticancer Chemosensitization and Radiosensitization by the Novel Poly(ADP-ribose) Polymerase-1 Inhibitor AG14361.**

Calabrese, Christopher R.; Almasy, Robert; Barton, Stephanie; Batey, Michael A.; Calvert, A. Hilary; Canan-Koch, Stacie; Durkacz, Barbara W.; Hostomsky, Zdenek; Kumpf, Robert A.; Kyle, Suzanne; Li, Jianke; Maegley, Karen; Newell, David R.; Notarianni, Elena; Stratford, Ian J.; Skalitzky, Donald; Thomas, Huw D.; Wang, Lan-Zhen; Webber, Stephen E.; Williams, Kaye J.; Curtin, Nicola J. Northern Institute for Cancer Research, Medical School, University of Newcastle upon Tyne, New Castle upon Tyne, UK. Journal of the National Cancer Institute (2004), 96(1), 56-67. Publisher: Oxford University Press, CODEN: JNCIEQ ISSN: 0027-8874. Journal written in English. CAN 141:220937 AN 2004:27281 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

#### Abstract

Background: Poly(ADP-ribose) polymerase-1 (PARP-1) facilitates the repair of DNA strand breaks. Inhibiting PARP-1 increases the cytotoxicity of DNA-damaging chemotherapy and radiation therapy in vitro. Because classical PARP-1 inhibitors have limited clin. utility, we investigated whether AG14361, a novel potent PARP-1 inhibitor (inhibition const. <5 nM), enhances the effects of chemotherapy and radiation therapy in human cancer cell cultures and xenografts. Methods: The effect of AG14361 on the antitumor activity of the DNA alkylating agent temozolomide, topoisomerase I poisons topotecan or irinotecan, or x-irradn. or  $\gamma$ -radiation was investigated in human cancer cell lines A549, LoVo, and SW620 by proliferation and survival assays and in xenografts in mice by tumor vol. detn. The specificity of AG14361 for PARP-1 was investigated by microarray anal. and by antiproliferation and acute toxicity assays in PARP-1<sup>-/-</sup> and PARP-1<sup>+/+</sup> cells and mice. After i.p. administration, the concn. of AG14361 was detd. in mouse

plasma and tissues, and its effect on PARP-1 activity was detd. in tumor homogenates. All statistical tests were two-sided. Results: AG14361 at 0.4  $\mu$ M did not affect cancer cell gene expression or growth, but it did increase the antiproliferative activity of temozolomide (e.g., in LoVo cells by 5.5-fold, 95% confidence interval [CI] = 4.9-fold to 5.9-fold;  $P = .004$ ) and topotecan (e.g., in LoVo cells by 1.6-fold, 95% CI = 1.3-fold to 1.9-fold;  $P = .002$ ) and inhibited recovery from potentially lethal  $\gamma$ -radiation damage in LoVo cells by 73% (95% CI = 48% to 98%). In vivo, nontoxic doses of AG14361 increased the delay of LoVo xenograft growth induced by irinotecan, x-irradn., or temozolomide by two- to threefold. The combination of AG14361 and temozolomide caused complete regression of SW620 xenograft tumors. AG14361 was retained in xenografts in which PARP-1 activity was inhibited by more than 75% for at least 4 h.

Conclusion: AG14361 is, to our knowledge, the first high-potency PARP-1 inhibitor with the specificity and in vivo activity to enhance chemotherapy and radiation therapy of human cancer.

Answer 19:

### Bibliographic Information

**Sensitization of pancreatic tumor xenografts to carmustine and temozolomide by inactivation of their O6-methylguanine-DNA methyltransferase with O6-benzylguanine or O6-benzyl-2'-deoxyguanosine.** Kokkinakis, Demetrius M.; Ahmed, Mansoor M.; Chendil, Damodaran; Moschel, Robert C.; Pegg, Anthony E. Department of Pathology and the Cancer Institute, The University of Pittsburgh, Pittsburgh, PA, USA. *Clinical Cancer Research* (2003), 9(10, Pt. 1), 3801-3807. Publisher: American Association for Cancer Research, CODEN: CCREF4 ISSN: 1078-0432. Journal written in English. CAN 140:210081 AN 2003:847716 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

### Abstract

Adenocarcinoma of the pancreas is refractory to chemotherapeutic agents, including BCNU and streptozotocin. We have previously shown that drugs, which adduct the O6-position of guanine, are ineffective against pancreatic tumor cell lines because of high expression of O6-methylguanine-DNA methyltransferase (MGMT). The effect of MGMT inactivation on the resistance of pancreatic tumors to carmustine (BCNU) and to temozolomide (TMZ) was examd. in five human pancreatic tumor xenografts in athymic mice. Tumor-bearing mice were treated: (a) with a single i.p. injection of BCNU or TMZ at the max.-tolerated doses of 75 and 340 mg/m<sup>2</sup>, resp.; and (b) with O6-benzylguanine (BG) or O6-benzyl-2'-deoxyguanosine (dBG) in combination with BCNU or TMZ. Pretreatment with the MGMT inactivators BG or dBG reduced the max.-tolerated doses of BCNU and TMZ to 35 and 170 mg/m<sup>2</sup>, resp. MIA PaCa-2, CFPAC-1, PANC-1, CAPAN-2, and BxPC-3 having MGMT levels of 890, 1680, 680, 900, and 330 fmol/mg protein, resp., were unresponsive to BCNU. MIA PaCa-2 and CFPAC-1 were also unresponsive to TMZ, whereas CAPAN-2 responded with a tumor delay of 32 days. BG or dBG sensitized all tumors to both BCNU and TMZ. BG plus BCNU treatment of MIA PaCa-2, CFPAC-1, PANC-1, CAPAN-2, and BxPC-3 induced tumor delays of 18, 16, 12, 14, and 16 days, resp. In comparison, dBG plus BCNU at doses that were equitoxic to BCNU plus BG yielded tumor delays of 30, 19, 16, 21, and 22 days, resp. The pancreatic tumors tested displayed functional mismatch repair that, however, may not be always sufficiently restrictive to prevent mutations under alkylation stress. Treatments with either BCNU or TMZ resulted in some degree of mutation in recurring tumors with the exception of CAPAN-2, the only wt-p53 xenograft. DBG, a weak MGMT inactivator in vitro as compared with BG, was markedly more effective than the latter in enhancing the efficacy of BCNU against pancreatic tumor xenografts.

Both BG and dBG also enhanced the efficacy of TMZ against pancreatic tumors, possibly because of the repression of MGMT, which cannot be achieved with TMZ treatments alone. These results suggest that pancreatic tumors, which are resistant to DNA alkylating agents, may be sensitized to such agents when pretreated with MGMT inactivators.

Answer 20:

### Bibliographic Information

**O6-benzylguanine-mediated enhancement of chemotherapy.** Friedman, Henry S.; Keir, Stephen; Pegg, Anthony E.; Houghton, Peter J.; Colvin, O. Michael; Moschel, Robert C.; Bigner, Darell D.; Dolan, M. Eileen. Department of Surgery, Duke University Medical Center, Durham, NC, USA. *Molecular Cancer Therapeutics* (2002), 1(11), 943-948. Publisher: American Association for Cancer Research, CODEN: MCTOCF ISSN: 1535-7163. Journal written in English. CAN 139:30329 AN 2003:69745 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

We have previously demonstrated (A. E. Pegg, *Cancer Res.*, 50: 6119-6129, 1990) that O6-benzylguanine (O6-BG) enhances nitrosourea, temozolomide, and cyclophosphamide activity in malignant glioma xenografts growing in athymic nude mice. More recently, we have demonstrated (V. J. Patel et al., *Clin. Cancer Res.*, 6: 4154-4157, 2000; P. Pourquier et al., *Cancer Res.*, 61: 53-58, 2001) that the combination of temozolomide plus irinotecan (CPT-11) displays a schedule-dependent enhancement of antitumor activity secondary to trapping of topoisomerase I by O6-methylguanine residues in DNA. These studies suggested that there might be favorable therapeutic interactions between O6-BG and combinations of 1,3-bis(2-chloroethyl)-1-nitrosourea (BCNU) plus cyclophosphamide or temozolomide plus CPT-11, resp. Our present results indicate that the combination of cyclophosphamide plus BCNU plus O6-BG produces growth delays modestly-to-markedly-superior to combinations of cyclophosphamide with BCNU. Although the combination of temozolomide and CPT-11 reveals a marked increase in activity compared with either agent used alone, the addn. of O6-BG to this combination dramatically increased the growth delay of the O6-alkylguanine-DNA alkyltransferase (AGT)-pos. malignant glioma D-456 MG. These results suggest that a Phase I trial of CPT-11 plus temozolomide plus O6-BG in AGT-pos. tumors may be an important intervention to maximize the therapeutic benefits of the combination of CPT-11 and temozolomide.

Answer 21:

**Bibliographic Information**

**Multifaceted resistance of gliomas to temozolomide.** Bocangel, Dora B.; Finkelstein, Sydney; Schold, S. Clifford; Bhakat, Kishor K.; Mitra, Sankar; Kokkinakis, Demetrius M. Department of Pathology, The University of Pittsburgh Cancer Institute, Pittsburgh, PA, USA. *Clinical Cancer Research* (2002), 8(8), 2725-2734. Publisher: American Association for Cancer Research, CODEN: CCREF4 ISSN: 1078-0432. Journal written in English. CAN 138:215003 AN 2002:682258 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

The contributions of O6-methylguanine-DNA-methyltransferase (MGMT), p53 status, mismatch repair, and apoptotic response to the resistance of glial tumors to temozolomide (TMZ) were tested using 7 established human glial tumor cell lines in culture and xenografts in athymic mice. Resistance to TMZ was only marginally dependent on MGMT activity, because subtoxic doses of TMZ easily eliminated MGMT reserves for at least 18 h after treatment. Resistance to TMZ varied most notably with the p53 status of the tumor. Tumors with wild-type (wt) p53 and a functional p53 response to DNA damage (SWB40 and SWB61) were most sensitive. The p21-related cell cycle arrest was intimately linked to TMZ toxicity because tumors with wt p53 but lacking a robust increase in p21 protein level (D-54) were resistant to TMZ. In contrast, tumors with a dysfunctional p53 cycle and a weak cell cycle response to DNA damage (SWB39 and SWB77) were extremely unresponsive to treatment even with the aid of MGMT inactivators. Notable exceptions to the above were obsd. with the p53 mutated tumors SWB33 and SWB95, which were arrested by TMZ in G1-S and consequently underwent apoptosis despite their failure to express p21. By testing a limited no. of glial tumors in cell culture and also as xenografts, the authors have shown that mobilization of the p53 in response to TMZ damage is likely to induce a cell cycle arrest and apoptosis in glial tumors. Addnl. pathways linking cell cycle arrest and apoptosis contribute to the efficacy of TMZ against p53 mutated glial tumors. The unusual resistance of tumors, of which the cell cycle was not arrested in response to TMZ treatment, was assocd. with allelic losses during regrowth of treated tumors. Nevertheless such resistance was not related to dysfunctional mismatch repair.

Answer 22:

**Bibliographic Information**

**Synergy between methionine stress and chemotherapy in the treatment of brain tumor xenografts in athymic mice.** Kokkinakis, Demetrius M.; Hoffman, Robert M.; Frenkel, Eugene P.; Wick, Jacquelynn B.; Han, Qinghong; Xu, Mingxu; Tan, Yuying; Schold, S. Clifford. Department of Neurological Surgery, University of Texas Southwestern Medical Center, Dallas, TX, USA. *Cancer Research* (2001), 61(10), 4017-4023. Publisher: American Association for Cancer Research, CODEN: CNREA8 ISSN: 0008-5472. Journal written in English. CAN 135:205104 AN 2001:401936 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

This study describes a novel approach to the treatment of brain tumors with the combination of recombinant L-methionine- $\alpha$ -deamino- $\gamma$ -lyase and chemotherapeutic regimens that are currently used against such tumors. The growth of Daoy, SWB77, and D-54 xenografts in athymic mice was arrested after the depletion of mouse plasma methionine (MET) with a combination of a MET- and choline-free diet and recombinant L-methionine- $\alpha$ -deamino- $\gamma$ -lyase. The treated tumor-bearing mice were rescued from the toxic effects of MET withdrawal with daily i.p. homocysteine. This regimen suppressed plasma MET to levels below 5  $\mu$ M for several days, with no treatment-related deaths. MET depletion for 10-12 days induced mitotic and cell cycle arrest, apoptotic death, and widespread necrosis in tumors but did not prevent tumor regrowth after cessation of the regimen. However, when a single dose of 35 mg/m<sup>2</sup> of N,N'-bis(2-chloroethyl)-N-nitrosourea (BCNU), which was otherwise ineffective as a single therapy in any of the tumors tested, was given at the end of the MET depletion regimen, a more than 80-day growth delay was obsd. for Daoy and D-54, whereas the growth of SWB77 was delayed by 20 days. MET-depleting regimens also trebled the efficacy of temozolomide (TMZ) against SWB77 when TMZ was given to animals as a single dose of 180 mg/m<sup>2</sup> at the end of a 10-day period of MET depletion. The enhanced responses of both Daoy and SWB77 to DNA alkylating agents such as BCNU and TMZ could be attributed to the down-regulation of O6-methylguanine-DNA methyltransferase activity. However, the synergy of MET depletion and BCNU obsd. with D-54 tumors, which do not express measurable O6-methylguanine-DNA methyltransferase protein, is probably mediated by a different mechanism. MET depletion specifically sensitizes tumors to alkylating agents and does not significantly lower the toxicity of either BCNU or TMZ for the host. In this regard, the combination approach of MET depletion and genotoxic chemotherapy demonstrates significant promise for

evaluation.

Answer 23:

**Bibliographic Information**

**Thresholds of O6-alkylguanine-DNA alkyltransferase which confer significant resistance of human glial tumor xenografts to treatment with 1,3-Bis(2-chloroethyl)-1-nitrosourea or temozolomide.** Kokkinakis, Demetrius M.; Bocangel, Dora B.; Schold, S. Clifford; Moschel, Robert C.; Pegg, Anthony E. Department of Neurosurgery, University of Texas Southwestern Medical Center at Dallas, Dallas, TX, USA. Clinical Cancer Research (2001), 7(2), 421-428. Publisher: American Association for Cancer Research, CODEN: CCREFA ISSN: 1078-0432. Journal written in English. CAN 135:174817 AN 2001:196443 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

Bis-2-chloroethylnitrosourea (BCNU) or temozolomide (TMZ) were tested alone or in combination with the AGT inhibitors O6-benzyl-2'-deoxyguanosine (dBG) or O6-benzylguanine (BG) against human glial tumor xenografts growing s.c. in athymic mice. Four glioblastoma (SWB77, SWB40, SWB39, and D-54) and one anaplastic oligodendroglioma (SWB61) xenografts having O6-alkylguanine-DNA alkyltransferase (AGT) activities of 75, 45, 10, <10, and 16 fmol/mg protein, resp., were used. BCNU at 35 mg/M2 was ineffective against these tumors, although 70 mg/M2 (LD10, 75 mg/M2) produced a marked tumor growth delay (T-C) in D54 but had no effect against SWB40 or SWB77. Coadministration of BG or dBG and BCNU necessitated redn. of the BCNU dose to a max. of 30 and 35 mg/M2, resp., because of increased toxicity. Optimized treatment with dBG (250 mg/M2) and BCNU (35 mg/M2) resulted in T-Cs of 30, 29, 11, 16, and 14 days for SWB77, SWB40, SWB39, D-54 and SWB61, resp. These delays were more pronounced than those induced with optimized, isotoxic treatments with BG (180 mg/M2) and BCNU (30 mg/M2). In comparison to BCNU, TMZ was less toxic, with an LD10 of 400 mg/M2. TMZ (300 mg/M2) was more effective than BCNU against SWB77, SWB40, and SWB61, inducing T-Cs of 23, 53, and 56 days, resp. BG and dBG enhanced the toxicity of TMZ in athymic mice by decreasing the LD10 from 400 to 200 mg/M2. TMZ (180 mg/M2) with either BG (180 mg/M2) or dBG (250 mg/M2) resulted in T-Cs of 31 and 49 days in SWB77, resp., as compared with 16 days for TMZ (180 mg/M2) alone. In SWB40, the combination of TMZ with dBG, but not with BG, was significantly more effective than the max. tolerated dose of TMZ (300 mg/M2) alone. The combination of TMZ with AGT inactivators had no benefit, as compared with TMZ alone, against xenografts with marginal AGT activity. In conclusion, at equimolar doses dBG was less toxic than BG in athymic mice when combined with either BCNU or TMZ.

In this regard, BCNU or TMZ can be used at higher doses in combination with dBG than with BG. This study further demonstrates that there is a significant benefit of depleting AGT with nonspecific AGT inhibitors prior to treatment with either BCNU or TMZ in tumors having AGT activity >45 fmol/mg protein.

Answer 24:

#### Bibliographic Information

**Schedule-dependent activity of temozolomide plus CPT-11 against a human central nervous system tumor-derived xenograft.**

Patel, Vikas J.; Elion, Gertrude B.; Houghton, Peter J.; Keir, Stephen; Pegg, Anthony E.; Johnson, Stewart P.; Dolan, M. Eileen; Bigner, Darell D.; Friedman, Henry S. Department of Surgery, Duke University Medical Center, Durham, NC, USA. *Clinical Cancer Research* (2000), 6(10), 4154-4157. Publisher: American Association for Cancer Research, CODEN: CCRE4 ISSN: 1078-0432. Journal written in English. CAN 134:320530 AN 2000:808805 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

#### Abstract

This work evaluated the activity of temozolomide plus CPT-11 (a camptothecin deriv.) against a malignant glioma-derived xenograft, D-54 MG, growing s.c. in athymic nude mice. The initial schedule of i.p. drug administration was temozolomide at 0.1 LD10 on day 1 and CPT-11 at 0.1 LD10 on days 1-5 and 8-14. The combination of these two agents produced greater than additive activity against D-54 MG. This enhanced activity was maintained when the initial administration of CPT-11 was delayed to day 3 or day 5. However, when CPT-11 was administered first, on day 1 at 0.5 LD10 (for the single dose schedule) followed by temozolomide (0.1 LD10) 5 h, 3 days, or 5 days later, the enhancement of activity was substantially reduced. Thus, the combination of temozolomide plus CPT-11 displays a schedule-dependent enhancement of antitumor activity, suggesting a mechanistic explanation for the enhanced activity, and providing the rationale for a Phase I trial of this regimen.

Answer 25:

#### Bibliographic Information

**O6-Benzylguanine enhances the sensitivity of a glioma xenograft with low O6-alkylguanine-DNA alkyltransferase activity to temozolomide and BCNU.**

Wedge, S. R.; Newlands, E. S. Department Medical Oncology, Charing Cross Hospital, London, UK. *British Journal of Cancer* (1996), 73(9), 1049-1052. Publisher: Stockton, CODEN: BJCAAI ISSN: 0007-0920. Journal written in English. CAN 125:25644 AN 1996:330180 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

#### Abstract

The effect of the O6-alkylguanine-DNA alkyltransferase (AGT) inhibitor O6-benzylguanine (O6-BG) on the antitumor activity of temozolomide or BCNU was evaluated in athymic mice bearing s.c. human glioma (U87MG) xenografts. The activity of AGT in U87MG xenografts was 4.3 fmol/mg protein. These xenografts were inherently sensitive to treatment with the alkylating compds. alone, with nontoxic doses of temozolomide (35 mg/kg) or BCNU (10 mg/kg) producing tumor growth delays of 23.3 and 11.8 days, resp. O6-BG (40 mg/kg) did not inhibit tumor growth when administered alone, but it enhanced the antitumor activity of temozolomide or BCNU when administered 1 h before therapy. AGT activity measured 24 h after the administration of 40 mg O6-BG/kg was only 0.9 fmol/mg protein. These results are in contrast to previous studies in vitro with tumor cell lines of low AGT activity (<15 fmol/mg protein), in which the cytotoxicity of temozolomide or BCNU was unaffected by AGT depletion.

Answer 26:

#### Bibliographic Information

**Activity of temozolomide in the treatment of central nervous system tumor xenografts.**

Friedman, Henry S.; Dolan, M. Eileen; Pegg, Anthony E.; Marcelli, Susan; Keir, Stephen; Catino, Joseph J.; Bigner, Darell D.; Schold, S. Clifford, Jr. Dep. of Pediatrics and Pathology and the Preuss Lab. for Brain Tumor Res., Duke Univ. Medical Center, Durham, NC, USA. *Cancer Research* (1995), 55(13), 2853-7. Publisher: American Association for Cancer Research, CODEN: CNREA8 ISSN: 0008-5472. Journal written in English. CAN 123:74364 AN 1995:658721 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

The activity of 8-carbamoyl-3-methylimidazo(5,1-d-1,2,3,5-tetrazine-4,3H)-one (temozolomide) in the treatment of a panel of xenografts derived from ependymoma, medulloblastoma, and childhood and adult high-grade glioma was evaluated in athymic nude mice bearing s.c. and intracranial tumors. Temozolomide administered daily for a total of five doses demonstrated marked activity against a panel of Mer+ xenografts despite marginal to moderate activity of 1,3-bis(2-chloroethyl)-1-nitrosourea. The growth delays produced by temozolomide in these xenografts were 1.8-7.5-fold greater than those produced by procarbazine. Although temozolomide demonstrated marginal activity against the Mer+ cell line D341 Med when a 5-day schedule was used, a high-dose 1-day schedule resulted in moderate activity. Temozolomide produced increases in median survival of 1285% (adult glioma D-54 MG), 323% (childhood glioma D-456 MZG), and 68% (ependymoma D612 EP). Pretreatment of mice with P-benzylguanine increased temozolomide-induced mortality, requiring redn. of the dosage from 1200 to 750 mg/m<sup>2</sup> on the single-day regimen. O6-Benzylguanine pretreatment of mice bearing Mer+ D341 Med increased the growth delay of temozolomide, in duplicate expts., from -3.1 to 4.8 and 1.1 to 4.9 days. These studies suggest that temozolomide may be active in the treatment of a broad spectrum of central nervous system cancers, including Mer+ tumors resistant to 1,3-bis(2-chloroethyl)-1-nitrosourea.

Answer 27:

**Bibliographic Information**

**Preclinical antitumor activity of temozolomide in mice: efficacy against human brain tumor xenografts and synergism with 1,3-bis(2-chloroethyl)-1-nitrosourea.** Plowman, Jacqueline; Waud, William R.; Koutsoukos, Antonis D.; Rubinstein, Lawrence V.; Moore, Timothy D.; Grever, Michael R. Developmental Therapeutics Program, National Cancer Institute, Bethesda, MD, USA. Cancer Research (1994), 54(14), 3793-9. CODEN: CNREA8 ISSN: 0008-5472. Journal written in English. CAN 121:99274 AN 1994:499274 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

**Abstract**

Temozolomide, a methylating agent with clin. activity against brain tumors, demonstrated excellent antitumor activity following p.o. administration to athymic mice bearing human brain tumor xenografts. In the early stage s.c. implanted SNB-75 astrocytoma model, a 400-mg/kg dose administered on Day 5 produced 10 of 10 Day 54 tumor-free mice. In later staged s.c. U251 and SF-295 glioblastoma models, a single 600-mg/kg dose produced 9 of 10 Day 86 and 2 of 10 Day 40 tumor-free mice, resp. In the latter group, a tumor growth delay of >315% was attained. Similar levels of activity were attained with equal total doses on schedules of daily for 5 doses and every fourth day for 3 doses. A single 40-mg/kg i.v. dose of 1,3-bis(2-chloroethyl)-1-nitrosourea (BCNU) also demonstrated excellent activity, producing 9 of 10 tumor-free mice in the SNB-75 model and growth delays of 283 and 301% in the U251 and SF-295 models, resp. Temozolomide was also highly effective against intracerebral implants of U251 and SF-295 glioblastomas. Administration of either 600 mg/kg on Day 1 or 200 mg/kg on Days 1, 5, and 9 produced 7 of 9 Day 90 tumor-free mice in the U251 model. In the SF-295 model, a single 400-mg/kg dose or three 200-mg/kg doses produced 3 and 4 of 10 Day 90 tumor-free mice, resp., and prolonged survival by 127%. A single 40-mg/kg i.v. dose of BCNU was more effective in the intracerebral U251 model. The synergistic potential of temozolomide and BCNU in combination was evaluated in an advanced stage s.c. implanted SF-295 model. When temozolomide was administered 2 h after BCNU on a single treatment day, a dramatic synergistic therapeutic effect was obsd. in two expts. For example, single agent doses of temozolomide (600 mg/kg) and BCNU (60 mg/kg) and a combination (400 mg/kg + 27 mg/kg) demonstrating equiv. toxicity produced growth delays of 190, 258, and >492% (includes 5 of 10 Day 51 tumor-free mice), resp. Anal.

of the data by a quadratic dose response model indicated synergism with significance at  $P = 0.0001$  in both expts. Synergism also was demonstrated by the isobole method. The reverse sequence was more toxic, but at lower combination doses a synergistic effect was still obsd. ( $P = 0.0001$ ). Using the quadratic model, no confirmed modulation of the antitumor activity of temozolomide was demonstrated by i.p. administration of either 10 or 30 mg/kg O6-benzylguanine 1 h before or 1 h after temozolomide. These data provide a rationale for the clin. evaluation of temozolomide and BCNU combinations in patients with brain tumors.

Answer 28:

**Bibliographic Information**

**Enhancement of glioblastoma cell killing by combination treatment with temozolomide and tamoxifen or hypericin.** Gupta Vinay; Su Yuzhuang S; Wang Weijun; Kardosh Adel; Liebes Leonard F; Hofman Florence M; Schonthal Axel H; Chen Thomas C Department of Pathology, K. Norris Jr. Comprehensive Cancer Center, University of Southern California, Los Angeles, California 90089, USA Neurosurgical focus (2006), 20(4), E20. Journal code: 100896471. E-ISSN:1092-0684. Journal; Article; (JOURNAL ARTICLE); (RESEARCH SUPPORT, NON-U.S. GOV'T) written in English. PubMed ID 16709026 AN 2006281365 MEDLINE (Copyright (C) 2008 U.S. National Library of Medicine on SciFinder (R))

#### Abstract

OBJECT: The chemotherapeutic agent temozolomide has demonstrated antitumor activity in patients with recurrent malignant glioma. Because responses are not enduring and recurrence is nearly universal, further improvements are urgently needed. METHODS: In an effort to increase the clinical activity of temozolomide, the authors investigated whether its antitumor activity could be enhanced by adding tamoxifen or hypericin, two drugs that are known to inhibit the activity of protein kinase C. Human glioblastoma multiforme cell lines A172 and LA567 were treated with combinations of temozolomide and tamoxifen or hypericin in vitro, and cell survival was analyzed using various methods. Tamoxifen and hypericin were able to greatly increase the growth-inhibitory and apoptosis-stimulatory potency of temozolomide via the downregulation of critical cell cycle-regulatory and prosurvival components. Furthermore, with the use of an in vivo xenograft mouse model, the authors demonstrated that hypericin was able to enhance the antiglioma effects of temozolomide in the in vivo setting as well. CONCLUSIONS: Taken together, analysis of the results indicated that combination therapy involving temozolomide and tamoxifen or hypericin potently inhibited tumor growth by inducing apoptosis and provided an effective means of treating malignant glioma.

Answer 29:

#### Bibliographic Information

**Effect of O6-(4-bromothienyl)guanine on different temozolomide schedules in a human melanoma xenograft model.** Middleton Mark R; Thatcher Nicholas; McMurry T Brian H; McElhinney R Stanley; Donnelly Dorothy J; Margison Geoffrey P Cancer Research UK, Department of Medical Oncology, Christie Hospital NHS Trust, Manchester, United Kingdom International journal of cancer. Journal international du cancer (2002), 100(5), 615-7. Journal code: 0042124. ISSN:0020-7136. Journal; Article; (JOURNAL ARTICLE) written in English. PubMed ID 12124813 AN 2002378421 MEDLINE (Copyright (C) 2008 U.S. National Library of Medicine on SciFinder (R))

#### Abstract

The DNA repair protein O(6)-alkylguanine DNA alkyltransferase (ATase) is a major component of resistance to treatment with methylating agents and nitrosoureas. Inactivation of the protein, via the administration of pseudosubstrates, prior to chemotherapy has been shown to improve the latter's therapeutic index in animal models of human tumours. We have also shown that rational scheduling of temozolomide, so that drug is administered at the ATase nadir after the preceding dose, increases tumour growth delay in these models. We now report the results of combining these two approaches. Nude mice bearing A375M human melanoma xenografts were treated with vehicle or 100 mg/kg temozolomide ip for 5 doses spaced 4, 12 or 24 hr apart. Each dose was preceded by the injection of vehicle or 20 mg/kg 4BTG. All treatments resulted in significant delays in tumour quintupling time compared with controls: by 6.2, 5.9 and 16.8 days, respectively, for 24-, 12- and 4-hourly temozolomide alone and by 22.3, 21.3 and 22.1 days, respectively, in combination with 4BTG. Weight loss due to TMZ was unaffected by the presence of 4BTG. This was of the order of 6.2-10.6% with 24- and 12-hourly administration and 17.4-20.1% ( $p < 0.0001$ ) with 4-hourly treatment. In our model, combining daily temozolomide with 4-BTG confers increased antitumour activity equivalent to that achieved by compressing the temozolomide schedule but with less toxicity. Using temozolomide schedule compression with 4-BTG does not improve on this result, suggesting that ATase inactivation with pseudosubstrates is a more promising means of enhancing the activity of temozolomide than compressed scheduling. Copyright 2002 Wiley-Liss, Inc.

Answer 30:

**Bibliographic Information**

**Effect of single and multiple administration of an O6-benzylguanine/temozolomide combination: an evaluation in a human melanoma xenograft model.** Wedge S R; Porteous J K; Newlands E S Department of Medical Oncology, Charing Cross Hospital, London, UK Cancer chemotherapy and pharmacology (1997), 40(3), 266-72. Journal code: 7806519. ISSN:0344-5704. Journal; Article; (JOURNAL ARTICLE); (RESEARCH SUPPORT, NON-U.S. GOV'T) written in English. PubMed ID 9219512 AN 97363187 MEDLINE (Copyright (C) 2008 U.S. National Library of Medicine on SciFinder (R))

**Abstract**

The purpose of the present study was to examine the effect of O6-benzylguanine (O6-BG) on the antitumour activity and toxicity of 8-carbamoyl-3-methylimidazo [5, 1-d]-1,2,3,5-tetrazine-4(3H)-one (temozolomide) in a human malignant melanoma xenograft model following single and multiple administration of the combination. O6-BG irreversibly inactivates the DNA-repair protein O6-alkylguanine-DNA alkyltransferase (AGT), which confers resistance to temozolomide. Preadministration of O6-BG (35 mg/kg, i.p.) 1 h prior to temozolomide (i.p.) was examined using single and daily x5 dosing regimens in athymic mice bearing subcutaneous A375P xenografts. The AGT activity of A375P tumors was 95 +/- 8 fmol/mg protein (mean +/- SE, n = 4). O6-BG alone completely suppressed xenograft AGT activity within 1 h of administration but had no effect upon tumor growth. O6-BG did not significantly increase the tumor growth delay induced by a single 200-mg/ kg dose of temozolomide ( $P > 0.05$ , two-tailed Mann-Whitney test) but did increase the associated mean body weight loss ( $P < 0.025$ ). In contrast, when the same dose of temozolomide was divided into five equal fractions (40 mg/kg) and given with O6-BG on 5 consecutive days, a comparable increase in toxicity was accompanied by a very significant increase in tumor growth delay ( $P < 0.0025$ ), equivalent to that produced by a 3-fold greater dose of temozolomide alone. O6-BG with temozolomide also produced a greater antitumour effect than an equitoxic dose of temozolomide alone on this schedule ( $P < 0.005$ ). These data indicate that the enhancement of temozolomide antitumour activity by O6-BG preadministration is dependent upon the schedule of drug administration, with multiple dosing of O6-BG + temozolomide producing the greatest effect. The results also suggest that prolonged administration of the combination can lead to an increase in the therapeutic index of temozolomide.