EGFRvIII

- The EGFR pathway is frequently abnormally activated in human cancers, including glioblastoma. Activation of EGFR can induce cell proliferation, angiogenesis, invasion/metastasis, and the inhibition of apoptosis (Woodburn 1999; Baselga 2002).

- Many variants and mutations of EGFR have been reported in human glioblastoma (Frederick, Wang et al. 2000). The most frequent mutation in glioblastoma is an in-frame deletion of exons 2-7 that affects 801 base pairs at positions 275-1075, which is termed EGFRvIII (Libermann, Nusbaum et al. 1985; Bigner, Humphrey et al. 1990; Humphrey, Wong et al. 1990; Sugawa, Ekstrand et al. 1990).

- This EGFRvIII specific epitope is in the extracellular domain of the molecule where it is accessible to antibodies and is highly immunogenic. EGFRvIII is reported to be present with variable expression patterns in 25-30% of glioblastoma (Humphrey, Wong et al. 1990).

Protocol CDX110-04 (ACT IV Study)

Studio internazionale randomizzato, doppio cieco, controllato di Rindopepimut/GM-CSF con TMZ adiuvante in pazienti con glioblastoma di nuova diagnosi, trattati chirurgicamente, **EGFRvIII positivi**

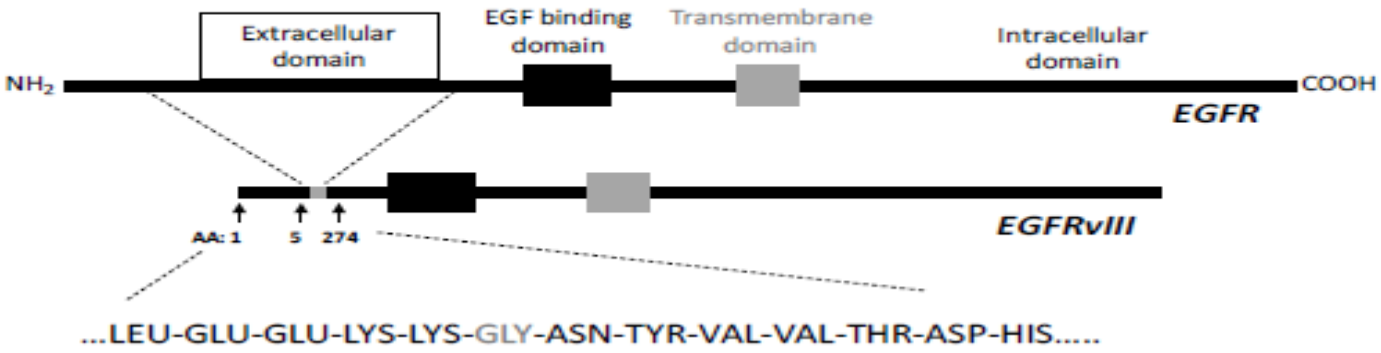
Numero di pazienti: 440

Numero di centri arruolatori: 200

Arruolamento: chiuso circa un anno fa

Rindopepimut

Figure 7. EGFRvIII Mutation and the Unique Extracellular Amino Acid Sequence Used in Rindopepimut Vaccine



- Rindopepimut consists of a 13-amino-acid peptide spanning the unique sequence created by EGFRvIII (Figure 7), covalently linked to the carrier protein Keyhole Limpet Hemocyanin (KLH), which acts as an immune stimulant to enhance immune responses against the EGFRvIII peptide

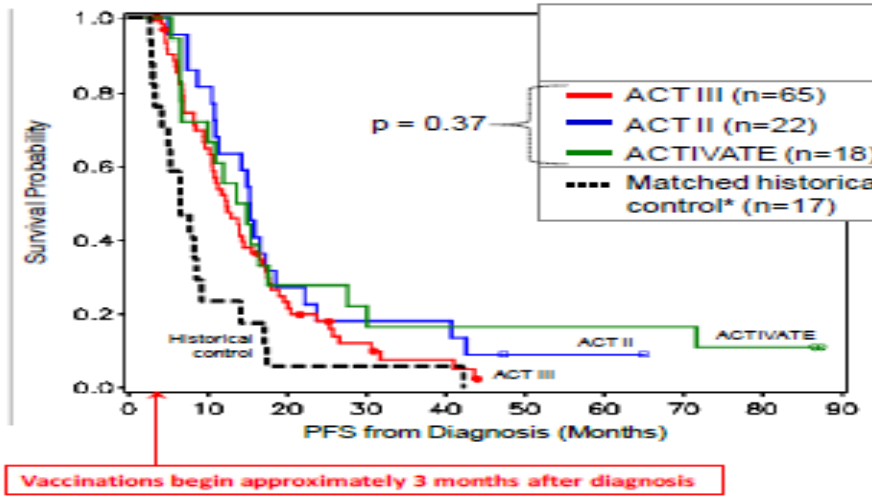
- Rindopepimut consists of a KLH subunit with ≥ 30 EGFRvIII peptides conjugated via the bifunctional linker sulfo-SMCC, resulting in an average molecular weight of 450-550 kDa for the monomer; however, the predominant form is a dimer

Rindopepimut: non-clinical Data

1) inducing a systemic immune response was effective against intracranial tumors expressing EGFRvIII
2) both humoral and cellular anti-EGFRvIII immune responses can participate in the anti-tumor effects
3) GM-CSF can effectively adjuvant the KLH-EGFRvIII vaccines

Rindopepimut: clinical Experience

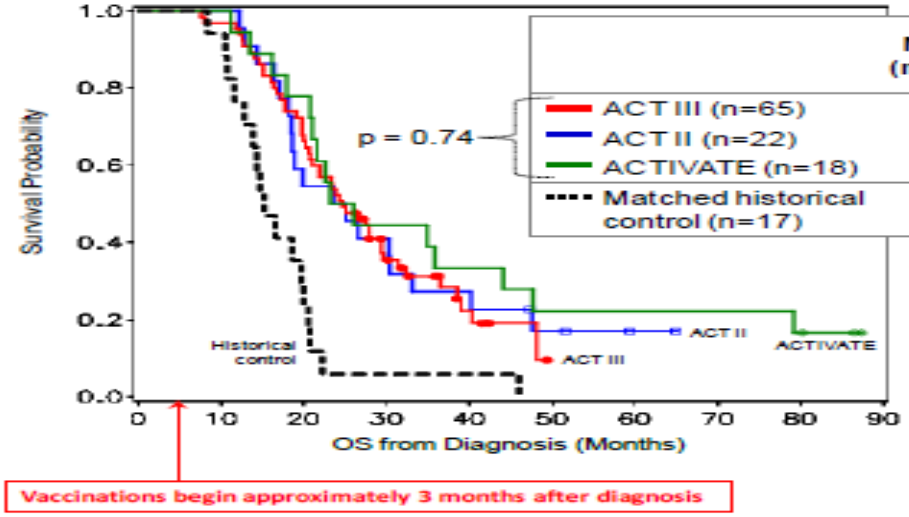
Figure 9. PFS from Diagnosis: ACTIVATE, ACT II and ACT III



	Median (months)	Comparison to Historical Control
ACT III (n=65)	12.3	p = 0.0088
ACT II (n=22)	15.3	p = 0.0029
ACTIVATE (n=18)	14.2	p = 0.0112
Matched historical control* (n=17)	6.4	

* Historical controls were treated at M.D. Anderson contemporaneously to the ACTIVATE trial and matched for eligibility (EGFRvIII-positive, gross-total resection, radiation/TMZ, no progression through ~3 months post-diagnosis)

Figure 10. OS from Diagnosis: ACTIVATE, ACT II and ACT III



	Median (months)	OS at 24 Months	Comparison to Historical Control
ACT III (n=65)	24.6	52%	p = <0.0001
ACT II (n=22)	24.4	50%	p = 0.0023
ACTIVATE (n=18)	24.6	50%	p = 0.0003
Matched historical control (n=17)	15.2	6%	

* Historical controls were treated at M.D. Anderson contemporaneously to the ACTIVATE trial and matched for eligibility (EGFRvIII-positive, gross-total resection, radiation/TMZ, no progression through ~3 months post-diagnosis)

Median duration of follow-up:
ACT III: 38.5 months
ACT II: 59.4 months
ACTIVATE: 86.5 months

Inclusion criteria:

1) Age > 18 years
2) Tumor tissue specimens (paraffin-embedded) from surgical resection must be available for central pathology review, MGMT status determination and analysis of EGFRvIII status.
3) Histologically confirmed, newly diagnosed, de novo glioma grade IV according to WHO 2007
4) Attempted surgical resection followed by conventional chemoradiation, consisting of radiotherapy at a minimally acceptable total dose of at least 90% of the planned radiation therapy dose (usually 60 Gy) and concomitant TMZ chemotherapy (75 mg/m2 body surface area per day).
5) Radiographic imaging from the post-operative period (ideally obtained within 72 hours of surgery, but acceptable if obtained up to the initiation of chemoradiation) and post-chemoradiation period (within 14 days of completion of chemoradiation) available for submission to the independent review committee.
6) Candidate for, and agrees to receive, adjuvant (maintenance) temozolomide therapy.
7) Systemic corticosteroid therapy at ≤2 mg of dexamethasone or equivalent per day for at least 3 days prior to randomization

7) WHO-ECOG Performance Status (Appendix 3) ≤ 2 throughout the week prior to randomization.

Figure 1. Pre-Study Treatment and Imaging Requirements

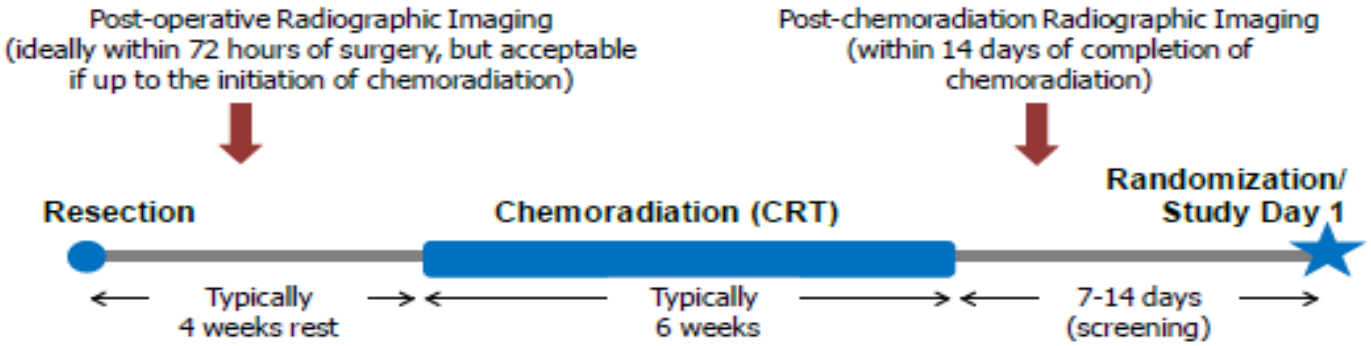
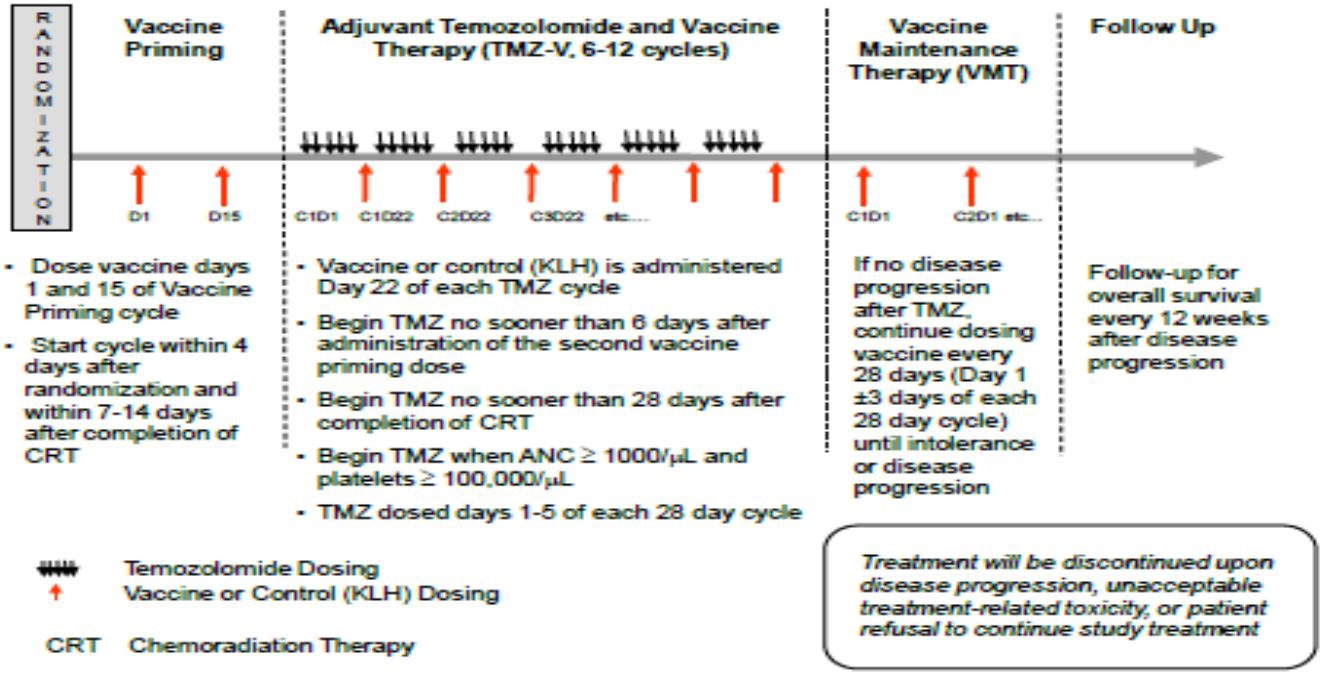


Figure 2. Trial Treatment Schema



Adverse effects:

- The most frequently reported treatment-related adverse events have been mild/moderate injection site reactions which generally resolved without the need for intervention

Table 3. Severity Assessment of Injection Site Reactions

Adverse Event	Grade 1	Grade 2	Grade 3	Grade 4	Grade 5
Injection Site Reaction	Tenderness with or without associated symptoms (e.g., warmth, erythema, itching)	Pain; lipodystrophy; edema; phlebitis	Ulceration or necrosis; severe tissue damage; operative intervention indicated	Life-threatening consequences; urgent intervention indicated	Death
Acneiform or Papulopustular Rash	Papules and/or pustules covering <10% BSA, which may or may not be associated with symptoms of pruritus or tenderness	Papules and/or pustules covering 10-30% BSA, which may or may not be associated with symptoms of pruritus or tenderness; associated with psychosocial impact; limiting instrumental ADL	Papules and/or pustules covering >30% BSA, which may or may not be associated with symptoms of pruritus or tenderness; limiting self care ADL; associated with local superinfection with oral antibiotics indicated	Papules and/or pustules covering any % BSA, which may or may not be associated with symptoms of pruritus or tenderness and are associated with extensive superinfection with IV antibiotics indicated; life-threatening consequences	Death
Maculo-papular rash	Macules/papules covering <10% BSA with or without symptoms (e.g., pruritus, burning, tightness)	Macules/papules covering 10-30% BSA with or without symptoms (e.g., pruritus, burning, tightness); limiting instrumental ADL	Macules/papules covering >30% BSA with or without associated symptoms; limiting self care ADL		
Pustular rash		Localized; local intervention indicated (e.g., topical antibiotic, antifungal, or antiviral)	IV antibiotic, antifungal, or antiviral intervention indicated; radiologic or operative		
Pain	Mild pain	Moderate pain; limiting instrumental ADL	Severe pain; limiting self care ADL		
Induration	Mild induration, able to move skin parallel to plane (sliding) and perpendicular to skin (pinching up)	Moderate induration, able to slide skin, unable to pinch skin; limiting instrumental ADL	Severe induration, unable to slide or pinch skin; limiting joint movement or orifice (e.g., mouth, anus); limiting self care ADL	Generalized; associated with signs or symptoms of impaired breathing or feeding	Death

BSA = Body Surface Area

ADL = Activities of daily living.

- Fatigue (26%), rash (17%), nausea (12%), pruritus (9%) and headache (8%).

Protocol PUMA-NER-5201

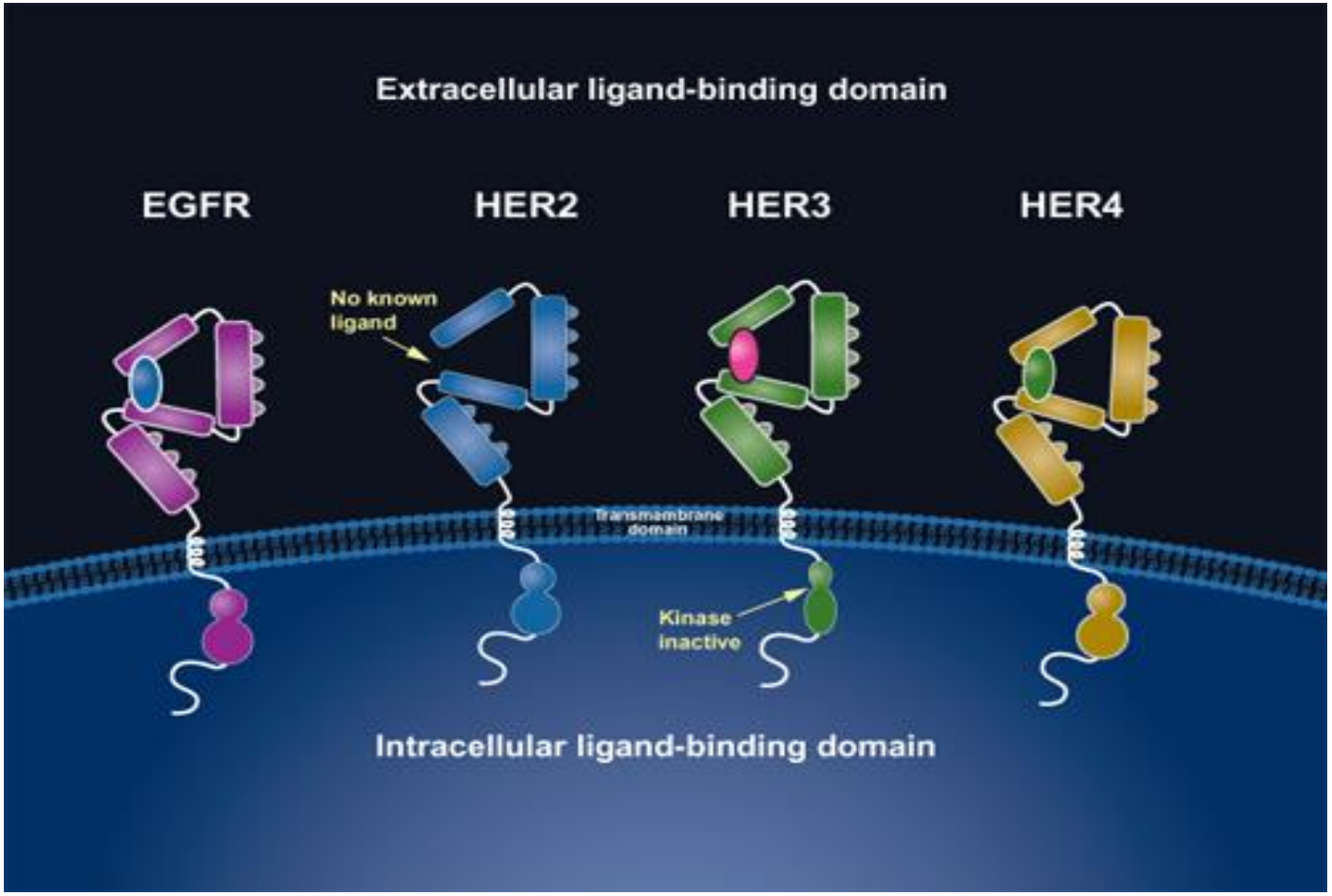
Studio internazionale multicentrico aperto di fase II con neratinib in pazienti con tumore solido con mutazione e/o amplificazione di Human Epidermal Growth Factor Receptor (EGFR, HER2, HER3)

Numero di pazienti: 35-180 (attualmente totale:100; 24 gliomi)
Torino unico centro in Europa

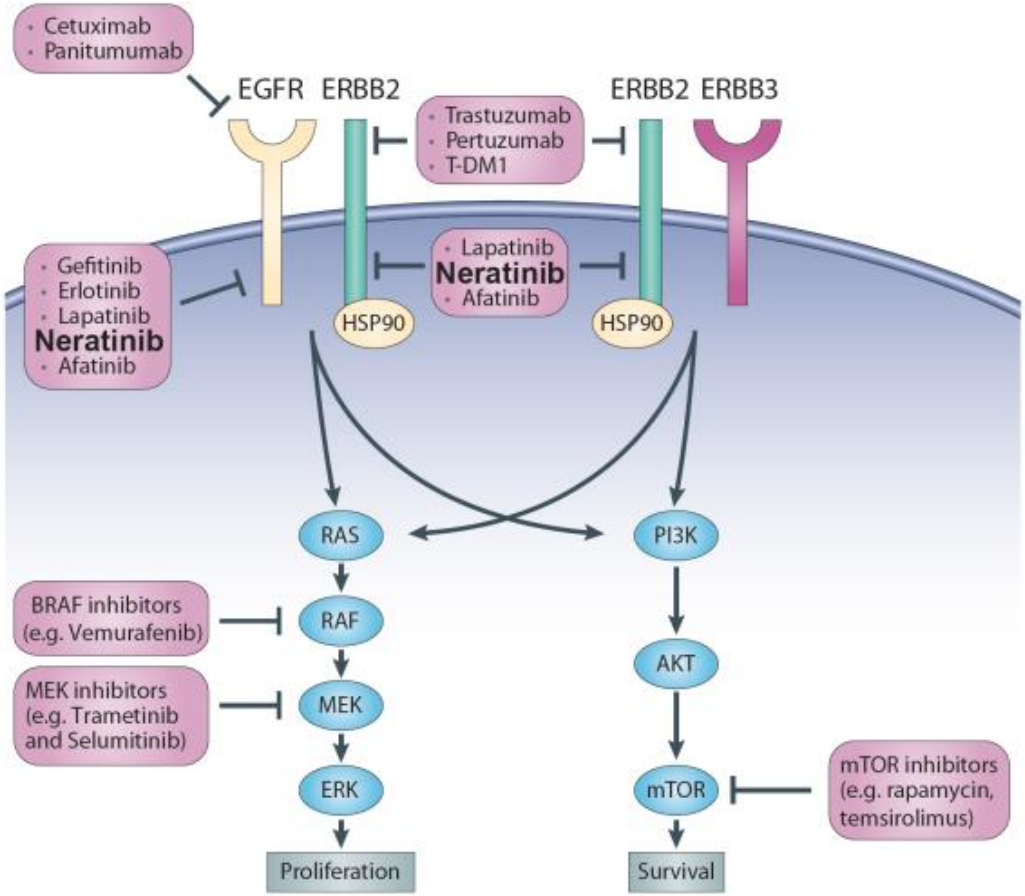
Numero di centri arruolatori: > 20

Arruolamento: in corso

EGFR/ERBB receptor family



Neratinib Inhibits ERBB-Dependent Signaling



Adapted from Yarden and Pines, Nature Reviews Cancer 2012

- ERBB family of receptors are often overexpressed or mutated in many types of cancer including lung, breast, head and neck, and colorectal cancers, including in gliomas.
- More than 90% of all solid tumors express at least one receptor from the ERBB family1
- Over-activation of the ERBB receptor tyrosine kinases leads to uncontrolled cell proliferation, inhibits apoptosis (programmed cell death) and promotes tumor growth and spread

Hynes, Nature Reviews Cancer 2005
Ami Citri , Nature Reviews Molecular Cell Biology

EGFR Mutations in Primary Brain Tumors

- ~40% tumors EGFR amplification (1)
- ~50% express active EGFRvIII mutation (2,3,4)
- Missense or point mutations noted in 14% GBM (5)
- Mutations are sensitive to EGFR kinase inhibitors (5)

1. Libermann, Nature 1985
2. Yamazaki, Mol Cell Biol 1988
3. Wong, Proc Natl Acad Sci USA 1992
4. Ekstrand, Proc Natl Acad Sci USA 1992
5. Lee, PLOS Medicine 2006

Treatment Cohorts

- The study will include 9 cohorts

1) HER2 mutant

- Cohort 1a: Bladder/urinary tract cancer
- Cohort 1b: Colorectal cancer
- Cohort 1c: Endometrial cancer
- Cohort 1d: Gastric/Esophageal cancer
- Cohort 1e: Ovarian cancer
- Cohort 1f: Breast
- Cohort 1z: Solid tumors NOS

2) EGFR mutant or amplified

- Cohort 2a: Primary brain tumors (GBM, gliosarcoma, glioma)

3) HER3 mutant

- Cohort 3z: Solid tumors NOS

Study Objectives 1

Primary Objective

Objective Response Rate at 8 weeks

Secondary Objectives

To determine the best confirmed overall response rate (ORR)

To determine the clinical benefit rate (CBR), defined as the percentage of patients with complete response (CR) + partial response (PR) + stable disease (SD) ≥ 16 weeks

To determine progression-free survival (PFS).

To determine the duration of response (DOR)

To determine overall survival (OS) for each cohort

To assess the safety profile and tolerability of neratinib therapy in patients with HER2/HER3 mutation-positive solid tumors and EGFR-amplified/mutant primary brain tumors

Study Objectives 2

Exploratory Objectives

- To collect and retrospectively evaluate ERBB (EGFR, HER2, HER3) or EGFR amplification/mutation status in pre-treatment archival tissue at a central laboratory
- To explore genetic modifiers of sensitivity to neratinib in solid tumors with mutations in ERBB genes using next-generation sequencing in pre-treatment archival tumor specimens and paired normal whole blood
- To utilize circulating tumor DNA (ctDNA) from plasma specimens collected during the course of treatment to explore mechanisms of primary and acquired resistance to neratinib-containing therapy.

Key Inclusion Criteria

- >18; male or female
- Histologically confirmed cancer for which no curative therapy exists
- Documented EGFR, HER2 or HER3 mutation, or EGFR mutant/amplified primary brain tumor
- At least one measurable or evaluable lesion by RECIST v1.1
- If not measurable by RECIST, disease must be evaluated using another accepted response criteria (eg, RANO, GCIG CA125 Response Criteria, PCWG2 Criteria, mPERCIST)
- LVEF >50% by MUGA or ECHO
- ECOG status of 0, 1 or 2

Adverse effects

System Organ Class MedDRA Preferred Term	All Grades n	 (%)	Grade ≥3 n	 (%)
Any Adverse Event	**507**	**(99.0)**	**281**	**(54.9)**
Gastrointestinal disorders	**482**	**(94.1)**	**157**	**(30.7)**
Diarrhoea	456	(89.1)	135	(26.4)
Nausea	245	(47.9)	18	(3.5)
Vomiting	179	(35.0)	24	(4.7)
Abdominal pain	117	(22.9)	7	(1.4)
Constipation	52	(10.2)	1	(0.2)
General disorders and administration site conditions	**326**	**(63.7)**	**46**	**(9.0)**
Fatigue	184	(35.9)	21	(4.1)
Asthenia	80	(15.6)	8	(1.6)
Pyrexia	60	(11.7)	1	(0.2)
Metabolism and nutrition disorders	**238**	**(46.5)**	**47**	**(9.2)**
Decreased appetite	164	(32.0)	17	(3.3)
Dehydration	57	(11.1)	19	(3.7)
Skin and subcutaneous tissue disorders	**236**	**(46.1)**	**5**	**(1.0)**
Rash	101	(19.7)	1	(0.2)

Protocol BMS-CA209-143

Studio internazionale multicentrico aperto di fase III con nivolumab versus bevacizumab in pazienti alla prima recidiva di GBM dopo trattamento standard

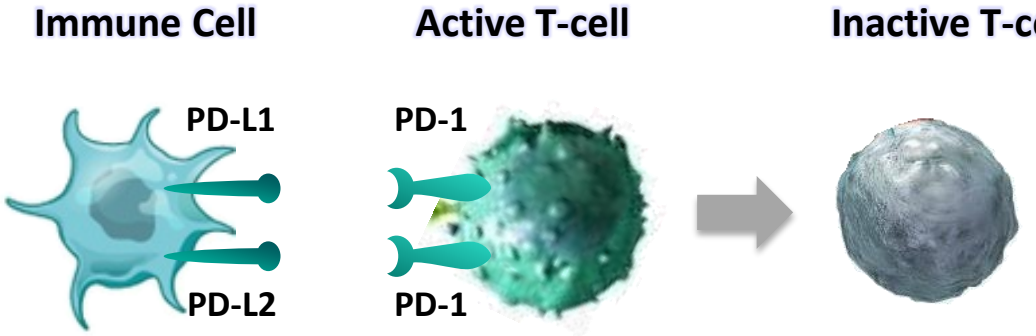
Numero di pazienti: 395

Numero di centri arruolatori: 65

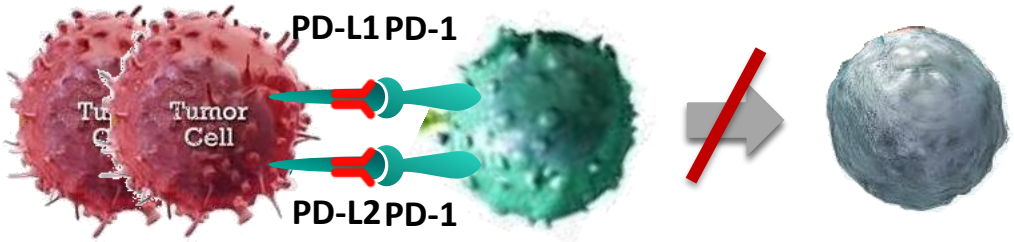
Arruolamento: chiuso ad aprile 2015

Programmed Death Pathway and Nivolumab

Normal Homeostatic Mechanism



Tumor Microenvironment



OPDIVO (nivolumab)

- Normal function of PD-1 pathway is to attenuate immune response to avoid immune system attack of "self"
- A "brake" to prevent overreaction & overproliferation

- Tumor cells "co-opt" the PD-1 pathway to evade T-cell immune responses

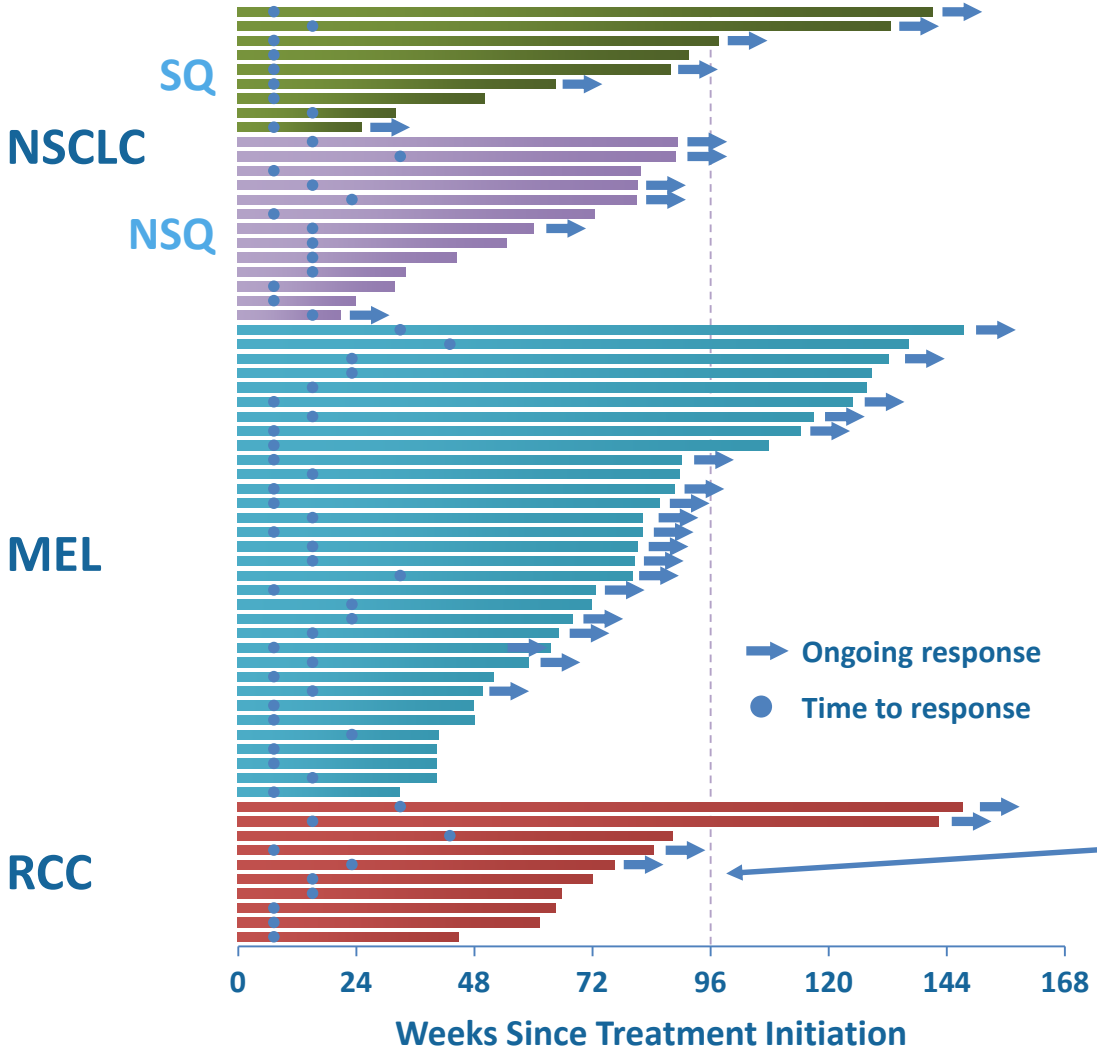
- Nivolumab occupies the PD-1 receptor of T-cells → prevents inhibitory ligand binding & T-cell inactivation

Clinical Experience with Nivolumab (CA209-003) Nivolumab single agent (Solid Tumors)

- Approximately 1,500 patients have been exposed to Nivolumab (IB Version 12.0; 21 July 2013
- Phase 1 study CA209003 provides largest experience with nivolumab monotherapy to date
- 03-Jul-2012 Database Lock:
  - 304 subjects received treatment (evaluable for safety)
  - 294 subjects began treatment by 01-Jul-2011 (evaluable for response)

Tumor Type	**Safety Population (N = 304)**	**Efficacy Population (N = 294)**
Melanoma	107 (35%)	106 (36%)
Nonsquamous NSCLC	73 (24%)	73 (25%)
Squamous NSCLC	53 (17%)	48 (16%)
RCC	34 (11%)	34 (12%)
Colorectal Cancer	19 (6%)	19 (6%)
CRPC	17 (6%)	13 (6%)
Unknown Histology NSCLC	1 (1%)	1 (1%)

CA209-003: Durability of Objective Responses Induced by Nivolumab in Patients With Advanced NSCLC, MEL, and RCC



Sixty-five of 306 patients had ORs (CR/PR):

- **30 of 65 (46%) responses were evident at first tumor evaluation (8 weeks)**
- **42 of 65 (65%) patients had responses lasting >1 year**
- **35 of 65 (54%) responses were ongoing at time of data analysis**
- **Responses persisted off-drug**

1. Topalian SL et al. Oral presentation at ASCO 2013. 3002.

CA209 -143 Primary and Secondary Objectives

Primary

Cohort 1 and 1b (Safety Lead-in):

✓ Evaluate the safety of nivolumab and nivolumab in combination with ipilimumab

Cohort 2 (Efficacy):

✓ Compare the overall survival (OS) of nivolumab versus bevacizumab

Secondary

Compare:

- ✓ OS at 12 months (OS12) of nivolumab vs bevacizumab
- ✓ Progression free survival (PFS) of nivolumab versus bevacizumab
- ✓ The objective response rate (ORR) between nivolumab and bevacizumab

Study Design Cohort 2 3.1

Cohort 2
Randomized, 2-Arm

Primary endpoint: Overall Survival, Secondary endpoints: OS12, PFS, ORR, of Nivo versus Bev – 1:1Randomization

Screening & Randomization Phase → **Treatment Phase** → **Follow-Up Phase**

Screening:
First recurrence of GBM after previous RT and temozolomide

1:1 Randomization (n = **220**)

Treatment Arm N:
Nivolumab 3mg/kg
every 2 weeks

Treatment Arm B
Bevacizumab 10 mg/kg
every 2 weeks

Treatment until confirmed progression or study discontinuation for any other reason. Post treatment follow-up for safety, overall survival, and progression.

Primary Endpoint:
OS

Secondary Endpoint:
OS12
PFS
ORR