NF110: Open-label, Phase 2 Clinical Trial of Crizotinib for Children and Adults with Neurofibromatosis Type 2 and Progressive Vestibular Schwannomas

Study Protocol & Statistical Analysis Plan

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ABBREVIATIONS:

ABI	Auditory brainstem implant
ABL	Abelson murine leukemia viral oncogene homolog 1
AE	Adverse event
ALK	Anaplastic lymphoma kinase
ALT	Aspartate aminotransferase
ANC	Absolute neutrophil count
AST	Alanine aminotransferase
ALT	Alanine transaminase/alanine transaminase
ATP	Adenosine triphosphate
AUC	Area under the curve
BCR	B-cell receptor
BRAF	v-Raf murine sarcoma viral oncogene homolog B
BSR	Biostatistics Shared Resource
CBC	Complete blood count
CDMRP	Congressionally Directed Medical Research Programs
CI	Confidence interval
C-kit	v-kit Hardy-Zuckerman 4 feline sarcoma viral oncogene homolog
CK	Creatinine phosphokinase
Cmax	Maximum (peak) concentration
C-met	MET proto-oncogene, receptor tyrosine kinase
CNS	Central nervous system
CR	Complete response
CRF	Case report form
CSF	Cerebrospinal fluid
CTCAE	Common terminology criteria for adverse events
CYP3A4	Cytochrome P450, family 3, subfamily A, polypeptide 4
DCC	Data Coordinating Center
DLCO	Diffusing lung capacity for carbon monoxide
DMSO	Dimethyl sulfoxide
DOD	Department of Defense
DPOAE	Distortion product otoacoustic emissions
DSMB	Data and Safety Monitoring Board
ECG	Electrocardiogram
EGFR	Epidermal growth factor receptor
EML4	Echinoderm microtubule-associated protein-like 4
ENT	Ear, nose and throat
ErbB2	Epidermal growth factor receptor 2
EU	European Union
FDA	Food and Drug Administration
HER-2	Human epidermal growth factor receptor 2

HGFR	Hepatocyte growth factor receptor
HRPO	Human Research Protection Official
I.B.	Investigator Brochure
IAC	Internal auditory canal
IRB	Institutional Review Board
MLV	Monitored live voice
MRI	Magnetic resonance imaging
MTD	Maximum tolerated dose
NCI	National Cancer Institute
NF2	Neurofibromatosis type 2
NFTI-QOL	Neurofibromatosis 2 impact on quality of life
NIH	National Institutes of Health
NOB	Neuro-Oncology Branch (NCI)
NOD/SCID	Non-obese diabetic/severe combined immunodeficiency
NPM	Nucleophosmin
NSCLC	Non-small-cell lung carcinoma
OAE	Otoacoustic emissions
PD	Progressive disease
PI	Principal Investigator
PK	Pharmacokinetics
PO	Per os (by mouth)
PR	Partial response
PS	Performance score
PTA	Pure tone average
QTc	Corrected QT interval
RON	Receptor originated from Nantes
ROS	ROS proto-oncogene 1, receptor tyrosine kinase
RT	Radiation therapy
SAE	Serious adverse event
SDS	Speech discrimination score (interchangeable with WRS)
SGOT	Serum glutamic oxaloacetic transaminase
SGPT	Serum glutamic pyruvic transaminase
SRT	Speech reception threshold
TRQ	Tinnitus reaction questionnaire
ULN	Upper limit of normal
VEGF	Vascular endothelial growth factor
VEGFR2	Vascular endothelial growth factor receptor 2
VS	Vestibular schwannoma
WBC	While blood count
WRS	Word recognition score (interchangeable with SDS)

PRECIS

Background

- Bilateral vestibular schwannomas (VS) are the hallmark of neurofibromatosis 2 (NF2). As these tumors enlarge, they cause sensorineural hearing loss and, ultimately, complete hearing loss.
- NF2-related vestibular schwannomas express Focal Adhesion Kinase 1 (FAK1)
- Crizotinib, a small molecule kinase inhibitor, reduces NF2 schwannoma cell growth and tumor formation in preclinical schwannoma models via inhibition of FAK1

Primary Objective

- To estimate the best objective volumetric response rates to crizotinib in pediatric and adult NF2 patients with VS

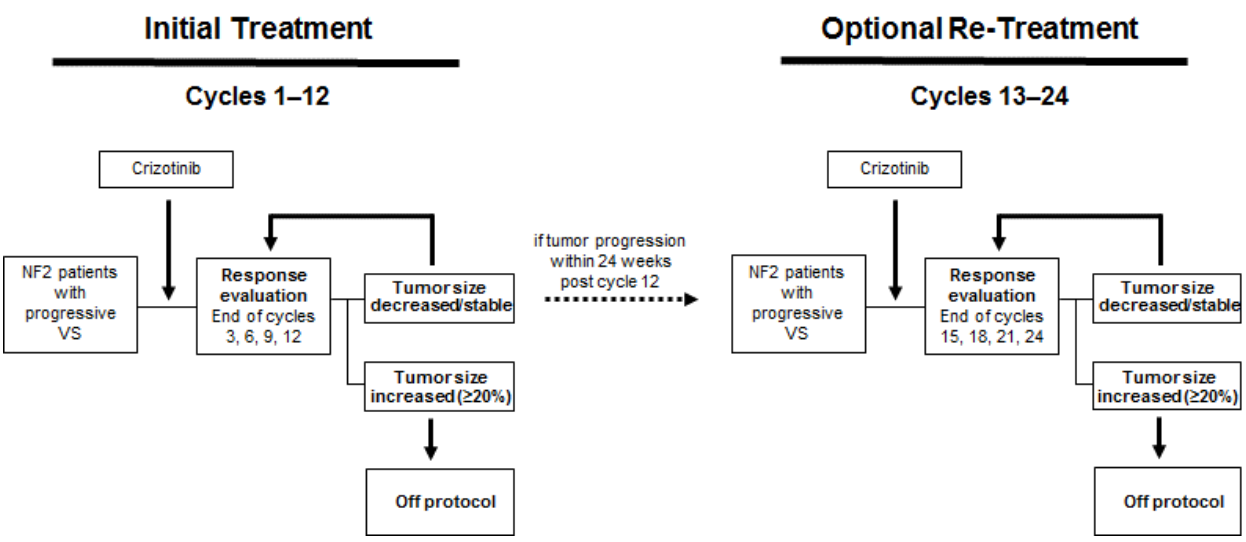
Eligibility

- NF2 patients ≥6 years of age on day 1 of treatment
- Subjects must have a target VS with the following qualities:
    - volumetrically measurable and ≥0.75 ml in size
    - volumetric evidence of progression over the past 18 months (defined as ≥20% annualized increase in volume)

Design

- Multi-Center, 2-stage, phase II open-label study
- Planned treatment: crizotinib is taken continuously for up to 12 cycles (48 weeks) until disease progression or unacceptable toxicity, with re-treatment option of patients who complete 12 cycles of treatment without disease progression, but progress within the following 24 weeks after stopping treatment with crizotinib.
- Response Evaluations: MRI brain (with 3D volumetrics, primary endpoint) and audiograms (if applicable, exploratory endpoint) at baseline and at the end of every 3rd cycle, i.e. every 12 weeks. Volumetric measurements by central review will be used to determine eligibility and continuation on treatment.

SCHEMA



Patients who complete 12 cycles of treatment without disease progression, but within the following 24 weeks show disease progression (defined as ≥20% increase in target tumor volume compared to off-treatment volume), will be eligible for re-treatment on study, provided they still meet study eligibility criteria

TABLE OF CONTENTS

1. OBJECTIVES

1.1 Study Design

Subjects with Neurofibromatosis Type 2 (NF2) and progressive vestibular schwannoma (VS) will be treated with crizotinib administered orally. Crizotinib will be taken continuously until disease progression or unacceptable toxicity, in continuous treatment cycles of 28 days each, for a maximum of 12 cycles. Clinical response will be assessed by MRI (volumetrics, primary objective) and audiology at the end of every 3rd cycle. Subjects with volumetric tumor progression will be taken off protocol. Patients who complete 12 cycles of treatment without disease progression, but within the following 24 weeks show subsequent disease progression (defined as >20% increase in target tumor volume compared to off-treatment volume), will be eligible for re-treatment on study for up to 48 additional weeks, provided they still meet study eligibility criteria.

1.2 Primary Objective

The primary objective of this study to estimate the best objective volumetric response rates (i.e. maximum tumor shrinkage) to crizotinib in NF2 patients with VS during up to 12 cycles (48 weeks) of treatment.

1.3 Exploratory Objectives (including re-treatment phase)

Exploratory objectives are:

(1) to describe the frequency of adverse events (possibly, probably, or definitely) related to crizotinib use in this patient population;

(2) to determine the durability of volumetric response, as measured from the time of first response to tumor progression (defined as an increase in volume of >20% compared to lowest tumor volume during treatment);

(3) to estimate hearing response rates (defined as an increase in word recognition score above the upper limit of the 95% critical difference from the baseline word recognition score, Appendix B) and durability of hearing response, as measured by freedom from hearing loss from the time of hearing response (defined as a decrease in word recognition score below the upper limit of the 95% critical difference of the baseline word recognition score, Appendix B);

(4) to explore effects of treatment on quality of life using a validated questionnaire for NF2 patients (NFTI-QOL);

(5) to explore association of volumetric or audiologic response with baseline and on-treatment plasma levels of crizotinib targets and candidate biomarkers including FAK, MET, ROS and ALK;

(6) to explore volumetric responses to crizotinib in NF2-associated non-VS tumors, such as meningiomas and ependymomas.

(7) to learn about the durability of imaging and hearing responses after discontinuation of study drug.

2. BACKGROUND

2.1 Neurofibromatosis 2

Neurofibromatosis type 2 (NF2) is an autosomal-dominant genetic disease with an incidence of approximately 1/40,000. The NF-2 gene is located on chromosome 22 and its gene product is named Merlin. Merlin's function is not well understood, but it appears to act as a tumor suppressor. The majority of NF2 patients develop progressive hearing loss in adolescence or young adulthood due to unilateral or bilateral vestibular schwannomas (VS).[1] Spinal tumors, including meningiomas, schwannomas and ependymomas, as well as intracranial tumors, mostly meningiomas, are also highly prevalent. NF2-related tumors, although mostly slow growing, cause considerable morbidity and mortality, particularly when first diagnosed at a young age. The available treatment options for these neoplasms, which often occur at multiple sites simultaneously, are non-curative and mostly limited to surgery and radiation therapy. As a result, all NF2 patients suffer from major morbidity, mortality and significantly reduced life expectancy.

Although classic chemotherapy is effective treatment for low-grade brain tumors, no such therapy has been validated for NF2 patients with multiple tumors for a variety of reasons. Most chemotherapeutic agents are mutagenic and there is reluctance to use them in patients with loss of tumor-suppressor function, such as NF2. Many chemotherapeutic drugs are neuro– and/or ototoxic, making them unsuitable for NF2 patients. Surgery remains the mainstay of therapy for VS, but carries major risks including complete deafness, facial palsy, stroke and CSF leak. Bevacizumab has emerged as a treatment option for NF2 patients, and may result in tumor shrinkage and/or hearing improvement in a subset of patients. In a recent prospective phase 2 study,[2] fourteen adult and pediatric patients with NF2 and progressive hearing loss in the target ear were enrolled. The primary end point, confirmed hearing response (improvement maintained ≥3 months), occurred in five (36%) of 14 patients (95% CI, 13–65%; P <0.001). Eight patients (57%) had transient hearing improvement above the 95% CI for word recognition scores, and imaging response was seen in six (43%) of target VS. However, responses to bevacizumab are not sustained off treatment,[3] and cumulative toxicities including hypertension and renal damage including proteinuria limit the long-term use of this therapy in responders. In addition, a subset of patients do not respond to therapy at all, or will progress after initial disease stabilization on bevacizumab.

We have previously conducted prospective NF2 phase 2 clinical trials with molecular targeted agents, including lapatinib and everolimus.[4,5] In the lapatinib study, four of 17 evaluable patients experienced an objective volumetric response, defined as ≥15% decrease in VS volume (23.5%; 95% confidence interval [CI], 10%-47%), with median time to response of 4.5 months (range, 3-12). In responders, reduction in VS volumes ranged from -15.7% to -23.9%. Four of 13 patients evaluable for hearing met hearing criteria for response (30.8%; 95% CI, 13–58%), but only one patient experiences a sustained hearing response exceeding 9 months in duration. Median time to overall

progression (i.e., volumetric progression or hearing loss) was 14 months. The estimated overall progression-free survival and volumetric progression-free survival at 12 months were 64.2% (95% CI, 36.9–82.1%) and 70.6% (95% CI, 43.1–86.6%), respectively. We concluded that lapatinib is well tolerated and has objective activity in a subset of NF2 patients with progressive VS, although the hearing and volumetric responses appeared to be inferior compared to published results with bevacizumab.[2,6,7] In our everolimus study, no objective imaging or hearing responses were observed in 9 evaluable patients with on stage 1 of the trial, and the study was closed according to predefined stopping rules.[5] We concluded from this data that everolimus is ineffective for the treatment of progressive VS in NF2 patients, and a subsequent study suggested that everolimus may have modest activity in some patients by decreasing tumor growth velocity.[8]

Taken together, additional effective medical treatment options for NF2 patients are urgently needed,[9] including for those who do not respond or progress on bevacizumab, or are unable to tolerate bevacizumab due to toxicities.

2.2 Study Agent(s)

2.2.1 Crizotinib (NSC# 749005)

Crizotinib (Xalkori®) is an ATP-competitive small-molecule oral inhibitor of the anaplastic lymphoma kinase (ALK), c-Met/hepatocyte growth factor receptor (HGFR), Recepteur d'Origine Nantais (RON), and ROS receptor tyrosine kinases and their oncogenic variants (e.g., c-Met/HGFR mutations and ALK or ROS1 fusion proteins).

Crizotinib has been studied in a variety of in vitro and in vivo model systems to determine potency for inhibition of ALK, c-Met/HGFR, RON, or ROS1 RTK activity, kinase selectivity, antitumor efficacy, pharmacodynamics, pharmacokinetics, and mechanism of action (reviewed by Rodig, Shapiro and Ou[10,11]). Crizotinib demonstrated potent concentration-dependent inhibition of the kinase activity of ALK, c-Met/HGFR, RON, and ROS1 in biochemical assays and inhibited phosphorylation and kinase dependent function in cell-based assays. Crizotinib demonstrated potent and selective growth inhibitory activity and induced apoptosis in tumor cell lines exhibiting ALK fusion variants (EML4-ALK or NPM-ALK), ROS1 fusion variants, or exhibiting amplification of the ALK or c-Met/HGFR gene locus. *In vivo*, crizotinib demonstrated antitumor efficacy, including marked cytoreductive antitumor activity, in mice bearing tumor xenografts that expressed ALK fusion variants or activated c-Met/HGFR. The antitumor efficacy of crizotinib was dose-dependent and correlated to pharmacodynamic inhibition of phosphorylation of ALK fusion variants (EML4-ALK or NPM-ALK) or c-Met/HGFR in tumors *in vivo*. The collective rationale for investigation of crizotinib in clinical studies is built on genetic alteration of its molecular targets, its predicted ability to target multiple processes that are common to cancer progression, and preclinical efficacy data. Crizotinib received regular marketing approval in the United States (US) for the treatment of patients with metastatic NSCLC that is ALK-positive as detected by a Food and Drug Administration (FDA)-approved test. Crizotinib also received regular and conditional approvals in Switzerland and the European Union (EU), respectively, for the treatment of adults with previously treated ALK-positive advanced NSCLC. In early 2016, the FDA has approved a supplemental New Drug Application for crizotinib to treat patients with metastatic non-small cell lung cancer (NSCLC) whose tumors are ROS1 gene rearrangement positive.

The safety and activity of crizotinib has also been investigated in pediatric patients in a Children's Oncology Group phase 1 consortium study, which included patients with refractory solid tumors or anaplastic large-cell lymphoma (ALCL).[12] Seventy-nine patients were enrolled in the study with a median age of 10.1 years (range 1.1-21.4). Crizotinib was well tolerated with a recommended phase 2 dose of 280 mg/m^2 twice daily. Grade 4 adverse events in cycle 1 were neutropenia (two) and liver enzyme elevation (one). Grade 3 adverse events that occurred in more than one patient in cycle 1 were lymphopenia (two), and neutropenia (eight). The mean steady state peak concentration of crizotinib was 630 ng/mL and the time to reach this peak was 4 h (range 1-6). Objective tumor responses were documented in 14 of 79 patients (nine complete responses, five partial responses); and the anti-tumor activity was enriched in patients with known activating ALK aberrations (eight of nine with anaplastic large-cell lymphoma, one of 11 with neuroblastoma, three of seven with inflammatory myofibroblastic tumor, and one of two with NSCLC).

In a study of 121 patients under the age of 21, patients with ALCL and inflammatory myofibroblastic tumor (IMT) reported visual disorders in 65% and 50% of these patients. One pediatric patient with IMT did experience grade 3 myopic optic nerve disorder. The onset of visual disorders was approximately 1 week and the most common symptoms were blurred vision and visual impairment. Other symptoms included photopsia, biterous floaters, and photophobia.

The pediatric recommended phase 2 dose of 280 mg/m^2 twice daily is equivalent to nearly twice the standard recommended adult dose of 250 mg twice daily. In adult subjects, geometric mean AUC and C_{max} continued to increase with doses above the standard dose of 250 mg BID. In the initial adult phase 1 studies, crizotinib was escalated to 300 mg orally twice daily, at which dose two patients experienced grade three fatigue. The dose of crizotinib was eventually reduced to 250 mg orally twice daily, found to be tolerable and was determined to be the recommended Phase II dose. In order to maximize long-term tolerability of crizotinib in NF2 patients on this study, we will cap crizotinib at the standard adult dose on 250 mg twice daily for all patients, including children.

Crizotinib (XALKORI®) Safety/Toxicity Summary

Safety evaluation of XALKORI is based on more than 1,200 patients with ALK-positive metastatic NSCLC who received XALKORI as monotherapy at a starting oral dose of 250 mg twice daily continuously. The most common adverse reactions (≥25%) of XALKORI are vision disorder, nausea, diarrhea, vomiting, constipation, edema, elevated transaminases, and fatigue. In the post-marketing experience, there is a report of increased blood creatine phosphokinase, but no frequency or adverse reaction was documented.

ALK-positive metastatic NSCLC-Study 1: Patients in the XALKORI arm (n=172) received XALKORI 250 mg orally twice daily until documented disease progression, intolerance to therapy, or the investigator determined that the patient was no longer experiencing clinical benefit. A total of 171 patients in the chemotherapy arm received pemetrexed 500 mg/m^2 (n=99) or docetaxel 75 mg/m2 (n=72) by intravenous infusion every three weeks until documented disease progression, intolerance to therapy, or the

investigator determined that the patient was no longer experiencing clinical benefit. Patients in the chemotherapy arm received pemetrexed unless they had received pemetrexed as part of first-line or maintenance treatment. The median duration of study treatment was 7.1 months for patients who received XALKORI and 2.8 months for patients who received chemotherapy. Across the 347 patients who were randomized to study treatment (343 received at least one dose of study treatment), the median age was 50 years; 84% of patients in the XALKORI arm and 87% of patients in the chemotherapy arm were younger than 65 years. A total of 57% of patients on XALKORI and 55% of chemotherapy patients were female. Forty-six percent (46%) of XALKORI-treated and 45% of chemotherapy-treated patients were from Asia. Serious adverse reactions were reported in 64 patients (37.2%) treated with XALKORI and 40 patients (23.4%) in the chemotherapy arm. The most frequent serious adverse reactions reported in patients treated with XALKORI were pneumonia (4.1%), pulmonary embolism (3.5%), dyspnea (2.3%), and interstitial lung disease (ILD; 2.9%). Fatal adverse reactions in XALKORI-treated patients in Study 1 occurred in 9 (5%) patients, consisting of: acute respiratory distress syndrome, arrhythmia, dyspnea, pneumonia, pneumonitis, pulmonary embolism, ILD, respiratory failure, and sepsis. Dose reductions due to adverse reactions were required in 16% of XALKORI-treated patients. The most frequent adverse reactions that led to dose reduction in the patients treated with XALKORI were alanine aminotransferase (ALT) elevation (7.6%) including some patients with concurrent aspartate aminotransferase (AST) elevation, QTc prolongation (2.9%), and neutropenia (2.3%). Discontinuation of therapy in XALKORI-treated patients for adverse reactions was 17.0%. The most frequent adverse reactions that led to discontinuation in XALKORI-treated patients were ILD (1.7%), ALT and AST elevation (1.2%), dyspnea (1.2%), and pulmonary embolism (1.2%). Additional adverse reactions occurring at an overall incidence between 1% and 30% in patients treated with XALKORI included decreased appetite (27%), fatigue (27%), neuropathy (19%; dysesthesia, gait disturbance, hypoesthesia, muscular weakness, neuralgia, peripheral neuropathy, parasthesia, peripheral sensory neuropathy, polyneuropathy, burning sensation in skin), rash (9%), ILD (4%; acute respiratory distress syndrome, ILD, pneumonitis), renal cyst (4%), and hepatic failure (1%).

ALK-positive metastatic NSCLC- Study 2: The safety analysis population in Study 2 included 934 patients with ALK-positive metastatic NSCLC who received XALKORI in a clinical trial. The median duration of treatment was 23 weeks. Dosing interruptions and reductions due to treatment-related adverse events occurred in 23% and 12% of patients, respectively. The rate of treatment-related adverse events resulting in permanent discontinuation was 5%. The most common adverse reactions (≥25%) included vision disorder (55%), nausea (51%), vomiting (46%), diarrhea (46%), edema (39%), constipation (38%), and fatigue (26%). Description of selected adverse drug reactions. Vision disorders, most commonly visual impairment, photopsia, blurred vision, or vitreous floaters, occurred in 691 (56%) patients across clinical trials (n=1225). The majority (99%) of these patients had Grade 1 or 2 visual adverse reactions. Across clinical studies, one patient had a treatment-related grade 3 vision abnormality. Based on the Visual Symptom Assessment Questionnaire (VSAQ-ALK), patients treated with XALKORI in Study 1 reported a higher incidence of visual disturbances compared to patients treated with chemotherapy. The onset of vision disorders generally started within the first week of drug administration. The majority of patients on the XALKORI arm in

Study 1 (> 50%) reported visual disturbances; these visual disturbances occurred at a frequency of 4-7 days each week, lasted up to 1 minute, and had mild or no impact (scores 0 to 3 out of a maximum score of 10) on daily activities as captured in a patient questionnaire. Neuropathy, most commonly sensory in nature, occurred in 235 (19%) of 1225 patients. Most events (95%) were Grade 1 or Grade 2 in severity. Renal Cysts Renal cysts occurred in 7 (4%) patients treated with XALKORI and 1 (1%) patient treated with chemotherapy in Study 1. The majority of renal cysts in XALKORI-treated patients were complex. Local cystic invasion beyond the kidney occurred, in some cases with imaging characteristics suggestive of abscess.

Pediatric Safety/Toxicity Data

The safety and activity of crizotinib has also been investigated in pediatric patients in a Children's Oncology Group phase 1 consortium study, which included patients with refractory solid tumors or anaplastic large-cell lymphoma.[12] Seventy-nine patients were enrolled in the study with a median age of 10.1 years (range 1.1-21.4). Crizotinib was well tolerated with a recommended phase 2 dose of 280 mg/m2 twice daily. Grade 4 adverse events in cycle 1 were neutropenia (two) and liver enzyme elevation (one). Grade 3 adverse events that occurred in more than one patient in cycle 1 were lymphopenia (two), and neutropenia (eight). The mean steady state peak concentration of crizotinib was 630 ng/mL and the time to reach this peak was 4 h (range 1-6). Objective tumor responses were documented in 14 of 79 patients (nine complete responses, five partial responses); and the anti-tumor activity was enriched in patients with known activating ALK aberrations (eight of nine with anaplastic large-cell lymphoma, one of 11 with neuroblastoma, three of seven with inflammatory myofibroblastic tumor, and one of two with NSCLC).

The pediatric recommended phase 2 dose of 280 mg/m^2 twice daily[12] ***is equivalent to nearly twice*** the standard recommended adult dose of 250 mg twice daily. In adult subjects, geometric mean AUC and Cmax continued to increase with doses above the standard dose of 250 mg BID. In the initial adult phase 1 study,[13] crizotinib was escalated to 300 mg orally twice daily, at which dose two patients experienced grade three fatigue. The dose of crizotinib was eventually reduced to 250 mg orally twice daily, found to be tolerable and was determined to be the recommended Phase II dose.

Further information, including detailed toxicity data, is provided by Pfizer in the Crizotinib Investigator Brochure.

2.3 Rationale

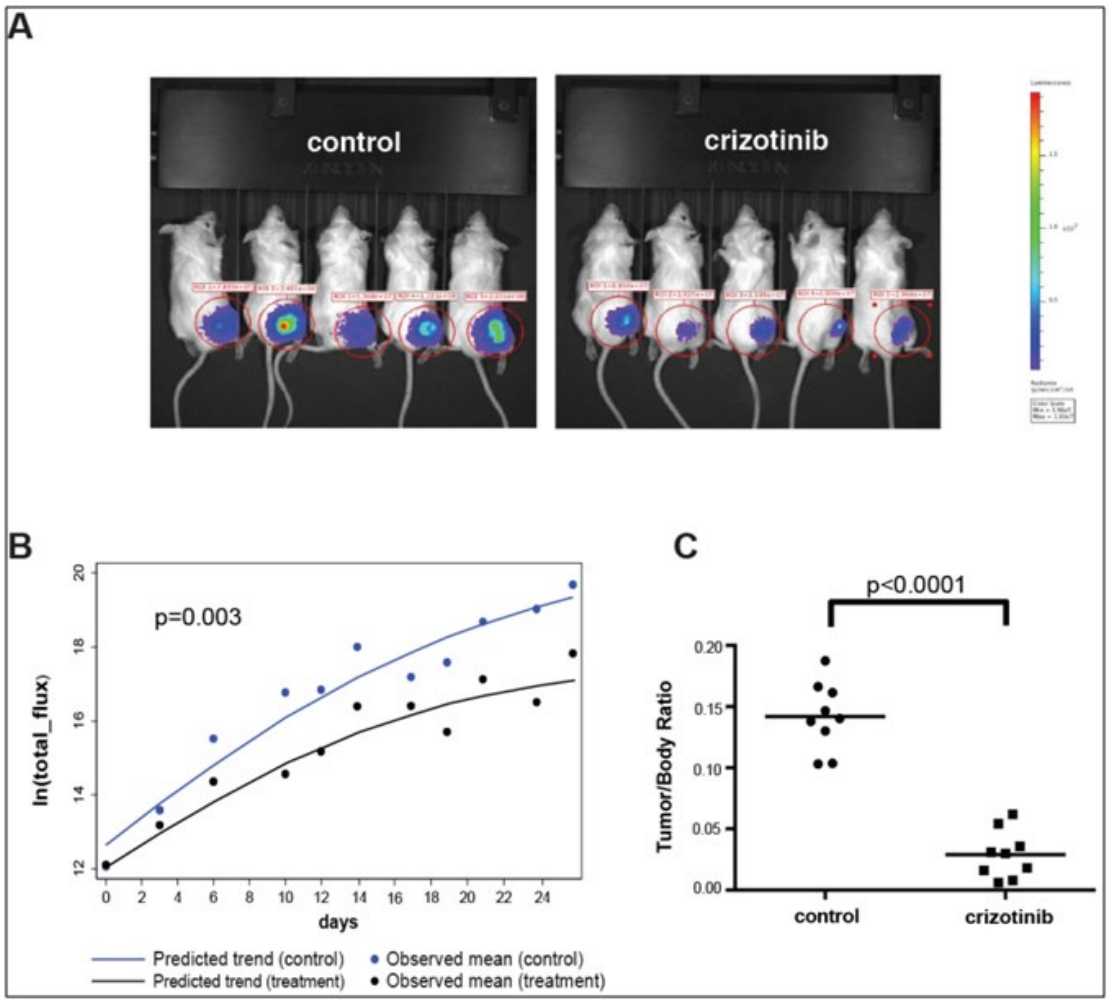
Recent preclinical data from the Kissil lab (Scripps Research Institute, FL)[14] provide a strong rationale for exploring crizotinib for the treatment of NF2 related tumors, including VS.

Crizotinib inhibits schwannoma cell proliferation in vitro and tumor formation in vivo:

Testing activity of different small molecule inhibitors in SC4 (mouse *NF2*-null Schwann cells) and HEI193 cells (*NF2*-null, derived from a schwannoma of an NF2 patient), identified the ALK/c-Met inhibitor crizotinib[15,16] as a dose-dependent suppressor of cell proliferation. To assess if crizotinib inhibits tumor growth in vivo, an orthotopic model of

NF2 was employed that recapitulates the tumor microenvironment of schwannomas, by injection of luciferase-tagged NF2-null schwannoma cells into a myelinated nerve.[17-20] Tumor progression was monitored every three days by bioluminescence imaging (BLI) and total flux counts were recorded for each animal. Ten days post injection, similar flux readings for all animals were validated and animals were enrolled randomly into control (vehicle only) or drug-treated cohorts and were treated (50mg/kg, PO, once daily) for a period of 24 days. Crizotinib treated mice had a significantly slower tumor growth rate than control animals (Figure 1A–B). After 24 days of treatment the animals were sacrificed and the tumors removed and weighed. Comparison of the cohorts demonstrates significantly lower average tumor weight in the crizotinib-treated group compared to control group (Figure 1C). Taken together, these data demonstrate that crizotinib has significant anti-proliferative activity against NF2-null Schwann cells *in vitro* and anti-tumor activity *in vivo*.

Figure 1: Crizotinib inhibits tumor growth *in vivo.*[14] (A) Representative images from bioluminescence imaging (BLI) of mice carrying orthotopic tumors treated with crizotinib (50mg/kg) or vehicle control at day 14 of treatment. NOD/ SCID mice were injected intraneurally with $5x10^4$ SC4/pLuc-mCherry cells and were enrolled into treatment after 10 days. Mice were treated daily for 25 days and imaged every 3 days to follow tumor development. (B) Quantitative analysis of the flux reading from treated cohorts. A mixed-effects regression analysis indicated that the speed of tumor growth in treatment group is significantly slower than that in control group (p=0.003). (C) Distribution of tumor/body weight ratio in the cohorts treated with crizotinib or vehicle control. The results of t-test with equal variances show that the crizotinib-treated group has significant lower average tumor weight than that observed in control group (p<0.0001). For the *in vivo* experiments the N=9 in each cohort.



Using pharmacological and genetic approaches, the Kissil lab demonstrated that the activity of crizotinib was mediated through focal adhesion kinase 1 (FAK1), a previously recognized target in NF2.[21] SC4 and HEI193 cells were treated and FAK1 activation was assessed by immunoblot analysis using a phospho-specific antibody against FAK-Y397, which is phosphorylated in activated FAK1. Treatment with crizotinib resulted in decreased FAK-Y397 phosphorylation, consistent with inhibition FAK1 (Figure 3A). To determine if FAK1 is required for proliferation of SC4 or HEI193 cells, FAK1 expression was knocked down using shRNA or siRNA, respectively. Indeed, the knockdown of FAK1 significantly impaired cell proliferation. Subsequently, the effects of treating SC4 or HEI193 cells with small-molecule FAK1 inhibitors were examined. Treatment of SC4 or HEI193 cells with defactinib (VS6303) or PND-1186 (VS-4718) significantly impaired cell proliferation in a dose-

dependent manner. Additional experiments using gain of function approaches showed that crizotinib-resistant FAK1 mutants rescue proliferation of treated NF2-null schwann cells, and together with the loss of function data strongly implicate FAK1 inhibition as the main mechanism through which crizotinib mediates its effects in NF2-null schwannoma cells. In addition, other studies have shown that c-Met, may also represent a molecular target in schwannoma cells.[22,23]

Expression of activated FAK in human VS tissue:

To confirm activated target expression in human VS tissue from NF2 patients, fresh frozen tumor samples obtained from the NYU Brain Tumor Bank were subjected to Western blot analysis. Strong expression of phospho-FAK was observed in 6/6 (100%) of samples (Figure 2, unpublished data).

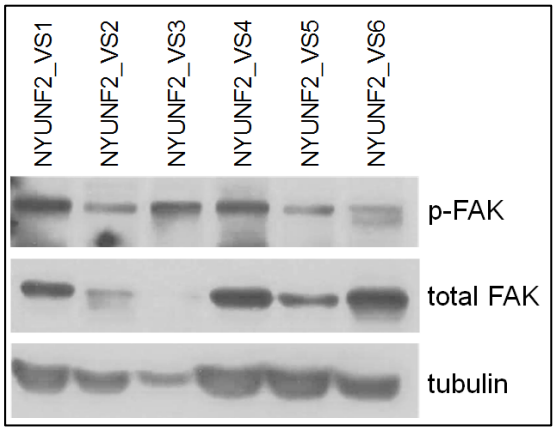


Figure 2: Expression of activated FAK in human VS tissue. Western blot analysis for total FAK and phospho-FAK(Tyr397) in VS frozen tissue lysates from NF2 patients. Tubulin was used as loading control.

Blood-brain barrier considerations:

Although crizotinib is considered not to cross the intact blood brain barrier, this is not expected to negatively impact on the therapeutic efficacy in NF2-related tumors. Meningiomas and VS are anatomically outside the blood brain barrier, and we have observed tumor tissue to plasma ratio of 4.3 ±1.9 (mean ±SD) in human VS tissue treated with the EGFR/ErbB2 receptor tyrosine kinase inhibitor lapatinib (Karajannis, et. al, Neuro-Oncology 14:i19, 2012 [abstract]). In addition, clinical activity of crizotinib has been reported in other contrast-enhancing intracranial tumors, such as brain metastases and even primary brain tumors.[24-26]

Prior clinical studies and competing clinical trials:

Over the past years, we and others have performed clinical studies, including prospective clinical trials, to investigate the potential of "molecular targeted" drugs to stop tumor growth and/or improve hearing in NF2 patients.[9] The most effective therapy to date, bevacizumab, results in radiologic and hearing response rates of approximately 50%, respectively.[27] In general, the treatment effect is only maintained with continued bevacizumab administration, which is time-limited due to cumulative toxicity, including hypertension and renal toxicity. Despite the success of bevacizumab for many NF2 patients with vestibular schwannomas, alternative therapies remain urgently needed for NF2 patients with progressive tumor growth. This includes non-responders to bevacizumab, or patients who cannot tolerate continued administration due to acute or

cumulative toxicity. In addition, bevacizumab is ineffective for NF2 patients with progressive meningiomas.[28] Attractive drug candidates for these populations therefore would have a target profile and mechanism of action that is different from bevacizumab, and ideally have non-overlapping toxicities. Over the past several years, we have shown that we are able to successfully conduct and complete single-institution Phase 2 clinical trials in the NF2 population using molecular targeted agents, including lapatinib and everolimus, in a short time frame.[4,5] The lapatinib trial showed only modest anti-tumor activity and the everolimus trial did not result in any objective responses, and there remains a need for novel therapies with better efficacy, ideally based on preclinical data. For this phase 2 clinical trial with crizotinib, we will follow a similar study design and endpoints, which are consistent with current consensus recommendations for NF2 clinical trials endpoints.[29,30]

A Phase 2 clinical trial for NF2 patients with axitinib, a multi-kinase inhibitor with the VEGF receptor VEGFR2 as the dominant target (ClinicalTrials.gov identifier NCT02129647), is currently ongoing at Memorial Sloan Kettering Cancer Center and NYU Langone Medical Center. This study excludes NF2 patients who received prior bevacizumab. No other phase 2 clinical trials with molecular targeted therapy specifically for NF2 patients with progressive VS are currently open for accrual in North America based on ClinicalTrials.gov search. Defactinib, an investigational FAK inhibitor, is included in the NF2 mutant tumors stratum of the NCI-Molecular Analysis for Therapy Choice (NCI-MATCH) trial. This clinical trial will require biopsy and confirmation of NF2 mutation in tumor tissue. Although this trial is aimed at patients with advanced refractory solid tumors or lymphomas, some NF2 patients may be eligible for this study, which does not include NF2-specific eligibility criteria, endpoints and recommended assessments, such as volumetric analysis or hearing. The safety and toxicity of defactinib (VS-6063), which is not currently FDA approved, was investigated in a phase I study of in adult patients with advanced solid tumors,[31] with the most frequent adverse events including nausea (37%), fatigue (33%), vomiting (28%), diarrhea (22%) and headache (22%). The safety of defactinib has not been studied in children. Of note, a ten-fold higher concentration of defactinib compared to crizotinib was required to inhibit growth in HEI193 cells.[14]

In summary, the preclinical data from the Kissil lab identified crizotinib as a promising drug candidate for clinical study in NF2 patients with progressive VS.[14] The favorable toxicity profile and tolerability of crizotinib, including in children,[12] and compared to other multi-kinase inhibitors, is advantageous for potential long-term administration in NF2. In addition, the non-overlapping molecular target and toxicity profiles of crizotinib, compared to bevacizumab, render crizotinib an attractive agent for NF2 patients, including non-responders to bevacizumab, or patients who develop dose-limiting toxicities on bevacizumab. Additional candidates for crizotinib include patients who have pre-existing contraindications to bevacizumab, such as difficult-to-control hypertension or prior CNS hemorrhage.

Neurofibromatosis type 2 (NF2) is an autosomal-dominant genetic disease with an incidence of approximately 1/40,000. The NF-2 gene is located on chromosome 22 and its gene product is named Merlin. Merlin's function is not well understood, but it appears to act as a tumor suppressor. The majority of NF2 patients develop progressive hearing loss in adolescence or young adulthood due to unilateral or bilateral vestibular

schwannomas (VS). Other intracranial and spinal tumors, including meningiomas, schwannomas and ependymomas, are also highly prevalent. NF2-related tumors, although generally slow growing, cause considerable morbidity and mortality, particularly when first diagnosed at a young age. The available treatment options for these neoplasms, which often occur at multiple sites simultaneously, are non-curative and mostly limited to surgery and radiation therapy. As a result, all NF2 patients suffer from major morbidity, mortality and significantly reduced life expectancy.

Recently published preclinical data from the Kissil lab shows that crizotinib inhibits NF2 schwannoma cell proliferation and tumor formation *in vivo*, providing a strong rationale for exploring crizotinib for the treatment of NF2 related tumors, including VS. We hypothesize that crizotinib is effective in treating vestibular schwannomas in NF2 patients, and propose to test this hypothesis in a prospective phase 2 clinical trial. Crizotinib is currently FDA approved for the treatment of patients with metastatic non-small cell lung carcinoma that is anaplastic lymphoma kinase (ALK) positive. The favorable toxicity profile and tolerability of crizotinib compared to other multi-kinase inhibitors, including in children, is advantageous for potential long-term administration in NF2 patients.

2.4 Correlative Studies Background

Prior studies, including in NF2 patients, have shown that circulating biomarkers can help predict tumor progression and response to molecular targeted therapies. For example, bevacizumab treatment was associated with decreased free vascular endothelial growth factor (not bound to bevacizumab) and increased placental growth factor in plasma. Hearing responses were inversely associated with baseline plasma hepatocyte growth factor ($P = 0.019$). Imaging responses were associated with high baseline tumor vessel permeability and elevated blood levels of vascular endothelial growth factor D and stromal cell-derived factor 1alpha ($P = 0.037$ and 0.025, respectively). [2]

There is no published data on molecular biomarkers for crizotinib in patients with NF2, and this study proposes to analyze baseline biomarkers with respect to outcomes (response) as well as the possible effect of crizotinib on biomarker levels over time (see Section 8.3.1).

3. PARTICIPANT SELECTION

3.1 Eligibility Criteria

Participants must meet the following criteria on screening examination to be eligible to participate in the study:

3.1.1 Patients must have a confirmed diagnosis of neurofibromatosis 2 by fulfilling National Institute of Health (NIH) criteria or Manchester criteria, or by detection of a causative mutation in the *NF2* gene.

The NIH criteria[32] include presence of:

- Bilateral vestibular schwannomas, **OR**
- First-degree relative with NF2 and **EITHER** unilateral eighth nerve mass

OR two of the following: neurofibroma, meningioma, glioma, schwannoma, juvenile posterior subcapsular lenticular opacity.

The Manchester criteria[33] include presence of:

- Bilateral vestibular schwannomas, **OR**
- First-degree relative with NF2 and **EITHER** unilateral eighth nerve mass **OR** two of the following: neurofibroma, meningioma, glioma, schwannoma, juvenile posterior subcapsular lenticular opacity, OR
- Unilateral vestibular schwannoma AND any two of: neurofibroma, meningioma, glioma, schwannoma, juvenile posterior subcapsular lenticular opacity, **OR**
- Multiple meningiomas (two or more) **AND** unilateral vestibular schwannoma **OR** any two of: schwannoma, glioma, neurofibroma, cataract.

3.1.2 Patients must have progressive and measurable disease, defined as at least one VS with the following qualities:

- ≥ 0.75 ml (on volumetric analysis) that can be accurately measured by contrast-enhanced cranial MRI scan with fine cuts through the internal auditory canal (1 mm slices, no skip)
- MRI evidence of progression over the past 18 months (defined as ≥20% annualized increase in volume)

  Note: pre-baseline MRI to be submitted together with baseline MRI for central volumetric review and confirmation of eligibility.

3.1.3 Age ≥ 6 years on day 1 of treatment.

3.1.4 Life expectancy of greater than 1 year.

3.1.5 Lansky/Karnofsky performance status ≥ 60 (see Appendix D).

3.1.6 Organ and marrow function as defined below:

- Absolute neutrophil count ≥ 1,500/ µl
- Platelets ≥ 100,000/ µl
- Total bilirubin within ≤ 1.5 X institutional upper limit of normal
- AST (SGOT)/ALT (SGPT) ≤ 2.5 X institutional upper limit of normal
- Patients must have a creatinine clearance or radioisotope GFR ≥60ml/min/1.73 m^2 or a normal serum creatinine based on age/gender described in the table below:

Age	**Maximum Serum Creatinine (mg/dL)**	
	Male	**Female**
6 to < 10 years	1	1
10 to < 13 years	1.2	1.2
13 to < 16 years	1.5	1.4
≥ 16 years	1.7	1.4

The threshold creatinine values in this Table were derived from the Schwartz formula for estimating GFR (Schwartz et al. J. Peds, 106:522, 1985) utilizing child length and stature data published by the CDC.

3.1.7 Fully recovered from acute toxic effects of any prior chemotherapy, biological modifiers or radiotherapy

3.1.8 Any neurologic deficits must be stable for ≥1 week

3.1.9 Patient or parent/legal guardian must be able to provide signed informed consent and assent (as applicable for minors)

3.2 Exclusion Criteria

Participants who exhibit any of the following conditions at screening will not be eligible for admission into the study.

3.2.1 Patients who previously received crizotinib

3.2.2 Patients currently receiving medical anticancer therapies or who have received medical anticancer therapies within 4 weeks of the start of study drug (including chemotherapy and molecular targeted agents), as these may interfere with the study drug

3.2.3 Monoclonal antibody treatment and/or agents with prolonged half-lives: At least three half-lives must have elapsed from the last dose prior to enrollment

3.2.4 Radiation therapy to a study target tumor within 1 year prior to enrollment, or any radiation therapy within 4 weeks prior to enrollment, as these may interfere with our ability to assess response to study drug

3.2.5 Prior treatment with any investigational drug within the preceding 4 weeks, as they may interfere with the study drug

3.2.6 Unstable or rapidly progressive disease, including patients who require glucocorticoids for symptomatic control of brain or spinal tumors, as this would represent a high risk for inability to comply with the study requirements

3.2.7 Use of drugs or foods that are known potent CYP3A4 inhibitors, including but not limited to ketoconazole, itraconazole, miconazole, clarithromycin, erythromycin, ritonavir, indinavir, nelfinavir, saquinavir, amprenavir, delavirdine, nefazodone, diltiazem, verapamil, and grapefruit juice, as this would interfere with study drug metabolism

3.2.8 Use of drugs that are known potent CYP3A4 inducers, including but not limited to carbamazepine, phenobarbital, phenytoin, rifabutin, rifampin, tipranavir, ritonavir, and St. John's wort, as this would interfere with study drug metabolism

3.2.9 Use of drugs that are CYP3A4 substrates with narrow therapeutic indices, including but not limited to pimozide, aripiprazole, triazolam, dihydroergotamine, ergotamine, astemizole, cisapride, terfenadine and halofantrine, as this would interfere with study drug metabolism

3.2.10 Ongoing cardiac dysrhythmias of CTCAE grade ≥2, uncontrolled atrial fibrillation of any grade or prolonged QTc interval (>480 msec), as patients with these conditions would be expected to have an increased risk for cardiac toxicity related to study drug

3.2.11 Patients who have any severe and/or uncontrolled medical conditions or other conditions that could affect their participation in the study such as:

- symptomatic congestive heart failure of New York heart Association Class III or IV
- unstable angina pectoris, symptomatic congestive heart failure, myocardial infarction within 6 months of start of study drug, serious uncontrolled cardiac arrhythmia or any other clinically significant cardiac disease
- severely impaired lung function as defined as spirometry and DLCO that is 50% of the normal predicted value and/or O2 saturation that is 90% or less at rest on room air
- active (acute or chronic) or uncontrolled severe infections
- liver disease, such as cirrhosis or severe hepatic impairment (Child-Pugh class C).

3.2.12 Impairment of gastrointestinal function or gastrointestinal disease that may significantly alter the absorption of crizotinib (e.g., ulcerative disease, uncontrolled nausea, vomiting, diarrhea, malabsorption syndrome or small bowel resection)

3.2.13 Female patients who are pregnant or breast feeding, or adults of reproductive potential who are not using effective birth control methods. Adequate contraception must be used throughout the trial and for 90 days after the last dose of study drug, as the effects of crizotinib on an unborn fetus are not known. Females of childbearing potential must have a negative serum pregnancy test within 7 days prior to administration of crizotinib.

3.2.14 Male patients whose sexual partner(s) are women of child bearing potential, who are not willing to use adequate contraception during the study and for 90 days after the last dose of study drug.

3.2.15 History of significant noncompliance to medical regimens that would jeopardize compliance with study therapy

3.2.16 Patients unwilling to or unable to comply with the study protocol

3.3 Inclusion of Women, Minorities and Other Underrepresented Populations

Both men and women, and members of all races and ethnic groups are eligible for this trial.

4. SCREENING AND STUDY ENROLLMENT PROCEDURES

The NF Operations Center should be contacted to ensure availability of a treatment slot.

4.1 Informed Consent/Assent

The investigational nature and objectives of the trial, the procedures and treatments involved and their attendant risks and discomforts, and potential alternative therapies will be carefully explained to the subject or the subject's parent(s) or guardian if the subject is a child, and a signed informed consent and assent will be obtained according to institutional and federal guidelines. In addition, study participants should sign the institution's HIPAA Consent if not already included in the study consent document.

4.2 Screening Procedures

Diagnostic or laboratory studies performed exclusively to determine eligibility for this trial must only be done after obtaining written informed consent. Documentation of the informed consent for screening will be maintained in the subject's research chart. Studies or procedures that were performed for clinical indications (not exclusively to determine eligibility) may be used for baseline values even if the studies were done before informed consent was obtained.

Before the subject can be enrolled, the responsible institutional investigator must sign and date the completed eligibility checklist. The completed eligibility checklist and de-identified supporting clinical documentation should be scanned and emailed for review and signed off by the Study PI and/or Study Co-PI and the NF Operations Center to confirm eligibility prior to subject enrollment.

4.3 Study Enrollment

Subjects may be enrolled on the study once all eligibility requirements for the study have been met and a treatment number for subject treatment has been confirmed by the NF Consortium Operations Center as well as either the study chair or study co-chair. Subjects who give informed consent for the protocol in order to undergo screening for eligibility are not considered enrolled and should not be enrolled until the screening is completed and they are determined to meet all eligibility criteria. **Treatment must start within 14 days of enrollment. Subjects must not receive any protocol therapy prior to enrollment. Baseline evaluations should be repeated if time between first screening and start of protocol therapy exceeds 28 days.**

4.4 General Guidelines for all Participating Institutions

The required forms for subject registration can be found on the UAB online entry system.

Following registration, participants should complete screening procedures and verify all eligibility criteria prior to enrollment into the study. Participants should begin protocol treatment within 14 days of enrollment. Issues that would cause treatment delays should be discussed with the Principal Investigator. If a participant does not receive protocol therapy following registration and enrollment, the participant's protocol status must be changed. The Operations Center should be notified of participant status changes as soon as possible by email.

Each participating institution will order the study agent from the UAB Research Pharmacy and the UAB NFCTC Operations Center (section 7.1.7). A participating site may order the agent only after all initial IRB approvals have been obtained (site, UAB and DOD).

4.5 Registration and Enrollment Process for All Participating Institutions

To register a participant, the following forms should be entered and locked in the electronic data collection system (eDCS) by the study coordinator or designee:

- Registration and Demographics Form
- IRB Consent Form
- MRI Transmittal Form

To complete enrollment, the following forms should be entered and locked in eDCS:

- MRI Acknowledgement (to be entered by Central Imaging at DFCI)
- Inclusion and Exclusion Forms must be completed and locked prior to enrollment in eDES. However, the Exclusion Form cannot be locked until approval to enroll is received from the study chairs performing the eligibility review.

NOTE: Registration with NF Consortium's web-based data entry system can be conducted anytime, but the DCCsupport can only be provided during the business hours of 8am – 5pm EST Monday through Friday.

5. TREATMENT PLAN

Crizotinib will be taken continuously until disease progression or unacceptable toxicity, in continuous treatment cycles of 28 days each, for a maximum of 12 cycles of initial therapy. Clinical response will be assessed by MRI (volumetrics, primary objective) and audiology at the end of every 3rd cycle. Subjects with volumetric tumor progression will be taken off protocol. Patients who complete 12 cycles of treatment without disease progression, but within the following 24 weeks show subsequent disease progression (defined as >20% increase in target tumor volume compared to end of cycle tumor volume), will be eligible for re-treatment on study for up to 12 cycles/48 additional weeks, provided they still meet study eligibility criteria (See Section 5.2).

The study drug crizotinib will be administered orally. The investigator will instruct the patient/family to take/administer the study drug exactly as specified in the protocol, and the

subjects will receive detailed instructions for drug administration along with a drug diary (see Appendix I).

All subjects will be provided with drug diaries and asked to record each dose of crizotinib. Drug diaries will be collected by the investigator after each cycle.

Crizotinib should be administered orally once or twice daily, depending on prescribed dose level. For twice daily administration, crizotinib should be taken approximately 12 hours apart, preferably in the morning and evening, at approximately the same time every day with or without food. If the patient vomits or misses a dose, an additional dose should not be taken. The next prescribed dose should be taken at the usual time.

Crizotinib will be provided by Pfizer, Inc., and is available as 200 mg and 250 mg hard gelatin capsules. Patients are advised to take crizotinib with or without food and whole. Crizotinib will be taken continuously until disease progression or unacceptable toxicity, in continuous treatment cycles of 28 days each (maximum of 12 cycles):

Patients who are unable to swallow capsules may take crizotinib as described in Appendix H: Disintegration of Capsules for Patients Unable to Swallow Capsules.

Starting dose for adults (≥18 years of age): standard adult dose of 250 mg PO twice daily (dose level D).

Starting dose for children (<18 years of age): based on body surface area (BSA) according to the dose level table below (equivalent to 280 mg/m^2/dose twice daily), with a maximum dose of 250 mg PO twice daily (dose level D):

Dose Level	BSA [m^2]	Total Daily Dose	Dose
A	**<0.45**	**200 mg**	**200 mg PO once daily**
B	**0.45-0.70**	**250 mg**	**250 mg PO once daily**
C	**0.71-0.88**	**400 mg**	**200 mg PO twice daily**
D*	**≥0.89**	**500 mg**	**250 mg PO twice daily**

*adult starting dose level (standard adult dose)

5.1 Safety Measures and Special Precautions:

Prior to each treatment cycle, the patient should be carefully assessed for toxicities. Decisions for retreatment or dose modification/interruption should follow the dose modification guidelines in Section 6.2.

Patients who have an ongoing study agent-related serious adverse event upon study completion or at discontinuation from the study will be contacted by the investigator or his/her designee periodically until the event is resolved or determined to be irreversible.

5.1.1 CYP3A4/5 inhibitors/inducers

As crizotinib is an inhibitor of CYP3A4, patients chronically receiving medications known to be metabolized by CYP3A4 and with narrow therapeutic indices including

pimozide, aripiprazole, triazolam, dihydroergotamine, ergotamine, astemizole, cisapride, terfenadine and halofantrine are not eligible. The topical use of these medications (if applicable) is allowed.

CYP3A4 Inhibitors: Patients chronically receiving drugs that are known potent CYP3A4 inhibitors within 7 days prior to study enrollment, including but not limited to ketoconazole, itraconazole, miconazole, clarithromycin, erythromycin, ritonavir, indinavir, nelfinavir, saquinavir, amprenavir, delavirdine, nefazodone, diltiazem, verapamil, and grapefruit juice are not eligible. The topical use of these medications (if applicable), e.g. 2% ketoconazole cream, is allowed.

CYP3A4 Inducers: Patients chronically receiving drugs that are known potent CYP3A4 inducers within 12 days prior to study enrollment, including but not limited to carbamazepine, phenobarbital, phenytoin, rifabutin, rifampin, tipranavir, ritonavir, and St. John's wort are not eligible. The topical use of these medications (if applicable) is allowed.

This list should not be considered all inclusive: please refer to other resources, such as http://medicine.iupui.edu/clinpharm/ddis/main-table, for additional information.

Steroids: If clinically indicated, short (≤14 days) courses of steroids, such as prednisone or dexamethasone, are allowed for patients with vestibular schwannomas and sudden-onset, transient hearing loss on protocol therapy. All such treatments must be documented in the clinical chart and study records.

5.1.2. Suicidal Risk

Question 24 of the Tinnitus Reaction Questionnaire (TRQ; See Appendix G) elicits possible suicidal ideation. A clinician should review this questionnaire immediately after it is completed and assess for any possible suicidal risk if the answer score is >2. Any concerns of suicidality should be immediately addressed with the responsible institutional investigator.

5.2 Duration of Therapy

Treatment will be administered in continuous treatment cycles of 28 days duration. Temporary treatment interruptions as per protocol, e.g. for toxicity, will not alter the length of cycles or timing of required observations. Initial treatment may continue for up to 12 cycles/48 weeks, or until one of the following criteria applies (whichever occurs first):

- **Primary target tumor** enlarges by ≥20% (as defined in Section 10.1.4.1). **Additional tumors may be monitored for growth or response, but any progression in those tumors will not trigger discontinuation of therapy.** Audiologic responses will be determined, but will not be used as primary response criteria.
- **New or worsening existing medical condition that prevents further protocol-specific administration of treatment**
- Unacceptable adverse event(s), including, but not limited to:
  - Any Grade 4 non-hematological toxicity

  - Grade 4 febrile neutropenia
  - Grade 3 or 4 left ventricular systolic dysfunction
  - Grade 3 or 4 penumonitis
  - Grade 4 prolonged QTc interval
  - Any hematological or non-hematological toxicity requiring treatment interruption for ≥3 weeks
  - Pregnancy during the study period (for female participants)
  - All Grade 4 events thought to be related to crizotinib by the investigator (NOTE: Patients who have an ongoing crizotinib-related Grade 4 or serious adverse event at the time of discontinuation from study treatment will continue to be followed until resolution of the event or until the event is considered irreversible)

- Participant decides to withdraw from the study; or
- General or specific changes in the participant's condition render the participant unacceptable for further treatment in the opinion of the treating investigator.

Re-treatment option: Patients who complete 12 cycles of treatment without disease progression, but within the following 24 weeks show subsequent disease progression (defined as >20% increase in target tumor volume compared to off-treatment volume), will be eligible for re-treatment on study for up to 12 additional cycles/48 additional weeks, provided they still meet study eligibility criteria.

Withdrawal of full consent for a study means that the subject does not wish to receive further investigational treatment and does not wish to or is unable to continue further study participation; subject data up to withdrawal of consent will be included in the subject's study data. Any subject may withdraw full consent to participate in the study at any time during the study. The investigator will discuss with the subject appropriate procedures for withdrawal from the study.

Withdrawal of partial consent means that the subject does not wish to take protocol-specified product any longer but is still willing to collaborate in providing further data by continuing on study (eg, participate in any or all subsequent study visits or procedures). Subjects may decline to continue receiving protocol-specified product at any time during the study. If this occurs, the investigator will discuss with the subject appropriate procedures for withdrawal from protocol-specified therapy. These subjects, as well as those who have stopped receiving protocol-specified product(s) for other reasons (e.g., investigator or sponsor concern) should continue the schedule of study observations at the discretion of the principal investigator.

NOTE: Hearing represents an exploratory endpoint, and therefore Progressive Hearing Loss (PHL) as defined in Section 10.3 in a target or non-target ear, does not require discontinuation of study therapy. A course of high dose glucocorticoids (typically prednisone 60 mg daily for 10 days followed by a taper of 10 mg every day) is permitted at the discretion of the treating physician in cases of sudden hearing loss during treatment (defined as hearing loss with onset over a period of less than 72 hours).

5.3 Duration of Follow-Up

Patients discontinued from the treatment phase of the study for any reason will be evaluated approximately 30 days (30–35 days) after the last dose of study drug taken. Follow-up of adverse event is required until the event or its sequelae resolve or stabilize at a level acceptable to the investigator and sponsor. To learn about the durability of imaging and hearing responses, the study team may contact the local study investigator and request results from MRIs, audiograms and clinical assessments obtained during a period of up to 18 months after discontinuation of study drug.

5.4 Criteria for Removal from Study

Participants will be removed from study when any of the criteria listed in Section 5.2 applies. The reason for study removal and the date the participant was removed must be documented in the data entry system. Alternative care options will be discussed with the participant.

6. EXPECTED TOXICITIES AND DOSING DELAYS/DOSE MODIFICATIONS

A number of measures will be taken to ensure the safety of patients participating in this trial. These measures will be addressed through exclusion criteria (see Section 3.2) and routine monitoring as follows.

Patients enrolled in this study will be evaluated clinically and with standard laboratory tests before and at regular intervals during their participation in this study. Safety evaluations will consist of medical interviews, recording of adverse events, physical examinations, blood pressure, and laboratory measurements. Patients will be evaluated for adverse events (all grades), serious adverse events, and adverse events requiring study drug interruption or discontinuation at each study visit for the duration of their participation in the study.

Dose delays and modifications will be made using the recommendations outlined in Sections 6.2 and 6.3. Toxicity assessments will be done using the CTEP Version 5.0 of the NCI Common Terminology Criteria for Adverse Events (CTCAE) which is identified and located on the CTEP website at https://evs.nci.nih.gov/ftp1/CTCAE/About.html.

If possible, symptoms should be managed symptomatically. In the case of toxicity, appropriate medical treatment should be used (including anti-emetics, anti-diarrheals, etc.).

6.1 Anticipated Toxicities

Toxicities with a possible association to crizotinib treatment are listed in Section 2.2.1.

6.2 Toxicity Management and Dose Modifications/Delays

Any toxicities associated or possibly associated with crizotinib treatment should be managed according to standard medical practice. There is no available antidote for crizotinib.

For patients who are unable to tolerate the protocol-specified dosing schedule, dose adjustments are permitted in order to keep the patient on study drug. If administration of crizotinib must be interrupted because of unacceptable toxicity, drug dosing will be

interrupted or modified according to rules described below. For patients who experience unacceptable toxicity at dose level A (Section 5.0), crizotinib will be discontinued permanently.

Toxicity will be assessed using the NIH-NCI Common Terminology Criteria for Adverse Events, version 5.0.

Recommendations for action based on type of toxicity are shown in the following table:

Toxicity	Actions
Non-hematological toxicity (with the exception of those listed in Section 6.3 Precautions and management of specific toxicities)	
Grade 1–2	**If the toxicity is tolerable to the patient, maintain the same dose. If the toxicity is intolerable to patient, interrupt crizotinib until recovery to grade ≤1. Then reintroduce crizotinib at same dose. If event returns to grade 1-2 and is intolerable to the patient, then interrupt crizotinib until recovery to grade ≤1. Then reintroduce crizotinib at the lower dose level.**
Grade 3	**Interrupt crizotinib until recovery to grade ≤1 or baseline. Then reintroduce crizotinib at a lower dose level.**
Grade 4	**Discontinue crizotinib.**
Hematological toxicity	
Grade 2 Thrombocytopenia (platelets <75, ≥50x10^9/L)	**Interrupt crizotinib until recovery to grade ≤1 (>75 x10^9/L). Then reintroduce crizotinib at initial dose. If thrombocytopenia again returns to grade 2, interrupt crizotinib until recovery to grade ≤1. Then reintroduce crizotinib at the lower dose level.**
Grade 3 Thrombocytopenia (platelets <50, ≥25 x109/L)	**Interrupt crizotinib until recovery to grade ≤1 (platelets ≥75 x10^9/L). Then resume crizotinib at one dose level lower. If grade 3 thrombocytopenia recurs, discontinue crizotinib.**
Grade 4 Thrombocytopenia (platelets <25 x10^9/L)	**Discontinue crizotinib.**
Grade 3 Neutropenia (neutrophils <1, ≥0.5 x10^9/L)	**Interrupt crizotinib until recovery to grade ≤1 (neutrophils ≥1.5 x 10^9/L). Then resume crizotinib at the initial dose. If ANC again returns to Grade 3, hold crizotinib until the ANC ≥1.5 x 10^9/L. Then resume crizotinib dosing at the lower dose level. Discontinue patient from study therapy for a third episode of grade 3 neutropenia.**
Grade 4 Neutropenia (neutrophils <0.5 x10^9/L)	**Interrupt crizotinib until recovery to grade ≤1 (neutrophils ≥1.5 x 10^9/L). Then resume crizotinib at the lower dose level. If grade 3 or grade 4 neutropenia occurs despite this dose reduction, discontinue crizotinib.**
Grade 3 febrile neutropenia (not life-threatening)	**Interrupt crizotinib until resolution of fever and neutropenia to grade ≤1. Hold further crizotinib until the ANC ≥1,500/mm^3 and fever has resolved. Then resume crizotinib at the lower dose level. If febrile neutropenia recurs, discontinue crizotinib.**
Grade 4 febrile neutropenia (life-threatening)	**Discontinue crizotinib.**
Any hematological or non-hematological toxicity requiring interruption for ≥3 weeks	**Discontinue crizotinib**

For additional precautions and management of specific toxicities see Section 6.3.

6.3 Precautions and Management of Specific Toxicities:

Pneumonitis (in absence of infection other pulmonary disease)

Grade 1: Continue at the same dose level.

Grade 2: If suspected, strongly consider administration of 100 mg of oral or intravenous prednisolone in single daily or two divided doses. The suggested dose for patients who develop pulmonary toxicity is methylprednisolone 1 mg/kg IV every 12 hours for a minimum of seven days.
Upon occurrence of pneumonitis, study therapy should be held, and the Study Chair and Research Manager should be notified within 48 hours.
Grades 3–4: If suspected, strongly consider administration of 100 mg of oral or intravenous prednisolone in single daily or two divided doses. The suggested dose for patients who develop pulmonary toxicity is methylprednisolone 1 mg/kg IV every 12 hours for a minimum of seven days. Discontinue crizotinib and do not retreat.

Prolonged QTc interval (as confirmed by a cardiologist)
Grade 1: Continue at the same dose level.
Grade 2: Continue at the same dose level. Assess electrolytes (particularly Ca^{++}, K^{+}, Mg^{+}) and concomitant medications. Correct any electrolyte abnormalities.
Grade 3: Withhold current dose until recovery to Grade ≤1. Assess and correct electrolytes (particularly Ca^{++}, K^{+}, Mg^{+}) and concomitant medications. Upon recovery to Grade ≤1, resume treatment by reducing the dose by 1 dose level if no other cause for QTc prolongation is found, otherwise resume at the same dose.
Grade 4: Discontinue treatment and do not retreat.

Vision disorder/loss of vision
Monitor patients for vision changes while on crizotinib, and withhold crizotinib in patients who develop Grade 2 vision disorder until recovery to less than or equal to Grade 1, then resume crizotinib after dose reduction by 1 dose level. Ophthalmologic examination, including visual acuity and slit lamp examination, should be performed at screening, end of cycle 1, 3, 6, 9, 12, and at the time a visual event is reported, and again if vision disorder persists or worsens in severity. Permanently discontinue crizotinib in patients who develop Grade 3 vision disorder or vision loss.

Increased Creatine phosphokinase level
Grade 1: Continue at the same dose level.
Grade 2: Continue at the same dose level. Assess for any symptoms of muscle pain, muscle cramping, electrolytes (particularly Ca^{++}, K^{+}, Mg^{+}) and concomitant medications. Correct any electrolyte abnormalities.
Grade 3: Withhold current dose until recovery to Grade ≤1. Assess and correct electrolytes (particularly Ca^{++}, K^{+}, Mg^{+}) and concomitant medications. Upon recovery to Grade ≤1, resume treatment by reducing the dose by 1 dose level if no other cause for increased creatine phosphokinase is found, otherwise resume at the same dose.
Grade 4: Discontinue treatment and do not retreat.

7. DRUG FORMULATION AND ADMINISTRATION

7.1 Crizotinib

7.1.1 Description

Other Names. PF-02341066, XALKORI®

Chemical Name. R)-3-[1-(2,6-Dichloro-3-fluorophenyl)ethoxy]-5-[1-(piperidin-4-yl)-1Hpyrazol-4-yl]pyridin-2-amine

Molecular Weight. 450.34 Daltons

Mode of Action. Crizotinib is an orally bioavailable, ATP-competitive, small-molecule multi-kinase inhibitor including c-met, ALK and FAK

Physical Description. White to pale yellow powder

Route of Administration. Oral

For further details and molecule characterization, see the crizotinib Investigator Brochure.

7.1.2 Form

Crizotinib is supplied as 250 and 200 mg hard gelatin capsules. The drug product consists of crizotinib and compendial excipients in a hard gelatin capsule. The capsules are packaged in high-density polyethylene (HDPE).

7.1.3 Storage and Stability

Crizotinib is stored according to the labeled conditions.

7.1.4 Handling

Qualified personnel, familiar with procedures that minimize undue exposure to themselves and the environment, should undertake the preparation, handling, and safe disposal of the chemotherapeutic agent in a self-contained and protective environment.

7.1.5 Availability

Distribution: Crizotinib will be supplied free of charge by Pfizer, Inc. and will be distributed to investigators by a designated central pharmacy (UAB Pharmacy).

7.1.6 Administration

The study drug crizotinib will be self-administered or administered by a parent/legal guardian. The investigator will instruct the patient to take the study drug exactly as specified in the protocol. For patients who are prescribed crizotinib twice daily, it should be administered approximately 12 hours apart, preferably in the morning and evening, at approximately the same time every day with or without food. If the patient vomits or misses a dose, an additional dose should not be taken. The next prescribed dose should be taken at the usual time. Patients who are unable to swallow capsules may take crizotinib as described in Appendix H: Disintegration of Capsules for Patients Unable to Swallow Capsules.

7.1.7 Ordering

Crizotinib will be provided by Pfizer, Inc. and shipped to The UAB Investigational Drug Service (IDS), which is designated as the Central Pharmacy. The UAB IDS is responsible for the transfer and transportation of shipment of crizotinib to study

sites. Transport of crizotinib to study sites must be pre-approved by the sponsor and IRB before any study drug will be transported. Crizotinib will be transported in accordance with specific storage requirements.

When study drug is needed at a study site, an electronic ordering system is utilized for planned transport of study drug, with date and time specifications included in the request. To initiate an order, the UAB IDS ordering form will be sent to nfcop@uab.edu. Once received, NFCTC will approve the order and forward to UAB IDS. UAB IDS will arrange the transportation to the requesting site. Crizotinib will be shipped out 3-7 business days after the UAB IDS ordering form is received. Once the study site receives the shipped product, the form must be completed and sent back to nfcop@uab.edu.

7.1.8 Accountability

The investigator, or a responsible party designated by the investigator, must maintain a careful record of the inventory and disposition of the agent (investigational or free of charge) using the NCI Drug Accountability Record or another comparable drug accountability form. (See the CTEP website at http://ctep.cancer.gov/protocolDevelopment for the "Policy and Guidelines for Accountability and Storage of Investigational Agents" or to obtain a copy of the drug accountability form.)

Sites will be required to send site drug accountability records to Operations Center on a quarterly basis. The site study drug accountability records will be checked against UAB Pharmacy study drug administration data accountability records.

Drug accountability records should be submitted to nfcop@uab.edu.

7.1.9 Destruction and Return

At the end of the study, unused supplies of crizotinib should be destroyed according to institutional policies. Destruction will be documented in the Drug Accountability Record Form.

8. CORRELATIVE/SPECIAL STUDIES

8.1 Pharmacokinetic Studies

N/A

8.2 Pharmacodynamic Studies

N/A

8.3 Laboratory correlative markers

8.3.1 Candidate Biomarkers

There is no published data on molecular biomarkers for crizotinib in patients with NF2, and this study proposes to analyze baseline biomarkers with respect to outcomes (response) as well as the possible effect of crizotinib on biomarker levels over time.

We will collect blood samples from the patients before and during the course of treatment (see study table). Analysis of plasma levels of total and phosphorylated MET, FAK, ROS and ALK, which represent known molecular targets of crizotinib, as well as HGF, will be performed using ELISA assays. The level of these candidate biomarkers during treatment with crizotinib will be compared to the baseline level. Samples will not be collected during the retreatment therapy.

8.3.2 Collection of specimen(s)

Sample Collection Time Points

Blood samples will be obtained for protein analysis of candidate biomarkers for crizotinib therapy when clinical labs are drawn at the following time points:

- Prior to initiating therapy (cycle 1)
- End of cycles 1, 3 and 12

Blood collection (needed for each time point sample)

- Collect 8 ml of blood in a polypropylene tube containing the anticoagulant EDTA. Tubes should be pre-cooled in an ice bath.
  - SARSTEDT Monovette® EDTA KE (9 ml), Part # 02.1333.001 ***or***
  - Becton-Dickinson Vacutainer™ K2E (10 ml), Part # 367525 ***or***
  - Greiner Bio-One Vacuette® K3E EDTA K3 (9 ml), Part 455036
- Blood tubes must be gently inverted several times after collection to ensure thorough mixing of EDTA with the sample to prevent clotting.
- Cool all tubes in an ice bath immediately after collection.
- **Glass tubes MUST NOT be used** as they may break during transport and freeze-thaw cycles.
- **Heparin MUST NOT be used** as an anticoagulant as it may interfere with downstream methodology.

8.3.3 Handling of specimens

Centrifuge tubes collected at 700 G for 20 minutes at 4C° (without brakes) within 30 minutes of collection.

Prepare two labels each printed with Study-No., patient ID, and day/time of sample collection (24-hour clock format, *i.e.*, 6:30 pm = 18:30). A label example is provided below:

Study-No.: NF110	Investigator:
Patient-ID:	Sample Type: *Plasma*
Date of sampling: *(mm/dd/yy)*	Time of Sampling: *(hh:mm) (24-hr format)*

- Plasma is pipetted in 1 ml aliquots into two red-labeled Nalgene cryovials.
- Clearly label tubes as "plasma" and store at -80°C.
- When samples from all time-points have been collected (after the patient goes off trial) the plasma samples should be shipped to the Kissil Laboratory on DRY ICE in a Styrofoam box. If a deep freezer is not available on site, the plasma sample should be kept and shipped on dry ice on the same day.
- Specimens should be shipped Monday to Wednesday only by overnight FedEx to the Kissil Laboratory at the address provided below.

8.3.4 Shipping and analysis of specimen(s)

Multiple specimens may be batched together for shipping.

Specimens should be shipped Monday to Wednesday only by overnight FedEx to the following address:

H. Lee Moffitt Cancer Center
University of South Florida
Attn: Kissil Lab, SRB 22234
3011 W Holly Drive
Tampa, FL 33612
Phone: 813-745-0019
Scott.Troutman@moffitt.org

On the day of shipment, the study coordinator will notify the Kissil Laboratory via email: Scott.Troutman@moffitt.org to expect the upcoming shipment and include the estimated date of arrival and FedEX tracking number.
All analysis will be performed within the Kissil Laboratory.

8.3.5 Future use of specimen(s)

Leftover blood specimens will be stored for future use in NF studies approved by the IRB. Specimens will be labeled as noted above (see 8.3.3) and stored indefinitely in the Kissil Laboratory. Neither cell lines nor commercial products will be developed from the blood specimens. Specimens will be banked under the current study protocol. Access to specimen samples will be limited to research personnel approved by the IRB. Subjects who wish to have their samples removed from storage (e.g., withdraw consent for biomarker storage) should contact the study PI who will work with the Kissil Laboratory to have the sample destroyed.

9. SCHEDULE OF EVALUATIONS

Baseline evaluations are to be conducted within 28 days prior to start of protocol therapy.

The baseline (prestudy) visit includes drug dispensing, and repeat baseline evaluations if time between first screening and dispensing exceeds 28 days. *Otherwise, baseline evaluations need not be repeated, unless there is a significant change in the patient's clinical condition.*

Each cycle is 28 days. ***Interruptions of study drug (e.g., due to toxicity) do not affect the length of cycles***. All evaluations should be obtained at the end of a cycle, or within 7 days *prior* to starting a new cycle.

The schedule of evaluations, including all required observations, is shown in Appendix A.

10. MEASUREMENT OF EFFECT

10.1 Antitumor Effect– Solid Tumors

For the purposes of this study, patients should be re-evaluated for response every 12 weeks. Response and progression will be evaluated in this study using the criteria proposed by Dombi and colleagues[29] for neurofibromatosis-associated lesions.

10.1.1 Definitions

Evaluable for toxicity: All patients will be evaluable for toxicity from the time of their first treatment with crizotinib.

Evaluable for objective response: Only those patients who have received at least one cycle (4 weeks) of therapy and have had their disease re-evaluated will be considered evaluable for response. These patients will have their response classified according to the definitions stated below.

10.1.2 Disease Parameters

Measurable disease: Measurable lesions are defined as those that can be accurately measured using volumetric analysis of cranial MRI scans. All study MRI scans should include standard brain imaging sequences as well as fine cuts through the internal auditory canal (1 mm slice, no gaps) to image small tumors. In patients who have had surgery for tumors in the cerebellopontine angle, fat-saturation should be performed with the post-contrast sequences to compensate for the possible presence of post-operative fat packing.

NOTE: Tumor lesions that are situated in a previously irradiated area are considered measurable.

Non-measurable disease: Non-measurable lesions include skull-base lesions that are obscured by artifact from auditory brainstem implants (ABIs) or lesions whose margins are completely obscured by neighboring tumors (*i.e.*, "collision" tumors).

Target lesions: Investigators should identify a single target lesion in all subjects. The target lesion in this study is the *progressive* VS (*e.g.*, the lesion that is enlarging) that led to enrollment in the protocol. In cases where subjects have bilateral progressive tumors that fulfill eligibility criteria, the target lesion should be the larger of the two tumors on imaging. Target lesions should be identified at baseline

and measured using volumetric analysis of the baseline MRI scan. The baseline volumetric MRI scan will be used as reference for comparison of all future MRI scans to characterize the objective tumor response.

Non-target lesions: Non-target lesions in this study include VS contralateral to the target lesion (if present). The baseline contralateral VS will be used as reference by which to characterize the objective tumor response.

NOTE: Histologic confirmation of tumor type is not required. Designation of tumor type will be determined by the radiographic appearance by the study radiologist.

10.1.3 Methods for Evaluation of Measurable Disease

All measurements should be taken and recorded in metric notation [*i.e.*, cubic centimeters (cm^3) and in millimeters (or decimal fractions of centimeters)] for linear measures. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.

The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up.

Cranial MRI. Volumetric analysis of MRI scans should be performed on post-contrast Gradient Echo 3D imaging with fine cuts **(1 mm slices, no skip)** preferably through the entire brain, and at the minimum including the entire target tumor (see Appendix F).

10.1.4 Response Criteria

10.1.4.1 Evaluation of Target Lesions

For the purpose of this study, radiographic response will be defined by the change in tumor volume compared to baseline, as previously defined in previous studies in NF1.[15] Volumetric measurements by central review (Appendix F) will be used to determine eligibility and continuation on treatment. Hearing will be measured in standardized fashion as described in Appendix E. Hearing response (Section 10.3) will be defined by the change in word recognition scores, taking as reference the baseline word recognition score (Appendix B and Section 10.3).

Radiographic Response (RR): At least a 20% decrease in the volume of the target lesion, taking as reference the baseline volume.

Progressive Disease (PD): At least a 20% increase in the volume of the target lesion, taking as reference the lowest tumor volume during treatment volume.

Stable Disease (SD): Does not meet criteria for radiographic response or for progressive disease.

Unknown (UN): Assessment of target lesions cannot be made due to insufficient or unevaluable data. In

this case, a concise explanation must be given.

10.1.4.2 Evaluation of Non-Target Lesions

Radiographic evaluations should be calculated separately for non-target lesions (contralateral VS).

Radiographic Response (RR): At least a 20% decrease in the non-target lesions, taking as reference the baseline volumes.

Stable Disease (SD): Does not meet criteria for radiographic response or progressive disease.

Progressive Disease (PD): At least a 20% increase in the volume of the non-target lesions, taking as reference the lowest tumor volume during treatment volume.

Although a clear progression of "non-target" lesions only is exceptional, the opinion of the treating physician should prevail in such circumstances, and the progression status should be confirmed at a later time by the review panel (or Principal Investigator).

10.1.4.3 Evaluation of Best Overall Response

The best overall response is the best response for the target vestibular schwannoma recorded from the start of the treatment until disease progression (taking as reference for progressive disease the hearing measurements recorded at baseline and the minimal tumor volume during treatment).

Note: Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be reported as "*symptomatic deterioration"*. Every effort should be made to document the objective progression even after discontinuation of treatment.

10.1.5 Duration of Response

Hearing evaluations should be performed for non-target (contralateral) VS if present and if hearing is present in the ipsilateral ear.

Duration of hearing response: The duration of hearing response (HR) is measured from the time that measurement criteria are met for HR until the first date that the word recognition score decreases beneath the upper limit of the 95% critical difference of the baseline word recognition score.

Duration of stable hearing: Stable disease is measured from the start of the treatment until the criteria for progression are met, taking as reference the word recognition score recorded at baseline.

Duration of radiographic response: The duration of radiographic response is measured from the time measurement criteria are met for RR until the first date that progressive disease is objectively documented (taking as reference for progressive disease the lowest tumor volume during treatment).

Duration of stable disease: Stable disease is measured from the start of the treatment until the criteria for progression are met, taking as reference the measurements recorded at baseline.

10.1.6 Progression-Free Survival: Freedom from tumor progression and freedom from progressive hearing loss

The proportion of patients alive and free from progressive tumor growth after 3, 6, 12, and 12 cycles from the start of treatment will be determined. The proportion of patients alive and free from progressive hearing loss after 3, 6, 9, and 12 cycles will be determined.

10.1.7 Response Review

Central review of audiologic and radiologic data will be performed at MSKCC and MGH. Audiograms collected during the study period will be sent to the Operations Center and to the team at MSKCC for analysis. A study member (under the guidance of lead study audiologist Kerri O'Connor, AuD, CCC-A) will extract data for review by the Principal Investigator and lead audiologist.

MRI images from enrolled subjects will be transmitted by the Partners Secure File Transfer (sFTP) to the Dana-Farber/Harvard Cancer Center Tumor Imaging Metric Core (TIMC) for central analysis of tumor volume. The procedures for clinical sites to upload MRI scans are provided in Appendix F.

Enhancing lesions will be outlined using a volumetric approach that includes outlining each enhancing voxel on post-contrast scans and then summing the voxels to calculate an overall lesion volume. A report will be generated and then sent to the Principal Investigator at each study site within 2 business days of receiving the files.

A secondary review of volumetric image analyses (identified by participant code) done by TIMC may be performed at MSKCC through the TIMC website. If necessary, re-review of volumetric measurements by TIMC may be requested.

10.2 Antitumor Effect – Hematologic Tumors

N/A

10.3 Other Response Parameters: Hearing Response

Hearing Response (HR): Improvement in word recognition score above the 95% critical difference, taking as reference the baseline word recognition score (Appendix B).

Stable Hearing (SH): Persistence of word recognition score within the 95% critical difference, taking as reference the baseline word recognition score (Appendix B).

Progressive Hearing Loss (PHL): Decline in word recognition score below the 95% critical difference, taking as reference the baseline word recognition score (Appendix B).

11. ADVERSE EVENT REPORTING REQUIREMENTS

11.1 Definitions

11.1.1 Specification of Safety Variables

Safety assessments will consist of monitoring and reporting adverse events (AEs) and serious adverse events (SAEs) that are considered related to crizotinib, all events of death, and any study specific issue of concern.

11.1.2 Adverse Events

An AE is any unfavorable and unintended sign, symptom, or disease temporally associated with the use of an investigational medicinal product (IMP) or other protocol-imposed intervention, regardless of attribution.

This includes the following:

- AEs not previously observed in the subject that emerge during the protocol-specified AE reporting period, including signs or symptoms associated with neurofibromatosis 2 that were not present prior to the AE reporting period.
- Complications that occur as a result of protocol-mandated interventions (*e.g.*, invasive procedures such as cardiac catheterizations).
- If applicable, AEs that occur prior to assignment of study treatment associated with medication washout, no treatment run-in, or other protocol-mandated intervention.
- Preexisting medical conditions (other than the condition being studied) judged by the investigator to have worsened in severity or frequency or changed in character during the protocol-specified AE reporting period.

Abnormal laboratory values or diagnostic test results constitute adverse events only if they are accompanied by clinical symptoms, result in a change in study treatment or require a medical intervention, or are considered to be clinically significant in the investigator's judgment.

Abnormal vital signs constitute adverse events only if they are accompanied by clinical symptoms, results in a change in study treatment, or require a medical

intervention, or are considered to be clinically significant in the investigators judgment.

In the event of an adverse event the first concern will be for the safety of the subject.

11.1.3 Serious Adverse Event (SAE)

Investigators are required to report to the Operations Center ANY serious treatment emergent adverse event as soon as possible. The Operations Center will report ANY serious treatment emergent adverse event to Pfizer Drug Safety, FDA, UAB IRB and the Protocol Chair.

An AE should be classified as an SAE if the following criteria are met:

- It results in death (*i.e.*, the AE actually causes or leads to death).
- It is life threatening (*i.e.*, the AE, in the view of the investigator, places the subject at immediate risk of death. It does not include an AE that, had it occurred in a more severe form, might have caused death.).
- It requires or prolongs inpatient hospitalization.
- It results in persistent or significant disability/incapacity (*i.e.*, the AE results in substantial disruption of the subject's ability to conduct normal life functions).
- It results in a congenital anomaly/birth defect in a neonate/infant born to a mother exposed to the IMP.
- It is considered a significant medical event by the investigator based on medical judgment (*e.g.*, may jeopardize the subject or may require medical/surgical intervention to prevent one of the outcomes listed above).

Events **not** considered to be serious adverse events are hospitalizations for:

- routine treatment or monitoring of the studied indication, not associated with any deterioration in condition, or for elective procedures;
- elective or pre-planned treatment for a pre-existing condition that did not worsen;
- emergency outpatient treatment for an event not fulfilling the serious criteria outlined above and not resulting in inpatient admission; and/or respite care.

11.1.4 Expectedness

Adverse events can be 'Expected' or 'Unexpected.'

11.1.4.1 Expected adverse event

Expected adverse events are those that have been previously identified as resulting from administration of the agent. For the purposes of this study, expected adverse events are those adverse events that are listed or characterized in the Package Insert or current Investigator Brochure (I.B).

11.1.4.2 Unexpected adverse event

For the purposes of this study, unexpected adverse events are those not listed in the Package Insert or current Investigator Brochure or not identified. This includes adverse events for which the specificity or severity is not consistent with the description in the package insert or I.B. For example, under this definition, hepatic necrosis would be unexpected if the package insert or I.B. only referred to elevated hepatic enzymes or hepatitis.

11.2 Methods and Timing for Assessing and Recording Safety Variable

The investigator is responsible for ensuring that all AEs and SAEs that are observed or reported during the study, are collected and reported to the FDA, appropriate IRB(s), the Sponsor, and Pfizer, Inc. in accordance with 21 CFR 312.32 (IND Safety Reports) and with the most recent approved protocol document.

11.2.1 Adverse Event Reporting Period

The study period during which all AEs and SAEs must be reported begins after the time of first dose of study drug. The reporting period for both AEs and SAEs ends 30 days following the last administration of study treatment or study discontinuation/termination, whichever is earlier. After this period, investigators should only report SAEs that are attributed to prior study treatment.

11.2.2 Assessment of Adverse Events

All AEs and SAEs whether volunteered by the subject, discovered by study personnel during questioning, or detected through physical examination, laboratory test, or other means will be reported in the participant's medical record and on the appropriate study-specific case report forms and entered into the NF Consortium's web-based data entry system. Each reported AE or SAE will be described by its duration (*i.e.*, start and end dates), regulatory seriousness criteria if applicable, suspected relationship to the crizotinib (see following guidance), and actions taken. To ensure consistency of AE and SAE causality assessments, investigators should apply the following general guideline:

Yes: There is a plausible temporal relationship between the onset of the AE and administration of crizotinib; and the AE cannot be readily explained by the subject's clinical state, intercurrent illness, or concomitant therapies; and/or the AE follows a known pattern of response to crizotinib; and/or the AE abates or resolves upon discontinuation of the crizotinib or dose reduction and, if applicable, reappears upon re-challenge.

No: Evidence exists that the AE has an etiology other than the crizotinib (*e.g.*, preexisting medical condition, underlying disease, intercurrent illness, or concomitant medication); and/or the AE has no plausible temporal relationship to crizotinib administration (*e.g.*, cancer diagnosed 2 days after first dose of study drug).

11.3 Procedures for Eliciting, Recording, and Reporting Adverse Events

11.3.1 Eliciting Adverse Events

A consistent methodology for eliciting AEs at all subject evaluation time points should be adopted. Examples of non-directive questions include:

- "How have you felt since your last clinical visit?"
- "Have you had any new or changed health problems since you were last here?"

11.3.2 Specific Instructions for Recording Adverse Events

Investigators should use correct medical terminology/concepts when reporting AEs or SAEs. Avoid colloquialisms and abbreviations. The descriptions and grading scales found in the CTEP Version 5.0 of the NCI Common Terminology Criteria for Adverse Events (CTCAE v. 5.0) will be utilized for AE reporting. The CTEP Version 5.0 of the CTCAE is identified and located on the CTEP website at https://evs.nci.nih.gov/ftp1/CTCAE/About.html.
All appropriate treatment areas should have access to a copy of the CTEP Version 5.0 of CTCAE.

11.3.2.1 Diagnosis vs. Signs and Symptoms

If known at the time of reporting, a diagnosis should be reported rather than individual signs and symptoms (*e.g.*, record only liver failure or hepatitis rather than jaundice, asterixis, and elevated transaminases). However, if a constellation of signs and/or symptoms cannot be medically characterized as a single diagnosis or syndrome at the time of reporting, it is ok to report the information that is currently available. If a diagnosis is subsequently established, it should be reported as follow-up information.

11.3.2.2 Deaths

All deaths that occur during the protocol-specified AE reporting period, regardless of attribution, will be reported to the appropriate parties. When recording a death, the event or condition that caused or contributed to the fatal outcome should be reported as the single medical concept. If the cause of death is unknown and cannot be ascertained at the time of reporting, report "Unexplained Death".

11.3.2.3 Preexisting Medical Conditions

A preexisting medical condition is one that is present at the start of the study. Such conditions should be reported as medical and surgical history. A preexisting medical condition should be re-assessed throughout the trial and reported as an AE or SAE only if the frequency, severity, or character of the condition worsens during the study. When reporting such events, it is important to convey the concept that the preexisting condition has changed by including applicable descriptors (*e.g.*, "more frequent headaches").

11.3.2.4 Hospitalizations for Medical or Surgical Procedures

Any AE that results in hospitalization or prolonged hospitalization should be documented and reported as an SAE. If a subject is hospitalized to undergo a medical or surgical procedure as a result of an AE, the event

responsible for the procedure, not the procedure itself, should be reported as the SAE. For example, if a subject is hospitalized to undergo coronary bypass surgery, record the heart condition that necessitated the bypass as the SAE.

Hospitalizations for the following reasons do not require reporting:

- Hospitalization or prolonged hospitalization for diagnostic or elective surgical procedures for preexisting conditions;
- Hospitalization or prolonged hospitalization required to allow efficacy measurement for the study; or
- Hospitalization or prolonged hospitalization for scheduled therapy of the target disease of the study.

11.3.2.5 Pregnancy

If a female subject becomes pregnant while receiving investigational therapy or within 28 days after the last dose of study drug, a report should be completed and expeditiously submitted to the Pfizer, Inc. as an SAE. Follow-up to obtain the outcome of the pregnancy should also occur. Abortion, whether accidental, therapeutic, or spontaneous, should always be classified as serious, and expeditiously reported as an SAE. Similarly, any congenital anomaly/birth defect in a child born to a female subject exposed to the crizotinib should be reported as an SAE.

11.3.2.6 Post-Study Adverse Events

The investigator should expeditiously report any SAE occurring after a subject has completed or discontinued study participation if attributed to prior crizotinib exposure. If the investigator should become aware of the development of cancer or a congenital anomaly in a subsequently conceived offspring of a female subject who participated in the study, this should be reported as an SAE.

11.3.2.7 Reconciliation/Pfizer

The Sponsor agrees to conduct reconciliation for the product. Pfizer and the Sponsor will agree to the reconciliation periodicity and format, but agree at minimum to exchange monthly line listings of cases received by the other party. The responsible individuals for each party shall handle the matter on a case-by-case basis until satisfactory resolution.

11.4 SAE Expedited Reporting/Pfizer

All serious adverse events that occur after the initial dose of study treatment, during treatment, or within 30 days of the last dose of treatment must be reported to the NF Consortium using the SAE reporting form in eDCS, and MedWatch Form 3500a. This includes events meeting the criteria outlined in Section 11.1.3, as well as the following:

- Grade 3 (severe) events that are unexpected and at least possibly related/associated with the intervention.

- All Grade 4 (life-threatening or disabling) events that are unexpected or not specifically listed in the protocol as not requiring reporting.
- All Grade 5 (fatal) events while the participant is enrolled and actively participating in the trial OR when the event occurs within 30 days of the last study intervention.

Note: If the participant is in long term follow-up, report the death at the time of continuing review.

All SAEs should be entered on the SAE CRF in the eDCS and recorded on a MedWatch 3500a Form and sent to NF Consortium Operations Center (Attn: Research Nurse Manager; nfcop@uab.edu) and Dr. Matthias Karajannis (karajanm@mskcc.org) within 24 hours.

The NF Consortium is also responsible for notifying the UAB IRB, DSMB and appropriate Regulatory agencies, including any Pfizer-related reporting requirements.

MedWatch 3500a Reporting Guidelines:
For Investigator-Sponsored Studies, some additional reporting requirements for the FDA apply in accordance with the guidance set forth in 21 CFR 314.80.

The report should include the following information within the Event Description (Section B.5) of the MedWatch 3500a form:

- Treatment regimen (dosing frequency, combination therapy)
- Protocol description (and number, if assigned)
- Description of event, severity, treatment, and outcome, if known
- Supportive laboratory results and diagnostics
- Investigator's assessment of the relationship of the adverse event to each investigational product and suspect medication

All related adverse events that are both serious and unexpected will be sent to the FDA on Form FDA 3500A MedWatch Form (http://www.fda.gov/medwatch/getforms.htm) by Dr. Bruce Korf and the Operations Center. MedWatch Form will be faxed to:

MedWatch
Fax: 1-800-FDA-0178

MedWatch 3500A (Mandatory Reporting) form is available at:
http://www.fda.gov/downloads/AboutFDA/ReportsManualsForms/Forms/UCM048334.pdf

Additional Pfizer Expedited Reporting Guidelines

To ensure patient safety, every SAE, regardless of suspected causality, occurring after the subject receives first crizotinib dose and until at least 30 days after the last administration of crizotinib must be reported to Pfizer.

Even though there may not be an associated SAE, exposure to the Pfizer Product during pregnancy and exposure to the Pfizer Product during lactation are reportable.

SAEs that occur after completion of the reporting time period as defined above are reportable to Pfizer if the Investigator suspects a causal relationship between the Pfizer product and the SAE.

Information about all SAEs will be collected and recorded on the MedWatch Form 3500a. All applicable sections of the form must be completed in order to provide a clinically thorough report. The investigator must assess and record the relationship of each SAE to each specific study treatment on the MedWatch Form 3500a and email to NF Operations Center (nfcop@uab.edu) immediately for a death or life-threatening event, and within 24 hours for all other reportable SAEs.

The NF Operations Center will forward any SAE report within 24 hours (or immediately if the event is fatal or life-threatening) of learning of the event to Pfizer followed by a complete written report within 24 hours of the initial 24-hour report.

The original copy of the MedWatch Form must be kept with the case report form documentation at the site.

When new, updated, or corrected information about a previously reported SAE is obtained, a follow-up report should be submitted on a new MedWatch Form that includes the data that are new or revised from the previous report. Follow-up information should never be added to a previously submitted report form. Ensure that any new events included on a follow-up report are marked as serious and a causality assessment is provided for each of them. The follow-up information should describe whether the event has resolved or continues, if and how it was treated, and whether the patient continued or withdrew from study participation.

Pfizer US Clinical Trial Department Fax: 1-866-997-8322

11.5 Adverse Event Reporting to the US Army

This trial receives funding by a Grant of the U.S. Army. Per US Army request, adverse events will therefore also be reported by the NF Consortium Operations Center to the US Army as follows: All adverse events, which require prompt reporting (as defined in Section 11.1.3 will be immediately reported by telephone to the USAMRMC Deputy Chief of Staff for Regulatory Compliance and Quality (301-619-2165) (non-duty hours call 301-619-2165, email or by facsimile to (301-619-7803). A written report will follow the initial contact within three working days to the NF Consortium Operations Center. The NF Operations Center will notify all appropriate study regulatory personnel and send a written report to the U.S. Army Medical Research and Materiel Command within seven

working days of the initial contact to ATTN: MCMR-RCQ, 504 Scott Street, Fort Detrick, Maryland 21702-5012 or email.

11.6 Reporting Requirements

The study must be conducted in compliance with FDA regulations, local safety reporting requirements, and reporting requirements of the Principal Investigator.

Each investigative site will be responsible for reporting SAEs that occur at that institution to their respective IRB. It is the responsibility of the Operations Center to report serious adverse events (SAEs) to the study sponsor and/or others as previously described.

11.6.1 Non-Serious Adverse Event Reporting

Non-serious adverse events will be reported to the NF Consortium via the web-based data entry system on the adverse event screen(s). All non-serious adverse events will be forwarded, at most, on a quarterly report to Pfizer by the NF Consortium.

11.7 Reporting to the Institutional Review Board (IRB)

All participating study sites will report all serious adverse events directly to their institutional regulatory agencies according to their institutional policies and procedures.

A copy of the submitted institutional SAE documentation should be forwarded to:

NF Consortium Operations Center
Attn: Juliette Southworth
Phone: (205) 975-4075
Email: jsouthworth@uab.edu

All SAE reports will be reported to Study PI by the Operations Center via emails from the data entry system.

11.8 Reporting to the NIH Office of Biotechnology Activities (OBA)

N/A

11.9 Reporting to the Institutional Biosafety Committee (IBC)

N/A

11.10 Reporting to Hospital Risk Management

Participating investigators will report to their local Risk Management office any subject safety reports or sentinel events that require reporting according to institutional policy.

12. DATA AND SAFETY MONITORING

12.1 Data Reporting

12.1.1 Method

The NF Consortium Data Coordinating Center at CHOP will collect, manage, and monitor data for this study.

12.1.2 Data Collection

The trial is being conducted by the NF Consortium. Case reports forms developed by the NF Consortium Operations Center will be used for submitting clinical data to the Operations Center. Data must be submitted to the Operations Center within two weeks of completing each required evaluation while the subject is on study.

The NF Consortium DCC is responsible for compiling and providing the data to the Principal Investigator for review.

12.2 Safety Meetings

The trial PI and clinical coordinator will review the study progress regularly. Patients entered on the trial and adverse events will be reviewed to ensure that the study is implemented as outlined in the protocol. Monthly reports will be generated by the NF Consortium to assess completeness and timeliness of data. There will be monthly phone conferences between the NF Consortium and the Principal Investigator to address QA issues. A Data Safety Monitoring Board has been established for the purpose of ensuring data compliance and regular monitoring of this trial.

A research monitor has been selected for this study (Diana Osorio, MD, MPH, see roster at beginning of protocol). The research monitor is a qualified physician and is not associated with this particular protocol. The monitor will work closely with the DSMB and Protocol Chair to monitor the participants' treatment while on this study.

The research monitor is required to review all unanticipated problems involving risk to subjects or others, serious adverse events and all subject deaths associated with the protocol and provide an unbiased written report of the event. At a minimum, the research monitor should comment on the outcomes of the event or problem, and in the case of a serious adverse event or death, comment on the relationship to participation in the study. The research monitor should also indicate whether he/she concurs with the details of the report provided by the study investigator. Reports for events determined by either the investigator or research monitor to be possibly or definitely related to participation and reports of events resulting in death should be promptly forwarded to the USAMRMC ORP OHRO.

12.3 Monitoring

Involvement in this study as a participating investigator implies acceptance of potential audits or inspections, including source data verification, by representatives designated by the Protocol Chair or NF Consortium Operations Center. The purpose of these audits or inspections is to examine study-related activities and documents to determine whether these activities were conducted and data were recorded, analyzed, and accurately

reported in accordance with the protocol, institutional policy, Good Clinical Practice (GCP), and any applicable regulatory requirements and to ensure subject safety.

All data will be monitored for timeliness of submission, completeness, and adherence to protocol requirements.

12.4 Privacy

Data provided must be treated in strictest confidence. No information provided from individual subject's records may be discussed with anyone other than those individuals mentioned in the collaborative research agreement. Data may not be released in any form except as provided in the agreement.

12.5 Confidentiality

Each subject enrolled will, from that point forward, be identified by a unique identifier (study number). This study number will also be used for any research specimens collected and shipped to analysts outside of the NF Consortium. All records generated will be stored in a locked office area, only accessible to study personnel. Clinical information will be accessed, according to HIPAA requirements, by study personnel to complete study documents, as needed**.** For standard of care procedure reimbursement, clinical information may be shared with insurers.

13. REGULATORY CONSIDERATIONS

13.1 Protocol Review and Amendments

This protocol, the proposed informed consent and all forms of participant information related to the study (*e.g.*, advertisements used to recruit participants) and any other necessary documents must be submitted, reviewed and approved by a properly constituted IRB governing each study location.

Any changes made to the protocol must be submitted as amendments and must be approved by the UAB IRB and the DOD prior to implementation. Any changes in study conduct must be reported to the IRB. The NF Consortium Operations Center will disseminate protocol amendment information to all participating investigators.

All decisions of the IRB concerning the conduct of the study must be made in writing.

13.2 Informed Consent

All participants must be provided a consent form describing this study and providing sufficient information for participants to make an informed decision about their participation in this study. The formal consent of a participant, using the IRB approved consent form, must be obtained before the participant is involved in any study-related procedure. The consent form must be signed and dated by the participant or the participant's legally authorized representative, and by the person obtaining the consent. The process of informed consent should also be documented in the subject research record. The participant must be given a copy of the signed and dated consent document.

The original signed copy of the consent document must be retained in the medical record or research file.

13.3 Ethics and Good Clinical Practice (GCP)

This study is to be conducted according to the following considerations, which represent good and sound research practice:

- E6 Good Clinical Practice: Consolidated Guidance
  www.fda.gov/downloads/RegulatoryInformation/Guidances/UCM129515.pdf

- US Code of Federal Regulations (CFR) governing clinical study conduct and ethical principles that have their origin in the Declaration of Helsinki

    - Title 21 Part 11 – Electronic Records; Electronic Signatures
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr11_02.html

    - Title 21 Part 50 – Protection of Human Subjects
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr50_02.html

    - Title 21 Part 54 – Financial Disclosure by Clinical Investigators
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr54_02.html

    - Title 21 Part 56 – Institutional Review Boards
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr56_02.html

    - Title 21 Part 312 – Investigational New Drug Application
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr312_02.html

State laws

It is understood that deviations from the protocol should be avoided, except when necessary to eliminate an immediate hazard to a research participant. In such case, the deviation must be reported to the IRB according to the local reporting policy.

13.4 Study Documentation

The investigator must prepare and maintain adequate and accurate case histories designed to record all observations and other data pertinent to the study for each research participant, including all protocol deviations. This information enables the study to be fully documented and the study data to be subsequently verified.

Original source documents supporting entries in the electronic data entry system include but are not limited to hospital records, clinical charts, laboratory and pharmacy records, recorded data from automated instruments, microfiches, photographic negatives, microfilm or magnetic media, and/or x-rays. To standardize data collection and entry, study staff can complete a hard copy of the eCRFs prior to entry into the web-based data collection system, but it is not mandated.

13.5 Records Retention

All study-related documents must be retained for the maximum period required by applicable federal regulations and guidelines or institutional policies.

13.6 Multi-center Guidelines

This protocol will adhere to the policies and requirements of NF Consortium IRB and the Sponsor. The specific responsibilities of the Protocol Chair, NF Consortium Operations Center, and Participating Institutions are presented in the Multi-Center Data and Safety Monitoring Plan.

- The Protocol Chair and NF Consortium Operations Center are responsible for distributing all IND Action Letters or Safety Reports to all participating institutions for submission to their individual IRBs for action as required.
- Mechanisms will be in place to ensure quality assurance, protocol compliance, and adverse event reporting at each site.
- Drug Distribution Center - UAB Research Pharmacy

  According to 21 CFR 312.6, UAB IDS will label the packaging of the investigational drug with the following statement: "Caution: New Drug - Limited by Federal law to investigational use".

  UAB IDS will maintain records showing the receipt, shipment, and other pertinent information of the investigational drug including:

  - Name of the investigator to whom the drug is shipped

  - Date, quantity, and batch number of each shipment

  - Return, destruction, or other appropriate disposition of unused study drug

To request study drug, the UAB Ordering Form will be used and all correspondence regarding ordering be sent to nfcop@uab.edu.

13.7 Cooperative Research and Development Agreement (CRADA)/Clinical Trials Agreement (CTA)

N/A

14. STATISTICAL CONSIDERATIONS

14.1 Study Design/Endpoints

To evaluate efficacy, imaging response is treated as a binary variable whereby patients who achieve a volumetric response in the target tumor at any point within 12 months from beginning of therapy are considered responders and all other patients are nonresponders. A 2-stage Simon design will be used.[34] Crizotinib will be considered not of sufficient interest for further evaluation if the true response rate in *evaluable subjects* (as defined below) is 5% (p0) or less and considered active if the true response rate is 25% or greater (p1). With alpha (Type I error, probability of rejecting crizotinib with a true response rate 5% under H0) set at 0.05 and beta (Type II error, probability of failing to reject a treatment

with true response rate 25% under Ha) set at 0.2, the required sample size is 9 patients for stage 1 and an additional 8 patients for stage 2. If there are no responses after the first stage is completed, crizotinib will be deemed ineffective and the trial terminated. Recruitment will be halted after the 9th evaluable patient is enrolled (stage 1) until at least one response is observed during the first 12 cycles. Crizotinib will be considered effective and of interest for further study if after successful completion of both stages, the cumulative number of responses is ≥3. Using this 2-stage design, the probability of early termination is 0.63 if the true response rate to crizotinib is 5% or lower.

Stage	Cumulative number of patients with volumetric responses	Decision
Stage 1: enter 9 patients	0	Terminate the trial: agent **ineffective**
	1 or more	**Inconclusive** result: continue trial (proceed to Stage 2)
Stage 2: enter 8 additional patients	2 or less	Terminate the trial: agent **ineffective**
	3 or more	Continue the trial to obtain full sample size: agent **effective**

14.2 Sample Size/Accrual Rate

Subjects of both genders, from all racial and ethnic groups are eligible for this trial if they meet the criteria outlined in Section 3.1. To date, there is no information that suggests differences in drug metabolism or disease response among racial or ethnic groups or between the genders, indicating that results of the trial will be applicable to all groups. Efforts will be made to extend the accrual to a representative population, but in a phase II study with limited accrual, a balance must be struck between subject safety considerations and limitations on the number of individuals exposed to potentially toxic or ineffective treatments on the one hand, and the need to explore gender, racial, and ethnic aspects of clinical research on the other. If differences in outcome that correlate to gender, age, racial, or ethnic identity are noted, accrual may be expanded or additional studies may be performed to investigate those differences more fully.

A maximum of 19 eligible subjects will be accrued into the trial, allowing for up to 10% of non-evaluable patients. We believe this 10% rate is reasonable given our prior experience with NF2 phase 2 clinical trials.

The expected accrual rate will be 2 subjects/month.

14.3 Stratification Factors

None

14.4 Analysis of Exploratory Endpoints (including re-treatment phase)

- Frequency of adverse events (possibly, probably, or definitely) related to crizotinib use in this patient population will be summarized.
- Audiologic responses will be summarized over time using descriptive statistics and graphical displays. Similarly, quality of life will be summarized over time in these patients. Additional descriptive analyses will be carried out for adults versus children.
- The association between audiometric and volumetric responses will be summarized over time with graphical displays and the association between best binary responses will be summarized using contingency tables over time, using Chi-square as a measure of association.
- In the analysis of the TRQ and NFTI-QOL questionnaire data, compliance with questionnaire administration (defined as the proportion of questionnaires actually received out of the expected number) will be calculated, and questionnaires will be scored as recommended in the instructions for the instruments.[35,36]

14.5 Reporting and Exclusions

14.5.1 Evaluation of toxicity. All patients will be evaluable for toxicity from the time of their first treatment with crizotinib.

14.5.2 Evaluation of response. Only those patients who have received at least one cycle of therapy, and have had their disease re-evaluated will be considered evaluable for response. Each participant should be assigned one of the following categories: 1) hearing response, 2) stable hearing, 3) progressive hearing loss, 4) early death from NF2-related condition, 5) early death from toxicity, 6) early death because of other cause, or 9) unknown (not assessable, insufficient data). By arbitrary convention, category 9 usually designates the "unknown" status of any type of data in a clinical database.

Participants in response categories 5-9 should be considered to have a treatment failure (disease progression). Thus, an incorrect treatment schedule or drug administration does not result in exclusion from the analysis of the response rate.

All conclusions should be based on all eligible participants. Sub-analyses may then be performed on the basis of a subset of participants, excluding those for whom major protocol deviations have been identified (*e.g.*, early death due to other reasons, early discontinuation of treatment, major protocol violations, etc.). However, these sub-analyses may not serve as the basis for drawing conclusions concerning treatment efficacy, and the reasons for excluding participants from the analysis should be clearly reported. The 95% confidence intervals should also be provided.

15. PUBLICATION PLAN

The results of this study will be made public within 12 months of the end of data collection. If a report is planned to be published in a peer-reviewed journal, then that initial release may be an abstract that meets the requirements of the International Committee of Medical Journal

Editors. A full report of the outcomes should be made public no later than 1 year after the end of data collection.

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Appendix A: Schedule of Evaluations

STUDIES TO BE OBTAINED	Baseline	Cycle 1	Subsequent cycles	Off-Treatment Follow-Up (30–35 days post treatment)
Informed Consent	X			
Inclusion/Exclusion Criteria	X			
Ophthalmologic examination	X		End of cycle 1, 3, 6, 9, 12	
Oxygen Saturation (SaO_2)	X			
History	X	Prior to start †	End of cycle 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12	X
Physical Exam	X	Prior to start †	End of cycle 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12	X
Vital Signs	X	Prior to start †	End of cycle 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12	X
Performance Status	X	Prior to start †	End of cycle 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12	X
CBC, diff, comprehensive metabolic panel*, Mg, Phos, CK	X	Prior to start †	End of cycle 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12	X
Biomarker blood sample	X		End of cycle 1, 3, 12	
Serum Pregnancy test **	X	Prior to start **	End of cycle 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12	X
Subject Diary Review			End of cycle 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12	
ECG (QTc)	X	Prior to start †	End of cycle 3, 6, 9, 12	
MRI Brain with volumetric analysis	X	Prior to start †	End of cycle 3, 6, 9, 12	
Audiogram ***	X	Prior to start †	End of cycle 3, 6, 9, 12	
NFTI-QOL *(subjects age ≥16 years only)*	X	Prior to start †	End of cycle 6, 12	
TRQ Tinnitus Assessment *(subjects age ≥18 years only****)*	X	Prior to start †	End of cycle 6, 12	

Each cycle is 28 days, with no extension for drug interruptions. All required evaluations should be obtained within days 21-28 of a cycle. Assessments at the end of cycle 12 may be obtained within 1 week after completion of cycle 12.

† Includes drug dispensing, and repeat baseline evaluations if time between first screening and dispensing exceeds 4 weeks. *Otherwise, baseline evaluations need not be repeated, unless there is a significant change in the patient's clinical condition.*

[#] *Includes glucose, calcium, albumin, total protein, sodium, potassium, bicarbonate, chloride, blood urea nitrogen, creatinine, alkaline phosphatase, alanine amino transferase, aspartate amino transferase, bilirubin **Required for women of childbearing potential within 7 days of starting crizotinib. ***Not required for patients with no measurable hearing.

****Question 24 of the Tinnitus Reaction Questionnaire (TRQ; See Appendix G) elicits possible suicidal ideation.** A clinician should review this questionnaire immediately after it is completed and assess for any possible suicidal risk if the answer score is >2. Any concerns of suicidality should be immediately addressed with the responsible institutional investigator.

Retreatment Option- Schedule of Evaluations

STUDIES TO BE OBTAINED	Baseline	Cycle 13	Subsequent cycles	Off-Treatment Follow-Up (30–35 days post treatment)
Informed Consent				
Inclusion/Exclusion Criteria	X			
Ophthalmologic examination	X		End of cycle 13, 15,18, 21, 24	
Oxygen Saturation (SaO_2)	X			
History	X	Prior to start †	End of cycle 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24	X
Physical Exam	X	Prior to start †	End of cycle 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24	X
Vital Signs	X	Prior to start †	End of cycle 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24	X
Performance Status	X	Prior to start †	End of cycle 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24	X
CBC, diff, comprehensive metabolic panel*, Mg, Phos, CK	X	Prior to start †	End of cycle 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24	X
Serum Pregnancy test **	X	Prior to start ***	End of cycle 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24	X
Subject Diary Review			End of cycle 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24	
ECG (QTc)	X	Prior to start †	End of cycle 15, 18, 21, 24	
MRI Brain with volumetric analysis	X	Prior to start †	End of cycle 15, 18, 21, 24	
Audiogram ***	X	Prior to start †	End of cycle 15, 18, 21, 24	
NFTI-QOL *(subjects age ≥16 years only)*	X	Prior to start †	End of cycle 18, 24	
TRQ Tinnitus Assessment *(subjects age ≥18 years only)***	X	Prior to start †	End of cycle 18, 24	

Each cycle is 28 days, with no extension for drug interruptions. All required evaluations should be obtained within days 21-28 of a cycle. Assessments at the end of cycle 24 may be obtained within 1 week after completion of cycle 24.

† Includes drug dispensing, and repeat baseline evaluations if time between first screening and dispensing exceeds 4 weeks. *Otherwise, baseline evaluations need not be repeated, unless there is a significant change in the patient's clinical condition.*

*Includes glucose, calcium, albumin, total protein, sodium, potassium, bicarbonate, chloride, blood urea nitrogen, creatinine, alkaline phosphatase, alanine amino transferase, aspartate amino transferase, bilirubin. **Required for women of childbearing potential within 7 days of starting crizotinib. ***Not required for patients with no measurable hearing.

****Question 24 of the Tinnitus Reaction Questionnaire (TRQ; See Appendix G) elicits possible suicidal ideation.** A clinician should review this questionnaire immediately after it is completed and assess for any possible suicidal risk if the answer score is >2. Any concerns of suicidality should be immediately addressed with the responsible institutional investigator.

Appendix B: Hearing Response Guidelines

Clinical criteria for definition of hearing response based on a 50-word hearing test. Upper and lower limits for the 95% critical differences for percentage scores are adapted from Thornton and Raffin.[37]

Baseline word recognition score (%)	95% critical difference (%)	Hearing Response (%)	Progressive hearing loss (%)
0	0–4	≥ 6	n/a
2	0–10	≥ 12	n/a
4	0–14	≥ 16	n/a
6	2–18	≥ 20	0
8	2–22	≥ 24	0
10	2–24	≥ 26	0
12	4–26	≥ 28	≤ 2
14	4–30	≥ 32	≤ 2
16	6–32	≥ 34	≤ 4
18	6–34	≥ 36	≤ 4
20	8–36	≥ 38	≤ 6
22	8–40	≥ 42	≤ 6
24	10–42	≥ 44	≤ 8
26	12–44	≥ 46	≤ 10
28	14–46	≥ 48	≤ 12
30	14–48	≥ 50	≤ 12
32	16–50	≥ 52	≤ 14
34	18–52	≥ 54	≤ 16
36	20–54	≥ 56	≤ 18
38	22–56	≥ 58	≤ 20
40	22–58	≥ 60	≤ 20
42	24–60	≥ 62	≤ 22
44	26–62	≥ 64	≤ 24
46	28–64	≥ 66	≤ 26
48	30–66	≥ 68	≤ 28
50	32–68	≥ 70	≤ 30
52	34–70	≥ 72	≤ 32
54	36–72	≥ 74	≤ 34
56	38–74	≥ 76	≤ 36
58	40–76	≥78	≤ 38
60	42–78	≥ 80	≤ 40
62	44–78	≥ 80	≤ 42
64	46–80	≥ 82	≤ 44
66	48–82	≥ 84	≤ 46
68	50–84	≥ 86	≤ 48

70	52–86	≥ 88	≤ 50
72	54–86	≥ 88	≤ 52
74	56–88	≥ 90	≤ 54
76	58–90	≥ 92	≤ 56
78	60–92	≥ 94	≤ 58
80	64–92	≥ 94	≤ 62
82	66–94	≥ 96	≤ 64
84	68–94	≥ 96	≤ 66
86	70–96	≥ 98	≤ 68
88	74–96	≥ 98	≤ 72
90	76–98	100	≤ 74
92	78–98	100	≤ 76
94	82–98	100	≤ 80
96	86–100	n/a	≤ 84
98	90–100	n/a	≤ 88
100	96–100	n/a	≤ 94

Appendix C: NFTI-QOL (Neurofibromatosis 2 impact on quality of life)

To be administered to patients ≥16 years only

SUBJECT IDENTIFIER: _____

INSTRUCTIONS FOR COMPLETING THE NFTI-QOL

Please complete the following information:

Age: _____ years

Gender: Male □1 Female □2 (please tick)

For each of the questions on the next page, please tick the one box that describes how you feel today

Usual activities include: work; housework; study; sport; social; family or leisure activities

Guy's NFTI-QOL

Q1. Do balance or dizziness problems stop you performing your usual activities?

- No balance problems or dizziness ☐0
- Balance or dizziness problems but no difficulties ☐1
- Balance or dizziness problems cause me some difficulties ☐2
- Balance or dizziness problems stop my usual activities ☐3

Q2. Do hearing problems stop you performing your usual activities?

- No hearing problems ☐0
- Hearing problems but no difficulty ☐1
- Hearing problems cause me some difficulty ☐2
- Hearing problems stop my usual activities ☐3

Q3. Does facial weakness stop you performing your usual activities?

- No facial weakness ☐0
- Facial weakness, but no difficulty ☐1
- Facial weakness causes some difficulty ☐2
- Facial weakness stops my usual activities ☐3

Q4. Do problems with your sight stop you performing your usual activities?

- No problems with sight ☐0
- Sight problems, but no difficulty ☐1
- Sight problems cause me some difficulty ☐2
- Sight problems stop my usual activities ☐3

Q5. Do you have any problems in mobility and walking?

- No problems in mobility and walking ☐0
- Some difficulty but can manage on my own ☐1
- Unable to walk around without some help ☐2
- Unable to walk at all ☐3

Guy's NFTI-QOL

Q6. Has your medical condition affected your role and outlook on life? (e.g., confidence, vulnerability, relationships, caring for family, career, having children)

No effect or positive effect	☐0
Small negative effect	☐1
Moderately negative effect	☐2
Large negative effect	☐3

Q7. Pain; throughout our lives, most of us have had pain from time to time such as mild headaches, sprains and toothaches. Have you had pain *other than this* in the last week?

None	☐0
Mild pain	☐1
Moderate pain	☐2
Severe pain	☐3

Q8. Do you currently suffer from anxiety or depression?

No	☐0
Mild anxiety or depression	☐1
Moderate anxiety or depression	☐2
Extreme anxiety or depression	☐3

IF you have any further comments regarding the impact of NF2 on your quality of life, please write them here:

You have now completed the NFTI-QOL. Thank you for your input.

Appendix D: Performance Status Criteria

Karnofsky Scale (recipient age ≥ 16 years)	Lansky Scale (recipient age <16 years)
Able to carry on normal activity; no special care is needed	**Able to carry on normal activity; no special care is needed**
100 Normal, no complaints, no evidence of disease	**100** Fully active
90 Able to carry on normal activity	**90** Minor restriction in physically strenuous play
80 Normal activity with effort	**80** Restricted in strenuous play, tires more easily, otherwise active
Unable to work, able to live at home, cares for most personal needs, a varying amount of assistance is needed	**Mild to moderate restriction**
70 Cares for self, unable to carry on normal activity or to do active work	**70** Both greater restrictions of, and less time spent in active play
60 Requires occasional assistance but is able to care for most needs	**60** Ambulatory up to 50% of time, limited active play with assistance/supervision
50 Requires considerable assistance and frequent medical care	**50** Considerable assistance required for any active play, fully able to engage in quiet play
Unable to care for self, requires equivalent of institutional or hospital care, disease may be progressing rapidly	**Moderate to severe restriction**
40 Disabled, requires special care and assistance	**40** Able to initiate quite activities
30 Severely disabled, hospitalization indicated, although death not imminent	**30** Needs considerable assistance for quiet activity
20 Very sick, hospitalization necessary	**20** Limited to very passive activity initiated by others (e.g., TV)
10 Moribund, fatal process progressing rapidly	**10** Completely disabled, not even passive play

Appendix E: Audiology Procedures

1.0 INTRODUCTION

The purpose of this appendix is twofold: In the first section, procedures for local audiologists will be outlined. In the second section, procedures for transfer of the audiometric data from the local audiogram to the data form will be specified.

2.0 AUDIOLOGY PERSONNEL

The primary source of reliability and validity will be the qualifications of the clinical audiologists performing the tests, and their adherence to standard practices. These practices will be specified in this section. A secondary source of reliability and validity will be the oversight of the local Lead Audiologist appointed for each clinical site. The Lead Audiologist will compile information assuring standard calibration, installation of reference-calibrated equipment, etc. The local Lead Audiologist will also act as a stable contact on behalf of their center. Finally, the Senior Study Audiologist (MSKCC) will provide a final layer of assurance by developing and updating the procedures in this manual in response to any problems and by maintaining regular contact with each site's Lead Audiologist.

2.1 Lead Audiologist

Each Clinical Site will designate a Lead Audiologist who will be the contact for study-related issues with the Senior Study Audiologist. This audiologist will oversee local audiology operations and also communicate with the site PI. The lead audiologist may train other audiologists at the site for testing.

2.2 Qualifications

Each evaluation will be performed by a fully qualified audiologist. The precise definition of qualification can vary from state to state depending on licensure laws, etc. For the purposes of this study, full qualification is defined as the highest local level of qualification, certification or licensure. Basic requirements for Lead Audiologist will be no different, but only one audiologist per center will be designated for this duty.

2.3 Training

Each site's Lead Audiologist will be responsible for local training of any audiologist performing assessments for this protocol. This training will be based on this appendix, and the appendix will remain available to all trained audiologists as a resource. All Lead Audiologists will be contacted in advance of the initiation of participant enrollment at the site, and the Senior Study Audiologist will discuss and demonstrate the procedures contained in this Appendix. These will include personnel, test protocols, data cross checking, procedures for correcting or completing evaluations, and reporting results.

2.4 Contacts

A system of regular contact between the Senior Study Audiologist and each Lead Audiologist will be initiated before any participant's enrollment. This will take place primarily by e-mail, with documents faxed as necessary. As audiologic issues arise, the local audiologists will be asked to contact the Lead Audiologist, who will act as liaison with the Senior Study

Audiologist. Other local issues are expected to be addressed by contact among the clinical site PIs, their Lead Audiologists, and the CRCs.

3.0 AUDIOLOGY FILES

Each clinical site's Lead Audiologist will, at the commencement of the study, create a filing system for audiology facility records and correspondence. System features will be at the discretion of the Lead Audiologist, incorporating the local filing protocols. However, at minimum this system will contain three recognizable sections: general information, correspondence, and worksheets.

3.1 General Information Section

This section will include site-specific information as to the identification of the PI, the Lead Audiologist, Senior Study Audiologist, and others associated with the study, along with extensive contact and coverage information. This section will also include the local location of this appendix, and the data reporting protocol along with copies of calibrations and other equipment records sufficient to document validity. Finally, the general section will include a comment log, in which dated and initialed entries by any audiologist may make note of any events or concerns.

3.2 Correspondence Section

This section will include records of conference calls and other correspondence regarding procedural issues, changes in equipment or personnel, and any adverse events.

3.3 Audiogram Section

This section will include audiograms generated for each audiologic evaluation related to this study.

4.0 AUDIOLOGY TESTING PROTOCOL OVERVIEW

Participants will be referred for each evaluation by the local site PI or CRC, who will determine the timing of return visits. At minimum, the referral will include designation of the target ear. When the participant arrives for each test, the audiologist will greet the participant and accompanying persons and briefly and privately discuss progress if the participant wishes. Study questions may be referred to the local PI or CRC. Audiologists will not be formally blinded to any aspect of the study.

The audiologist will seat the participant in a sound-treated room. No more than one person will be allowed to accompany the participant to this area and this person will not be allowed to sit in the booth or to be in the line of sight of the participant. As much as possible, light levels in the participant and the tester sides will be adjusted to provide a good view of the participant and a poorer view of the audiologist (*i.e.*, participant side bright, tester side dark). The participant should be seated perpendicular to the audiologist to minimize cues. The room door will be fully closed. The participant will be asked to respond by hand raise or by button push, whichever is customary at the clinical site.

The Lead Audiologist at each site will ensure that threshold tests are performed in the standard manner. This includes the Hughson-Westlake bracketing procedure. A 200 ms. ON versus

200 ms. OFF duty cycle for tone presentation is recommended with an opportunity to appreciate 4 tones per trial. Narrow band noises or FM modulated tones will not be substituted for standard pure tones. Thresholds will be transcribed on the Clinical Site's standard audiogram, using standard symbols. At the conclusion of the evaluation, the audiologist may briefly discuss the result with the participant and accompanying persons, again referring most study questions to the PI or CRC where possible. Discussion of helpful strategies and devices as indicated by the case is expected.

4.1 The Sound Pathway

The testing audiologist will perform a visual inspection of both ears using an otoscope. If there is sufficient access to the tympanic membrane to allow testing, the audiologist will proceed. If there are concerns as to cerumen or other factors, the audiologist may elect to perform tympanometry. When complete, the outcome of tympanometry will be noted on the audiogram in the customary manner for that site. If occluding cerumen or similar factors cannot be resolved, testing will not proceed. The local PI has overall responsibility to ensure unoccluded ears if referrals are required.

4.2 Pure Tone Thresholds

Pure tone thresholds will be found in the standard manner by bone conduction at 250, 500, 1000, 2000 and 4000 Hz. Air conduction thresholds will be found at 250,500, 1000, 2000, 3000, 4000 and 8000 Hz. Masking will be used in any case where it is considered necessary to establish which ear is responding. Air-bone gaps larger than 10 dB will be masked. For air conduction, masking will be applied for each threshold where the possibility of crossover exists. The possibility of overmasking will be evaluated by the audiologist and may be addressed, for example, by using insert phones. A masking plateau procedure will be performed at the discretion of the audiologist. The local audiologists have discretion to test other frequencies or to repeat testing to ensure validity. In addition, further testing (Stenger's test; SRT; OAE) is at the discretion of the local audiologist to ensure validity. Auditory brainstem response testing is not indicated in these patients because their retrocochlear diagnosis is established prior to entry.

4.2.1 Transducer

Air conduction thresholds may be evaluated using both standard headphones (TDH-39, 49, 50) or calibrated insert phones (Etymotic ER-3A), as long as specific correction factors are in place. The transducer type (*i.e.*, TDH-49, ER 3-A) should be entered on the local clinical audiogram form reflecting the method by which the final recorded threshold was obtained.

4.3 Vibrotactile and Responses at Limits

Vibrotactile responses will never be reported as thresholds. On discussing the nature of the percept with the participant and determining that they are vibrotactile, the audiologist will mark the response as "out at limits" at the vibrotactile response level. Other "out at limits" responses will be noted at the highest actual stimulus level with the standard "down arrow" notation. None of the study frequencies should ever be left blank.

4.4 Word Recognition

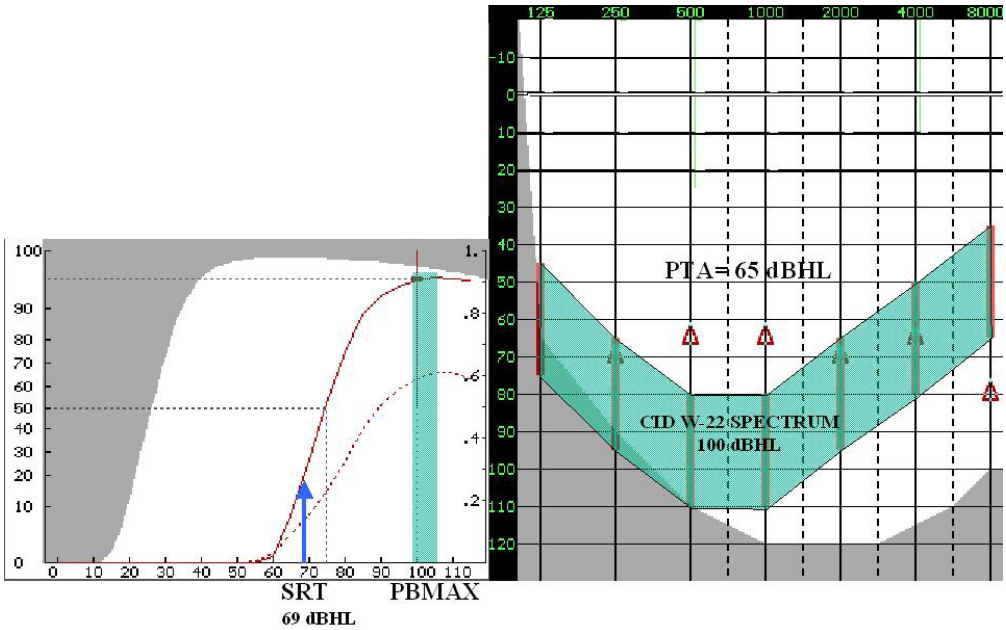
Speech intelligibility will be evaluated for each ear using standard word recognition of monosyllables. All tests will use recorded lists (CID-W22, Ira Hirsh recording, Q/MASS v 2) supplied by the Senior Study Audiologist, including standardized score sheets. The recording and list must be noted on the clinical audiogram. This is necessary to verify the list used in all cases when the report of this study is made. Since the results are analyzed as repeated measures, a participant may be evaluated using English materials if the local audiologist decides that the results reasonably reflect comparative (test to test) performance. In such a case, the participant may be instructed to produce the sounds of the words, even if they do not necessarily recognize the meaning. All lists will be recorded (no live voice tests) and 50 items always given.

4.4.1 Setting the "High" and "Low" speech test levels

The principal guiding the initial level setting for speech tests is: The level should be set where maximum performance is expected for each ear. Performance has been shown to exhibit "rollover" in some NF2 cases, so an empirical search using at least two levels will also be used. These will be designated the "High" and the "Low" levels. The "High" level will be the level calculated for maximum audibility, as in the standard method for level setting in non-tumor cases. The "Low" level will be the maximum level minus 10-15 dB as chosen by the audiologist to reflect a level with less chance of rollover.

Fletcher's Articulation Index will be used to assist in calculation of the initial ("High") presentation level. This formula can be used to draw a cumulative (ogive) optimum performance/intensity (P/I) function for any audiogram (Figure 1, below). If rollover was not a factor, the maximum performance would be expected at levels

Figure 1. Cumulative optimum performance/intensity (P/I) function.



on the asymptote of the curve. [13] Using software developed for this purpose, Table 1 was created for setting "High" speech levels:

Table 1. High Speech Level Settings:

PTA2	Monosyllable level
30	-
35	-
40	70
45	75
50	80
55	85
60	90
65	95

The PTA2 parameter designates Fletcher's 2-frequency average. PTA2 is the average of the two best (lowest) thresholds of the set 500 Hz, 1 KHz and 2 KHz. This value is easy to calculate and delivers better performance predicting speech reception threshold (SRT) and other speech level values than the more inclusive PTAs. The levels were developed Monte Carlo fashion by submitting a representative sample of audiograms to the P/I function generator and relating the PTA2 to the lower levels of the asymptotic portion of the curve. The step size is 5 dB. Smaller gradations are available on some audiometers and may be used at the discretion of the audiologist. No "High" level list will ever be given lower than 70 dB HL. The advantage of this minimum level is that normal, mild, and moderate loss cases can all be tested at the same physical level.

Table 1 is designed to replace rule-based level setting schemes since they may not be optimal, especially given severe losses (PTA ≥ 50 dB HL). For example, a common rule requires a further test (SRT) to allow levels set at SRT + 40 dB. While this provides reasonable maximum performance for milder cases, with severe losses the dynamic range is reduced below the 40 dB constant. On the other hand, no rule or table can replace good clinical sense when very high levels are required. In abandoning the table values, the audiologist will be asked to decrease levels minimally below the participant's Uncomfortable Level (UCL) rather than set the level at a determined Most Comfortable Level (MCL). No levels should exceed 95 dB HL.

As described above, after performing a word list at the "High" level, the Audiologist will reduce the intensity by 10-15 dB and present a 50-item list at the "Low" level. If the speech stimulus exceeds most frequencies at a level 15 dB below the "High" level then that level should be used. If, on the other hand, many important frequencies are no longer audible, then the level 10 dB below should be used. This procedure will be used for the target and the contralateral ear, with the exception that the "High" level may be used alone for the contralateral ear if there is no vestibular schwannoma on that side.

4.4.2 Participant responses

Participants with poor speech intelligibility are expected to initially report no intelligibility (*i.e.*, "I can't get it"). The audiologist is required to explain that guessing is very helpful and that, in order for the test to proceed, the participant must guess a monosyllable each time the carrier phase is heard. Instead of reporting no recognition, a "wild guess" should be substituted. This strategy will optimize responses in cases of

low speech intelligibility and is surprisingly effective with participants who are really only uncertain. Dropped plurals are to be scored wrong. If the participant responds with several options, the first one said will be scored. Items will not be repeated. Small variations in regional dialect (*i.e.*, vowel boundaries) are scored at the discretion of the audiologist. Fifty items will be given unless the audiologist is convinced that the participant has no speech-like percept at all. Fifty wrong guesses will be scored as 0% and this represents an optimal test by both the participant and the audiologist. If there is no speech-like percept at all, the test can be scored CNT (could not test). Skipping the word recognition test and entering the notation "DNT" (did not test) is never acceptable. The audiology evaluation will be considered incomplete if the notation DNT appears. Under no circumstances will half-lists or screening lists be accepted.

4.4.3 Masking for word recognition

Masking will be applied to the contralateral ear during speech tests if there exists a possibility of significant contribution due to crossover. To facilitate this application for this study, the Articulation Index was again used to calculate the maximum score possible by crossover given the difference in PTA2 and the effective masking required to reduce this contribution to zero (Table 2).

Table 2. Calculation/Crossover Scores

Speech Level minus Non-test PTA2	Speech Noise TDH	Speech Noise Inserts
50	15	-
55	20	-
60	25	-
65	30	-
70	35	10
75	40	15
80	45	20
85	50	25
90	55	30
95	60	35
100	65	40

Table 2 includes values based on published interaural attenuation values for both TDH phones and insert phones. Standard speech-shaped noise is assumed in this calculation and will be the masker used at the study centers.

4.5 Validity

If the audiologist determines that the test is valid she/he will indicate this in the space used for this purpose on the local audiogram form and need not enter any further information regarding validity. If the audiologist decides to perform other tests (for example SRT, to rule out functional loss) this will be noted along with the outcome in the text section. In most cases, the principles and procedures outlined above will allow reliable and valid data. One example of an exception requiring different tests would be when functional hearing loss (pseudohypoacusis) is suspected. Hesitant threshold responses or a history of litigation

for loss are some of the many warning signs audiologists use to initiate tests to rule out functional loss. In this study, audiologists, supported by their local Lead Audiologist will rule out functional loss on a case-by-case basis. For example, SRTs may be done to evaluate the match with the pure tones. Stenger's test may be performed. The text section of the local audiogram will also be used to convey any testing issues such as participant cooperation, noise, equipment issues, etc., which might adversely affect validity.

4.6 The Target Ear

All referrals to the local audiologist from the local investigator site must include a designation of the target ear. Designation of the target ear is noted to occur redundantly on many forms of documentation in this study. The local audiologist will ensure that the special "High" and "Low" level word recognition testing is performed on the target ear at minimum during the evaluation. This strategy should also be applied to the contralateral ear if that ear is known to have a vestibular schwannoma or its status is unknown. The audiologist will specify the target ear on their audiogram at every evaluation.

4.7 Example Audiogram

Figure 2 shows an example audiogram which satisfies the needs of this study protocol. It is for example only and local variation is expected.

AUDIOLOGIC EVALUATION

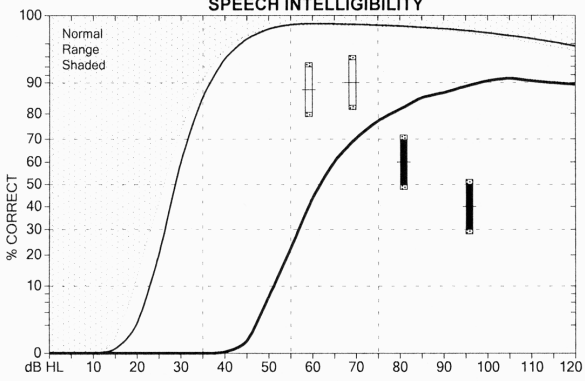
Massachusetts Eye and Ear Infirmary
Audiology Department

NAME: BEVA CIZUMAB **UNIT:** 9999999
DOB: 1/21/1990
REFERRED BY: PLOTKIN **AGE:** 20 **SEX:** M

AUDIOLOGIST: Chris Halpin, Ph.D., CCC-A

ID: 111 **DATE:** 09/16/2010 **TIME:** 14:51 **ROOM:** CFH

SPEECH INTELLIGIBILITY



WORD RECOGNITION

RIGHT			LEFT		
LIST	dB	(%)	LIST	dB	(%)
W22 3B	70	90	W22 1A	95	40
W22 4B	60	88	W22 2A	80	60

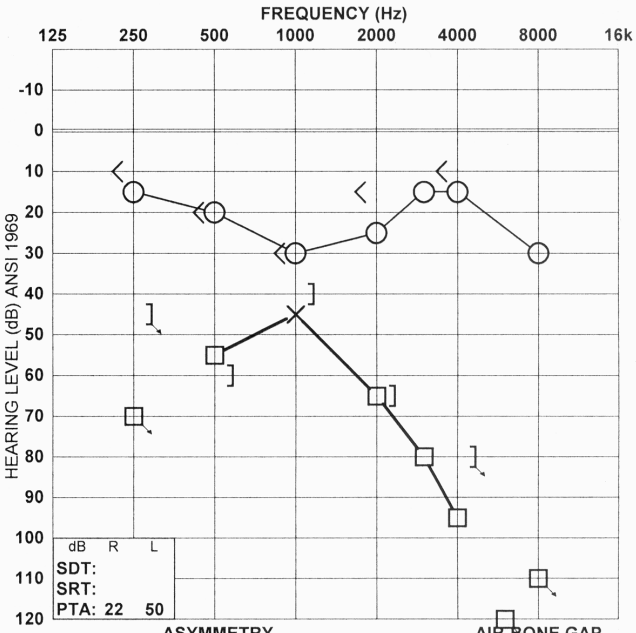
SYMBOLS	RIGHT	LEFT
Speech Intelligibility Index Prediction Speech Score 84%CI 95%CD 90%CI 98%CD		
Air Conduction		
AC Masked		
Bone Conduction		
BC Masked		
Response at Limit		
No Response		
Acoustic Reflex Ipsilateral Probe		
Acoustic Reflex Contralateral Probe		

TYMPANOMETRY

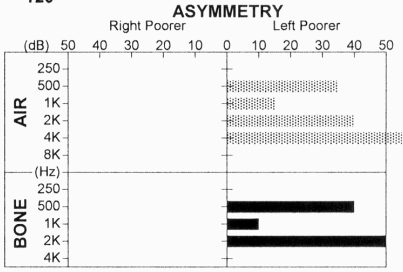
FEATURE	RIGHT	LEFT
VOLUME	Normal	Normal
PRESSURE	Normal	Normal
MOBILITY	Normal	Normal
SHAPE	Normal	Normal
RESONANCE		

REFLEX DECAY

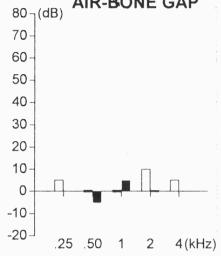
FREQUENCY	RIGHT	LEFT
500 Hz		
1000 Hz		
2000 Hz		
4000 Hz		



ASYMMETRY



AIR-BONE GAP



REASON FOR TEST (from physician requisition):
vestibular schwannoma

HISTORY
Patient reports a diagnosis of NF2 with bilateral vestibular schwannomas. The patient is in the bevacizumab NF2 study with a target ear: Left.

HEARING MEASUREMENT
Some cerumen noted on the left, but test was completed following normal tympanometry. Word recognition was done using study CD, lists 3B and 4B right; and 1A and 2A left. Results considered valid.

IMPRESSIONS
There is a large sensory loss on the left.

CONSULTATION
Result discussed with patient and his family.

FORM ID AU 4 '92

AUDIOLOGY

5.0 DATA CROSS CHECKS

This section will describe data cross check activities performed by the testing audiologist and the Lead Audiologist, as well as the mechanism for reporting unforeseen problems or concerns.

5.1 Testing Audiologist

The primary data cross checks will be the responsibility of the testing audiologist. Specifically, equipment and training will be maintained which will allow such procedures as masking plateau verification, tympanometry, SRT, Stenger's test, etc. These tests will be applied at the discretion of the testing audiologist to verify results and to rule out functional or retrocochlear hearing loss. None of these tests will be used as data, but will be noted on the local audiogram form along with the overall judgment of the audiologist as to validity.

5.2 Lead Audiologist

The audiogram will then be given to the Lead Audiologist or placed in the study "Audiograms" file section for data awaiting review. The Lead Audiologist will verify the audiologic aspects of the data. The Lead Audiologist will verify validity and completeness, or will contact the CRC for rescheduling for other testing if necessary. The completed form will be faxed or emailed (scanned) to the NF Consortium Operations Center.

5.3 Problems and Concerns

The Senior Study Audiologist will be responsible for resolution of problems in audiology data interpretation. If these arise at the office of the PI, they will be communicated to the Senior Study Audiologist (*i.e.*, not the local Lead Audiologist directly), who will have discretion as to resolution study-wide or site-specific. If problems arise at an individual site, they will be communicated by the local Lead Audiologist to the Senior Study Audiologist, who will again be responsible for resolution either study-wide or site-specific. The anticipated mechanism for resolution of study-wide issues will be communication with all local Lead Audiologists and changes or additions to this Appendix. The Senior Study Audiologist will be responsible for informing and receiving advice and approval from the Study Chair as appropriate.

6.0 AUDIOGRAM ACCEPTANCE

After the audiogram is faxed (or emailed) to the NF Consortium Operations Center, it will be forwarded to the Senior Audiologist in the office of the overall study PI for review and approval for use in the study. This is particularly critical in the case of the initial audiometric evaluation. All subsequent evaluations and outcomes will be compared to this initial audiometric evaluation and, following intent-to-treat principles, patients will not be removed from the study after the fact if the audiogram is found to be invalid. If the audiogram is not accepted, it must be re-scheduled by the study local PI's office, re-done and re-submitted for acceptance. The Senior Study Audiologist will be responsible for providing any required materials and advice in order to ensure audiogram acceptance before the fact. It is necessary that each site make contact with the Study Audiologist (Kerri O'Connor, oconnok7@mskcc.org) when they are contacted by referral sites in order to initiate this support. The following sections will describe some factors which will cause the audiogram not to be accepted.

6.1 Improper Referral

If a patient arrives at the audiologist's office without that office being previously contacted about the study, or provided full materials and adequate time to have questions addressed, this evaluation is not expected to be possible and the result will be a request by the Senior Audiologist and the Study PI to the referring center for adequate referral procedures.

6.2 Ear Occluded With Cerumen

If the audiologist finds the ear occluded with cerumen (it is recommended that this be confirmed using tympanometry), the patient's audiologic evaluation cannot be performed and the patient should be returned to the referral source for remediation if the cerumen cannot be managed in the audiologists' office. If a notation is found that either ear was occluded with cerumen, the audiogram will not be accepted.

6.3 Word Recognition

If word recognition is recorded as "DNT" (Did Not Test) on either side, the patient's audiologic evaluation cannot be accepted. This is also true if a half (25-item) or screening list was given, or if the word recognition test was delivered using monitored live voice (MLV). Word recognition must be positively noted as having been done using the provided CD, 50 items and done at high and low levels as described in section 5.6.1 in order to be accepted. The software used to create Table 2 may be used to evaluate the potential for influence of the contralateral ear, though significant crossover is unlikely.

6.4 Pure Tone Thresholds

If pure tone thresholds do not include all frequencies specified in section 5.4, for example if bone conduction was not done, the audiogram cannot be accepted. With regard to masking, thresholds may be input to the Harvard Audiometry software for evaluation of crossover. If significant chance that the non-test ear was responding is found, the audiogram cannot be accepted.

7.0 EQUIPMENT

The following section contains specifications for equipment used in this study.

7.1 Sound-treated Enclosure

A single- or double-walled sound-treated enclosure that meets American National Standard Criteria for Maximum Permissible Ambient Noise Levels for Audiometric Test Rooms shall be used to conduct pure tone air and bone conduction thresholds and word recognition testing.

An illuminated otoscope is used to examine a participant's ear canals. If any possible contraindications to audiometric testing (such as excess cerumen, eardrum abnormalities, etc.) are detected, the participant must be referred for medical evaluation before audiometric testing can proceed.

7.2 Audiometer

Audiometers that meet the American National Standard Specifications for Audiometers and have two channels are used to conduct pure tone air and bone conduction threshold, SRT and word recognition testing. One channel of the audiometer generates and delivers the test signals, either pure-tones or prerecorded speech. The second channel delivers narrow-band or

speech-band masking noise simultaneously with the test signal, but to the non-test ear whenever necessary. The audiometer must have an input jack for external equipment such as a compact disc player or tape player, which will be used to present speech stimuli for word recognition testing.

7.3 Audiometer Transducers

Earphones mounted in supra-aural cushions and calibrated according to the American National Standard Specification for Audiometers are used to deliver the test material from the audiometer to the participant. The earphones are designated as "right" and "left" and will be placed comfortably over the participant's right and left ears, respectively. Bone vibrators calibrated according to the same standards are used to obtain bone conduction thresholds. During the testing, the bone vibrator is positioned over the mastoid area of the participant's test ear, taking care that it is not in contact with the posterior part of the pinna.

7.4 Compact Disc Player

A compact disc player must be used to deliver pre-recorded speech material to the audiometer and subsequently to the transducers positioned over the participant's ears. A cable extends between the output jack of the compact disc player and the input jack of the audiometer.

7.5 Acoustic Immittance Equipment

Immitance is not required by this study protocol, but may be used at the audiologists' discretion. An immittance device that meets the American National Standard Specifications for Instruments to Measure Aural Acoustic Impedance and Admittance is used to conduct tympanometry and acoustic reflex threshold testing. Test results will be printed directly from the immittance device or recorded manually at the conclusion of testing on each ear. Probe tips must be appropriate in size to seal the participant's ear canal tightly during tympanometry and acoustic reflex testing. Clinical centers must have an adequate variety of sizes of probe tips to accommodate ear canals of varying dimensions.

7.6 Maintenance

Each clinical center is responsible for the proper operation and maintenance of its audiometric equipment. Responsibility for proper maintenance is assumed by the Lead Audiologist, and all staff are instructed to report promptly any real or suspected equipment problems to that person. All checks, inspections, and repairs are documented and recorded by date in a permanent log. The Study PI and Study Senior Audiologist may review this log at periodic site visits. All study test equipment including audiometers and acoustic immittance devices must be calibrated according to the American National Standards Institute. Listening checks may help to identify problems that could influence participants' test behavior and audiometric results in between scheduled physical calibrations. Study audiologists should perform a listening check on any day when a participant enrolled in the protocol will be tested.

8.0 Data Reporting

Audiograms done using the procedures described above will be faxed or emailed from the NF Consortium Operations Center to the office of the Study Audiologist (Kerri O'Connor) and the Principal Investigator. The Senior Study Audiologist and others at the PI's office, will have direct access to the eCRF software used for data collection in the study. The clinical audiograms will be checked and the data transferred to the study eCRF system. The PI's office

(Senior Study Audiologist) will keep copies of each of the study audiograms and records of the transfer process.

Appendix F: MRI Protocols

Image Acquisition

Each scanning session must include the following sequences (although additional sequences can be performed per institutional protocol):

1) Scout sequence
2) T2-weighted imaging
3) Fluid-attenuated inversion recovery (FLAIR) imaging
4) T1-weighted pre-contrast imaging
5) Diffusion weighted imaging (DWI)
6) Post-contrast T1-weighted imaging
7) Post-contrast Gradient Echo 3D imaging with fine cuts (1 mm slices, no skip) preferably through the entire brain, and at the minimum including the entire target tumor.

NOTE: The post-contrast Gradient Echo 3D imaging with fine cuts **(1 mm slices, no skip)** and volumetric analysis will be used to assess the endpoint of radiographic response (defined as ≥ 20% decrease in tumor volume by MRI scan). Suitable post-contrast Gradient Echo 3D imaging sequences include BRAVO (GE), MPRAGE (Siemens), 3D-T1 TFE (Philips), 3D GEIR (Hitachi) and 3D Fast FE (Toshiba).

All images used for volumetric analysis will be reviewed by the PI or co-PI to verify the following:

- Prior to patient enrollment, the PI or co-PI will review volumetric assessments to confirm that *1)* baseline volumetrics were performed on post-contrast Gradient Echo 3D imaging with fine cuts (1 mm slices, no skip), *2)* target tumors were volumetrically measurable and ≥0.75 ml in size, and *3)* protocol defined criteria were met for progression (i.e., volumetric evidence of progression over the past 18 months, defined as ≥20% annualized increase in volume)
- For imaging response assessment on study, the PI or co-PI will review volumetric assessments to verify whether *1)* volumetrics were performed on post-contrast Gradient Echo 3D imaging with fine cuts (1 mm slices, no skip), *2)* protocol defined criteria were met for imaging response or progression (defined as ≥20% decrease or increase in volume compared to lowest tumor volume during treatment, respectively)

Image Analysis

Enhancing lesions will be quantitatively analyzed by an experienced neuroradiologist blinded to the order of the scans, patient identity and treatment status of the patients. Bi-dimensional diameters will be created and outlined using electronic calipers in accordance with the Macdonald criteria [48]. The lesions will also be outlined using a volumetric approach described previously [52] that includes outlining each enhancing voxel on post-contrast scans and then summing the voxels to calculate an overall lesion volume.

Secure File Transfer

The procedures for uploading MRI scans to the Tumor Imaging Metrics Core are listed below.

MRI images from enrolled subjects should be de-identified and will be transmitted electronically to the Dana-Farber/Harvard Cancer Center Tumor Imaging Metric Core (TIMC) under the direction

of Dr. Gordon Harris for central analysis of tumor volume. (see section 10.1.7). Sites that are unable to de-identify scans without removing scan date and scanner information may upload images without removal of PHI if institution allows. Once uploaded, TIMC will de-identify scans prior to archiving in their system. TIMC will then inform PI or co-PI to confirm proper sequences by email.

Non-Partners Access

1. Email TIMC Help Desk and provide two email addresses for account registration (a primary and a backup, if necessary)
2. TIMC will send an invite via the Partners Healthcare Secure File Sharing service
3. Click the link in the email to accept the invitation and register your account
   **Please note: Invitations expire after 7 days*
   a. Check Spam and Junk folders for invite if not received in Inbox
4. Create a password
5. Click Register

Partners Access

1. Go to https://transfer.partners.org/courier/web/1000@/wmLogin.html
2. Click I don't have an account yet
3. Enter E-mail address and click Register

Send Files

1. Create a folder on your desktop
2. Copy DICOM data from CD into the desktop folder
   a. Remove any image viewers as this will interfere with data upload to the local analysis software
3. Right click folder > Send To > Compressed (zipped) folder
4. Log in to the system: https://transfer.partners.org/courier/web/1000@/wmLogin.html
5. Select Send File tab
6. Enter tumormetrics@partners.org into the To field
7. Select Choose File > browse for the zipped file on your desktop
8. Add any notes pertaining to the scan or assessment (optional)
9. Select notification preferences from the Additional Options field (optional)
10. Click Send
11. Click OK after receiving confirmation that the files have been sent

Please contact the TIMC Help Desk at tumormetrics@partners.org for any questions.

Appendix G: Tinnitus Reaction Questionnaire (TRQ) [Wilson et al., J Speech Hear Res 34, 1991]

To be administered to patients ≥18 years only

SUBJECT IDENTIFIER : _____

*0 = not at all, 1 = a little of the time, 2 = some of the time, 3 = a good deal of the time, and 4 = almost all of the time

Number	Item	Scores*				
1	My tinnitus has made me unhappy.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
2	My tinnitus has made me feel tense.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
3	My tinnitus has made me feel irritable.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
4	My tinnitus has made me feel angry.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
5	My tinnitus has led me to cry.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
6	My tinnitus has led me to avoid quiet situations.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
7	My tinnitus has made me feel less interested in going out.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
8	My tinnitus has made me feel depressed.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
9	My tinnitus has made me feel annoyed.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
10	My tinnitus has made me feel confused.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
11	My tinnitus “driven me crazy”.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
12	My tinnitus interfered with my enjoyment of life.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4

13	My tinnitus made it hard for me to concentrate.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
14	My tinnitus has made it hard for me to relax.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
15	My tinnitus has made feel distressed.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
16	My tinnitus has me feel helpless.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
17	My tinnitus has made me feel frustrated with things.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
18	My tinnitus has interfered with my ability to work.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
19	My tinnitus has led me to despair.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
20	My tinnitus has led me to avoid noisy situations.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
21	My tinnitus has led me to avoid social situations.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
22	My tinnitus has made me feel hopeless about the future.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
23	My tinnitus has interfered with my sleep.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
24	My tinnitus led me to think about suicide.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
25	My tinnitus has made me feel panicky.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4
26	My tinnitus has made me feel tormented.	☐ 0	☐ 1	☐ 2	☐ 3	☐ 4

Appendix H: Disintegration of Capsules for Patients Unable to Swallow Capsules

PRESCRIBING INFORMATION

Crizotinib (Xalkori) capsules are intended for twice daily administration and are not a delayed-, extended-, or sustained-release formulation. It is supplied commercially as a hard shell gelatin capsule with a pink opaque cap and body (250 mg strength) and a pink opaque cap and white opaque body (200 mg strength).[1] The solubility of crizotinib in aqueous media decreases over the range pH 1.6 to pH 8.2 from > 10 mg/mL to < 0.1 mg/mL. The log of the distribution coefficient (octanol/water) at pH 7.4 is 1.65. Capsules should be swallowed whole and patients should not crush, dissolve or open capsules.[1]

LITERATURE SEARCH

As of September 16, 2016, a search of the published medical literature failed to identify any data regarding the disintegration of capsules for ease oral administration. However, we are aware of unpublished data that discusses this topic. This information is presented below.

BACKGROUND

Xalkori is an immediate-release formulated capsule (not a delayed-, extended-, or sustained-release formulation).[2] A proportion of the patient population may not be able to swallow capsules. Specific pharmacokinetic and stability studies have not formally evaluated the method of capsule disintegration in water as described in this letter; however, the 1001 and PROFILE 1005 studies allowed capsules to be disintegrated in water and administered orally as necessary.[3] The instructions that follow below may be applied to commercial crizotinib supply.[4]

CLINICAL DATA

Unpublished Data

Preparation of Liquid Suspension Formulation Using Clinical Study Supply

Administration of crizotinib (clinical study supply) using a liquid suspension formulation was made possible within the clinical trials for patients unable to swallow whole capsules. The instructions provided below applied to the administration of clinical supply crizotinib; however, the instructions that follow below may be applied to commercial crizotinib supply. Oral suspension dosing is accomplished via the disintegration of a capsule in 2 tablespoonfuls of boiling water, followed by the addition of 1 tablespoon of room temperature or cold water in a 6-8 ounce drinking glass. Dosing of the suspension is followed by three 1 tablespoonful water rinses of the glass.[3]

Once the suspension preparation process was begun, all steps were conducted in the order as outlined below. Suspensions should be prepared at the time of dosing and may NOT be stored. The suspension is dosed immediately after preparation. All preparation was performed in a clean working space.

Inspection of glass and spoons prior to each dose preparation was performed to ensure that the supplies are clean, dry and particulate-free.[3]

No specific safety precautions were described; however, during preparation, handlers were instructed not to breathe in crizotinib dust and to avoid crizotinib contact with skin, eye, and clothing. The preparation area and supplies were cleaned immediately after completion of the dosing process. Local disposal and drug accountability practices were utilized. The patient and/or caregiver should go through practice preparation of the instructions to gain familiarity with this procedure.[3]

Supplies Used

Pfizer cannot make specific recommendations regarding the types of supplies used. The following was used in the clinical study program.[3]

- Crizotinib capsule(s)
- 6-8 ounce drinking glass
- Tap water or bottled water
- Stirring spoon
- Tablespoon (1 TBSP=15 mL) measuring spoon
- Mint candies (optional and used for masking any unpleasant taste)

Instructions

Preparation and Administration of Oral Liquid Suspension Formulation as Used in Clinical Studies[3]

Step 1	One cup of boiling water (tap or bottled water) was prepared.
Step 2	1 TBSP of the above boiling water was measured and poured into the drinking glass.
Step 3	The prescribed crizotinib dosage was added to the drinking glass. Note: The capsule was not opened.
Step 4	Using the spoon, the solution in Step 3 was stirred continuously for at least 2 minutes.
Step 5	A mint candy was administered to the patient (optional) allowing it to dissolve in mouth to aid in masking undesirable taste.
Step 6	Another 1 TBSP of boiling water was measured and poured into the drinking glass containing the crizotinib dosage.
Step 7	This solution was stirred with a spoon continuously for another 2 minutes.
Step 8	An additional 1 TBSP of room temperature or cold water was poured into the solution, rinsing the stirring spoon in the process such that any solids on the spoon were removed and rinsed into the glass.
Step 9	Any undissolved candy in the mouth of the patient was removed.
Step 10	Immediately before dosing, the drinking glass's contents were swirled vigorously for 10 seconds.
Step 11	The suspension was administered to the patient.
Step 12	In the same drinking glass, 1 TBSP of room temperature water was added, rinsing the inside walls of the glass in the process. The contents were swirled vigorously for 10 seconds. This was administered to the patient for drinking.
Step 13	Repeat Step 12 two more times.
Step 14	One mint candy was administered to the patient (optional).
Note: Specific pharmacokinetic studies have not been performed to evaluate the method of capsule disintegration for ease of oral administration as described in this letter using the clinical supply crizotinib formulation. Therefore, Pfizer Inc cannot guarantee that the use of this method will result in comparable safety or efficacy outcomes to that observed with oral administration of the intact commercially available crizotinib capsule formulation.	

In-Use Shelf-Life and Storage Requirements

The crizotinib suspension is dosed immediately after preparation. A stability study conducted showed no chemical degradation during dose delivery using the above instructions. A shelf life of 2 hours has been validated. No extended stability or temperature excursion data for this formulation is available.[3]

REFERENCES

1. Xalkori® (crizotinib) capsules. Package Insert. Pfizer.
2. Data on file (57). Pfizer.
3. Data on file (11). Pfizer.
4. Data on file (14). Pfizer.

Instructions for Patients

Preparation and Administration of Oral Liquid Suspension of Crizotinib (Xalkori)

- **Crizotinib capsule(s)**
- **6-8 ounce drinking glass**
- **Tap water or bottled water**
- **Stirring spoon**
- **Tablespoon (1 TBSP=15 mL) measuring spoon**
- **Mint candies (optional and used for masking any unpleasant taste)**

Instructions

Step 1	Prepare one cup of boiling water (tap or bottled water).
Step 2	Pour 1 TBSP of the above boiling water into the drinking glass.
Step 3	Add prescribed crizotinib dose to the drinking glass. Note: do not open capsule.
Step 4	Using the spoon, stir the solution prepared in Step 3 for at least 2 minutes.
Step 5	Optional: dissolve mint candy in patient's mouth to aid in mask undesirable taste.
Step 6	Pour another 1 TBSP of boiling water into the drinking glass containing the crizotinib dose.
Step 7	Stir solution with a spoon continuously for another 2 minutes.
Step 8	Pour and additional 1 TBSP of **room temperature or cold** water into the solution, rinsing the stirring spoon in the process such that any solids on the spoon are removed and rinsed into the glass.
Step 9	Remove any undissolved mint candy from patient's mouth.
Step 10	Swirl drinking glass's contents vigorously for 10 seconds.
Step 11	Immediately administer suspension to the patient.
Step 12	In the same drinking glass, add 1 TBSP of room temperature water, rinsing the inside walls of the glass in the process. Swirl the contents vigorously for 10 seconds, and administer to the patient for drinking.
Step 13	Repeat Step 12 two more times.
Step 14	Optional: Administer one mint candy to the patient.

APPENDIX I: SUBJECT DIARY ON NF2 CRIZOTINIB PROTOCOL

Patient Name: _____ Cycle #: _____ Start Date: ___/____/_____

Dose Level (Circle one)	BSA [m^2]	Dose
A	**<0.45**	**200 mg once daily**
B	**0.45-0.70**	**250 mg once daily**
C	**0.71-0.88**	**200 mg twice daily**
D	**≥0.89**	**250 mg twice daily**

Instructions:

1) *Every day, record each dose of crizotinib taken*
2) *Crizotinib should be taken once or twice daily, with or without food, depending on prescribed dose level. For twice daily administration, crizotinib should be taken approximately 12 hours apart, preferably in the morning and evening.*
3) *If missing a dose (e.g., due to vomiting), do not take another dose. Resume dosing with the next scheduled dose.*

	Date	*AM Dose taken (check)*	*PM Dose taken (check)*	*Comments*
Day 1				
Day 2				
Day 3				
Day 4				
Day 5				
Day 6				
Day 7				
Day 8				
Day 9				
Day 10				
Day 11				
Day 12				
Day 13				
Day 14				

Day 15				
Day 16				
Day 17				
Day 18				
Day 19				
Day 20				
Day 21				
Day 22				
Day 23				
Day 24				
Day 25				
Day 26				
Day 27				
Day 28				