FDA Briefing Document

NDA 215500

Drug name: eflornithine (DFMO)

Applicant: USWM, LLC

Oncologic Drugs Advisory Committee Meeting

October 4, 2023

Division of Oncology 2/Office of Oncologic Diseases

DISCLAIMER STATEMENT

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Glossary

AC	Advisory Committee
ASCT	Autologous stem cell transplant
BD	Briefing Document
BRF	Benefit-Risk Framework
CBER	Center for Biologics Evaluation and Research
CDER	Center for Drug Evaluation and Research
CDTL	Cross-Discipline Team Leader
ECT	Externally controlled trial
EFS	Event-free survival
ELDA	extreme limiting dilution analysis
FDA	Food and Drug Administration
HRNB	High-risk neuroblastoma
IA	integrated assessment
INRC	International Neuroblastoma Response Criteria
ODC	Ornithine decarboxylase
OS	Overall survival
PD	pharmacodynamic
REMS	risk evaluation and mitigation strategy
RPM	Regulatory Project Manager
SAP	Statistical Analysis Plan
SD	standard deviation

1 Executive Summary/Draft Points for Consideration by the Advisory Committee

On November 21, 2022, US World Meds submitted a New Drug Application (NDA) pursuant to Section 505(b)(2) of the Federal Food, Drug, and Cosmetic Act for eflornithine (DFMO) for the following proposed indication:

- To reduce the risk of relapse in pediatric patients with high-risk neuroblastoma who have completed multiagent, multimodality therapy

DFMO is an oral irreversible inhibitor of the enzyme ornithine decarboxylase (ODC), the first and rate-limiting enzyme in the biosynthesis of polyamines and a transcriptional target of *MYCN*. In the proposed product label, eflornithine is administered orally twice daily for two years or until recurrence of disease or unacceptable toxicity. The proposed recommended dosage is based on body surface area (BSA).

The U.S. Food and Drug Administration (FDA) is convening the Oncologic Drugs Advisory Committee (ODAC) to discuss the strengths and limitations of the evidence provided by the Applicant to conclude that DFMO, administered after completion of multimodality treatment, improves event-free survival (EFS) in pediatric patients with high-risk neuroblastoma (HRNB).

The Applicant submitted results of a single externally controlled trial (ECT) and supportive data to provide evidence of effectiveness for DFMO to reduce the risk of relapse in pediatric patients with HRNB who have completed multiagent, multimodality therapy. The ECT was designed to compare data from Study 3(b), a multi-center, open-label, single arm trial of eflornithine monotherapy for up to 2 years in pediatric patients with HRNB in remission after immunotherapy, to an external control arm constructed from patients with HRNB enrolled on Study ANBL0032, a multi-center, open-label, randomized trial of immunotherapy plus standard up-front therapy vs. up-front therapy alone.

The efficacy population for the comparative analysis of Study 3(b) and ANBL0032 included patients from both studies who were less than 21 years of age with histologic verification of HRNB and who demonstrated at least a partial response based on imaging, with no evidence of disease in the bone marrow at the end of immunotherapy, and did not experience an EFS event prior to starting eflornithine therapy (for Study 3(b)), or for at least 30 days from the end of immunotherapy (for ANBL0032). Eligible patients on Study 3(b) received immunotherapy while enrolled on ANBL0032 or were treated according to the ANBL0032 protocol off study; notably, 94% of patients in the primary matched population from Study 3(b) received immunotherapy on ANBL0032 within the preceding 1 to 4 months (the remaining 6% were treated as per ANBL0032 off study).

A total of 90 DFMO-treated patients who met the criteria for the comparison and had complete data for specified clinical covariates were matched (1:3) using propensity scores to 270 ANBL0032 control patients for a comparison of outcomes. The primary endpoint was EFS,

defined as the time from the end of immunotherapy until the first occurrence of disease progression, relapse, secondary cancer, or death due to any cause. Overall survival (OS), defined as death due to any cause, was a secondary endpoint.

The Applicant's primary analysis resulted in a hazard ratio (HR) for EFS of 0.48 (95% Confidence Interval [CI]: 0.27, 0.85) and 0.32 (95% CI: 0.15, 0.70) for OS, favoring the DFMO arm. Generally similar results were observed in multiple sensitivity and supportive analyses conducted to assess the impact of potential differences between the investigational and control arms.

In addition to the results of the single externally controlled trial, the Applicant provided supportive evidence including nonclinical data and data from exploratory clinical studies and an expanded access program of DFMO in related populations. The supportive clinical data includes results of an analysis of EFS at 2 years in 35 patients with relapsed/refractory neuroblastoma enrolled in a separate cohort of Study 3(b) (Stratum 2), data from a dose escalation study of DFMO alone and in combination with oral etoposide in patients with relapsed or refractory HRNB, as well as data from an expanded access program. Interpretation of the potential supportive clinical evidence is limited by the small number of patients in each study; single arm nature of Study 3(b) Stratum 2; and the lack of prespecified, standardized response evaluation in the expanded access program. In some cases, supportive evidence may be found in evidence of effectiveness in other tumor types; however, there are no approved oncology indications for DFMO, and investigations in cancer over decades of study have not produced robust evidence of efficacy in other tumor types. Preliminary evidence of a non-statistically significant improvement in OS in patients with anaplastic glioma who received chemotherapy with DFMO compared to patients who received chemotherapy alone, has led to further study in this disease.

Randomized controlled trial (RCT) designs are the gold standard for evaluation of time to event endpoints, and FDA initially recommended that an RCT be conducted to evaluate efficacy in this disease setting. Subsequently, a large single arm trial (Study 3[b]) was conducted by an investigator sponsor, and the data appeared to suggest a substantive improvement over a benchmark historic control. The investigator sponsor proposed an ECT design given the challenges of conducting an RCT in the face of this preliminary information, in addition to pre-existing challenges in conducting an RCT in a timely manner in a rare disease. The use of an external controlly controlled trial to support a marketing application may be acceptable in the setting of a rare disease with a well-defined natural history and poor prognosis if the expected treatment effect is estimated to be large, particularly in a setting where conduct of an RCT may be infeasible. Although a randomized trial may have been initially feasible in patients with HRNB, the review team considered the use of an external control in this unique setting to be reasonable given the external control data source (e.g., patient-level data from a large, randomized clinical trial), similarity of the propensity score matched Study 3(b) and ANBL0032 populations, and difficulty in conducting a new randomized controlled trial in light of the published results of DFMO for the proposed indication. FDA review has noted that the ECT

appears adequate and well-controlled with a clear statement of objectives, an appropriately constructed design with appropriate patient selection, and reliably conducted clinical assessments to evaluate the effect of the drug.

However, while the propensity score matched external control and treatment arm patient populations appear sufficiently similar with respect to major baseline demographic and disease characteristics for HRNB to permit a comparative analysis, there are inherent limitations in interpreting the estimated treatment effect of a single non-randomized, externally controlled trial (e.g., factors leading to residual and unmeasured confounding). These potential biases may impact the estimated treatment effect in ways that may not be directly quantifiable. Examples of such factors include social determinants of health; supportive care, which may improve over time or differ by geographic location; and biological factors, such as tumor cytogenetics, which were not collected on ANBL0032.

FDA is seeking the opinion of the committee on the strengths and limitations of the evidence of effectiveness provided by the externally controlled trial to support the use of DFMO in pediatric patients with high-risk neuroblastoma.

FDA Guidance for Industry, "Demonstrating Substantial Evidence of Effectiveness for Human Drug and Biological Products" (FDA, 2019), states that "in general, substantiation of a drug's effectiveness obtained with two trials, especially with complementary design, will provide more convincing evidence of effectiveness than would a single trial." However, "under certain circumstances and consistent with FDAMA, FDA can conclude that one adequate and well-controlled clinical investigation plus confirmatory evidence is sufficient to establish effectiveness," particularly in the setting of a large multicenter trial where no single trial site is the main contributor to observed effect. To support an oncology drug application, FDA typically receives at least one randomized trial with additional confirmatory evidence based on early clinical studies demonstrating an objective response rate. In the case of DFMO, the primary clinical trial uses an external control, and no persuasive clinical data on objective response rate has been provided. While there is nonclinical data supporting a cytostatic mechanism of action (MOA) in neuroblastoma and two independent animal models demonstrating delay in tumor progression, the lack of objective response data is a significant limitation for any cancer drug application and makes it more difficult to assess a drug's effectiveness. Given the reliance on a single externally controlled clinical trial, the additional nonclinical and clinical data submitted by the Applicant are particularly important to support efficacy in this application.

FDA considers independently conducted nonclinical data as well as clinical data from early phase studies and an expanded access program provided by the Applicant to be sources of potential confirmatory evidence.

FDA is seeking the opinion of the committee on the strengths and limitations of the potential confirmatory evidence.

FDA acknowledges the high unmet need and the practical challenges in conducting a new randomized clinical trial in the proposed indication at this time. The quality of clinical evidence to establish effectiveness and the resulting level of certainty about the demonstration of substantial evidence is impacted by the selection of trial design and endpoints, as well as statistical considerations. Given the nature of the ECT and cytostatic mechanism of action of DFMO, the overall evidentiary submission provides a higher level of uncertainty than is typically seen in oncology applications. FDA is seeking input from the committee regarding the strength of the single externally controlled trial and the additional supportive clinical and nonclinical data provided, in the context of the rarity and unmet need of the proposed indication.

1.1 Purpose/Objective of the AC Meeting

The FDA Division of Oncology 2 is convening this ODAC meeting to discuss the following key issues, which the FDA considers relevant to a determination regarding whether substantial evidence of effectiveness has been established for DFMO for the treatment of patients with high-risk neuroblastoma following multimodality therapy.

1. Discuss the strengths and limitations of the externally controlled trial results to support the use of DFMO in pediatric patients with high-risk neuroblastoma.

2. Discuss the strengths and limitations of the additional nonclinical and clinical data to support the use of DFMO in pediatric patients with high-risk neuroblastoma.

1.2 Context for Issues to Be Discussed at the AC

Disease Background

Neuroblastoma is a childhood cancer that originates in the sympathetic nervous system, typically occurring in or near the adrenal glands. It accounts for 8-10% of childhood cancers, with 700-800 cases diagnosed per year in the US (ASCO, 2023). Approximately half of these patients have high-risk neuroblastoma based on age and tumor characteristics, including presence of tumor *MYCN* amplification (DuBois, 2022). *MYCN* amplification, defined as a greater than 4-fold increase in the *MYCN* signal number compared with a reference probe by fluorescence in situ hybridization (Ambros, 2009), is associated with a poor prognosis in HRNB.

Typical front-line multi-modality therapy for HRNB includes induction chemotherapy, surgical resection if indicated for any residual tumor, consolidative high dose chemotherapy with subsequent hematopoietic autologous stem cell transplantation (ASCT) and radiation, followed

by immunotherapy and differentiating therapy with 13-cisretinoic acid (cis-RA) (Yu, 2021). Patients in remission receive no further pharmacologic therapy; however, there is a high risk of relapse, with most relapses occurring within the first 2 years after up-front treatment. The historical EFS rate after standard up-front therapy is 66% at 2 years and 50% at 5 years (Irwin, 2021). After relapse, survival is poor, with a 5-year OS rate of less than 10% (Moreno, 2020). There are no FDA-approved therapies to reduce the risk of relapse in patients with HRNB after front-line therapy.

Information Relevant to the Mechanism of Action

The enzyme ornithine decarboxylase (ODC) is the first and rate-limiting enzyme in the biosynthesis of polyamines and a transcriptional target of *MYCN.* This enzyme is particularly relevant in NB because the ODC gene is found upstream of *MYCN*. Elevated polyamines act as oncometabolites in neuroblastoma (Casero, 2018). Overexpression of *ODC1,* the gene encoding ODC, has been correlated with poor therapeutic outcomes in *MYCN*-amplified neuroblastoma, with inferior survival rates observed in patients overexpressing *ODC1* compared to those with low *ODC1*, even when stratified by *MYCN* status (Hogarty, 2008; Bassiri, 2015; Geerts, 2010). In addition, high expression of the miRNA binding protein *LIN28B* has been reported to be associated with worse survival outcomes in patients with neuroblastoma (Diskin, 2012). These results are consistent with mechanistic data demonstrating that LIN28B upregulates *MYCN* post-transcriptionally in neuroblastoma cells (Balzeau, 2017), and polyamines regulate LIN28 expression in cancer (Paz, 2014).

Drug Product Information and Mechanism of Action

DFMO is an oral irreversible inhibitor of ODC. DFMO-induced inhibition of polyamine synthesis restores the balance of the LIN28/Let-7 metabolic pathway, which is involved in regulation of cancer stem cells and glycolytic metabolism (Lozier, 2014). DFMO also induces in vitro senescence and suppresses neurosphere formation in both *MYCN*-amplified and *MYCN* non-amplified neuroblastoma cells (data submitted by the Applicant). Thus, DFMO is cytostatic rather than cytotoxic and targets tumor-initiating cells, thereby preventing or delaying tumor formation and improving survival in two animal models of *MYCN*-amplified neuroblastoma (Hogarty, 2008; Rounbehler, 2009; data submitted by the Applicant).

In the proposed product label, DFMO is administered orally twice daily for two years or until recurrence of disease or unacceptable toxicity. The proposed recommended dosage is based on body surface area (BSA) as follows:

Body Surface Area (m^2)	Dosage
>1.5	768 mg orally twice a day
0.75 to 1.5	576 mg orally twice a day
0.5 to < 0.75	384 mg orally twice a day
0.25 to < 0.5	192 mg orally twice a day

Regulatory Considerations

FDA approval requires substantial evidence of effectiveness to be established by two or more adequate and well-controlled trials or by a single adequate and well-controlled trial with supportive evidence (FDA draft guidance for industry, Demonstrating Substantial Evidence of Effectiveness for Human Drug and Biological Products, 2019). The quality of clinical evidence to establish effectiveness and the resulting level of certainty about the demonstration of substantial evidence is impacted by the selection of trial design and endpoints, as well as statistical considerations. The "substantial evidence" of effectiveness standard in the statute refers to both the quality and quantity of evidence. In 1962, Congress defined substantial evidence as "evidence consisting of adequate and well-controlled investigations…on the basis of which it could fairly and responsibly be concluded by such experts that the drug will have the effect it purports or is represented to have…" (FD&C Act section 505(d) [21 U.S.C. § 355(d)]).

In this NDA, the Applicant submitted the results of one ECT with potential supportive clinical and nonclinical data. To determine whether the results provide substantial evidence of effectiveness of DFMO for the proposed indication, the FDA review team considered the following critical issues:

1. Whether the external control is appropriate for use,
2. Whether the single ECT is adequate and well-controlled, and
3. Whether the substantial evidence of effectiveness can be established using the results of the single ECT with confirmatory clinical and nonclinical evidence.

If substantial evidence of effectiveness is established by the results of the single trial with confirmatory evidence, FDA would then make an overall benefit:risk assessment that considers the safety of the drug in the intended population.

Appropriateness of Externally Controlled Trial

An externally controlled trial is defined by the International Conference on Harmonisation (ICH) of Technical Requirements for Registration of Pharmaceuticals for Human Use as a trial which

"compares a group of subjects receiving the test treatment with a group of patients external to the study, rather than to an internal control group consisting of patients from the same population assigned to a different treatment" (ICH, 2000). As also noted by the ICH Guideline, "tests of statistical significance carried out in such studies are less reliable than in randomized trials."

For regulatory purposes, randomized trials are generally required to assess the effect of a drug on a time-to-event endpoint (e.g., event-free survival, overall survival) because randomization controls for both known and unknown prognostic factors. In specific circumstances, such as when randomized trials are infeasible or impractical, an adequate and well-controlled trial may utilize an external control; however, regulations stipulate that a valid analysis of the treatment effect must be based on "comparable patients or populations" (21 CFR 314.126). As discussed in the 2019 FDA Draft Guidance for Industry: Demonstrating Substantial Evidence of Effectiveness for Human Drug and Biological Products, externally controlled trials differ in several important ways from randomized trials. Most notably, randomization is not a feature of external control designs and is the most powerful design feature to facilitate balance in known and unknown confounding influences. As a result, there may be differences in baseline patient characteristics or concomitant treatments in the trial population compared to the external control population that may lead to differences in outcomes that are independent of the investigational treatment. The Substantial Evidence draft guidance also states that the level of support for effectiveness provided by an externally controlled trial is strengthened if the following conditions are present:

- the natural history of a disease is well defined,
- the external control population is very similar to that of the treatment group,
- concomitant treatments that affect the primary endpoint are not substantially different between the external control population and the trial population, and
- the results provide compelling evidence of a change in the established progression of disease (such as partial or complete response in a disease where spontaneous regression is not observed).

Based on these factors and given the design and estimated treatment effect observed, FDA considers the use of the submitted ECT to be acceptable support for the primary outcome measure of EFS for this marketing application.

Adequate and Well-Controlled Studies

As described in 21 CFR 314.126, "the purpose of conducting clinical investigations of a drug is to distinguish the effect of a drug from other influences, such as spontaneous change in the course of the disease, placebo effect, or biased observation…Reports of adequate and well-controlled investigations provide the primary basis for determining whether there is

'substantial evidence' to support the claims of effectiveness for new drugs." According to 21 CFR 314.126, an adequate and well-controlled trial has key features including:

- A clear statement of objectives and methods of analysis
- A design which permits a valid comparison with a control
- Adequate assurance that subjects have the condition being studied
- Adequate measures to minimize bias in subject assignment to treatment group
- Adequate measures to minimize bias on the part of subjects, observers, and analysts of the data
- Well-defined and reliable methods to assess response, and
- Adequate analysis of the results of the study to assess the effect of the drug.

FDA's initial review has considered the externally controlled trial in this Application to be adequate and well-controlled, permitting an assessment of efficacy. The FDA will solicit additional perspective from the oncology drug advisory committee regarding strengths and limitations of the externally controlled trial.

Substantial Evidence of Effectiveness: Single Trial with Confirmatory Evidence

The FDA Substantial Evidence of Effectiveness Guidance (2019) states that "in general, substantiation of a drug's effectiveness obtained with two trials, especially with complementary design, will provide more convincing evidence of effectiveness than would a single trial." The Food and Drug Administration Modernization Act (FDAMA) of 1997 amended the statutory provision on substantial evidence of effectiveness such that "under certain circumstances…FDA can conclude that one adequate and well-controlled clinical investigation plus confirmatory evidence is sufficient to establish effectiveness," taking into consideration the persuasiveness of the single trial, robustness of the confirmatory evidence, and the seriousness of the disease.

Examples of evidence that may serve as confirmatory evidence are provided in the 2019 FDA Guidance, and include:

- An adequate and well-controlled investigation demonstrating effectiveness of a drug in a closely related approved indication.
- Earlier phase clinical results or testing that provide compelling mechanistic evidence in the setting of well-understood disease pathophysiology. The guidance states that generally clinical testing using a relevant and well understood pharmacodynamic endpoint would be used to provide mechanistic support, but data from relevant animal models may be used alone or in combination with clinical data supporting the mechanism of action.

- Well-established natural history of the disease which reinforce a very persuasive finding.
- Scientific knowledge of the effectiveness of other drugs in the same pharmacological class, obtained through adequate and well-controlled trials.

When considering this Application, FDA recognizes that it is appropriate to exert regulatory flexibility in applying the statutory standards of safety and effectiveness in the evaluation of therapies intended to treat persons with life-threatening illnesses, particularly when there is no satisfactory alternative therapy, as outlined in 21 CFR 312, subpart E (21 CFR 312.8). The FDA also considers that a higher degree of uncertainty may be acceptable given the poor prognosis of pediatric patients with high-risk neuroblastoma given the high likelihood of relapse after up-front therapy.

Nevertheless, the requirement for substantial evidence of effectiveness to be established by an adequate and well controlled trial with supportive evidence or two or more adequate and well-controlled trials applies irrespective of the degree of unmet medical need. In order to render an approval decision, FDA must reach the conclusion that the application contains substantial evidence of effectiveness, taking into account the level of uncertainty and degree of regulatory flexibility that are appropriate in the context of the strength of the scientific evidence, in addition to risks of the drug and degree of unmet medical need.

1.3 Brief Description of Issues for Discussion at the AC

FDA approval requires substantial evidence of effectiveness to be established (FD&C Act section 505(d) [21 U.S.C. § 355(d)]); such evidence must be generated by one or more adequate and well-controlled investigations. To establish a drug's effectiveness, it is essential to distinguish the effect of the drug "from other influences, such as spontaneous change in the course of the disease, placebo effect, or *biased observation* (emphasis added)" (21 CFR 314.126).

The Division of Oncology 2 is seeking an ODAC meeting to facilitate discussion regarding the NDA for DFMO, which was submitted based on results of a single-arm, multicenter trial with a primary endpoint of EFS, employing an external control arm to interpret the event-free survival results. Specifically, FDA requests discussion on the strengths and limitations of the evidence of effectiveness of DFMO, including the strengths and limitations of the single externally controlled clinical trial, and the supportive clinical and nonclinical data.

1.4 Draft Points for Consideration

1. Discuss the strengths and limitations of the evidence of the externally controlled trial to support the use of DFMO in pediatric patients with high-risk neuroblastoma.

2. Discuss the strengths and limitations of the additional nonclinical and clinical data to support the use of DFMO in pediatric patients with high-risk neuroblastoma.

3. Has the Applicant provided sufficient evidence to conclude that DFMO improves event-free survival in patients with high-risk neuroblastoma?

2 Introduction and Background

2.1 Background of the Condition/Standard of Clinical Care

Neuroblastoma is a childhood cancer that originates in the sympathetic nervous system, typically occurring in or near the adrenal glands. It accounts for 8-10% of childhood cancers, with 700-800 cases diagnosed per year in the US (ASCO, 2023). Approximately half of these patients have HRNB based on age and tumor characteristics (DuBois, 2022).

Typical front-line multi-modality therapy for HRNB includes induction chemotherapy, surgical resection if indicated for any residual tumor, consolidative high dose chemotherapy with subsequent hematopoietic ASCT and radiation, followed by immunotherapy and differentiating therapy with 13-cisretinoic acid (cis-RA) (Yu, 2021). Patients in remission receive no further pharmacologic therapy; however, there is a high risk of relapse, with most relapses occurring within the first 2 years after up-front treatment. The historical EFS rate is 66% at 2 years and 50% at 5 years (Irwin, 2021). After relapse, survival is poor, with 5-year overall survival (OS) of less than 10% (Moreno, 2020). There are no FDA-approved therapies to reduce the risk of relapse in patients with HRNB after front-line therapy.

2.2 Pertinent Drug Development and Regulatory History

Eflornithine (Ornidyl) was FDA-approved in 1990 in an intravenously administered injectable formulation for West African Sleeping Sickness, but never marketed due to commercial viability reasons unrelated to safety or efficacy. The current 505(b)(2) application for DFMO relies upon some of FDA's prior nonclinical and clinical safety findings from its review of Ornidyl, the reference listed drug. Eflornithine (Vaniqa) topical cream was approved for the reduction of unwanted facial hair in women in 2000. There are no existing oncologic indications for eflornithine/DFMO. The drug also has a long history of evaluation in patients with cancer.

In neuroblastoma, DFMO was initially developed by an investigator sponsor through multiple interactions with the FDA. In a 2015 meeting, FDA stated that a "randomized controlled trial is required to scientifically assess the effect of DFMO as a maintenance therapy."

Despite efforts by the sponsor to assess the feasibility of a randomized trial in this setting, the sponsor elected to develop the drug in single arm cohorts. The primary study supporting efficacy in this application, Study 3(b), was originally conducted under a research IND and

subsequently transferred to the Applicant for further development. On November 22, 2017, Orphan Drug Designation was granted for the treatment of neuroblastoma. In 2018, a preliminary Breakthrough Therapy Designation (BTD) discussion was held with FDA to discuss the results of the pivotal trial, Study 3(b), compared to a historical control rate from Children's Oncology Group (COG) Study ANBL0032. The sponsor contended that a randomized trial would lack equipoise given public knowledge of the results. FDA recommended that the sponsor provide patient-level data from the studies intended to support the request, including Study ANBL0032. On April 3, 2020, FDA granted BTD to DFMO for the treatment of pediatric patients with high-risk neuroblastoma with no evidence of disease or no active disease after first-line multiagent, multimodality therapy based on propensity score matched external control data from Study ANBL0032.

The FDA held multiple meetings with the Applicant throughout development to discuss the DFMO program. FDA considered that although a randomized trial may have been initially feasible in patients with HRNB, the use of an external control in this unique setting could be reasonable given the external control data source (e.g., patient-level data from a large, randomized clinical trial), similarity of the propensity score matched Study 3(b) and ANBL0032 populations, and difficulty in conducting a new randomized controlled trial in light of the published results of DFMO for the proposed indication.

Table 1: Key Regulatory History Relevant to NDA 215500

Date	Key Interactions
6/1/2012	Study 3(b) was initiated by an investigator sponsor.
11/18/2015	FDA and the investigator sponsor held a Type B meeting to discuss a proposed Phase II study design and a potential registration pathway for DFMO in pediatric high-risk neuroblastoma. **FDA stated that a "randomized controlled trial is required to scientifically assess the effect of DFMO as a maintenance therapy."**
2/23/2016	FDA and the sponsor held a Type C meeting to discuss the design of proposed pivotal studies and a potential registration pathway for DFMO in pediatric high-risk neuroblastoma. Discussion focused on Study NMTRC014, a single-arm, open-label study of patients with high-risk neuroblastoma in remission initiated after Study 3(b), and a proposed multicenter, randomized controlled trial of DFMO administered during immunotherapy.
12/19/2018	FDA and the sponsor held a Preliminary Breakthrough Therapy Designation Request Advice teleconference to discuss the results of Study 3(b) compared to a historical control rate; FDA recommended comparison to patient-level data from the studies intended to support the request.
4/3/2020	FDA granted **Breakthrough Therapy Designation** (BTD) for the treatment of pediatric patients with high-risk neuroblastoma with no evidence of disease or no active disease after first-line multiagent, multimodality therapy based the

	results of Study 3(b) using a propensity score matched analysis with external control data from Study ANBL0032.
9/24/2020	FDA and the Applicant held a Type B BTD meeting to discuss clinical, nonclinical, biopharmaceutic and clinical pharmacology components of the DFMO program.
10/15/2020	FDA and the Applicant held a Type B CMC-focused meeting to discuss plans for an accelerated CMC development program and specific CMC-related questions regarding the nonclinical package proposed for the NDA.
2/19/2021	FDA and the Applicant held an informal teleconference to discuss the Statistical Analysis Plan for the Comparison of Study 3(b) to ANBL0032 External Control Database to DFMO IND 144875
5/3/2021	FDA and the Applicant held a Type B BTD meeting to discuss the final proposal for the statistical analysis plan prior to finalization.
6/29/2021	FDA and the Applicant held a Type B BTD meeting to discuss the final proposal for the statistical analysis plan.
10/25/2021	FDA and the Applicant held a pre-NDA meeting to discuss the results of the propensity score-matched analysis and the planned NDA submission. **FDA stated that the proposed comparison to Study ANBL0032 appeared acceptable, but determination of substantial evidence of effectiveness would be based on an overall assessment of the results of multiple independent analyses.**
1/13/2022	FDA provided feedback on the Blinded Independent Central Review (BICR) Charter and Statistical Analysis Plan (SAP) Addendum via an advice letter.
7/19/2022 – 8/8/2022	FDA and the Applicant discussed issues related to the chemistry, manufacturing, and controls aspect of the proposed application via email.
9/2/2022	FDA requested additional information regarding BICR topline results provided August 26, 2022
10/7/2022	FDA provided agreement to the Applicant's request for participation in the Real-Time Oncology Review (RTOR) program and Project Orbis, and made requests related to efficacy package to be addressed in the NDA.
11/22/2022	505(b)(2) NDA 215500 was submitted.

3 Summary of Issues for the AC

3.1 Efficacy Issues

An oncology application with primary evidence based on an externally controlled trial with no confirmatory ORR data produces a higher level of uncertainty than is typically reviewed, and FDA considered three main efficacy issues. Two of these issues concern evaluation and interpretation of efficacy data from the externally controlled trial (ECT), and one is related to the evaluation of additional data outside of the ECT which may serve as confirmatory evidence. These issues are as follows:

1) the comparability of the external control arm to the investigational arm,
2) the magnitude of the treatment effect relative to potential sources of bias, and
3) the strength of the potential confirmatory clinical and nonclinical evidence.

3.1.1 Sources of Data for Efficacy

The Applicant submitted a 505(b)(2) NDA based on the results of an ECT comprised of a single-arm trial, Study 3(b), compared to an external control arm constructed from patients enrolled on the National Cancer Institute (NCI)/Children's Oncology Group (COG)-sponsored clinical trial, Study ANBL0032.

Table 2: Overview of Clinical Trials to Support Efficacy and Safety

Trial Identity, NCT Number and Trial Design	Study Population	Study Endpoints	Treatment Duration/ Follow Up	Number of Patients	Number of Centers and Countries
NMTRC003b (Study 3b) NCT02395666 Single-arm, multi-center, open-label Supports efficacy and safety evaluation	HRNB patients in remission after upfront (Stratum 1) or refractory/relapse (Stratum 2) therapy	**Primary Efficacy**: EFS at 2 years **Secondary Efficacy**: EFS at 4 years, OS at 2 years Safety and PK Endpoints: Safety: vital signs, audiograms, adverse events and tolerability; PK; biological correlates	2 years of DFMO treatment + 5 years of long term, off-therapy follow up (7 years total)	140 Stratum 1: n=105 Stratum 2: n= 35	22 US sites
NMTRC014 (Study 14) NCT02679144 Single-arm, multi-center, multi-cohort, open-label	HRNB patients in remission after upfront (Strata 1/1b and 2) or relapse/refractory (Strata 3 and 4) treatment	**Efficacy Endpoints:** N/A for application; single arm study contributing safety and PK data **Safety and PK Endpoints:** Safety (laboratory, vital signs,	2 years of DFMO treatment + 5 years of long term, off-therapy follow up (7 years total)	280 enrolled (as of NDA submission date) out of 441 planned Stratum 1: 132 Stratum 1b: 21 Stratum 2: 50 Stratum 3: 29	37 sites (33 in US and 4 in Canada)

Supports safety evaluation		ECGs, audiograms, adverse events) and tolerability, PK, biological correlates, PK and safety		Stratum 4: 48	

Source: Adapted from Assessment Aid submitted to NDA 215500

Study 3(b): Investigational Arm

Study 3(b) (NMTRC003(b); NCT02395666), entitled "A Phase II preventative study of DFMO as a single agent in patients with HRNB in remission," is a multi-center, open-label, single arm trial of eflornithine monotherapy for up to 2 years in pediatric patients with HRNB who completed standard of care up-front therapy including immunotherapy. The trial enrolled patients from June 2012 to February 2016 across 22 sites in the U.S.

Stratum 1, which provided the primary efficacy data for the experimental arm of the ECT, enrolled 105 patients who were in remission at the end of up-front therapy defined as chemotherapy (5-7 cycles), surgery as indicated, consolidation therapy as indicated, radiation therapy as indicated, or anti-ganglioside 2 (GD2) antibody therapy with retinoic acid up to 6 cycles. Remission was defined as patients achieving a disease response of at least partial response (by CT or MRI) at the time of study entry and histologically negative bone marrow aspirate/biopsy. Patients were considered to have no evidence of disease if negative CT/MRI and MIBG scan (or PET for patients with a history of MIBG non-avid disease). Patients were considered to have no active disease if they had stable residual tumor masses visible on CT/MRI provided the residual mass was either MIBG-negative or MIBG-positive without FDG-PET avidity, which was taken as evidence that the mass did not represent active disease and would otherwise not have received additional therapy after antibody therapy.

The primary endpoint was EFS at 2 years compared to a historical control rate of 70% based on clinical trial results from Study ANBL0032. Patients were treated until disease progression, unacceptable toxicity, withdrawal of consent, loss to follow-up, death, or discontinuation from the study treatment due to any other reason. Patients were followed for 5 years after completing DFMO for a total study duration of 7 years. During the off-therapy, long-term follow up, patient contact was required at approximately 6, 12, 18, 24, 36, 48, and 60 months from the end of DFMO treatment. EFS was assessed until disease progression, death, loss to follow-up, or withdrawal of study consent. Patients only withdrawing consent to continue DFMO treatment or who discontinued treatment for a reason other than relapse were still followed for outcomes. Overall survival (OS) was assessed until patients withdrew study consent, died, or were lost to follow-up.

Notably, Study 3 was originally initiated with 250 mg eflornithine hydrochloride tablets provided by Cancer Prevention Pharmaceuticals, Inc. (CPP). Amendment 5 (dated December 18, 2014) was initiated due to a change in source of study drug supply, from CPP to KC Pharmaceuticals, Inc. (KCP). The protocol was initiated as NMTRC003 and was converted to NMTRC003b at the time of the change in drug product supplier. The change to NMTRC003b

also resulted in the creation of a separate study database and separate informed consent from that of NMTRC003. Outcome data for 100 of the 140 total patients, comprising those patients that either transferred from NMTRC003 to NMTRC003b or directly enrolled in NMTRC003b, were reported in the NMTRC003b database. However, because the Applicant did not have access to the NMTRC003 database, Study 3(b) outcome data in the application for the remaining 40 patients (those patients that enrolled only under NMTRC003) were sourced from the "BCC001 database," a retrospective chart review of HRNB outcomes across Beat Childhood Cancer Research Consortium (BCC) sites.

Study 3(b) was not initially designed to support a marketing application for DFMO. However, the Applicant submitted results of Study 3(b) compared to the planned historical control as part of a preliminary breakthrough therapy designation discussion in 2018. Subsequently, as described in Table 1, multiple discussions were held between FDA and the Applicant regarding the potential for patient-level data from Study ANBL0032 to create an external control arm for comparison to Study 3(b) using a propensity score matching algorithm, ultimately with the intention to support an evaluation of the substantial evidence of effectiveness of DFMO for HRNB in remission after up-front therapy.

Study ANBL0032: External Control Data

Study ANBL0032 was a large, Children's Oncology Group (COG)-sponsored, multi-center, open-label, randomized trial of immunotherapy plus standard up-front therapy vs. up-front therapy alone in patients with newly diagnosed HRNB who had previously received induction and consolidation therapy, and who demonstrated at least a partial response prior to autologous stem cell transplant treatment. Patients were enrolled between October 2001 and July 2015 at 197 sites including 172 in the United States, as well as sites in Australia, New Zealand and Canada. The major efficacy outcome measure for ANBL0032 was investigator-assessed EFS, defined as the time from randomization to the first occurrence of relapse, progressive disease, secondary malignancy, or death.

In January 2009, the Data Safety Monitoring Committee performed a review of an interim analysis that demonstrated statistically significantly higher survival (both EFS and OS) in patients randomized to immunotherapy plus cis-RA (Regimen B) as compared to those randomized to standard cis-RA alone (Regimen A): EFS rate of 66 ± 5% vs. 46 ± 5% at 2 years, P=0.01 and OS rate of 86 ± 4% vs. 75 ± 5% at 2 years, P=0.02. As a result, randomization was halted and the study was amended to allow all patients enrolled to receive immunotherapy + cis-RA. Accrual under this amendment was opened on April 20, 2009. Publication of these findings by Yu et al. (2010) resulted in adoption of immunotherapy into the standard of care and the results of ANBL0032 supported the traditional approval of dinutuximab in the United States in 2015.

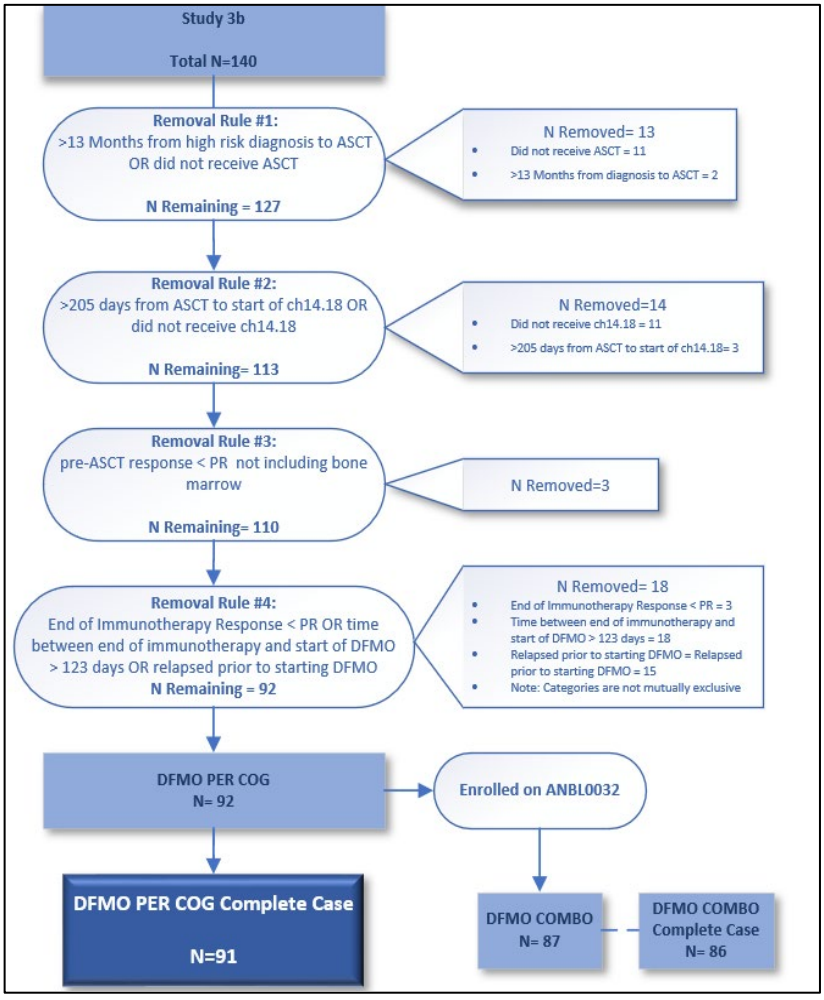
The efficacy population for the comparative analysis of Study 3(b) and ANBL0032 included patients from both studies who were less than 21 years of age with histologic verification of HRNB and who demonstrated at least a partial response based on imaging, with no evidence of disease in the bone marrow, at the end of immunotherapy, and who did not experience an EFS

event prior to starting DFMO therapy (for Study 3(b)), or for at least 30 days from the end of immunotherapy (for ANBL0032).

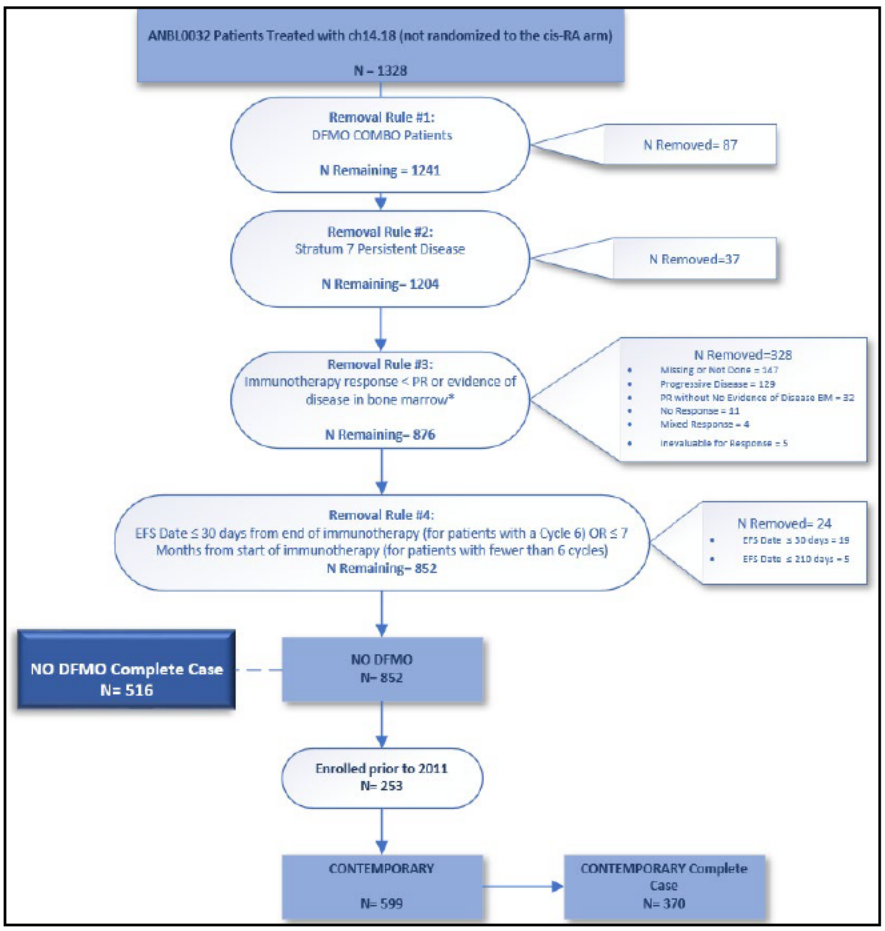
The selection algorithms for Study 3(b) and the ANBL0032 external control populations are provided as flow-charts in Figure 1.

Figure 1: Applicant Analysis of Patient Selection Process for (A) Study 3(b) and (B) ANBL0032 External Control Populations

A



B



Source: Applicant analysis submitted in NDA 215500, Module 5.3.5.4 Comparison of Study 3b to ANBL0032 External Control Database, pages 69 and 75.

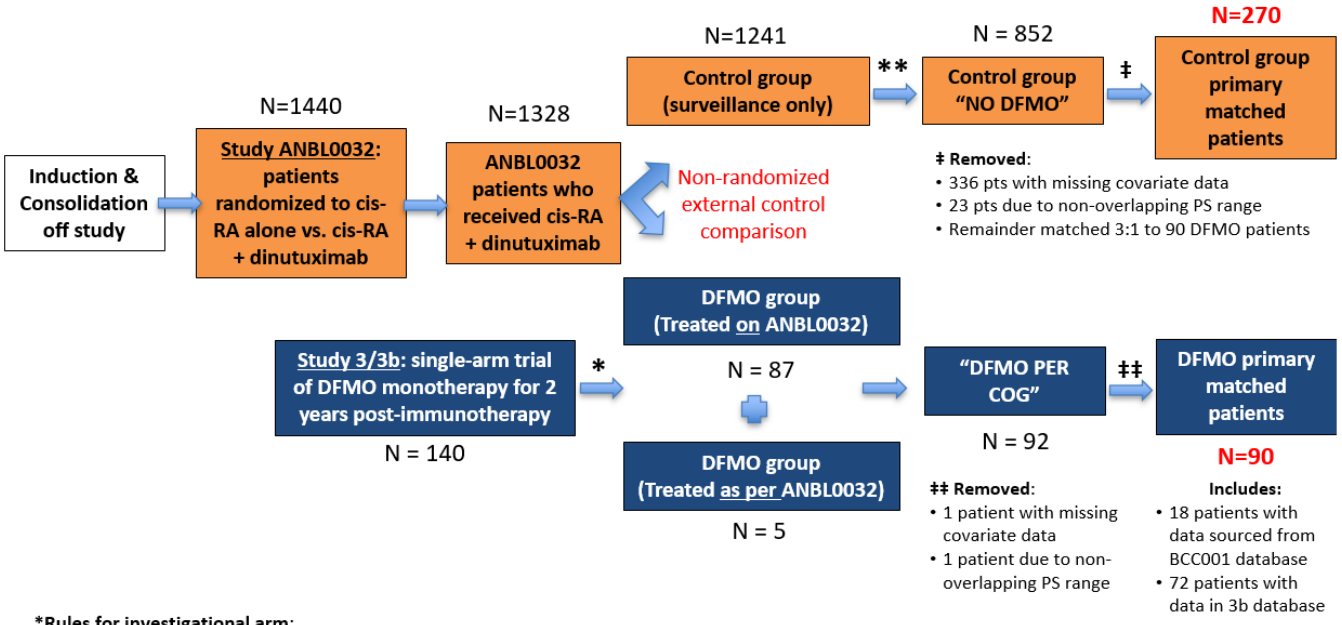
Eligible patients on Study 3(b) received immunotherapy on ANBL0032 or were treated according to the ANBL0032 protocol off study; notably, 94% of patients in the primary matched population from Study 3(b) had been treated on ANBL0032.

Of 140 patients who enrolled on Study 3(b) and received DFMO at the proposed dose, 87 were treated on ANBL0032 prior to enrolling on Study 3(b) and were included in the investigational treatment arm (referred to as DFMO COMBO patients in Figure 1) for the comparative analysis against the external control arm. An additional 5 patients who did not enroll on ANBL0032, but were treated as per the protocol, were also included in the investigational arm. Therefore, a total of 92 patients (referred to as DFMO PER COG patients in Figure 1) from the investigational arm contributed to the comparative analyses of outcomes.

Of 1,440 patients who enrolled on ANBL0032, 1,328 patients received up-front treatment including immunotherapy. After completion of up-front therapy, a total of 1,241 patients were observed with serial imaging and received no further treatment.

To permit an analysis of comparable populations, several pre-specified selection rules were applied to both arms; 852 patients who did not receive DFMO and 92 patients who received DFMO met all of the pre-specified selection rules. Of those, 516 and 91 patients, respectively, had no missing data for 11 pre-specified clinical covariates which were considered to be important predictors of relapse and survival outcomes. Of the 516 and 91 patients, a total of 270 ANBL0032 control patients and 90 DFMO-treated patients were matched (3:1) using propensity scores and an exact matching on *MYCN* status for a comparison of EFS and OS outcomes (Figure 2).

Figure 2: FDA Analysis of Patient Selection Process for Final Matched Populations in Externally Controlled Trial



HRNB = high-risk neuroblastoma; PS = propensity score; CR = complete response; VGPR = very good partial response; PR = partial response; ASCT = autologous stem cell transplantation; BM = bone marrow; EFS = event-free survival

Source: FDA Analysis based on datasets submitted in NDA 215500

A total of 90 DFMO-treated patients who met the criteria for the comparison and had complete data for specified clinical covariates were matched (1:3) using propensity scores and an exact matching on MYCN status to 270 ANBL0032 control patients for a comparison of outcomes. The primary endpoint was EFS, defined as the time from the end of immunotherapy until the first occurrence of disease progression, relapse, secondary cancer, or death due to any cause. A secondary endpoint was overall survival (OS), defined as death due to any cause.

3.1.2 Efficacy Summary

The Applicant proposes to meet the evidentiary standard to establish effectiveness based on the results of a single adequate and well-controlled study with confirmatory evidence.

Analysis of Efficacy Comparing Study 3(b) to Study ANBL0032

The goal of the comparison is to quantify the treatment benefit of DFMO based on the results of Study 3(b), leveraging external data from Study ANBL0032. The eligibility criteria for the two studies were similar and are described in Table 3.

Table 3: Key Eligibility Criteria for Comparative Analysis Populations

	Study 3(b) "A Phase II Preventative Trial of DFMO as a Single Agent in Patients with HRNB in Remission"	**ANBL0032** "Phase III Randomized Study of Chimeric Antibody 14.18 in HRNB Following Myeloablative Therapy and Autologous Stem Cell Rescue"
Age	0 to 21 years at the time of diagnosis	≤ 30.99 years of age at the time of diagnosis
Diagnosis	Histologic verification at the time of original diagnosis or previous relapse of HRNB	HRNB at time of diagnosis (exception: patients who are initially diagnosed as non high-risk neuroblastoma, but later converted (and/or relapsed) to high risk neuroblastoma are also eligible)
Disease Status	HRNB in remission ("no evidence of disease or having no active disease and who would not have otherwise received any further therapy per standard of care") **– Stratum 1:** Patients who are in remission at the end of up-front therapy (defined as chemotherapy [5-7 cycles], surgery as indicated, consolidation therapy as indicated, radiation therapy as indicated, anti-GD2 antibody therapy with retinoic acid up to 6 cycles) **– Stratum 2:** Patients who are in remission after any previous relapse or refractory therapy	• Prior to enrollment a determination of mandatory disease staging was to be performed (tumor imaging studies including CT or MRI, MIBG scan, bone marrow aspiration & biopsy) • At pre-ASCT evaluation patients must have met INRC for CR, VGPR, or PR for primary site, soft tissue metastases and bone metastases. Patients who meet those criteria must also meet the protocol specified criteria for bone marrow response as outlined below: – ≤ 10% tumor (of total nucleated cellular content) seen on any specimen from a bilateral bone marrow aspirate/biopsy. – Patients who had no tumor seen on the prior bone marrow, and then have ≤10% tumor on any of the bilateral marrow aspirate/biopsy specimens done at pre-ASCT and/or pre-enrollment evaluation will also be eligible (note that per INRC this would have

		been defined as “overall” response of progressive disease).
Prior Therapy	> 30 days from completion of cytotoxic and antibody therapy and < 120 days from previous therapy No other ongoing anticancer agents permitted	• Must have completed therapy including intensive induction followed by ASCT and radiotherapy. (Radiotherapy may be waived for patients who have small adrenal masses completely resected up-front or who never had identifiable primary tumor) • No more than 12 months from the first induction chemotherapy after diagnosis to the date of ASCT except for patients initially diagnosed as non HRNB but later converted or relapsed to high-risk. For tandem ASCT patients, this will be the date of the FIRST stem cell infusion. • The date protocol therapy is projected to start must have been no later than 10 days after the date of study enrollment. Patients were enrolled between Day 56 and Day 85 after PBSC infusion (day from 2nd stem cell infusion for tandem transplant). Patients were enrolled no later than Day 100 after PBSC infusion. Enrollment occurred after completion of radiotherapy, and after completion of tumor assessment post-ASCT and radiotherapy.
Performance status	• Lansky score ≥60% • Adequate organ function (ANC >500, PLT >50, AST/ALT <10x ULN, bilirubin <2.0 mg/dL, creatinine normal for age)	• Lansky or Karnofsky Performance Scale score of ≥50% and patients must have had a life expectancy of ≥2 months. • Adequate organ function (APC >1000, ALT <5x ULN, bilirubin <1.5x ULN, creatinine normal for age)

Source: Adapted from Applicant analysis submitted in NDA 215500, Module 5.3.5.4 Comparison of Study 3b to ANBL0032 External Control Database, page 53

ASCT: autologous stem cel transplant, CR: complete response, INRC: International Neuroblastoma Response Criteria, PBSC: peripheral blood stem cell; PR: partial response, VGPR: very good partial response.

For both arms, tumor assessments were required at baseline, 3, 6, 9, 12, 18, and 24 months after completion of immunotherapy and then per institutional standard. Study ANBL0032 had two additional required tumor assessments at 30 and 36 months after completion of immunotherapy. No anti-cancer therapies were permitted during study therapy for patients on

the investigational arm and there are no approved therapies in the maintenance setting after up-front therapy.

The primary endpoint for the comparative analysis was EFS per International Neuroblastoma Response Criteria (INRC) as assessed by investigator. EFS was defined as the period from the index date (end of immunotherapy study visit date) until the first occurrence of relapse, progressive disease, secondary cancer, or death due to any cause. If none of these events occurred, the patient was censored using the last day of contact. The secondary endpoint was OS, defined as the period from the index date until death from any cause. If death did not occur, the patient was censored using the last day of contact.

The Applicant proposed to use clinically important baseline covariates to build the propensity score model for the comparison of Study 3(b) to the external control group from Study ANBL0032. Propensity score and exact matching were used to ensure balance across 11 key clinical covariates, utilizing a 3:1 ratio match within the groups of 516 control patients and 91 patients treated with DFMO who were not missing any key covariate data (out of the 852 and 92 patients, respectively, described in Figure 2). The 10 covariates in the propensity score included: age at high-risk diagnosis, sex, race, stage at diagnosis, pre-ASCT response, transplant type (single vs. tandem), time from transplant to start of immunotherapy, response at end of immunotherapy, time from start of immunotherapy to end of immunotherapy, and time from diagnosis to end of immunotherapy. The Applicant used an exact match for *MYCN* status, as it was considered the most important predictor of relapse and survival outcomes. Overall populations of treated and control patients as well as propensity score matched cohorts of patients were compared for EFS and OS outcomes. The analysis is an unadjusted Cox proportional hazards model using the matched data, evaluating the effect of treatment (DFMO vs. no DFMO) on the primary outcome, EFS.

The index date was the end of immunotherapy, defined as the study visit date at the end of all completed cycles of immunotherapy in ANBL0032, which for most patients is the visit date following completion of 6 cycles of immunotherapy. For patients in the investigational arm (DFMO), if the last date of cis-RA administration was later than the last visit date in the immunotherapy phase of treatment, the date of cis-RA administration was used.

To account for potential bias in the local evaluator reported outcomes, a blinded independent central review (BICR) was conducted for patients in the investigational arm. Source imaging was not available to conduct a similar BICR for the control arm. As all images were available for the investigational arm patients, the same 90 treated patients and the same 270 control patients selected for the Applicant's proposed primary propensity score matched analysis for Study 3(b) vs. ANBL0032 were compared, using BICR reported outcomes rather than local evaluator reported outcomes for the treated patients.

Results

Externally Controlled Trial

The primary analysis included 90 patients treated with DFMO (investigational arm) and 270 patients observed without further treatment after immunotherapy (control arm). In the primary matched population, 85 of 90 patients on the investigational arm had completed immunotherapy on Study ANBL0032 within the prior 120 days with no other intervening anti-cancer therapy, and 5 patients had been treated according to the ANBL0032 protocol off study.The distribution of 11 matched clinical covariates used in the propensity score model and exact matching, as well as unmatched clinical and disease-related characteristics, are listed by arm in Table 4.

Study 3(b) was open at 22 U.S. sites and the 90 patients in the investigational arm were enrolled at 21 of those sites. Study ANBL0032 was open at 172 U.S. sites and 25 international sites, and the 270 patients in the external control arm were enrolled at 115 of those sites (99 in U.S., 11 in Canada, 4 in Australia, and 1 in New Zealand) . All Study 3(b) sites were also ANBL0032 sites. Approximately two-thirds of patients at the sites where both studies were open elected to enroll on Study 3(b) following enrollment on ANBL0032.

All patients in the control arm had an end of immunotherapy overall response recorded status of complete response (CR), very good partial response (VGPR), or partial response (PR); however, the ANBL0032 dataset was limited with regard to imaging and tumor assessments. The exact dates of tumor assessments were not recorded in the case report forms. A 6- to 12-month time window was recorded during which investigators could record “no disease present” or “disease present”, or “failed during period.”

Table 4: Matched and Unmatched Demographic, Disease and Treatment-Related Characteristics by Study Arm

MATCHED CHARACTERISTICS		**DFMO (N=90)**	**External Control (N=270)**
Age at diagnosis (years), median (range)		2.9 (0.1 – 15.8)	3.1 (0.2 – 20.1)
Sex, %	Male	60	58
	Female	40	42
Race, %	White	89	87
	Black or African American	7	6
	Asian	1	5
	Other	3	1
MYCN status (exact match), %	Amplified	44	44

	Not amplified	56	56
Stage at diagnosis, %	4	87	86
	4S	1	1
	Other (1, 2, 3)	12	13
Pre-ASCT response, %	CR	42	42
	VGPR	34	30
	PR	23	28
Single vs. Tandem ASCT, %	Single	91	91
	Tandem	9	9
End of immunotherapy overall response, %	CR	86	87
	VGPR	10	8
	PR	4	5
Days from ASCT to immunotherapy, median (range)		88 (60, 186)	89 (58, 201)
Duration of immunotherapy in days, median (range)		185 (108, 328)	185 (34, 259)
Days from diagnosis to end of immunotherapy, median (range)		460 (391, 1068)	466 (309, 2414)
UNMATCHED CHARACTERISTICS		**DFMO (N=90)**	**External Control (N=270)**
Demographics[a]			
Ethnicity, %	Hispanic/Latino	9	6
	Not Hispanic/Latino	19	93
	Missing	72	1
Geographic site of enrollment on ANBL0032[b], %	United States	99	86
	Canada	1	9
	Australia	0	5
	New Zealand	0	<1
Tumor Characteristics			
Histology, %	Favorable	7	5
	Unfavorable	83	85
	Missing	10	10

Primary Tumor Location[c], %	Adrenal	56	28
	Abdomen	13	13
	Retroperitoneum	14	9
	Other	16	6
	Missing	4	51
Tumor Cytogenetics, %	1p deletion	17	-
	11q deletion	9	-
	17q gain	3	-
	ALK mutation	3	-
	Missing	68	100
Treatment-Related Characteristics			
Days from Diagnosis to ASCT, median (range)		179.5 (137-804)	188 (139-2023)
Surgery during induction[d], %	Yes	93	71
	Missing	2	29
Radiation during consolidation[e], %	Yes	83	90
	Missing	17	10
Transplant regimen, %	Bu/Mel	36	11
	CEM	53	22
	TC and CEM	7	3
	Other/Missing	4	64
Cycles of immunotherapy, %	< 6 cycles	3	< 1
	6 cycles	97	99
End of immunotherapy bone marrow response, %	No evidence of disease	100	74
	Missing	0	1% no change; <1% improved; 25% missing[f]
Lansky performance status at end of immunotherapy, %	100	59	-
	80-90	18	-
	Missing	23	100

End of immunotherapy date, range	03/22/2012 to 01/25/2016	06/03/2005 to 01/28/2016

[a] Social determinants of health (e.g., socioeconomic status) were considered but data were unavailable; [b]All Study 3(b) sites were in the United States; [c]Patients may have multiple primary tumor locations; [d]Extent of surgery not specified; [e]Information regarding dose and type of radiation limited; [f]Optional field on ANBL0032 case report form and all patients with missing bone marrow response had an overall response documented of CR or VGPR
ASCT = autologous stem cell transplantation; Bu/Mel = busulfan/melphalan; CEM = carboplatin/etoposide/melphalan; CR = complete response; PR = partial response; TC = cyclophosphamide/thiotepa; VGPR = very good partial response
Source: FDA analysis based on datasets submitted in NDA 215500

Comparative Analysis:

Based on a data cut-off (DCO) date of June 30, 2021 for Study 3(b) and June 30, 2019 for ANBL0032, the Applicant's proposed primary analysis of event-free survival and overall survival demonstrated a hazard ratio of 0.48 (95% CI: 0.27, 0.85) and 0.32 (95% CI: 0.15, 0.70), respectively (Table 5). Figure 3 displays the Kaplan-Meier curves for EFS and OS for the Applicant's proposed primary propensity-score matched comparison.

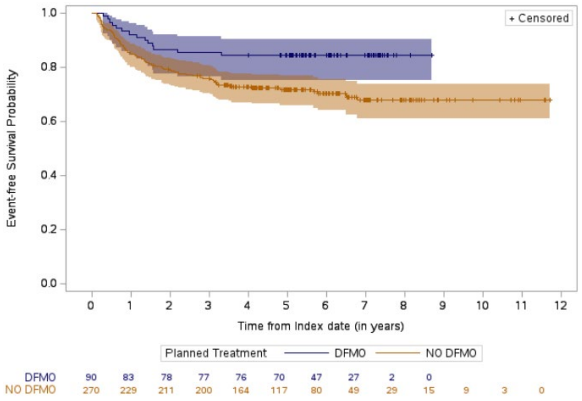
Table 5: Analysis of Primary Endpoint (EFS) and Secondary Endpoint (OS)

	DFMO (N=90)[1]	**NO DFMO** (N=270)[2]
Event-free Survival (EFS)[3,4]		
EFS events, n (%)[5]	14 (16)	79 (29)
Censored, n (%)	76 (84)	191 (71)
Hazard Ratio (95% CI)	0.48 (0.27, 0.85)	
Overall Survival (OS)[6]		
Deaths, n (%)	7 (8)	57 (21)
Censored, n (%)	83 (92)	213 (79)
Hazard Ratio (95% CI)	0.32 (0.15, 0.70)	

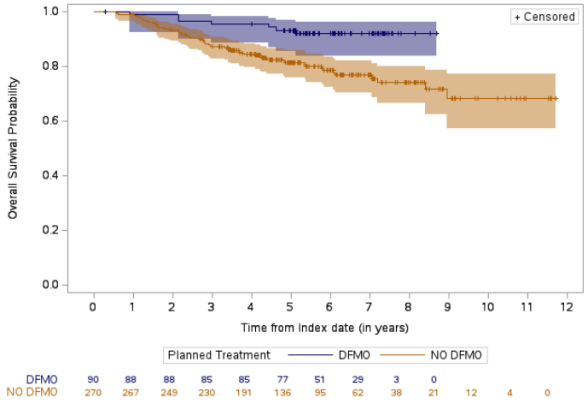
[1]Derived from 91 patients with no missing data out of 92 total eligible patients; [2]Derived from 516 patients with no missing data out of 852 total eligible patients; [3]Final analysis; DCO: Study NMRTC003b, June 2021; Study ANBL0032, June 2019 [4]Descriptive p-value from unstratified log-rank test = 0.0096; [5]2 events were deaths (both in the NO DFMO arm); [6]Descriptive p-value from unstratified log-rank test = 0.0027
Source: FDA Analysis of datasets submitted in NDA 215500

Figure 3: Kaplan-Meier Plot of EFS and OS by Treatment Group (Primary Analysis)

Event-Free Survival



Overall Survival



Source: FDA Analysis of datasets submitted in NDA 215500

3.1.3 Efficacy Issues in Detail

EFFICACY ISSUE 1: Evaluation of the comparability of the external control arm to the investigational arm.

Comparability of the external control arm to the investigational arm was a key review issue in order to assess interpretability of the externally controlled trial. FDA's assessment of the major strengths and limitations of the comparability of populations is outlined in Table 6. In the Applicant's proposed primary analysis population, 85 of 90 patients treated with DFMO in the investigational arm had completed immunotherapy on Study ANBL0032 within the prior 120 days with no other intervening anti-cancer therapy, and 5 patients had been treated according to ANBL0032 off study. As shown in Table 4, matched clinical characteristics were similar across arms. Notably, the Applicant used an exact match for *MYCN* status, given its important prognostic significance. Patients were required to be in remission at the end of immunotherapy, and although all patients were assessed as having at least a partial response, approximately 10% of patients in the control arm did not have available imaging assessments to corroborate the response data.

Non-matched clinical characteristics (also shown in Table 4) included geography. Since ANBL0032 enrolled patients at sites outside the US (including 14% of patients in the matched external control arm), sensitivity analyses which assessed the impact of country of enrollment were performed (refer to discussion under Issue #2). Other non-matched clinical characteristics include demographic information (ethnicity and social determinants of health), disease characteristics (tumor histology, primary tumor location, tumor cytogenetics), pre-immunotherapy treatment characteristics (surgery during induction, radiation during consolidation, and transplant regimen), and post-immunotherapy treatment characteristics (performance status at end of immunotherapy and end of immunotherapy date). These

variables were not included in the propensity score because they were either not collected or had substantial missingness across one or both study arms.

Given that patients who enrolled Study 3(b) elected to go onto an additional clinical trial, whereas patients in the control arm did not, it is possible that the groups differed in ways that may be associated with differences in outcome, such as social determinants of health (SDOH), performance status, or other related unmeasured factors. Since literature suggests that exposure to household poverty has an impact on patient outcomes (i.e., EFS and OS), FDA considered analyses to assess the potential impact of imbalances in SDOH between the investigational and external control arm on the treatment effect (see discussion of Issue #2 below) (Bona, 2021).

Data regarding cytogenetics were limited in ANBL0032. While all of the chromosomal alterations/genetic mutations recorded in patients in the DFMO arm (i.e., 1p deletion, 11q deletion, 17q gain, *ALK* mutations) are associated with worse prognosis, these data were not available for patients on the control arm. Sensitivity analyses performed by FDA assessed the potential impact of these unmeasured confounding factors on the estimated treatment effect in the ECT (see discussion of Issue #2).

Table 6: Assessment of Strengths and Limitations of Comparability of ECT Populations

Strengths	**Limitations**
Similar eligibility and tumor assessment criteria per protocol	Unknown factors in decision to enroll vs. not enroll on Study 3(b)
Patients matched on 11 pre-specified clinical characteristics	Unmeasured variables may result in confounding
Patients did not receive additional treatment after up-front therapy, except for DFMO	Non-contemporaneous index dates
Use of comparable index dates (end of immunotherapy)	Imaging pre-specified for 2 years post-immunotherapy and limited after 5 years
Study 3(b) sites were also ANBL0032 sites	14% of patients in control arm treated on ANBL0032 outside U.S.

FDA considers the populations of patients in the external control arm and the investigational arm to be adequately comparable to permit an inferential analysis while acknowledging the need to evaluate the impact of unmeasured confounders using statistical analyses.

EFFICACY ISSUE 2: Evaluation of the magnitude of the treatment effect in the context of potential threats to study validity.

FDA conducted three groups of analyses to evaluate the treatment effect in this ECT by addressing known sources of bias as well as potential unknown or unmeasured sources of bias.

- Group 1: sensitivity analyses that address the study design and data limitations

- Group 2: sensitivity analyses to provide an understanding of how treatment effect may vary in the presence of potential unmeasured confounders
- Group 3: sensitivity analyses using alternative statistical approaches to estimate the treatment effect to ensure that the observed results are not a result of the Applicant's specific approach

These groups of analyses are intended to address limitations previously described (Table 6), as outlined in Table 7.

Table 7: Sensitivity Analyses to Address Limitations in Comparability of ECT Populations

Limitations	**Sensitivity Analyses to Evaluate Limitations**
Unknown factors in decision to enroll vs. not enroll on Study 3(b)	Group 2 analyses
Unmeasured variables may result in confounding	
Non-contemporaneous index dates	Group 1 analyses
Imaging pre-specified for 2 years post-immunotherapy and limited after 5 years	
Patients treated on ANBL0032 outside U.S. (14% of control arm)	

FDA considers the results of these analyses collectively to evaluate the magnitude of the treatment effect in the ECT.

Group 1: Sensitivity analyses to address study design and data limitations

Table 8 displays a high-level summary of the sensitivity analyses performed by FDA.

A discussion of the concerns leading to the various sensitivity analyses follows the table.

Table 8: **Summary of FDA Group 1 Sensitivity Analyses**

Potential Source of Bias	**Adjustment in Analysis to Assess Impact**	**Results**		
		n	**EFS HR (95% CI)**	**OS HR (95% CI)**
Applicant's Proposed Primary Analysis		**360**	**0.48 (0.27, 0.85)**	**0.32 (0.15, 0.70)**
Differences in follow-up	Limit analysis to first 5 years of follow-up, which is similar across arms	360	**0.51** (0.29, 0.91)	**0.34** (0.14, 0.79)

Time period of treatment	Only include control arm patients with index dates in the same period as the DFMO arm	359	**0.63** (0.36, 1.11)	**0.45** (0.21, 0.98)
Investigator assessment of EFS	Use BICR assessment of EFS for DFMO arm (not available for control arm)	352	**0.49** (0.27, 0.89)	**0.30** (0.13, 0.71)
Immortal time from index to DFMO start	Exclude controls with earlier events (those prior to maximum observed "immortal time" of 123 days)	360	**0.54** (0.31, 0.96)	**0.43** (0.19, 0.96)
All of the above	FDA conservative approach to sensitivity analysis (with additive exclusions/ adjustments)*	152**	**0.59** (0.28, 1.27)	**0.16** (0.05, 0.57)

*Contemporary population per index date, including patients from U.S. sites only, uses equivocal events for BICR for patients with later unequivocal events, excludes 2 patients with BICR ineligible baseline scan; excludes all patients with treatment administration or index date related discrepancies; excludes control EFS dates prior to 75 days (75% of time between index and DFMO administration for calculation of immortal time bias)
**1:1 matching due to reduced sample size.
Source: FDA analysis based on datasets submitted in NDA 215500

Differences in follow-up

In Study 3(b) and Study ANBL0032, tumor assessments were required at the end of immunotherapy (ANBL0032)/baseline (Study 3(b)), at 3, 6, 9, 12, 18, and 24 months after completion of immunotherapy, and then per institutional standard. Study ANBL0032 also required imaging at 30 and 36 months after completion of immunotherapy. Subsequent imaging was performed per institutional standard. The imaging datasets for Study 3(b) and/or ANBL0032 include imaging through 2 years for >99% of patients in both arms and through 3 years in >95% of patients in both arms. At 5 years, 83% of patients in the investigational arm and 85% in the control arm had imaging recorded in the study database. At 7 years, 33% of patients in the investigational arm and 58% in the control arm had imaging recorded in the study database (see Table 9).

Table 9: FDA Analysis of Imaging Data Collected in ANBL0032 and/or Study 3(b) Datasets During Follow-Up Period

Number of years after end of immunotherapy	DFMO arm, Patients with EFS determination "alive with no event" (N=76) N (%)	Control arm, Patients with EFS determination "alive with no event" (N=191) N (%)
2 years	76 (100)	190 (99)
3 years	76 (100)	181 (95)
4 years	67 (88)	173 (91)
5 years	63 (83)	163 (85)
7 years	25 (33)	111 (58)

Source: FDA analysis of datasets submitted in NDA 215500 (ANBL0032 ADOPTF, NMTRC003 outcomes, NMTRC003 ADTTE)

To minimize the impact of differential follow-up times and differential imaging assessment frequencies or imaging availabilities in long-term follow-up of the studies, sensitivity analyses were performed restricting follow-up times at 2-, 3-, and 5-years, also known as administrative censoring. Efficacy results restricting follow-up times at 5-year are presented in Table 8. Analyses restricting follow-up to the first 2 or 3 years of follow-up resulted in similar hazard ratios for EFS as the analysis restricting to 5-year follow-up, with a HR of 0.60 (95% CI: 0.32, 1.11) with 2 years of follow-up and HR of 0.56 (95% CI: 0.31, 1.02) with 3 years of follow-up. OS results were also similar, with a HR of 0.16 (95% CI: 0.02, 1.16) with 2 years of follow-up and HR of 0.34 (95% CI: 0.12, 0.95) with 3 years of follow-up. Some numerical differences may be observed in the analyses with more restricted follow-up time due to the limited number of events available for estimation of treatment effect. Overall, the efficacy results using only the first few years of follow-up where imaging assessments are similar across arms appear to be consistent with the Applicant's proposed primary analysis results.

Index date

Because patients on Study ANBL0032 had no comparable date coinciding with the DFMO administration date on Study 3(b), the end of immunotherapy date was selected as the index date for both study arms. The end of immunotherapy date in Study 3(b) was reported as the last date of pharmacotherapy (e.g., last dose of cis-RA). The end of immunotherapy date reported in Study ANBL0032 is defined by the end of the last reporting period (cycle 6 in most patients), which allows for two weeks for scheduling of the imaging assessments required for disease evaluation.

Study 3(b) was designed to minimize variability by defining eligible patients as having completed antibody therapy > 30 days from the start of DFMO and completed all anti-cancer therapy < 120 days from the start of DFMO. In the primary analysis, for patients enrolled on Study ANBL0032 who went on to enroll in Study 3(b) (N = 85 of 90), the end of immunotherapy

date was defined as the later of the two dates recorded in Study 3(b) (last dose of cis-RA) or Study ANBL0032 (end of treatment visit).

To assess the impacts of variations in treatment administration times around the index date, FDA conducted a number of sensitivity analyses including changes in index date, analyses excluding patients who received the experimental treatment before the index date or received cis-RA after the index date, and analyses that added more exclusion rules in addition to the ones described here. The sensitivity analyses results are presented in Table 10.

Table 10: Sensitivity Analysis of EFS and OS to Assess the Treatment and Index Date Related Variations – Propensity Score Matched (3:1)

	Event-free Survival			Overall Survival		
	DFMO Events/N	NO DFMO Events/N	HR (95% CI)	DFMO Events/N	NO DFMO Events/N	HR (95% CI)
Primary Analysis	14/90	79/270	0.48 (0.27, 0.85)	7/90	57/270	0.32 (0.15, 0.70)
Index date is last administration of antibody treatment	14/90	79/270	0.48 (0.27, 0.85)	7/90	57/270	0.32 (0.15, 0.70)
Excluding DFMO patients receiving treatment prior to index	13/86	74/258	0.48 (0.27, 0.87)	7/86	50/258	0.38 (0.17, 0.85)
Excluding DFMO patients receiving cis-RA after index	13/85	69/255	0.52 (0.29, 0.94)	6/85	43/255	0.38 (0.16, 0.89)
Exclusions above + excluding patients who: were on Stratum 2, received < 6 cycles of immunotherapy, received DFMO on EC*	10/78	42/156	0.43 (0.21, 0.85)	4/78	28/156	0.26 (0.09, 0.73)

HR, hazard ratio; CI, confidence interval; EC, external control; * 2:1 match
Source: FDA Analysis based on Applicant submitted data: 3bvanbl0032 ADTTE

In general, the estimation of treatment effects on EFS and OS, when evaluating the impact of the choice of index date or variations in order of treatments received, seems to be consistent with the effect observed in the Applicant's proposed primary analyses of these endpoints.

Time period of treatment

Patients in the external control arm enrolled on Study ANBL0032 from 2004 to 2015, whereas patients in the investigational arm enrolled on Study 3(b) from 2012 to 2016. Due to potential differences in supportive care and outcomes by treatment era, analyses were performed including only patients on the control arm who were enrolled on Study ANBL0032 in 2011 or later, and patients completing an approximate 6-month course of immunotherapy (+ up to 120 days per Study 3(b) eligibility criteria) in approximately 2012 or later. FDA performed an analysis of contemporaneous populations across treatment arms by including only patients in Study ANBL0032 with index dates after March 22, 2012 (end of immunotherapy for first patient enrolled on Study 3(b)). Analyses of demographic and clinical characteristics of external control patients enrolled before and after March 22, 2012 do not indicate any substantial differences in these two populations. The estimation of treatment effects on EFS and OS endpoints in the two contemporaneous populations as defined by the Applicant and by the FDA per index date (Table 8), appeared to be consistent with the Applicant's proposed primary analyses results.

Investigator assessment of EFS and Blinded Independent Central Review (BICR) of Imaging

In order to account for potential bias in the local evaluator (LE) reported outcomes, a BICR was conducted for patients in the investigational arm. Source imaging was not available to conduct a similar BICR for the control arm.

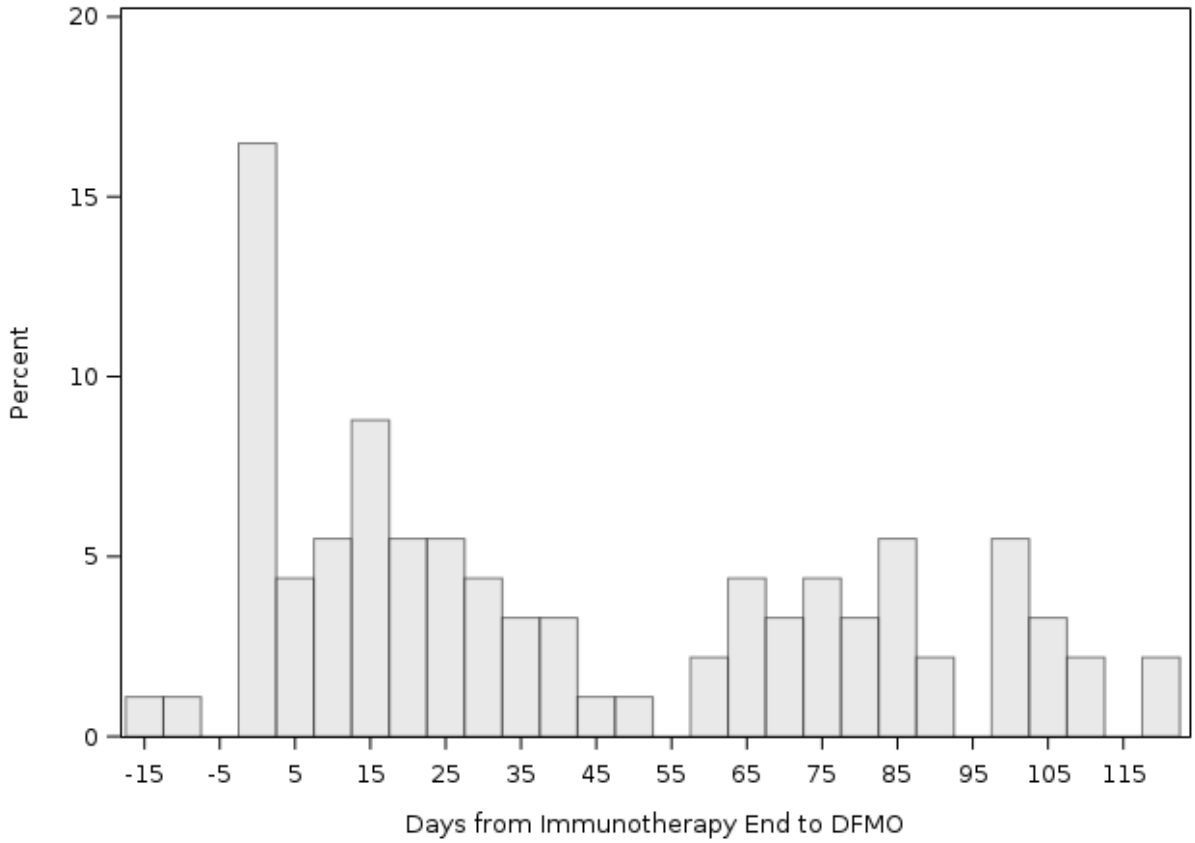
BICR was performed for all tumor imaging assessments on the investigational arm. There was agreement between the independent reviewers (IRs) and LE with the exception of 2 cases. In one case, the patient was determined to have a relapse event per LE, so no further imaging was done and the BICR determined the event to be equivocal based on the lack of confirmatory imaging. The second patient had an unequivocal event per BICR and no event per LE. The overall agreement between IRs across all timepoints is shown graphically in Figure 8 (Appendix).

In Study 3(b), the LE made only two types of determination when evaluating imaging assessments: "unequivocal event" or "no event". However, the BICR evaluators could make a determination of "equivocal EFS event" when, in their judgment, there was not sufficient clarity to definitively make a determination of disease recurrence or secondary cancer. Twenty-five patients had an equivocal imaging result at any time point. Of these, there were 7 instances in which the last event was equivocal per two readers and 4 instances when an equivocal event preceded an unequivocal event for the same lesion. The other equivocal events were isolated and only considered equivocal by one reader (the other reader determined no event). Overall, the results of the PFS analysis based on BICR-assessed PFS events appears similar to the Applicant's proposed primary analysis (Table 8).

Immortal Time Bias

When index date is not determined by randomization, there may be concerns regarding immortal time and its associated bias due the fact that there is an interval during which the outcome cannot occur. In the case of this ECT, for patients enrolled in Study 3(b), the period from end of immunotherapy until the start of DFMO is considered immortal time. Figure 4 shows the histogram of time from the end of immunotherapy to the start of DFMO treatment for all patients in the investigational arm.

Figure 4: Histogram of time from index date (end of immunotherapy) to DFMO initiation for patients on the investigational arm



Source: FDA Analysis of datasets submitted in NDA 215500 (3bvanbl0032 ADTTE)

If patients had an event during this window, they would not have qualified for enrollment on Study 3(b). To account for this concern when developing the protocol and SAP for the ECT, eligibility criteria were applied such that patients on the control arm would not be included if they had an EFS event within the median observed immortal time for the DFMO arm, which was calculated to be 31 days. The intent of this exclusion criterion was to limit the impact of immortal time bias; however, since the immortal time period during which a patient on the DFMO arm would be required to be event-free and alive was longer for half of the patients, an additional sensitivity analysis was conducted to explore this potential bias. The sensitivity

analysis presented in Table 8 provides a conservative approach to immortal time which excluded 16 patients from Study ANBL0032 who had an EFS time less than the maximum possible immortal time period (123 days from index date).

Analyses to assess impact of potential bias due to geographic region or site

Study ANBL0032 was open at 172 U.S. sites and 25 international sites, whereas all but one patient on the DFMO arm received immunotherapy in the US. In the matched EC arm, 86% of patients were treated on ANBL0032 in the US and 14% were treated in either Canada, Australia or New Zealand. Sensitivity analyses of EFS and OS (Table 10) were performed using the same approach as the Applicant's proposed primary analysis, however, matching was done after excluding non-US patients from the control arm and an exact matching of patients was done for census region in addition to *MYCN* status. The efficacy results appear to be consistent with the Applicant's proposed primary analysis result.

Table 11: Sensitivity Analysis of EFS and OS to Assess the Impact of Geographic Region – Propensity Score Matched (3:1)

	Event-free Survival			**Overall Survival**		
	DFMO Events/N	NO DFMO Events/N	HR (95% CI)	DFMO Events/N	NO DFMO Events/N	HR (95% CI)
Primary Analysis	14/90	79/270	0.48 (0.27, 0.85)	7/90	57/270	0.32 (0.15, 0.70)
Restricting control patients only from USA*	12/88	75/264	0.43 (0.23, 0.79)	5/88	48/264	0.29 (0.11, 0.72)

HR, hazard ratio; CI, confidence interval

*Patients were exactly matched on Census region and MYCN status

Source: FDA Analysis based on Applicant submitted data: 3bvanbl0032 ADTTE & ADSL

Summary of Group 1 Analyses

As shown in Table 7, FDA conducted a sensitivity analysis that concurrently adjusted for the previously described sources of bias (differences in follow-up, time period of treatment, investigator assessment of EFS, and immortal time bias). The estimated treatment effect with each individual sensitivity analysis, and the additive sensitivity analysis, remained generally consistent with the effect observed in the Applicant's proposed primary analysis. Additionally, analyses to minimize the bias due to geographic region resulted in similar treatment effects on EFS and OS as observed in the Applicant's proposed primary analysis.

Group 2: Analyses to provide an understanding of how the treatment effect may vary in the presence of potential unmeasured confounders

In the absence of randomization, it is especially important to evaluate the potential impact of unmeasured confounding on effect estimates due the threat of potentially false or spurious associations. The potential impact of missing clinical data and evolution of supportive care over time as it relates to these variables was a particular concern. As patients on Study 3(b) elected to go onto an additional clinical trial whereas patients in the control arm did not, it is possible that the groups differed with regard to social determinants of health, performance status, or other related unmeasured factors.

Both the comparability of cytogenetics results between arms and whether unknown genomic alterations in neuroblastoma tumors contribute to prognosis are unknown variables. These data were limited in both studies. While all of the chromosomal alterations/genetic mutations recorded in patients in the DFMO arm (i.e., 1p deletion, 11q deletion, q71 gain, ALK mutation) are associated with worse prognosis, these data were not available for patients on the control arm given that the study was initiated prior to routine clinical testing for these alterations.

To assess the impact of unmeasured confounding on the estimation of treatment effect, FDA conducted two types of sensitivity analyses. The first method considers the E-value (VanderWeele and Ding, 2017), which provides a straightforward estimate of the association an unmeasured confounder would have with both treatment and outcome for the observed results to be fully attributable to that confounder, much like a tipping point analysis. The second considers a method proposed by Lin et al (1998) which provides the expected effect size for the treatment on outcome given assumptions for the differential prevalence of the unmeasured confounder would have in each treatment arm and the expected size of the association of that unmeasured confounder with the outcome (which may or may not be differential by treatment arm).

FDA's analysis indicated that using various assumptions for the underlying true treatment effect, the E-value for EFS ranged from 2.7 to 3.6 and the E-value for OS ranged from 3.8 to 5.7. In other words, it would be expected that for an unmeasured confounder to tip the results of the EFS analysis from being positive to being neutral (HR=1), that variable would have to have a risk ratio of at least 2.7 to 3.6 for both treatment assignment and for EFS outcome. The E-value makes minimal assumptions, particularly regarding the underlying structure of the distribution of the unmeasured confounder with respect to the treatment or outcome. This, along with the ease of interpretation of the results, are advantages of the E-value method. However, this approach may also be criticized for being overly simplified and not practically applicable to real-world data scenarios.

The second method of sensitivity analyses, those proposed by Lin (1998), requires specification of prevalence of an unmeasured confounder in each treatment arm, as well as the relative risk of the outcome associated with that unmeasured confounder. The analytical solution to the proposed method assumes that the relative risk of outcome for the unmeasured confounder is the same for each treatment arm, that the distributions for unmeasured and measured confounders are approximately independent within each treatment arm, that the outcome

event is rare or that the association of the unmeasured confounder with time-to-event outcomes is small, and lastly, that the unmeasured confounder is binary.

FDA analyses using this methodology were conducted in two ways. First, FDA considered a variety of scenarios of unmeasured confounders with varying prevalence and strength of associations with the outcome. Second, FDA identified potential unmeasured confounders with evidence of prevalence and strength of association with outcome that emerged from a thorough literature review.

In the first set of analyses, FDA assumed an array for prevalence of the unmeasured confounder, with values in the DFMO arm ranging from 0% to 50% by 10% and values in the control arm ranging from 0 to 100% by 10%. Additionally, FDA assumed 3 potential strengths of association for the unmeasured confounder with the outcome variable: a moderate association (HR = 1.4), a strong association (HR = 2.0), and a very strong association (HR = 5.0), with the last scenario providing an unlikely extreme example of unmeasured confounding. For each of these strengths of association, the model provided the expected EFS hazard ratio for the effect of DFMO across the array of prevalence of the unmeasured confounder in each arm as presented in Tables 19 - 20 (see Appendix).

The results of these analyses indicate that with moderate or strong association between the unmeasured confounder and outcome (HR = 1.4 or 2), the EFS hazard ratios remained below 1.0 regardless of the disparity in the prevalence of the unmeasured confounder across treatment arms. For example, assuming a moderate association between the unmeasured confounder and EFS (HR = 1.4), even if the prevalence of the unmeasured confounder in the external control arm is 100% and prevalence in the DFMO arm is 0%, the hazard ratio for EFS comparing DFMO to the external control arm would be expected to be 0.69. Even with the most extreme assumption of very strong association between the unmeasured confounder (HR = 5.0), the hazard ratio for EFS comparing DFMO to the external control arm remains below 0.80 when the prevalence of the unmeasured confounder in the external control arm is less than double the prevalence of the DFMO arm.

Having such extreme differences in the prevalence of the unmeasured confounders would be unlikely, particularly for populations that are considered to be derived from the same underlying patient population as all patients were once enrolled on ANBL0032. The results indicate that the potential impact of an unidentified unmeasured confounding variable on the estimation of the treatment effect of this externally controlled study is likely to be low, in the case of reasonable assumptions of unmeasured confounder association with outcome and prevalence across arms. However, this method provides estimates for each confounder individually, and the composite effect of various multi-layered confounders on the estimate is unknown.

In the second set of analyses using the Lin et al (1998) method, FDA considered the impact of potential confounding factors that were not measured or available in the externally controlled trial which have a known impact on outcome. These analyses attempt to identify how a difference in prevalence of the potential confounding factor could change the estimated treatment effect. A key issue raised by external clinical experts consulted by FDA during review of the application was the absence of information on social determinants of health, which may

impact a family's ability to enroll on a clinical study following frontline therapy. Therefore, patients experiencing household poverty may be underrepresented in Study 3(b) compared to patients in ANBL0032. Further, household poverty been demonstrated in the literature to be associated with inferior EFS (HR = 1.9) in patients with HRNB (Bona, 2021).

FDA's approach to this set of analyses assumed a prevalence rate of the specific confounder published in the literature in the investigational arm and at least double the published prevalence in the control arm to evaluate a scenario for high unmeasured confounding. For the example of household poverty, if it is assumed that 35% of patients in the investigational arm and 70% of patients in the control arm experience household poverty, the resulting EFS HR would be 0.59 (95% CI: 0.53, 0.67).

As shown in Table 12, FDA considered the potential impact of other unmeasured potential confounding factors with a known impact on outcomes (EFS or OS) based on published literature. In certain cases, the control arm prevalence was capped at 100% if double the expected prevalence was greater than 100%, or the control arm prevalence was tripled due to low overall expected prevalence. Overall, the hazard ratios remain consistent with the primary analysis even when considering very large differences in prevalence of the confounding factors. Of note, these are based on individual published studies which provide a reasonable basis for estimates but are subject to individual study limitations.

Table 12: FDA Analysis of Impact of Potential Unmeasured Confounding Factors on Treatment Effect Estimates

Potential Unmeasured Confounder		Supporting Literature	Association with Outcome	Prevalence in DFMO arm[1]	Prevalence in Control Arm[1]	Adjusted DFMO HR (95% CI)
Applicant's Proposed Primary Analysis of EFS						0.48 (0.27, 0.85)
Social Determinants of Health	Household poverty	Bona (2021)	EFS HR = 1.9	35%	70%	0.59 (0.53, 0.67)
	Neighborhood & household poverty	Bona (2021)	EFS HR = 2.2	14%	42%*	0.62 (0.54, 0.71)
Primary Tumor Location	Adrenal vs. Non-adrenal	Kieuhoa (2014)	EFS HR = 1.1	47%	94%	0.50 (0.48, 0.52)
	Non-thoracic vs. Thoracic	Kieuhoa (2014)	EFS HR = 1.3	85%	100%**	0.50 (0.49, 0.50)
Applicant's Proposed Primary Analysis of OS						0.32 (0.15, 0.70)
Cytogenetics	Chromosome 1p deletion	Brown (1999)	OS HR = 1.9	47%	94%	0.42 (0.33, 0.48)
	Chromosome 17q gain	Brown (1999)	OS HR = 3.4	54%	100%**	0.47 (0.39, 0.52)

*Triple prevalence considered due to low expected prevalence; **If double prevalence exceeds 100%, the prevalence is capped to 100%.
[1]Prevalence in DFMO arm is the prevalence cited in the literature, and prevalence in the control arm adjusted to double prevalence, or triple prevalence in the case of neighborhood and household poverty.

Group 3: Alternative statistical approaches

Pre-specification of primary statistical methods for an externally controlled trial requires the choice of both a primary estimand as well as statistical method for creating balance among

observed patient characteristics in the comparative arms for estimation of treatment effect. These attributes of the statistical analysis plans have corresponding advantages and disadvantages with respect to assumptions required and application to the data, and therefore it is important to verify that the results of any non-randomized comparison is robust to the methodology prespecified in the statistical analysis plan.

The primary balancing method of this externally controlled trial was propensity score based matching, which allows for a population that intuitively similar to a randomized trial with 2 balanced arms. However, the matching process may exclude some patients from the final analysis population for comparison. A second approach to balancing the treatment arms by using inverse probability of treatment weighting (IPTW) was explored, allowing for all patients to contribute information to the efficacy analysis rather than just patients in the matched cohort.

These weighted analyses also considered two target populations for the causal estimand. The first considers the treated population (e.g., the population of Study 3(b) as the target population) and provides results for the average treatment effect on the treated (ATT). The second considers the overall population and is represented by the average treatment effect (ATE), which is the same target population and causal estimand as the propensity score matching approach used for the primary analysis.

The efficacy results from the Applicant's proposed primary analysis and analysis using weighting approaches with each estimand in the overall population are provided in Table 13, with Kaplan-Meier plots for the primary analysis and weighting corresponding to the ATT estimand provided in Figure 5 below.

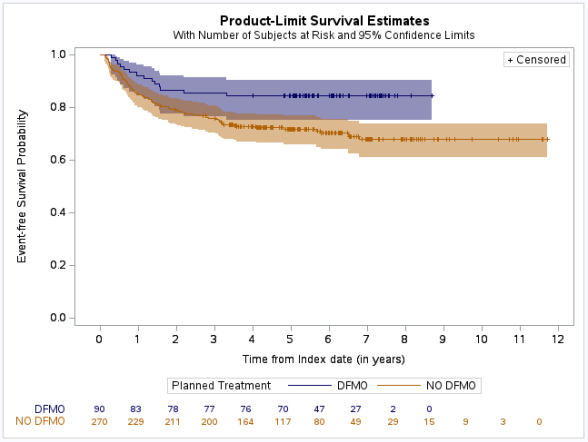
Table 13: Applicant's Proposed Primary Analysis and Alternative Analysis of EFS and OS using ATT Weight in the Overall Complete Case Population Meeting Eligibility Criteria

	Primary: Propensity Score Matching (ATE)[1]	Alternative: Propensity Score Weighting (ATT)[2]	Alternative: Propensity Score Weighting (ATE)[3]
EFS HR (95% CI)	0.48 (0.27, 0.85)	0.50 (0.26, 0.96)	0.39 (0.30, 0.52)
OS HR (95% CI)	0.32 (0.15, 0.70)	0.38 (0.16, 0.92)	0.34 (0.24, 0.49)

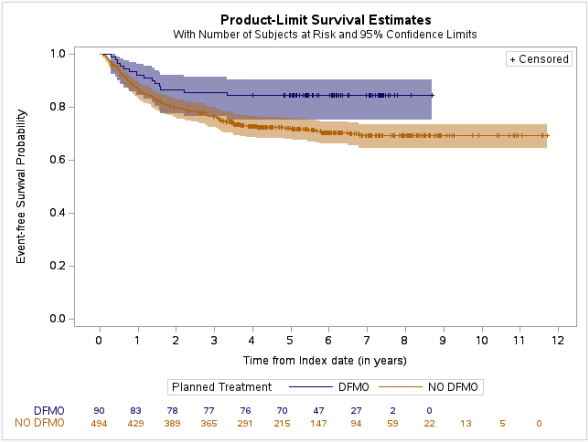
[1] N=360 (90 in DFMO arm, 270 in No DFMO arm); [2]N=180.5 (90 in DFMO arm, and 90.5 in no DFMO arm); [3]N= 1179.9 (595.4 in DFMO arm, and 584.5 in no DFMO arm)

Figure 5: Kaplan-Meier Plots of EFS for Propensity Score Matched and Weighted (ATT) Analysis in the Overall Complete Case Population Meeting Eligibility Criteria

a. Matched (3:1) analysis



b. Weighted (ATT) analysis



Source: FDA Analysis of datasets submitted in NDA 215500

In general, the point estimates of the hazard ratios are similar across statistical balancing methods. It is noteworthy that the confidence intervals are generally wider for the weighted analysis corresponding to the ATT estimand. This is expected as the control population is weighted to reflect the treated population, resulting in a small effective sample size (approximately equal to 1:1 matching). When sample size decreases, the variability in the data as measured by the standard error increases, resulting in a wider confidence interval. Similarly, as sample size increases, the standard error decreases, resulting in a narrower confidence interval. This latter scenario is applicable to the weighted analyses corresponding to the ATE estimand, which results in a larger effective sample size.

FDA also applied weighting approaches in two of the key sensitivity analyses described in the first group of sensitivity analyses (sensitivity analysis Group 1). In particular, weighting approaches were used in the analyses conducted in those patients who have contemporaneous index dates across arms (index dates of March 22, 2012 or later) and in the same population as that which provides the sensitivity analysis with simultaneous adjustment for multiple potential sources of bias. These analyses are provided in Table 14 and Table 15, respectively.

Table 14: Inverse Probability of Treatment Weighting Analysis of EFS and OS in the Contemporaneous Complete Case Population Meeting Eligibility Criteria

	Primary: Propensity Score Matching (ATE)[1]	Alternative: Propensity Score Weighting (ATT)[2]	Alternative: Propensity Score Weighting (ATE)[3]
EFS HR (95% CI)	0.63 (0.36, 1.11)	0.62 (0.32, 1.19)	0.53 (0.38, 0.73)
OS HR (95% CI)	0.45 (0.21, 0.98)	0.44 (0.18, 1.09)	0.45 (0.30, 0.70)

[1] N=359 (91 in DFMO arm, 268 in No DFMO arm); [2]N=179.82 (91 in DFMO arm, and 88.82 in no DFMO arm); [3]N= 790.49 (399.67 in DFMO arm, and 390.82 in no DFMO arm)

Table 15: **Inverse Probability of Treatment Weighting Analysis of EFS and OS in the Contemporaneous Complete Case Population Meeting Eligibility Criteria and with Simultaneous Adjustment for Multiple Potential Sources of Bias**

	Primary: Propensity Score Matching (ATE)[1]	Alternative: Propensity Score Weighting (ATT)[2]	Alternative: Propensity Score Weighting (ATE)[3]
EFS HR (95% CI)	0.59 (0.28, 1.27)	0.58 (0.27, 1.23)	0.45 (0.31, 0.65)
OS HR (95% CI)	0.16 (0.05, 0.57)	0.21 (0.06, 0.78)	0.18 (0.10, 0.34)

[1]N=152 (76 in DFMO arm, 76 in No DFMO arm); [2]N=150 (76 in DFMO arm, and 74 in no DFMO arm); [3]N= 729.53 (375.53 in DFMO arm, and 354.00 in no DFMO arm)

Overall, the observed results of the analyses using weighting approaches, with both the same and alternative estimand (corresponding to the different target population), were supportive of the results observed in the Applicant's proposed primary analyses as well as the several sensitivity analyses conducted by the Applicant and the FDA.

EFFICACY ISSUE 3: Evaluation of the strength of the supportive clinical and nonclinical evidence.

In addition to the single externally controlled trial, the Applicant submitted supportive data as part of the NDA. The supportive data included nonclinical data and clinical data from activity-estimating studies of eflornithine in related populations.

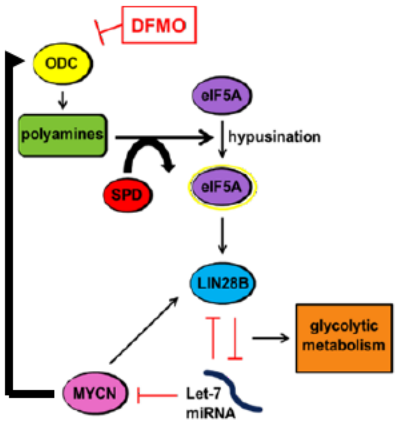
As previously summarized, the FDAMA Act of 1997 amended the statutory provision on substantial evidence of effectiveness such that under certain circumstances, "FDA can conclude that one adequate and well-controlled clinical investigation plus confirmatory evidence is sufficient to establish effectiveness." In order to determine if the application provides substantial evidence of effectiveness of DFMO in the proposed indication, it is necessary for FDA to evaluate whether the additional clinical and nonclinical data are sufficiently robust to serve as confirmatory evidence considering the strengths and limitations of the evidence of effectiveness provided by the ECT.

As discussed in the 2019 FDA draft Guidance for Industry: Demonstrating Substantial Evidence of Effectiveness for Human Drug and Biological Products, a single adequate and well-controlled clinical investigation, together with earlier phase clinical results and/or testing that provide compelling mechanistic evidence in the setting of well-understood disease pathophysiology, may be sufficient to provide substantial evidence of effectiveness of a new drug. According to the Guidance, mechanistic evidence would generally be obtained from clinical testing using a relevant and well understood pharmacodynamic (PD) endpoint; however, it could also be collected from other sources such as animal studies (e.g., those using an established, relevant animal model to study the effect of the drug on a PD marker of known relevance to humans). Therefore, FDA evaluated the potential for nonclinical mechanistic data to contribute to the

confirmatory evidence needed to establish substantial evidence of effectiveness for this application. To facilitate FDA's review, and in recognition of DFMO's limited utility as an anti-cancer agent over several decades of clinical investigation, the FDA nonclinical team also conducted an independent scientific literature-based assessment evaluating the effect of DFMO in neuroblastoma. Notably, the published literature identified in the FDA's assessment was generally consistent with the studies and published literature provided by the Applicant, with both data sources supportive of a cytostatic mechanism of action in neuroblastoma. DFMO-induced inhibition of polyamine synthesis in in vitro assays restored the balance of the LIN28/Let-7 metabolic pathway (Lozier et al. 2014; Figure 6A), which is involved in regulation of cancer stem cells and glycolytic metabolism, by decreasing protein expression of the oncogenic drivers MYCN and LIN28B and increasing expression of the tumor suppressor Let-7 in *MYCN*-amplified neuroblastoma cells. In vitro, DFMO inhibited polyamine synthesis, induced G1 cell cycle arrest (Wallick et al. 2005; Koomoa et al. 2013), induced cellular senescence at clinically relevant concentrations, and suppressed neurosphere formation in both *MYCN*-amplified (Figure 6B) and *MYCN* non-amplified (not shown) neuroblastoma cells, indicating a cytostatic effect in neuroblastoma irrespective of *MYCN* amplification status. In contrast, DFMO is not cytotoxic and did not affect in vitro cell viability or apoptosis.

Figure 6: Effect of DFMO on the LIN28/Let-7 metabolic pathway and in vitro neurosphere formation

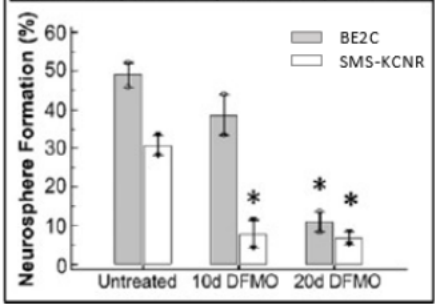
A



B

MYCN amplified neuroblastoma cell lines

	Vehicle	10d DFMO	20d DFMO
BE(2)-C	49.0%	38.5%	10.9%
SMS-KCNR	30.7%	7.8%	6.8%

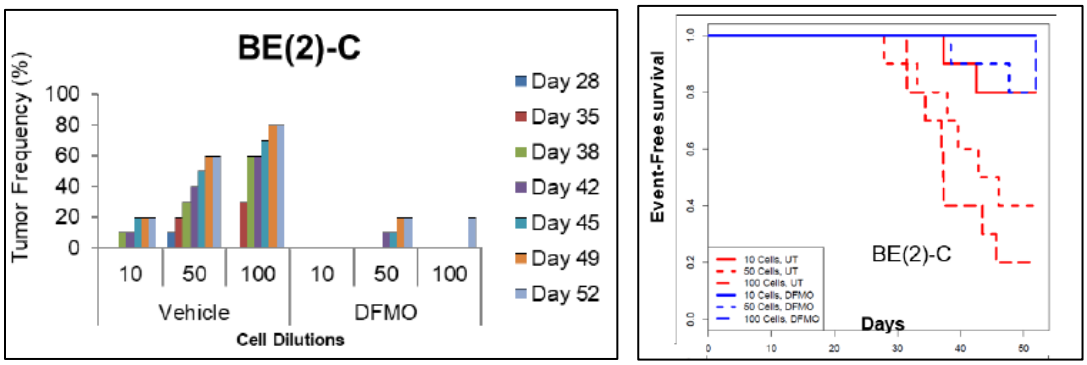


A, Source: Adapted/modified from figure in 1) NDA 215500, Summary of Clinical Pharmacology, page 62; and 2) Lozier et al. Oncotarget. 2014; 6(1):196-206. B, Source: NDA 215500, Applicant Information Amendment, submitted 1/30/2023; pages 31-32

To further evaluate the cytostatic action of DFMO in mouse models, the Applicant used extreme limiting dilution analysis (ELDA), an established method to measure the frequency of tumor-initiating cells within a tumor cell population and tumorigenicity in subpopulations of cells (den Hollander et al. 2022). In an ELDA experiment that FDA considers to relevantly model the proposed clinical indication, limited numbers of *MYCN*-amplified BE(2)-C neuroblastoma

cells were injected in mice and treatment with 2% DFMO was initiated on the day of injection when there were no tumors present yet. DFMO decreased the frequency of tumor-initiating neuroblastoma cells in vivo, thereby preventing or delaying tumor formation and improving EFS compared to untreated control (Figure 7). Similar results were seen with another *MYCN*-amplified neuroblastoma cell line (SMS-KCNR; data not shown). In addition, western blot analysis of excised tumors demonstrated that DFMO reduced LIN28B and MYCN expression in vivo compared to untreated controls, indicating on-target PD activity.

Figure 7: Effect of DFMO on in vivo neuroblastoma formation and EFS in an ELDA mouse model

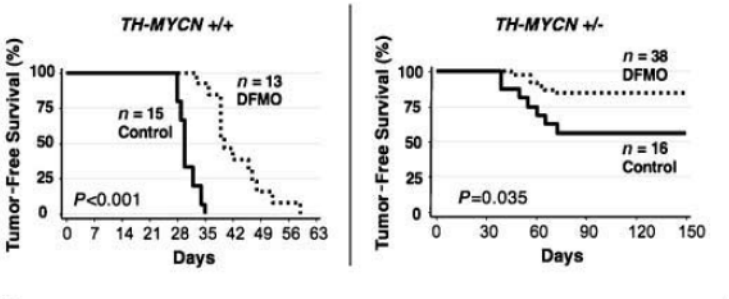


Source: NDA 215500, Applicant Information Amendment, submitted 1/30/2023; pages 31-32

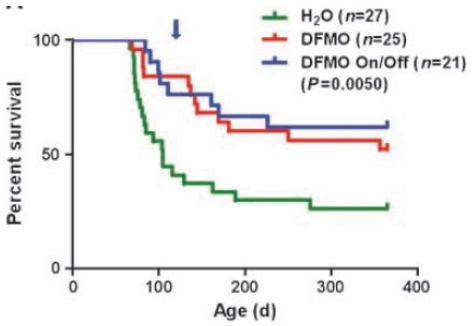
As part of the FDA review team's scientific literature search, two publications from two independent research groups were identified that evaluated the effect of DFMO on tumor prevention in a well-characterized, published transgenic mouse model (Weiss, 1997; Chesler, 2007; Moore, 2008). In these two publications, investigators used *TH-MYCN* transgenic mice, which overexpress human *MYCN* in neural crest cells and represent a well-established animal model of spontaneous neuroblastoma that shares biochemical features, orthologous genomic alterations, and histologic features with *MYCN*-amplified human neuroblastoma. Hogarty et al. (2008) demonstrated that giving mice 1% DFMO in the drinking water from birth onward increased tumor-free survival in homozygous (*TH-MYCN +/+*) mice and prevented tumor formation in ~84% of treated hemizygous (*TH-MYCN +/-*) mice (Figure 8A). DFMO-treated tumors harvested from *TH-MYCN +/+* homozygous mice exhibited decreased polyamine levels, demonstrating on-target PD activity. As shown in Figure 8B, a separate research group (Rounbehler et al. 2009) demonstrated that giving mice 1% DFMO in the drinking water beginning at 3 weeks of age delayed the onset/incidence of neuroblastoma formation in *TH-MYCN* mice and improved survival. In conclusion, these data demonstrate that DFMO can prevent or delay the formation of neuroblastoma and increase survival in a well-established transgenic mouse model of neuroblastoma.

Figure 8: Effect of DFMO on tumor-free survival in *TH-MYCN* transgenic neuroblastoma mouse model

A



B



A, Source: Hogarty et al. Cancer Res. 2008; 68(23): 9735-45. B, Source: Rounbehler et al. Cancer Res. 2009; Jan 15; 69(2): 547-53.

Overall, the DFMO pharmacology data submitted by the Applicant demonstrate a cytostatic, and not a cytotoxic, mechanism of action in neuroblastoma, in which DFMO targets tumor-initiating cells, and are further strengthened by supportive published data in the literature. Therefore, the available nonclinical data may provide a rationale for the lack of objective response rates in patients given DFMO as a monotherapy. DFMO is not expected to inhibit the growth of established tumors and traditional xenograft mouse models are not appropriate to test its efficacy. Relevant to the potential use of nonclinical data as confirmatory evidence for establishing substantial evidence of effectiveness for this application, in vitro mechanistic data demonstrates that DFMO targets drivers of neuroblastoma pathophysiology (e.g., ODC, MYCN, LIN28). In addition, data from two independently conducted, well-established, relevant animal models of neuroblastoma (ELDA and *TH-MYCN* transgenic mice) demonstrate that DFMO prevents or delays tumor formation in mice with no initial evidence of disease. Importantly, these mouse models evaluated clinically relevant endpoints such as EFS and provided PD evidence of on-target DFMO activity. Overall, the FDA nonclinical review team concluded that the available nonclinical data generally support the proposed mechanism of action of DFMO to prevent or delay HRNB relapse in patients who are in remission. Even though the activity of most investigational drugs is supported by nonclinical data, this nonclinical activity does not always translate to clinical efficacy. Thus, nonclinical data has not generally been used as confirmatory evidence to support approval of cancer drugs.

Clinical Studies

Study NMTRC002 (NCT 01059071) was a multi-center, single-arm, dose-escalation study of DFMO monotherapy administered for one cycle followed by DFMO plus oral etoposide, enrolled between 2010 and 2014 (Saulnier Sholler et al, 2015). A total of 21 patients were treated with DFMO, with 4 patients at 500 mg/m^2, five patients at 750 mg/m^2, three patients at 1000 mg/m^2, and nine patients at 1500 mg/m^2. There were no confirmed dose-limiting

toxicities (DLTs). The study enrolled 18 evaluable pediatric patients. Three patients had partial responses, although only one was actively progressing at the time of enrollment. Three patients had either bone marrow positivity or PET avid disease at study entry which improved after 1 cycle of DFMO alone (with an overall best disease response on study of stable disease or progressive disease); however, the timing of prior therapy with respect to initiation of DFMO is not known. Each of these three patients subsequently progressed. There are three long-term survivors from this study. The contribution of any anti-tumor effect of DFMO is challenging to interpret given it was primarily administered as combination therapy and patients had received multiple prior therapies.

Study NMTRC006 (NCT03581240) is an ongoing expanded access study for patients with relapsed rare tumors with increased LIN28 expression, MYCN amplification, or upregulation of ornithine decarboxylase, including high-risk neuroblastoma. As of January 2023, a total of 97 patients were enrolled, including 27 patients with HRNB in remission, defined as either in remission following upfront therapy (n=13) or in subsequent remission following refractory or relapse disease (n=14). As an expanded access study, there were no defined response criteria. Of the 13 patients with HRNB in initial remission, 12 (92%) remained in remission at the data cut-off, with eight completing two years of DFMO therapy and four continuing on DFMO therapy (range 180 to 629 days on therapy); one patient relapsed after 453 days on DFMO therapy. Of 15 patients with HRNB in remission after therapy for relapsed/refractory disease, 10 (67%) remained in remission at the data cut-off, with five patients completing 2 years of DFMO therapy; one patient stopped DFMO after 86 days; three patients continue on DFMO therapy (range 285 to 685 days on therapy); and five patients relapsed during DFMO therapy (start of DFMO therapy to relapse: 49 to 170 days). Expanded access data regarding tumor responses or duration of remission in HRNB and related tumor types are challenging to interpret given the lack of pre-specified response criteria and imaging assessments and of a control arm.

Study NMTRC003(b) also included Stratum 2. This stratum enrolled 35 patients with HRNB in remission after any previous relapse or refractory therapy. Patients had a median of one prior anti-cancer therapies (range 1-3) with a median of approximately one month between completion of last anti-cancer treatment and start of DFMO (range 4 to 124 days). Eligible patients could have relapsed at any point during their initial treatment course and specific drugs previously administered were not recorded. Most patients (63%) had experienced one relapse, with fewer (11% and 3%, respectively) having experienced two or three relapses. Patients in Study 3(b) Stratum 2 initiated DFMO between June 2012 and February 2016. In the Stratum 2 cohort, EFS at 2 years was 46% (95% CI: 29, 61) for patients treated with DFMO compared to a pre-specified historical control rate of 10%. The historical control rate was based on a published analysis of 91 patients with newly-diagnosed HRNBneuroblastoma enrolled on studies at a single institution between 1991 and 2002 (Santana, 2008). The calculated historical

control rate used a weighted average of EFS rates at 2 years considering median times from first relapse to second relapse, and second relapse to third relapse reported in the publication.

Given the lack of contemporaneity of Study 3b and data analyzed in the reference publication, FDA requested further justification for the control EFS rate of 10%. The Applicant provided reported outcomes from two studies conducted in a similar timeframe to Study 3b to evaluate maintenance therapies for patients in second or later remission, including NCT00072358 and NCT00911560, both of which found 5-year progression-free survival (PFS) rates of 32-33% (Kushner, 2015; Cheung, 2021). Both of these studies were conducted at a highly specialized single center and the outcomes may reflect both a potential treatment effect of the investigational treatments and an improvement from highly specialized care that is not widely available. Therefore, the appropriateness of these studies to serve as a historical benchmark for Stratum 2 of Study 3(b) is not certain.

FDA also considered that the combination of dinutuximab, irinotecan, and temozolomide has become a commonly used relapse regimen for patients with HRNB since the initial publication of the results of ANBL1221 (Mody, 2017; Mody, 2020). This study enrolled patients with HRNB at the time of first relapse or first designation of refractory disease between 2013 and 2015, with results published in 2017 and 2020. Patients who received dinutuximab, irinotecan and temozlomide had a 1-year PFS rate of 68% (95% CI, 55, 81) and the Kaplan-Meier curves (Figure 2) in the publication demonstrate a PFS rate of approximately 40% at 2 years. Given the timing of enrollment, patients on Study 3(b) initiated DFMO prior to the publication of the results of ANBL1221; however, it is not clear whether patients could have received dinutuximab, irinotecan, and temozolomide (on Study ANBL1221 or according to the study protocol) prior to enrollment on Study 3(b) since prior therapies are not known.

Due to limitations of data capture in ANBL0032 after initial relapse, a historical control rate for patients in a second or later remission after relapse from the ANBL0032 dataset cannot be reliably estimated . Based on the outcomes reported in published literature in more recent studies, it is possible that the proposed historical control rate of EFS at 2 years of 10% is lower than would be currently observed in the US population, but the anticipated EFS rate in a contemporary population is unclear. Therefore, the EFS results of Study 3(b) Stratum 2 are difficult to interpret.

Additional Trials Evaluating DFMO

There are several ongoing trials with DFMO in patients with neuroblastoma, for which efficacy data is not provided in this application.

Study 14 is an ongoing multi-center, open label trial of DFMO monotherapy in patients with HRNB in remission after up-front (Strata 1/1b/2) or relapse/refractory (Strata 3/4) therapy. The primary endpoint is EFS rate at 4 years compared to a historical control rate. This study supports the safety evaluation for this application. At the time of NDA submission, efficacy data

were not available from this application. Since the design of Study 14 is similar to Study 3b, efficacy results which replicate findings from Study 3b could provide support for the proposed indication. However, in the absence of a comparator arm, interpretation of the findings of Study 14 would be limited by its single arm nature. Further, use of data from ANBL0032 as a comparator for Study 14 may be less appropriate given the non-overlapping time periods in which the studies were conducted, as well as other potential differences.

Two additional randomized studies for which data have not been provided in the current application are ongoing in the newly diagnosed and relapsed/refractory setting:

- Study NMTRC012 is an open-label, randomized trial which evaluates immunotherapy alone vs. immunotherapy with DFMO, with a primary endpoint of EFS. Following this randomized period during immunotherapy, all patients receive DFMO for 2 years in the post-immunotherapy setting; therefore, the study is not designed to evaluate the efficacy of DFMO in the maintenance setting. The trial is expected to complete enrollment in 2-5 years.
- COG Study ANBL1821 is an open-label, randomized (1:1) trial of irinotecan, temozolomide and dintuxutimab with or without DFMO in patients with relapsed/refractory high risk neuroblastoma. The study is anticipated to be complete in 2024.

A potential limitation of the confirmatory evidence available for DFMO is the lack of robust efficacy and anti-tumor activity data in other adult oncology disease settings despite decades of study. DFMO is being investigated for the treatment of adult patients with high grade glioma, in which tumorigenesis is regulated at least in part by LIN28 signaling. Breakthrough therapy designation was granted to DFMO in 2014 based on a non-statistically significant OS results from a randomized study of patients with anaplastic glioma who received procarbazine, CCNU, and vincristine with or without DFMO after completion of radiation therapy for newly diagnosed disease (Levin, 2003). Based on these results, a randomized trial of lomustine with or without DFMO is ongoing in patients with anaplastic astrocytoma with recurrence after prior irradiation and treatment with temozolomide. The study is closed to enrollment with results anticipated in 2023.

3.2 Safety Issues

Known safety issues with DFMO include the potential for new or worsening hearing loss during treatment. FDA is not seeking discussion of safety issues during the advisory committee meeting; a brief summary is provided below.

3.2.1 Sources of Data for Safety

Safety data in this application is derived from Study 3(b) and Study 14 (NCT02679144). Patients in Strata 1, 2, 3 and 4 of Study 14 received DFMO 750 ± 250 mg/m^2 BID for up to 2 years; patients in Stratum 1b of Study 14 received DFMO 2500 ± 250 mg/m^2 BID. Enrollment began in

February 2016 with 280 of 441 planned patients enrolled at 37 sites in the US and Canada as of June 30, 2021.

The pooled safety population (N=360) reflects patients with newly diagnosed or relapsed/refractory HRNB who were exposed to DFMO as a single agent taken orally twice daily at a dosage based on BSA for a maximum of 2 years from Study 3b (N=101) and Study 14 (N=259).

The primary safety population (N=85) includes a subset of patients who received DFMO at the proposed dose in Study 3(b) Stratum 1 (in which the population was reflective of the proposed indication) for whom safety data were available.

Safety data collection in Studies 3(b) and 14 was limited as these studies were investigator-initiated and not initially intended to support a marketing application. Study 3(b) collected Grade 2 or higher treatment-emergent adverse events (AEs) and laboratory data were not systematically collected. Study 14 collected only Grade 3 or higher AEs. The BCC001 database did not collect safety information, so there are no safety data for the 18 patients in the primary efficacy population who did not transfer onto Study 3(b) at the time of the change in drug supplier. The data cutoff for both studies was June 30, 2021. For Study 3(b), all patients either completed or discontinued DFMO therapy prior to the cutoff date.

3.2.2 Safety Summary

Among the 360 patients in the pooled safety population, the median age was 4 years (range: 1 to 19); 56% were male; 78% were White, 7% were Black, 5% were Asian; 9% were Hispanic or Latino; 85% had International Neuroblastoma Staging System Stage 4 disease; and 39% had neuroblastoma with known MYCN-amplification.

In the pooled safety population (N=360), the most common (≥5%) adverse reactions were hearing loss (11%), otitis media (10%), pyrexia (7%), pneumonia (5%) and diarrhea (5%). The most common (≥2%) Grade 3 or 4 laboratory abnormalities (based on adverse event reporting) were increased ALT (11%), increased AST (6%), decreased neutrophils (4.2%), and decreased hemoglobin (3.3%). There were no Grade 5 treatment-emergent adverse events.

In the primary safety population (N=85), the most common (≥5%) adverse reactions, including laboratory abnormalities, were otitis media, diarrhea, cough, sinusitis, pneumonia, upper respiratory tract infection, conjunctivitis, vomiting, pyrexia, allergic rhinitis, decreased neutrophils, increased ALT, increased AST, hearing loss, skin infection, and urinary tract infection (Table 13 and Table 14).

In the primary safety population (N=85), serious adverse reactions occurred in 12% of patients who received DFMO. Serious adverse reactions in > 1 patient included skin infection (3 patients). Permanent discontinuation of DFMO due to an adverse reaction occurred in 11% of patients. Adverse reactions which resulted in permanent discontinuation of DFMO in > 1

patient included hearing loss. Dose reductions of DFMO due to an adverse reaction occurred in 8% of patients. Adverse reactions which required dose reductions in > 1 patient included hearing loss.

Table 16: Adverse Reactions (≥5%) in Patients with HRNB Who Received DFMO in Study 3(b)

Adverse Reaction[a]	DFMO (n=85)	
	All Grades[b,c] (%)	Grade 3 or 4 (%)
Infections and Infestations		
Otitis media	32	2.4*
Sinusitis	13	0
Pneumonia	12	1.2*
Upper respiratory tract infection	11	0
Conjunctivitis	11	0
Skin Infection	7	4.7*
Urinary tract infection	6	1.2*
Gastrointestinal Disorders		
Diarrhea[1]	15	3.5*
Vomiting	11	1.2*
Respiratory Disorders		
Cough	15	0
Allergic rhinitis	11	0
General Disorders		
Pyrexia	11	1.2*
Ear and Labyrinth Disorders		
Hearing loss	7	7*

- a Adverse Reactions were graded using CTCAE Version 4.03.
- b Grade 1 adverse events were not comprehensively collected in Study 3b.
- c No Grade 5 adverse reactions were reported in clinical studies.
- * Events of Grade 3 only (no Grade 4 occurred).
- [1]Diarrhea include diarrhea and colitis.
- Source: FDA Analysis of ADAE datasets submitted in NDA 215500

Table 17: Select Laboratory Abnormalities (≥1%) in Patients with HRNB Who Received DFMO in Study 3(b)

Laboratory Abnormality[1]	DFMO[2] (n=85)	
	All Grades (%)	Grade 3 or 4 (%)

Chemistry		
Increased ALT	9	7*
Increased AST	8	6*
Increased alkaline phosphatase	4.7	2.4*
Decreased potassium	2.4	2.4*
Decreased glucose	2.4	1.1
Decreased sodium	2.4	0
Increased potassium	1.2	0
Increased glucose	1.2	0
Hematology		
Decreased neutrophils	9	8
Decreased hemoglobin	4.7	2.4*
Decreased white blood cells	2.4	0
Decreased platelets	1.2	0

[1] The table presents laboratory parameters reported as adverse events according to CTCAE Version 4.03.

[2] Grade 1 adverse events were not comprehensively collected in Study 3b.

* Events of Grade 3 only (no Grade 4 occurred).

Source: FDA Analysis of ADAE datasets submitted in NDA 215500

Proposed warnings for the product label include myelosuppression, hepatotoxicity, and hearing loss. In the pooled safety population, hepatotoxicity was limited to isolated liver enzyme (AST/ALT) elevations in < 10% of patients. There were no events of drug-induced liver injury or liver failure. Increased ALT/AST leading to dose interruption or reduction occurred in 2.5% of patients and DFMO was discontinued due to increased ALT/AST in 0.6% of patients.

Similarly, a relatively small (< 10%) proportion of patients in the pooled safety population experienced an event of myelosuppression, most commonly neutropenia; however, one patient experienced an event described as bone marrow failure which resolved.

Hearing loss is discussed in more detail below.

Hearing Loss

Hearing loss is an adverse event of special interest in the DFMO clinical program and has been identified in studies of DFMO in other populations including in patients with trypanosomiasis who received, Ornidyl, an IV formulation of DFMO. Most patients (81%) who received DFMO in Studies 3(b) and 14 had an abnormal audiogram at baseline likely related to platinum chemotherapy received during their initial treatment. In the pooled safety population, 13% had

new or worsening hearing loss; 12% of patients had worsening from baseline to Grade 3 (hearing loss sufficient to indicate therapeutic intervention) or 4 (audiologic indication for cochlear implant and additional speech-language services indicated). Hearing loss resulted in dose interruption or reduction in 7%, and discontinuation in 1.4% of patients. A total of 47 patients (13%) experienced worsening hearing loss from baseline; the event resolved in four patients (1.1%). FDA considers that receipt of previous platinum-based chemotherapy likely contributed to worsening or new hearing loss during DFMO therapy. However, given the single arm nature of the study, the contribution of prior therapy to evolving hearing loss during treatment with DFMO is not quantifiable. Given that hearing loss was observed in other patient populations who received DFMO but who did not receive similar prior therapy, it is also likely that new or worsening hearing loss is associated with treatment with DFMO.

3.3 Risk Mitigation

If substantial evidence of effectiveness is established and there is a clear potential for clinical benefit for patients with high-risk neuroblastoma who have completed multiagent, multimodality therapy, the safety issues addressed above can be characterized and managed by appropriate product labeling.

4 Summary

The Applicant submitted the results of a single externally controlled trial and supportive data to provide evidence for the proposed indication for DFMO to reduce the risk of relapse in pediatric patients with high-risk neuroblastoma (HRNB) who have completed multiagent, multimodality therapy. The externally controlled trial was designed to compare data from Study 3b, a multi-center, open-label, single arm trial of DFMO monotherapy for up to 2 years in pediatric patients with HRNB in remission after immunotherapy, to an external control constructed from patients with HRNB enrolled on Study ANBL0032, a multi-center, open-label, randomized trial of immunotherapy plus standard up-front therapy vs. up-front therapy alone.

In addition to the results of the single externally controlled trial, the Applicant provided supportive data derived from nonclinical data and clinical data from exploratory studies of DFMO in related populations.

The development program to support DFMO to reduce the risk of relapse in pediatric patients with high-risk neuroblastoma (HRNB) who have completed multiagent, multimodality therapy has been challenging. Randomized clinical trials are the gold standard for evaluation of time-to-event endpoints in oncology clinical trials. The importance of a well-matched contemporaneous comparison is magnified when the anticipated mechanism of action is cytostatic and objective response rate, which would increase confidence in an observed treatment effect, is not expected. The best design feature to achieve this comparability is randomization. Nonetheless, pediatric patients with high-risk neuroblastoma represent a population with high unmet medical need and FDA has been receptive to evaluating the current data, particularly given the unique nature of the external control. FDA considers the externally controlled trial to be adequate and well-controlled, and robust to sensitivity analyses. As has been previously stated, in order to establish substantial evidence of effectiveness, a single adequate and well controlled trial must be accompanied by sufficient confirmatory evidence, and there remains uncertainty regarding the evidentiary package submitted necessitating feedback from the oncology drug advisory committee.

FDA requests discussion of the evidence of effectiveness of DFMO for the proposed indication, including the strengths and limitations of a single externally controlled clinical trial that FDA considers to be adequate and well-controlled and of the adequacy of supportive clinical and nonclinical data as potential confirmatory evidence to support a determination of substantial evidence of effectiveness.

5 References

Ambros PF, Ambros IM, Brodeur GM, Haber M, Khan J, Nakagawara A, Schleiermacher G, Speleman F, Spitz R, London WB, Cohn SL, Pearson AD, Maris JM. International consensus for neuroblastoma molecular diagnostics: report from the International Neuroblastoma Risk Group (INRG) Biology Committee. Br J Cancer. 2009 May 5;100(9):1471-82. doi: 10.1038/sj.bjc.6605014. PMID: 19401703.

ASCO Cancer.Net Editorial Board, "Neuroblastoma – Childhoood: Statistics. Updated March 2023. Available at: https://www.cancer.net/cancer-types/neuroblastoma-childhood/statistics

Balzeau J, Menezes MR, Cao S, Hagan JP. The LIN28/let-7 Pathway in Cancer. Front Genet. 2017; 8:31. doi: 10.3389/fgene.2017.00031.

Bassiri H, Benavides A, Haber M, Gilmour SK, Norris MD, Hogarty MD. Translational development of difluoromethylornithine (DFMO) for the treatment of neuroblastoma. Transl Pediatr. 2015 Jul;4(3):226-38. doi: 10.3978/j.issn.2224-4336.2015.04.06. PMID: 26835380; PMCID: PMC4729051.

Casero RA Jr, Murray Stewart T, Pegg AE. Polyamine metabolism and cancer: treatments, challenges and opportunities. Nat Rev Cancer. 2018; 18(11):681-695.

Chesler L, Goldenberg DD, Seales IT, Satchi-Fainaro R, Grimmer M, Collins R, Struett C, Nguyen KN, Kim G, Tihan T, Bao Y, Brekken RA, Bergers G, Folkman J, Weiss WA. Malignant progression and blockade of angiogenesis in a murine transgenic model of neuroblastoma. Cancer Res. 2007; 67(19): 9435-42.

Cheung IY, Cheung NV, Modak S, et al. Survival Impact of Anti-GD2 Antibody Response in a Phase II Ganglioside Vaccine Trial Among Patients With High-Risk Neuroblastoma With Prior Disease Progression. *J Clin Oncol*. Jan 20 2021;39(3):215-226. doi:10.1200/jco.20.01892.

den Hollander P, Joseph R, Vasaikar S, Kuburich NA, Deshmukh AP, Mani SA. Limiting Dilution Tumor Initiation Assay: An In Vivo Approach for the Study of Cancer Stem Cells. Methods Mol Biol. 2022; 2429:547-554.

Diskin SJ, Capasso M, Schnepp RW, Cole KA, Attiyeh EF, Hou C, Diamond M, Carpenter EL, Winter C, Lee H, Jagannathan J, Latorre V, Iolascon A, Hakonarson H, Devoto M, Maris JM. Common variation at 6q16 within HACE1 and LIN28B influences susceptibility to neuroblastoma. Nat Genet. 2012; 44(10): 1126-30.

DuBois SG, Macy ME, Henderson TO. High-Risk and Relapsed Neuroblastoma: Toward More Cures and Better Outcomes. Am Soc Clin Oncol Educ Book. 2022 Apr;42:1-13. doi: 10.1200/EDBK_349783. PMID: 35522915.

Geerts D, Koster J, Albert D, Koomoa DL, Feith DJ, Pegg AE, Volckmann R, Caron H, Versteeg R, Bachmann AS. The polyamine metabolism genes ornithine decarboxylase and antizyme 2 predict aggressive behavior in neuroblastomas with and without MYCN amplification. Int J Cancer. 2010 May 1;126(9):2012-24. doi: 10.1002/ijc.25074. PMID: 19960435; PMCID: PMC2847573.

Hogarty MD, Norris MD, Davis K, Liu X, Evageliou NF, Hayes CS, Pawel B, Guo R, Zhao H, Sekyere E, Keating J, Thomas W, Cheng NC, Murray J, Smith J, Sutton R, Venn N, London WB, Buxton A, Gilmour SK,

Marshall GM, Haber M. ODC1 is a critical determinant of MYCN oncogenesis and a therapeutic target in neuroblastoma. Cancer Res. 2008; 68(23): 9735-45.

International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use, "Choice of Control Group and Related Issues in Clinical Trials: E10." Dated 20 July 2000. Available at: https://database.ich.org/sites/default/files/E10_Guideline.pdf

Irwin MS, Naranjo A, Zhang FF, Cohn SL, London WB, Gastier-Foster JM, Ramirez NC, Pfau R, Reshmi S, Wagner E, Nuchtern J, Asgharzadeh S, Shimada H, Maris JM, Bagatell R, Park JR, Hogarty MD. Revised Neuroblastoma Risk Classification System: A Report From the Children's Oncology Group. J Clin Oncol. 2021 Oct 10;39(29):3229-3241. doi: 10.1200/JCO.21.00278. Epub 2021 Jul 28. PMID: 34319759.

Koomoa DL, Geerts D, Lange I, Koster J, Pegg AE, Feith DJ, Bachmann AS. DFMO/eflornithine inhibits migration and invasion downstream of MYCN and involves p27Kip1 activity in neuroblastoma. Int J Oncol. 2013; 42(4):1219-28.

Kushner BH, Ostrovnaya I, Cheung IY, et al. Prolonged progression-free survival after consolidating second or later remissions of neuroblastoma with Anti-G(D2) immunotherapy and isotretinoin: a prospective Phase II study. Oncoimmunology. Jul 2015;4(7):e1016704. doi:10.1080/2162402x.2015.1016704.

Levin VA, Hess KR, Choucair A, et al. Phase III randomized study of postradiotherapy chemotherapy with combination alpha-difluoromethylornithine-PCV versus PCV for anaplastic gliomas. Clin Cancer Res. Mar 2003;9(3):981-90.

Lin DY, Psaty BM, and Kronmal RA. Assessing the sensitivity of regression results to unmeasured confounders in observational studies. Biometrics. 1998:948-963.

Lozier AM, Rich ME, Grawe AP, et al. Targeting ornithine decarboxylase reverses the LIN28/Let-7 axis and inhibits glycolytic metabolism in neuroblastoma. Oncotarget. 2014;6(1):196-206.

Moore HC, Wood KM, Jackson MS, Lastowska MA, Hall D, Imrie H, Redfern CP, Lovat PE, Ponthan F, O'Toole K, Lunec J, Tweddle DA. Histological profile of tumours from MYCN transgenic mice. J Clin Pathol. 2008; 61(10):1098-103.

Moreno L, Barone G, DuBois SG, Molenaar J, Fischer M, Schulte J, Eggert A, Schleiermacher G, Speleman F, Chesler L, Geoerger B, Hogarty MD, Irwin MS, Bird N, Blanchard GB, Buckland S, Caron H, Davis S, De Wilde B, Deubzer HE, Dolman E, Eilers M, George RE, George S, Jaroslav Š, Maris JM, Marshall L, Merchant M, Mortimer P, Owens C, Philpott A, Poon E, Shay JW, Tonelli R, Valteau-Couanet D, Vassal G, Park JR, Pearson ADJ. Accelerating drug development for neuroblastoma: Summary of the Second Neuroblastoma Drug Development Strategy forum from Innovative Therapies for Children with Cancer and International Society of Paediatric Oncology Europe Neuroblastoma. Eur J Cancer. 2020 Sep;136:52-68. doi: 10.1016/j.ejca.2020.05.010. Epub 2020 Jul 9. PMID: 32653773.

Paz EA, LaFleur B, Gerner EW. Polyamines are oncometabolites that regulate the LIN28/let-7 pathway in colorectal cancer cells. Mol Carcinog. 2014; 53 Suppl 1:E96-106.

Rounbehler RJ, Li W, Hall MA, Yang C, Fallahi M, Cleveland JL. Targeting ornithine decarboxylase impairs development of MYCN-amplified neuroblastoma. Cancer Res. 2009; Jan 15; 69(2): 547-53.

Santana VM, Furman WL, McGregor LM, Billups CA. Disease control intervals in high-risk neuroblastoma. Cancer. 2008 Jun 15;112(12):2796-801. doi: 10.1002/cncr.23507. PMID: 18429000.

U.S. Food and Drug Administration. Guidance Document: Considerations for the Design and Conduct of Externally Controlled Trials for Drug and Biological Products. February 2023. Available at: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/considerations-design-and-conduct-externally-controlled-trials-drug-and-biological-products.

U.S. Food and Drug Administration. Guidance Document: Demonstrating Substantial Evidence of Effectiveness for Human Drug and Biological Products: Draft Guidance for Industry. December 2019. Available at: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/demonstrating-substantial-evidence-effectiveness-human-drug-and-biological-products

VanderWeele TJ, Ding P. Sensitivity analysis in observational research: introducing the E-value. Ann Intern Med. 2017;167(4):268-274.

Wallick CJ, Gamper I, Thorne M, et al. Key role for p27Kip1, retinoblastoma protein Rb, and MYCN in polyamine inhibitor-induced G1 cell cycle arrest in MYCN-amplified human neuroblastoma cells. Oncogene. Aug 25 2005;24(36):5606-18.

Weiss WA, Aldape K, Mohapatra G, Feuerstein BG, Bishop JM. Targeted expression of MYCN causes neuroblastoma in transgenic mice. EMBO J. 1997; 16(11):2985-95.

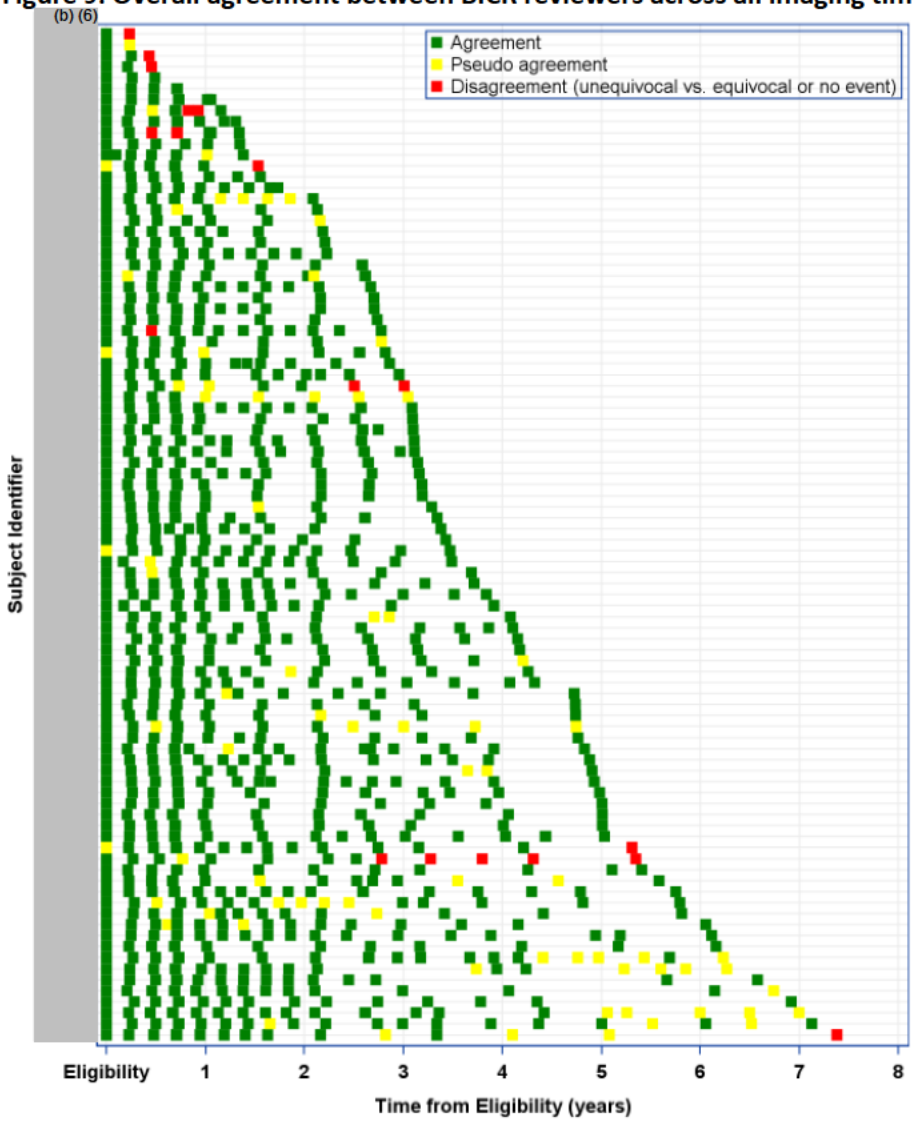
Yu AL, Gilman AL, Ozkaynak MF, et al. Anti-GD2 antibody with GM-CSF, interleukin-2, and isotretinoin for neuroblastoma. The New England journal of medicine. Sep 30 2010;363(14):1324-34. doi:10.1056/NEJMoa0911123

Yu AL, Gilman AL, Ozkaynak MF, et al. Long-Term Follow-up of a Phase III Study of ch14.18 (Dinutuximab) + Cytokine Immunotherapy in Children with High-Risk Neuroblastoma: COG Study ANBL0032. Clin Cancer Res. 2021;27(8):2179-2189. doi:10.1158/1078-0432.CCR-20-3909

6 Appendix

Figure 9 demonstrates agreement or lack of agreement between BIRC reviewers for the investigational arm.

Figure 9: Overall agreement between BICR reviewers across all imaging timepoints



Source: Applicant analysis submitted in NDA 215500, Module 5.3.5.4 Comparison of Study 3b to ANBL0032 External Control Database, page 177.

Table 18: Post-Relapse Therapies

	DFMO arm (N=90)	**Control arm** (N=270)
EFS events, N (%)	**14 (16)**	**79 (29)**
Relapse	14	76
Death	0	2
Secondary malignancy	0	1
Known Relapse Count, N		
Single	9	44
Multiple	5	22
Missing	-	13
Number of post-relapse therapies, median (range)	3 (1, 5)	Unknown
Chemotherapy	13	-
Antibody therapy	8	-
Radiation	7	-
Other (e.g., vaccine trial)	7	-
Number of patients alive at data cut-off[1], N (%)	6 (43)	22 (28)

[1]DFMO data cut-off: 6/30/2021; Control data cut-off: 6/30/2019

Source: Adapted from Applicant analysis submitted in NDA 215500, Module 5.3.5.4, 3bvANBL0032 Efficacy Report Appendix 9

Table 19: Adjusted Event-free Survival hazard ratios comparing DFMO vs. NO DFMO adjusting for an unmeasured binary confounder having a hazard ratio of 1.4 and the observed hazard ratio in the current trial of 0.48

	P_1					
P_0	0.0	0.1	0.2	0.3	0.4	0.5
0.0	**0.48**	0.46	0.44	0.43	0.41	0.40
0.1	0.50	**0.48**	0.46	0.44	0.43	0.41
0.2	0.52	0.50	**0.48**	0.46	0.44	0.43
0.3	0.54	0.52	0.50	**0.48**	0.46	0.45
0.4	0.56	0.54	0.52	0.50	**0.48**	0.46
0.5	0.58	0.56	0.54	0.52	0.50	**0.48**
0.6	0.60	0.58	0.56	0.53	0.52	0.50
0.7	0.62	0.60	0.57	0.55	0.53	0.51
0.8	0.64	0.62	0.59	0.57	0.55	0.53
0.9	0.67	0.64	0.61	0.59	0.57	0.55
1.0	0.69	0.66	0.63	0.61	0.59	0.56

Note. P_1 and P_0 are the prevalence of the unmeasured confounder in the DFMO arm and in the control arm, respectively.

Source: FDA analysis based on review of NDA 215500

Table 2020: Adjusted Event-free Survival hazard ratios comparing DFMO vs. NO DFMO adjusting for an unmeasured binary confounder having a hazard ratio of 2.0 and the observed hazard ratio in the current trial of 0.48

	P_1					
P_0	0.0	0.1	0.2	0.3	0.4	0.5
0.0	**0.48**	0.44	0.40	0.37	0.34	0.32
0.1	0.53	**0.48**	0.44	0.41	0.38	0.35
0.2	0.58	0.52	**0.48**	0.44	0.41	0.38
0.3	0.62	0.57	0.52	**0.48**	0.45	0.42
0.4	0.67	0.61	0.56	0.52	**0.48**	0.45
0.5	0.72	0.65	0.60	0.55	0.51	**0.48**

0.6	0.77	0.70	0.64	0.59	0.55	0.51
0.7	0.82	0.74	0.68	0.63	0.58	0.54
0.8	0.86	0.79	0.72	0.66	0.62	0.58
0.9	0.91	0.83	0.76	0.70	0.65	0.61
1.0	0.96	0.87	0.80	0.74	0.69	0.64

Note. P_1 and P_0 are the prevalence of the unmeasured confounder in the DFMO arm and in the control arm, respectively.

Source: FDA analysis based on review of NDA 215500