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ORIGINAL ARTICLE

Onset and resolution of ovarian toxicity with nirogacestat treatment in females with desmoid tumors: Updated safety analyses from the DeFi phase 3 study

Elizabeth T. Loggers MD, PhD[1] | Rashmi Chugh MD[2] | Noah Federman MD[3] | Lee Hartner MD[4] | Richard F. Riedel MD[5] | Sunny Cho PharmD[6] | David Hyslop MD[6] | Allison Lim PharmD[6] | Ana B. Oton MD[6] | Kutluk H. Oktay MD, PhD[7]

[1]Clinical Research Division, Fred Hutchinson Cancer Center/Division of Hematology and Oncology, University of Washington, Seattle, Washington, USA

[2]University of Michigan, Rogel Comprehensive Cancer Center, Ann Arbor, Michigan, USA

[3]Departments of Pediatrics and Orthopaedic Surgery, David Geffen School of Medicine, University of California, Los Angeles, California, USA

[4]University of Pennsylvania, Abramson Cancer Center, Pennsylvania Hospital, Philadelphia, Pennsylvania, USA

[5]Duke Cancer Institute, Duke University Medical Center, Durham, North Carolina, USA

[6]SpringWorks Therapeutics, Inc, Stamford, Connecticut, USA

[7]Department of Obstetrics, Gynecology, and Reproductive Sciences, Yale University School of Medicine, New Haven, Connecticut, USA

Correspondence
Elizabeth T. Loggers, 1100 Fairview Ave N, Seattle, WA 98109, USA.
Email: eloggers@fredhutch.org

Funding information
Springworks Therapeutics, Inc

Abstract

Introduction: Nirogacestat is a targeted gamma secretase inhibitor approved in the United States for adults with progressing desmoid tumors. In the phase 3 DeFi study (NCT03785964) of nirogacestat, ovarian toxicity (OT) was identified as a safety signal among females of reproductive potential (FORP). This analysis further describes the incidence, presentation, and resolution of OT.

Methods: Patients were randomized to twice-daily oral nirogacestat (150 mg) or placebo, taken in continuous 28-day cycles. Investigator-identified OT in FORP was based on abnormal reproductive hormone values or perimenopausal symptoms (or both). Adverse event follow-up was conducted to assess OT resolution. Post hoc analyses included return of menstruation and return of follicle-stimulating hormone (FSH) to within normal limits (WNL) (≤20.4 mIU/mL).

Results: Of 92 randomized females, 73 in the safety population were FORP ($n = 36$ nirogacestat, $n = 37$ placebo). OT was identified in 75% (27 of 36) receiving nirogacestat and 0% (0 of 37) receiving placebo. As of October 24, 2022, investigators reported OT resolution in 78% (21 of 27) of patients, with median OT duration of 19.1 weeks. Off-treatment resolution was reported in all 11 patients (100%) who stopped nirogacestat treatment; of these, all nine with available menstruation information experienced return of menstruation and eight had FSH WNL at last reported assessment. Resolution was reported in 10 of 14 (71%) while on nirogacestat; of these, all 10 experienced return of menstruation and seven had FSH WNL. Two patients were lost to follow-up.

Conclusion: Most FORP treated with nirogacestat experienced OT, with the majority resolving, including all who stopped treatment, suggesting that OT is transient.

The clinical trial registration is NCT03785964.

KEYWORDS
fertility, gamma secretase, GSI, oncology, ovarian dysfunction, women of childbearing potential

INTRODUCTION

Desmoid tumors (DT) are rare, soft tissue neoplasms (classified as intermediate tumors) characterized by locally aggressive growth without evidence of metastasis.[1,2] The disease course of DT can be unpredictable, and tumors can lead to substantial morbidity, pain, and functional limitation.[1,3] Prevalence is approximately 2- to 3-fold higher in females than males, and patients are often diagnosed in adulthood between the ages of 20 and 44 years,[4–6] when many females are of reproductive age.

Nirogacestat (OGSIVEO™, SpringWorks Therapeutics) is an oral, small-molecule, targeted gamma secretase inhibitor (GSI) that works by blocking proteolytic activation of Notch receptors.[7–9] Mutagenic or clastogenic effects have not been demonstrated with nirogacestat.[9] Recently, nirogacestat became the only US Food and Drug Administration-approved medicine for the treatment of adults with progressing DT who require systemic treatment.[10] The approval of nirogacestat was based primarily on the DeFi study, a global, randomized, double-blind, placebo-controlled, phase 3 study.[11] Treatment with nirogacestat demonstrated a significant benefit in progression-free survival (PFS) over placebo (hazard ratio, 0.29; 95% confidence interval, 0.15–0.55; $p < .001$), as well as significant improvements in objective response rate (ORR) (41% vs. 8%; $p < .001$) and patient-reported outcomes of pain, DT-specific symptom burden, physical and role functioning, and overall quality of life ($p \leq .01$, all).[11] The most common (≥15%) adverse reactions with nirogacestat were diarrhea, rash, nausea, fatigue, stomatitis, headache, abdominal pain, cough, alopecia, upper respiratory tract infection, and dyspnea.[9]

Ovarian toxicity (OT), also referred to as ovarian dysfunction, was a common adverse reaction reported with nirogacestat in females of reproductive potential that participated in DeFi.[9,11] As defined in a recent American Society of Clinical Oncology (ASCO) research statement, ovarian toxicity describes the spectrum of ovarian functional impairment potentially resulting from drug exposure.[12] In general, OT may present as menstrual cycle abnormalities (length and flow), hot flashes, and night sweats, and may be associated with changes in follicle-stimulating hormone (FSH), estradiol, luteinizing hormone (LH), and/or anti-Müllerian hormone (AMH).[12,13]

The risk of OT varies among drug classes and is influenced by several factors, including previous lines of therapy (number and type), dosage, patient age, and baseline fertility status.[13] Reversibility of OT depends in part on a drug's mechanism of action (MOA) and its effects (permanent or temporary) on the female reproductive system.[13] Specifically, an individual's finite primordial follicle pool—that is established in utero—decreases over a female's reproductive lifespan with age; menopause occurs when the primordial follicle reserve is nearly depleted.[14]

Some treatments, such as antimetabolite drugs, may only deplete growing follicles without damaging primordial follicles.[15] In this case, growing follicles that produce steroid hormones and support menstrual function will eventually be replaced from the intact primordial follicle pool.[16,17] Because the replacement of these follicles may take more than 3 months,[18] females may experience temporary amenorrhea and alteration of reproductive hormones without permanently losing their reproductive potential. Existing evidence suggests that GSIs impact follicular activation, maturation, and growth; angiogenesis; and subsequent steroid hormone secretion by the developing corpus luteum,[16,17,19] but do not lead to depletion of primordial follicles. However, treatments that partially or completely deplete the primordial pool, such as alkylating agents, may permanently affect reproductive potential by resulting in early menopause and infertility due to the inability to develop ovulatory follicles.[20]

Ovarian toxicity is rarely systematically assessed in drug development, resulting in its under-reporting in clinical trials.[12,21,22] Although initial data on OT incidence and resolution from DeFi were previously reported in the *New England Journal of Medicine*,[11] here we provide updated OT analyses from DeFi to further describe its incidence, presentation, and resolution.

MATERIALS AND METHODS

DeFi overview

DeFi (NCT03785964) was a phase 3 study evaluating the efficacy, safety, and tolerability of nirogacestat; study details have been previously published.[11] The study included patients at least 18 years of age with a histologically confirmed diagnosis of progressing desmoid tumors. In the double-blind treatment phase, patients were assigned (1:1) to receive oral nirogacestat (150 mg) or placebo taken twice daily in continuous 28-day cycles. The study met all primary and key secondary end points. The safety population includes all patients who received at least one dose of nirogacestat or placebo.

The DeFi study was conducted in accordance with ethical principles derived from the Declaration of Helsinki and all applicable laws, regulations, and scientific guidelines. All patients provided written informed consent before enrollment.

Reporting, description, and analysis of OT

Ovarian toxicity was evaluated as a safety signal and classified as an adverse event of special interest for safety reporting. Females of reproductive potential were defined as being between menarche and confirmed menopause (i.e., 12 months since last menstruation) with

intact ovaries, based on investigator's judgment; no prespecified age range was used to define this population.

Serum hormone levels were measured for all patients (female and male). Female hormone levels assessed included FSH, AMH, estradiol, LH, and progesterone. Blood samples were collected at: screening and baseline for all new study entrants; treatment cycle 1, cycle 2, cycle 4, and cycle 7, and every three cycles thereafter; the end of treatment; and follow-up. Prolactin and thyroid-stimulating hormone (TSH) were also collected at screening and end of treatment. For patients identified as having OT, data collection continued every 3 months until event resolution was reported or for at least 90 days after discontinuing study treatment.

Ovarian toxicity was identified in females of reproductive potential by investigators based on abnormal reproductive hormone values or perimenopausal symptoms (e.g., changes in menstrual regularity) or both. The verbatim terms for these events were coded to the Medical Dictionary for Regulatory Activities (MedDRA, version 24.0) to the preferred terms of ovarian failure, premature menopause, amenorrhea, and menopause. Adverse events were reported up to the data cutoff date (April 7, 2022), with additional follow-up through October 24, 2022 conducted to assess the resolution of OT as reported by investigators.

To further characterize the investigator-reported cases of OT resolution, the recently published ASCO recommendations to assess menstruation and reproductive hormone levels (such as FSH)[12] were applied to post hoc analyses of patient-level laboratory values and adverse events in the clinical database. Additional information, such as return of menstruation, was reported via pharmacovigilance follow-up.

TABLE 1 Ovarian toxicity events reported.

	Nirogacestat, 150 mg BID
Total safety population, *n*	69
Total females, *n*	44
Total females of reproductive potential, *n*	36
Total females of reproductive potential with OT events, *n* (%)[a]	27 (75)
Time to first onset of OT events, median, weeks	8.9
Duration of OT events, median, weeks[b]	19.1
Patients with OT events with dose modifications, *n* (%)[c]	
Interrupted	2 (7)
Reduced	2 (7)
Withdrawn	4 (15)
Concomitant medications to manage OT symptoms	
Use of concomitant medications, *n* (%)[c]	4 (15)[d]
Duration of use, median, weeks	44

Abbreviations: BID, twice daily; OT, ovarian toxicity.

[a]Denominator is out of the total number of females of reproductive potential.

[b]Median duration across OT events (i.e., resolution on and off nirogacestat treatment).

[c]Denominator is out of the total number of females of reproductive potential with OT.

[d]Comprises three patients who received progesterone and estrogen fixed combination and one patient who received gabapentin and venlafaxine (not concurrently).

RESULTS

Study population

From May 2019 through August 2020, 142 patients underwent randomization in DeFi. Of 92 females randomized, 74 were of reproductive potential; one female of reproductive potential randomized to receive nirogacestat discontinued the study before treatment. Onset and resolution results presented herein are based on data from 73 females of reproductive potential who received at least one dose of study treatment (safety population: $n = 36$ nirogacestat; $n = 37$ placebo).

Overview of OT events

Among females of reproductive potential, OT was identified by investigators in 75% (27 of 36) receiving nirogacestat and 0% receiving placebo (Table 1). Five patients experienced more than one event of OT. All OT events were grade 2. Events of OT were associated with increased levels of FSH and LH, and decreased levels of AMH, progesterone, and estradiol. In addition, OT events were associated with perimenopausal symptoms, such as menstrual irregularities or amenorrhea. No abnormalities were observed with prolactin or TSH, ruling out other potential causes of OT. The median time to first onset of OT events was 8.9 weeks after nirogacestat initiation, with median duration of OT events of 19.1 weeks (Table 1).

Among the 27 females of reproductive potential receiving nirogacestat with an OT event, OT led to nirogacestat dose reduction in 7% (2 of 27) and drug discontinuation in 15% (4 of 27). Four females with an OT event received concomitant medications to manage symptoms of OT, including hormone therapy or low-dose selective serotonin reuptake inhibitors (Table 1).

Patient characteristics

Characteristics of females of reproductive potential with and without OT are summarized in Table 2. Nine patients had a baseline medical history of amenorrhea and/or irregular menstruation, including one 50-year-old patient with baseline irregular menstruation who had an investigator-reported OT event of menopause after study entry. Nearly three-fourths (74%) of females of reproductive potential with OT events were younger than 34 years of age, consistent with their child-bearing status, whereas a larger percentage without OT (56%) were 34 years of age or older. More (85%; 23 of 27) patients with

TABLE 2 Baseline characteristics in females of reproductive potential with and without ovarian toxicity.

Characteristic, *n* (%)	Females of reproductive potential with OT (*n* = 27)	Females of reproductive potential without OT (*n* = 9)
Age, years		
<34	20 (74)	4 (44)
≥34 to <46	5 (19)	3 (33)
≥46	2 (7)	2 (22)
Body mass index[a]		
<25 kg/m^2	15 (56)	6 (67)
≥25 to <30 kg/m^2	5 (19)	2 (22)
≥30 kg/m^2	6 (22)	1 (11)
Genetic history		
CTNNB1 mutations	14 (52)	8 (89)
APC mutations	9 (33)	1 (11)
Radiation		
Any prior radiation treatment	6 (22)	2 (22)
Systemic therapies		
Any prior systemic therapy	23 (85)	6 (67)
≥2 prior systemic therapies	14 (52)	4 (44)
≥4 prior systemic therapies	6 (22)	1 (11)
Chemotherapy		
Doxorubicin	6 (22)	1 (11)
Methotrexate + vinblastine	8 (30)	1 (11)
Tyrosine kinase inhibitor		
Sorafenib	10 (37)	2 (22)

Abbreviations: *APC*, adenomatous polyposis coli; *CTNNB1*, β-catenin; OT, ovarian toxicity.

[a]One patient in the group with OT had missing data.

reported OT events received prior systemic therapy compared with 67% (6 of 9) of those without OT.

Resolution of OT events

Resolution of OT was reported by investigators based on reproductive hormone values or perimenopausal symptoms, or both. As of October 24, 2022, investigators reported that OT resolved in 78% (21 of 27) of females of reproductive potential with OT events (Figure 1). This resolution rate is higher than the 74% (20 of 27) reported in the primary DeFi publication[11] because one additional patient was reported to have resolved on treatment after the initial follow-up period. Resolution of OT was reported regardless of the coded MedDRA preferred term, with the exception of the single patient with investigator-reported menopause after study entry (Table 3).

Off-treatment resolution was reported in 11 of 11 patients (100%) who stopped nirogacestat for any reason. All 11 of these patients met at least one of the recently recommended ASCO criteria for assessing OT resolution: eight had FSH values within normal limits (≤20.4 mIU/mL) at the last reported assessment, all patients where menstruation information was available (9 of 9) experienced return of menses, and six patients had both FSH within normal limits and return of menses (Table 4). The two patients with unavailable menstruation information had FSH values within normal limits at the last available follow-up. The median time to off-treatment resolution was 76 days (range: 28–126 days), or approximately 11 weeks.

Resolution of OT was reported in 10 of 14 (71%) patients while on nirogacestat treatment, which included four patients with reported event resolution but who later discontinued nirogacestat (for any reason) and six patients with reported event resolution and who remained on nirogacestat through the primary analysis data cutoff date (Table 5). All 10 patients with on-treatment OT resolution had return of menses and seven had FSH values within normal limits at the last reported assessment. Median duration of time from reporting of OT to on-treatment resolution was 171 days (range: 10–770 days), or approximately 24 weeks. The four patients with ongoing OT continue to receive nirogacestat and are being followed in the ongoing open-label extension of DeFi.

DISCUSSION

Nirogacestat, the first GSI to be approved and the only therapy approved for progressing DT,[9] was associated with significant benefit with respect to PFS, ORR, pain, DT-symptom burden, physical/role functioning, and overall quality of life in the phase 3 DeFi study.[11] During DeFi, OT was deemed to be an adverse event of special interest for nirogacestat. Although OT commonly occurs with oncology drugs, the nature of OT and timeline for ovarian function recovery, if any, can vary greatly based on a variety of factors, including drug MOA, dosage, patient characteristics (e.g., age, fertility status, and family history), and prior therapy.[13]

Results from DeFi highlight the importance of evaluating ovarian function in oncology studies through clinical and laboratory assessments, especially when females of reproductive potential are participants. Including OT end points in clinical trials can benefit both patients and clinicians, as data may improve counseling on treatment options through evidence-based discussions.[12,21] Potential gaps in knowledge arise from the fact that, historically, ovarian function has been rarely assessed proactively in clinical trials.[12,21,22] Furthermore, current clinical trial coding schema (e.g., MedDRA, Common Terminology Criteria for Adverse Events) may not sufficiently collect nor reflect the current scientific understanding of, or the terminology used in, reproductive medicine.[12] Additional barriers to OT assessments in clinical trials may include lack of prioritization, limited resources, and lack of knowledge.[21] To address these gaps, the 2023 ASCO research statement provides recommendations for incorporating OT as a safety end point in relevant trials of anticancer agents as well as methods for conducting clinical and hormonal assessments.[12] Most of the ASCO

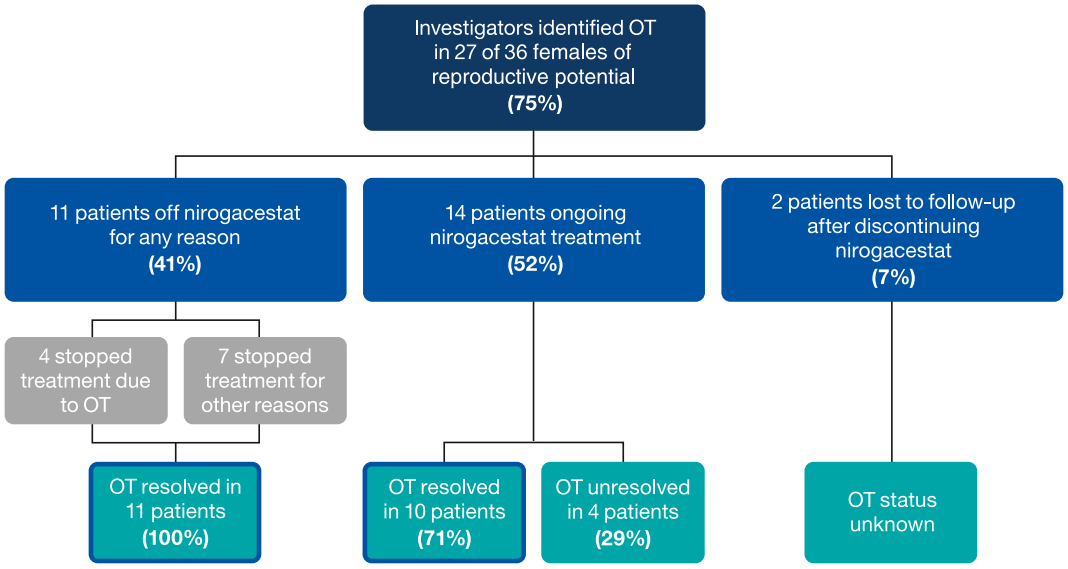

FIGURE 1 Ovarian toxicity frequency and resolution as of October 24, 2022. OT was identified in females of reproductive potential by investigators based on abnormal reproductive hormone values or perimenopausal symptoms (e.g., changes in menstrual regularity) or both. Follow-up through October 24, 2022, was conducted to assess the resolution of OT as reported by investigators. OT indicates ovarian toxicity.

TABLE 3 Investigator-reported ovarian toxicity and resolution by MedDRA preferred terms.

MedDRA preferred term	OT occurrence[b] (n = 27)	OT resolution[c] (n = 21)
Ovarian failure[a]	13	10
Premature menopause	11	9
Amenorrhea[a]	3	3
Menopause	1	0

Abbreviations: MedDRA, Medical Dictionary for Regulatory Activities; OT, ovarian toxicity.

[a]One patient reported two events that were coded to ovarian failure and amenorrhea. Both resolved per investigator.

[b]Identified in females of reproductive potential by investigators from the DeFi double-blind phase.

[c]Resolution of OT was reported by investigators in patients for whom follow-up data was available as of October 24, 2022.

recommendations had been used in the DeFi study, and as such, key learnings from this study emphasize the importance of monitoring patients for ovarian function in clinical trials.

Among females of reproductive potential treated with nirogacestat, 75% (27 of 36) were reported to have OT based on abnormal reproductive hormone levels or perimenopausal symptoms, or both, with resolution reported in 78% (21 of 27). Nirogacestat treatment was also associated with decreases in AMH, estradiol, and progesterone, and increases in FSH and LH.

Of those who stopped nirogacestat for any reason, investigator-reported resolution occurred in 100% (11/11), with a median time to resolution off treatment of approximately 11 weeks. This short time to resolution after discontinuation suggests that the effects of nirogacestat on ovaries are transient. For the 14 patients with OT who remained on treatment, resolution was reported in 10 (71%). Median duration of time from reporting of OT to resolution on treatment was longer in these patients (approximately 24 weeks), which is not unexpected given that patients remained on treatment. Post hoc analyses, applying the ASCO recommendations, further characterized the investigator-reported cases of OT resolution. For the 21 patients with off- or on-treatment OT resolution, all met at least one of the criteria (return of menses or FSH values within normal limits), with the majority achieving both criteria.

DeFi represents the most comprehensive OT data set in DT clinical trials to date, although study limitations must be noted. Concomitant or previous treatments, including hormonal contraception and/or prior systemic therapies, could have confounded results. In DeFi, 85% of females of reproductive potential with OT received prior systemic treatment, with 22% having received at least four prior therapies. The small number of nirogacestat-treated females without reported events of OT limits the assessment of the impact of baseline factors, including prior treatment, on OT incidence. For clinical measures, menstrual diaries were not included, and patients with amenorrhea or menstrual irregularities at baseline were still considered for OT assessments. For hormone measures, blood collections aligned with study visits, not with patients' menstrual cycles, and AMH assessments were not complete for all patients. Given some of these limitations, hormone results reported here focused primarily on FSH, in conjunction with return of menses. Further studies, including the ongoing DeFi open-label extension, will be used to advance understanding of hormonal fluctuations.

Although hormone assessments are important for use in clinical trials to assess potential events of OT, currently there is no standard clinical practice to monitor reproductive hormones in patients before or during systemic treatment.[13] This aligns with the prescribing information for nirogacestat, which recommends monitoring patients

TABLE 4 DeFi patients with events of ovarian toxicity resolving after stopping nirogacestat.

Patient	Age at BL (years)	OT AE		Last investigator-reported outcome	Duration of OT (days)	Days to resolution after treatment stopped	FSH resolved to ≤20.4 mIU/mL[a]			Resumption of menstruation (yes/no)
		Preferred term	Start day				Yes/no	Last reported value (mIU/mL)	Study day (days after stopping niro)	
1	19	Premature menopause	57	Resolved	518	35	Yes	14.6	574 (33)	Information on menstruation not provided; prior history of menstrual irregularities and amenorrhea
2	25	Premature menopause	118	Resolved	65	64	No	108	134 (14)	Yes
3	48	Premature menopause	56	Resolved	73	75	No	31.7	135 (80)	Yes
4	18	Ovarian failure	42	Resolved	211	NA	Yes	3.3	1113 (91)	Yes
		Ovarian failure	421	Resolved	601	92				
5	26	Ovarian failure	77	Resolved	125	126	Yes	4.8	198 (121)	Yes
6	37	Premature menopause	57	Resolved	133	NA	Yes	5.2	652 (64)	Yes
		Premature menopause	398	Resolved	30	NA				
		Premature menopause	551	Resolved	129	93				
7	23	Ovarian failure	257	Resolved	49	36	Yes	6.2	303 (32)	Yes
8	25	Ovarian failure	131	Resolved	102	126	Yes	3.5	233 (125)	Yes
9	29	Premature menopause	1[b]	Resolved	118	76	Yes	7.5	169 (125)	Yes
10	25	Premature menopause	52	Resolved	55	77	No	79.1	85 (54)	Yes
11	26	Premature menopause	102	Resolved[c]	27	28	Yes	7	488[c]	Menstruation info not provided

Abbreviations: AE, adverse event; BL, baseline; FSH, follicle-stimulating hormone; NA, not applicable; niro, nirogacestat; OT, ovarian toxicity.

[a]The reference range used for females of reproductive potential at any phase in the menstrual cycle in DeFi was 1.8–20.4 mIU/mL.

[b]Event was reported during the first month of treatment and the exact onset date for this event was not available; as such, the most conservative estimate of day 1 of treatment was used.

[c]Patient reported event resolution following an extended (~1 month) dose interruption and as such is included in this table as having resolved off study treatment. Patient remained on study treatment as of the data cut, therefore the last reported FSH value in the double-blind phase of the DeFi study provided.

TABLE 5 DeFi patients with events of ovarian toxicity resolving while on nirogacestat treatment.

		OT AE				FSH resolved to ≤20.4 mIU/mL[a]			
Patient	Age at BL (years)	Preferred term	Start day	Last investigator-reported outcome	Duration of OT (days)	Yes/no	Last reported value (mIU/mL)	Study day	Resumption of menstruation (yes/no)
Resolved on treatment and remained on treatment through primary analysis data cutoff (April 7, 2022)									
12[b]	28	Ovarian failure	26	Resolved	436	No	46.6	652	Yes
13	33	Ovarian failure	381	Resolved	590	Yes	6.2	1142	Yes
14	32	Amenorrhea	86	Resolved	649	Yes	12.8	882	Yes
15	39	Ovarian failure	148	Resolved	94	Yes	11.9	757	Yes
16[c]	34	Ovarian failure	57	Resolved	93	No	30	841	Yes
		Ovarian failure	194	Resolved	143				
17	23	Premature menopause	145	Resolved	32	No	52.9	671	Yes
		Premature menopause	493	Resolved	171				
Resolved on treatment but later discontinued treatment before primary analysis data cutoff (April 7, 2022)									
18	22	Amenorrhea	1[d]	Resolved	221	Yes	7.2	365	Yes
19	29	Premature menopause	69	Resolved	651	Yes	9.3	895	Yes
20	20	Ovarian failure	53	Resolved	770	Yes	5.2	823	Yes
21	44	Amenorrhea	50	Resolved	57	Yes	6.3	450	Yes
		Ovarian failure	97	Resolved	10				

Abbreviations: AE, adverse event; BL, baseline; FSH, follicle-stimulating hormone; OT, ovarian toxicity.

[a]The reference range used for females of reproductive potential at any phase in the menstrual cycle in DeFi was 1.8–20.4 mIU/L.

[b]Although chemical biomarker data demonstrated resolution around the time of event resolution (FSH [16.7 mIU/mL] values were within normal limits for age on day 421), this patient was not considered to have confirmed hormonal resolution while remaining on nirogacestat at the time of the primary analysis because the last reported values (day 652) in the double-blind phase of DeFi were outside the expected reference range for a female of childbearing potential.

[c]Although chemical biomarker data demonstrated resolution around the time of event resolution (FSH [18.1 mIU/mL] values were within normal limits for age on day 337), this patient was not considered to have confirmed hormonal resolution while remaining on nirogacestat at the time of the primary analysis because the last reported values in the double-blind phase of DeFi were outside the expected reference range for a female of childbearing potential.

[d]Event was reported during the first month of treatment and the exact onset date for this event was not available; as such, the most conservative estimate of day 1 of treatment was used.

for clinical measures of OT (e.g., changes in menstrual cycle regularity or the development of symptoms of estrogen deficiency, including hot flashes, night sweats, and vaginal dryness).[9]

The OT events observed with nirogacestat treatment are likely a class effect, as preclinical and clinical data have demonstrated that GSIs can impact normal ovarian function and follicular development.[17,23–25] The mechanism for OT with nirogacestat is not fully elucidated, although it is likely due to the role of gamma secretase in Notch signaling, which plays critical roles in follicular activation, maturation, and growth; angiogenesis; and subsequent steroid hormone secretion by the developing corpus luteum.[16,17,19]

Activation of the primordial follicle, which initiates follicular maturation/growth, is controlled within the oocyte in adults via the phosphoinositide 3-kinase (PI3K)/protein kinase B (AKT) signaling pathway.[26] Because Notch1 can activate AKT, gamma secretase inhibition has the potential to block the activation and further development of the primordial follicle,[27] and therefore may not negatively impact the oocyte reserve. This mechanism may more likely contribute to OT persistence, because effects would be expected in >3 months (the time needed for follicular growth from primordial to ovulatory stages)[18] whereas median time to OT onset with nirogacestat was 8.9 weeks (~2 months). Other mechanisms through which nirogacestat could lead to OT are potential interference of blood supply and reduction in cell proliferation. The development and growth of the corpus luteum relies on angiogenesis from preexisting vessels of the follicular theca layer,[28] and inhibition of angiogenesis leads to attenuated follicular growth and disrupted ovulation.[29] The Notch and vascular endothelial growth factor signaling pathways, of which gamma secretase is an integral part, are critically involved in angiogenesis in the ovary.[19,30] Furthermore, given its expression pattern, Notch2 signaling likely mediates

follicular granulosa and thecal cell growth, and estradiol production, at later stages of follicular development.[31] Together, these data suggest that OT observed with nirogacestat may impede cell proliferation in growing follicles and interfere with the angiogenesis needed to support the development of late-stage follicles and corpus luteum, rather than damage or destroy ovarian tissue or primordial follicles. The timeline for recovery of ovarian function in all patients after cessation of nirogacestat treatment (median, ~11 weeks; range: 28–126 days) supports this hypothesis, because resolution of OT after treatment with known gonadotoxic agents can take 1 to 2 years, or more.[32,33]

Although DeFi OT resolution data suggest that the impact of nirogacestat on the ovary does not persist, the long-term effects of nirogacestat on fertility are currently unknown, and further data collection may be warranted. ASCO and European guidelines support fertility preservation methods with treatments known to or that could potentially impact ovarian function[34–36]; therefore, health care providers should initiate discussions with their patients to address any fertility concerns before DT treatment.

In conclusion, OT associated with nirogacestat treatment was identified by investigators based on abnormal reproductive hormone values or the presence of perimenopausal symptoms, or both. Most females of reproductive potential treated with nirogacestat experienced OT, with investigators reporting the majority resolved (specifically, all patients with OT who stopped nirogacestat recovered, and nearly three-fourths of patient with OT who continued nirogacestat recovered while on therapy). These results, along with what is currently understood about the mechanisms associated with the effect of GSIs on ovarian function, suggest that OT associated with nirogacestat is transient. Future analyses, including data from the DeFi open-label extension, are planned to better characterize the incidence and resolution of OT during and following treatment with nirogacestat.

AUTHOR CONTRIBUTIONS

Elizabeth T. Loggers: Conceptualization, investigation, writing–review and editing, formal analysis, project administration, and resources. **Rashmi Chugh:** Conceptualization, investigation, writing–review and editing, formal analysis, project administration, and resources. **Noah Federman:** Conceptualization, investigation, writing–review and editing, formal analysis, project administration, and resources. **Lee Hartner:** Conceptualization, investigation, writing–review and editing, formal analysis, project administration, and resources. **Richard F. Riedel:** Conceptualization, writing–review and editing, formal analysis, and resources. **Sunny Cho:** Conceptualization, writing–review and editing, formal analysis, project administration, and supervision. **David Hyslop:** Conceptualization, writing–review and editing, formal analysis, and supervision. **Allison Lim:** Conceptualization, writing–review and editing, formal analysis, and supervision. **Ana B. Oton:** Conceptualization, writing–review and editing, formal analysis, and supervision. **Kutluk H. Oktay:** Conceptualization, formal analysis, and writing–review and editing.

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CONFLICT OF INTEREST STATEMENT

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DATA AVAILABILITY STATEMENT

SpringWorks Therapeutics is committed to data transparency and sharing data to further research while maintaining the privacy and confidentiality of research participants. Pertinent patient-level data from completed registrational clinical trials will be made available by SpringWorks to qualified researchers on approval of reasonable requests following de-identification/anonymization pursuant to applicable law. Requests for data must be sent to medinfo@springworkstx.com.

ORCID

Elizabeth T. Loggers https://orcid.org/0000-0003-3432-0443
Richard F. Riedel https://orcid.org/0000-0001-5412-8710
Kutluk H. Oktay https://orcid.org/0000-0003-0914-7757

REFERENCES

1. Kasper B, Baumgarten C, Garcia J, et al. An update on the management of sporadic desmoid-type fibromatosis: a European Consensus Initiative between Sarcoma PAtients EuroNet (SPAEN) and European Organization for Research and Treatment of Cancer (EORTC)/Soft Tissue and Bone Sarcoma Group (STBSG). *Ann Oncol*. 2017;28(10):2399-2408. doi:10.1093/annonc/mdx323
2. Sbaraglia M, Bellan E, Dei Tos AP. The 2020 WHO classification of soft tissue tumours: news and perspectives. *Pathologica*. 2021;113(2):70-84. doi:10.32074/1591-951x-213
3. Bektas M, Bell T, Khan S, et al. Desmoid tumors: a comprehensive review. *Adv Ther*. 2023;40(9):3697-3722. doi:10.1007/s12325-023-02592-0
4. van Broekhoven DL, Grunhagen DJ, den Bakker MA, van Dalen T, Verhoef C. Time trends in the incidence and treatment of extra-abdominal and abdominal aggressive fibromatosis: a population-based study. *Ann Surg Oncol*. 2015;22(9):2817-2823. doi:10.1245/s10434-015-4632-y
5. Anneberg M, Svane HML, Fryzek J, et al. The epidemiology of desmoid tumors in Denmark. *Cancer Epidemiol*. 2022;77:102114. doi:10.1016/j.canep.2022.102114
6. Nieuwenhuis MH, Casparie M, Mathus-Vliegen LM, Dekkers OM, Hogendoorn PC, Vasen HF. A nation-wide study comparing sporadic and familial adenomatous polyposis-related desmoid-type fibromatoses. *Int J Cancer*. 2011;129(1):256-261. doi:10.1002/ijc.25664
7. PubChem. *Nirogascestat (PubChem Compound Summary for CID 46224413)*. National Library of Medicine. National Center for Biotechnology Information; July 6, 2010. Accessed January 23, 2024. https://pubchem.ncbi.nlm.nih.gov/compound/Nirogacestat
8. Federman N. Molecular pathogenesis of desmoid tumor and the role of gamma-secretase inhibition. *NPJ Precis Oncol*. 2022;6(1):62. doi:10.1038/s41698-022-00308-1
9. Nirogacestat. *Prescribing information*. SpringWorks Therapeutics. Inc; 2023.
10. *FDA approves nirogacestat for desmoid tumors*. US Food and Drug Administration (FDA). November 28, 2023. Accessed January 23, 2024. https://www.fda.gov/drugs/resources-information-approved-drugs/fda-approves-nirogacestat-desmoid-tumors
11. Gounder M, Ratan R, Alcindor T, et al. Nirogacestat, a γ-secretase inhibitor for desmoid tumors. *N Engl J Med*. 2023;388(10):898-912. doi:10.1056/nejmoa2210140
12. Cui W, Rocconi RP, Thota R, et al. Measuring ovarian toxicity in clinical trials: an American Society of Clinical Oncology research statement. *Lancet Oncol*. 2023;24(10):e415-e423. doi:10.1016/s1470-2045(23)00390-x
13. Reynolds AC, McKenzie LJ. Cancer treatment-related ovarian dysfunction in women of childbearing potential: management and fertility preservation options. *J Clin Oncol*. 2023;41(12):2281-2292. doi:10.1200/jco.22.01885
14. Faddy MJ, Gosden RG, Gougeon A, Richardson SJ, Nelson J. Accelerated disappearance of ovarian follicles in mid-life: implications for forecasting menopause. *Hum Reprod*. 1992;7(10):1342-1346. doi:10.1093/oxfordjournals.humrep.a137570
15. Himpe J, Lammerant S, Van den Bergh L, Lapeire L, De Roo C. The impact of systemic oncological treatments on the fertility of adolescents and young adults—a systematic review. *Life*. 2023;13(5):1209. doi:10.3390/life13051209
16. Guo S, Quan S, Zou S. Roles of the notch signaling pathway in ovarian functioning. *Reprod Sci*. 2021;28(10):2770-2778. doi:10.1007/s43032-021-00610-6
17. Vanorny DA, Mayo KE. The role of Notch signaling in the mammalian ovary. *Reproduction*. 2017;153(6):R187-R204. doi:10.1530/rep-16-0689
18. Oktay K, Newton H, Mullan J, Gosden RG. Development of human primordial follicles to antral stages in SCID/hpg mice stimulated with follicle stimulating hormone. *Hum Reprod*. 1998;13(5):1133-1138. doi:10.1093/humrep/13.5.1133
19. Xie Q, Cheng Z, Chen X, Lobe CG, Liu J. The role of Notch signalling in ovarian angiogenesis. *J Ovarian Res*. 2017;10(1):13. doi:10.1186/s13048-017-0308-5
20. Titus S, Szymanska KJ, Musul B, et al. Individual-oocyte transcriptomic analysis shows that genotoxic chemotherapy depletes human primordial follicle reserve in vivo by triggering proapoptotic pathways without growth activation. *Sci Rep*. 2021;11(1):407. doi:10.1038/s41598-020-79643-x
21. Cui W, Phillips KA, Francis PA, et al. Understanding the barriers to, and facilitators of, ovarian toxicity assessment in breast cancer clinical trials. *Breast*. 2022;64:56-62. doi:10.1016/j.breast.2022.05.002
22. Cui W, Francis PA, Loi S, et al. Assessment of ovarian function in phase III (neo)adjuvant breast cancer clinical trials: a systematic evaluation. *J Natl Cancer Inst*. 2021;113(12):1770-1778. doi:10.1093/jnci/djab111
23. Simutis FJ, Sanderson TP, Pilcher GD, Graziano MJ. Nonclinical safety assessment of the γ-secretase inhibitor avagacestat. *Toxicol Sci*. 2018;163(2):525-542. doi:10.1093/toxsci/kfy048
24. Simutis FJ, Sanderson TP, Pilcher GD, Graziano MJ. Investigations on the relationship between ovarian, endocrine, and renal findings in nonclinical safety studies of the γ-secretase inhibitor avagacestat. *Toxicol Sci*. 2019;171(1):98-116. doi:10.1093/toxsci/kfz129
25. Gounder M, Jones RL, Chugh R, et al. RINGSIDE phase 2/3 trial of AL102 for treatment of desmoid tumors (DT): phase 2 results. *J Clin Oncol*. 2023;41(suppl 16):11515. doi:10.1200/jco.2023.41.16_suppl.11515
26. John GB, Gallardo TD, Shirley LJ, Castrillon DH. Foxo3 is a PI3K-dependent molecular switch controlling the initiation of oocyte growth. *Dev Biol*. 2008;321(1):197-204. doi:10.1016/j.ydbio.2008.06.017
27. Hales EC, Taub JW, Matherly LH. New insights into Notch1 regulation of the PI3K-AKT-mTOR1 signaling axis: targeted therapy of γ-secretase inhibitor resistant T-cell acute lymphoblastic leukemia. *Cell Signal*. 2014;26(1):149-161. doi:10.1016/j.cellsig.2013.09.021
28. Woad KJ, Robinson RS. Luteal angiogenesis and its control. *Theriogenology*. 2016;86(1):221-228. doi:10.1016/j.theriogenology.2016.04.035
29. Robinson RS, Woad KJ, Hammond AJ, Laird M, Hunter MG, Mann GE. Angiogenesis and vascular function in the ovary. *Reproduction*. 2009;138(6):869-881. doi:10.1530/rep-09-0283
30. Boulton ME, Cai J, Grant MB. γ-Secretase: a multifaceted regulator of angiogenesis. *J Cell Mol Med*. 2008;12(3):781-795. doi:10.1111/j.1582-4934.2008.00274.x
31. Jing J, Jiang X, Chen J, et al. Notch signaling pathway promotes the development of ovine ovarian follicular granulosa cells. *Anim Reprod Sci*. 2017;181:69-78. doi:10.1016/j.anireprosci.2017.03.017
32. Goldfarb SB, Turan V, Bedoschi G, et al. Impact of adjuvant chemotherapy or tamoxifen-alone on the ovarian reserve of young women with breast cancer. *Breast Cancer Res Treat*. 2021;185(1):165-173. doi:10.1007/s10549-020-05933-7
33. Oktay KH, Turan V, Bedoschi G, Abdo N, Bang H, Goldfarb S. A prospective longitudinal analysis of the predictors of amenorrhea after breast cancer chemotherapy: impact of BRCA pathogenic variants. *Cancer Med*. 2023;12(18):19225-19233. doi:10.1002/cam4.6527
34. Lambertini M, Peccatori FA, Demeestere I, et al. Fertility preservation and post-treatment pregnancies in post-pubertal cancer patients: ESMO Clinical Practice Guidelines. *Ann Oncol*. 2020;31(12):1664-1678. doi:10.1016/j.annonc.2020.09.006

35. Loren AW, Mangu PB, Beck LN, et al. Fertility preservation for patients with cancer: American Society of Clinical Oncology clinical practice guideline update. *J Clin Oncol*. 2013;31(19):2500-2510. doi:10.1200/jco.2013.49.2678
36. Oktay K, Harvey BE, Partridge AH, et al. Fertility preservation in patients with cancer: ASCO clinical practice guideline update. *J Clin Oncol*. 2018;36(19):1994-2001. doi:10.1200/jco.2018.78.1914

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