

## Appendix G - FDA-REQUESTED SUPPLEMENTAL ANALYSES

FDA requested IceCure perform additional analyses to evaluate outcomes in different subpopulations of ICE3 clinical study patients based on adjuvant treatments and biologic characteristics.

### 1.1.Subpopulations Intended to Align with LUMINA

#### 1.1.1. Subpopulation with Endocrine Therapy Only

##### Patient Selection

This subpopulation includes 124 patients from the ICE3 clinical study who received adjuvant endocrine therapy without adjuvant radiotherapy.

##### IBTR Rate

The subpopulation who received endocrine therapy only demonstrated an estimated local IBTR five-year recurrence rate of 3.7% at year five follow-up.

*Please note: One patient returned late for their five-year visit (63 months post-treatment) and was observed to have recurred. Typically, Kaplan Meier survival analysis truncates the data contribution at the 5-year anniversary (Month 60 or Day 1825) and would not have included this event. The analysis shown in Table 1 conservatively includes this recurrence observed during the 5-year visit which occurred after the 5-year treatment anniversary.*

**Table 1: ICE3 Clinical Study Local IBTR: ICE Subpopulation with Endocrine Therapy Only (N=124)**

Time	N start*	At Risk**	Cumulative Recurrence	Survival Estimate†	Recurrence Estimate†	1-sided 95% CIUB	2-sided 95% CILB	2-sided 95% CIUB
Operative	124	124	-	-	-	-	0.0%	-
Month 6	124	123	0	100.0%	0.0%	0.0%	0.0%	0.0%
Year 1	123	123	0	100.0%	0.0%	0.0%	0.0%	0.0%
Year 2	123	120	0	100.0%	0.0%	0.0%	0.0%	0.0%
Year 3	120	115	0	100.0%	0.0%	0.0%	0.0%	0.0%
Year 4	115	108	2	98.2%	1.8%	5.6%	0.4%	6.9%
Year 5	108	87	4	96.3%	3.7%	8.2%	1.4%	9.6%

\*N start: number of patients at the beginning of the follow-up time interval.

\*\*At risk: number of patients that completed the follow-up time interval with no IBTR event.

†Kaplan-Meier (product-limit) estimate with 1-sided 95% CI upper bound (UB) and 2-sided 95% lower and upper bounds (LB and UB).

Source: Table 4\_1 Local IBTR\_Radiation No - Hormone Yes.sas; Analyzed: 30MAR2024

## Secondary Effectiveness Endpoint

Secondary effectiveness, including disease free survival, overall survival, and breast cancer survival are show below in **Table 2**. Disease free survival is presented according to the pre-specified definition in the ICE3 protocol and, separately, according to the National Cancer Institute (NCI) definition. Definitions are as follows:

- ICE3 protocol defines as local (Ductal Carcinoma In Situ (DCIS) or invasive), regional, or distant breast cancer recurrence, second primary cancer, DCIS or invasive contralateral breast cancer, or death due to any cause.
- National Cancer Institute defines as local (Ductal Carcinoma In Situ (DCIS) or invasive), regional, or distant breast cancer recurrence.

Overall survival includes patients who died from any cause, regardless of relationship to breast cancer. Breast cancer survival considers only patients who died from breast cancer.

The disease free survival according to the ICE3 protocol definition was 83.0% and 95.3% according to the NCI definition. The overall survival and breast cancer survival were 89.5% and 96.1%, respectively.

**Table 2: ICE3 Clinical Study Summary of Secondary Effectiveness Endpoints - Survival Analysis - ICE3 Subpopulation with Endocrine Therapy Only (N=124)**

<b>Secondary Effectiveness Endpoint</b> <i>(see outcome definition in Section 5.5)</i>	<b>Year 5 Survival Rate</b>	<b>2-sided 95% CI LB</b>	<b>2-sided 95% CI UB</b>
<b>Disease Free Survival - Protocol Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	83.0%	74.7%	88.9%
<b>Disease Free Survival - NCI Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	95.3%	89.1%	98.0%
<b>Overall Survival Estimate</b>	89.5%	82.2%	93.9%
<b>Breast Cancer Survival Estimate</b>	96.1%	89.9%	98.5%

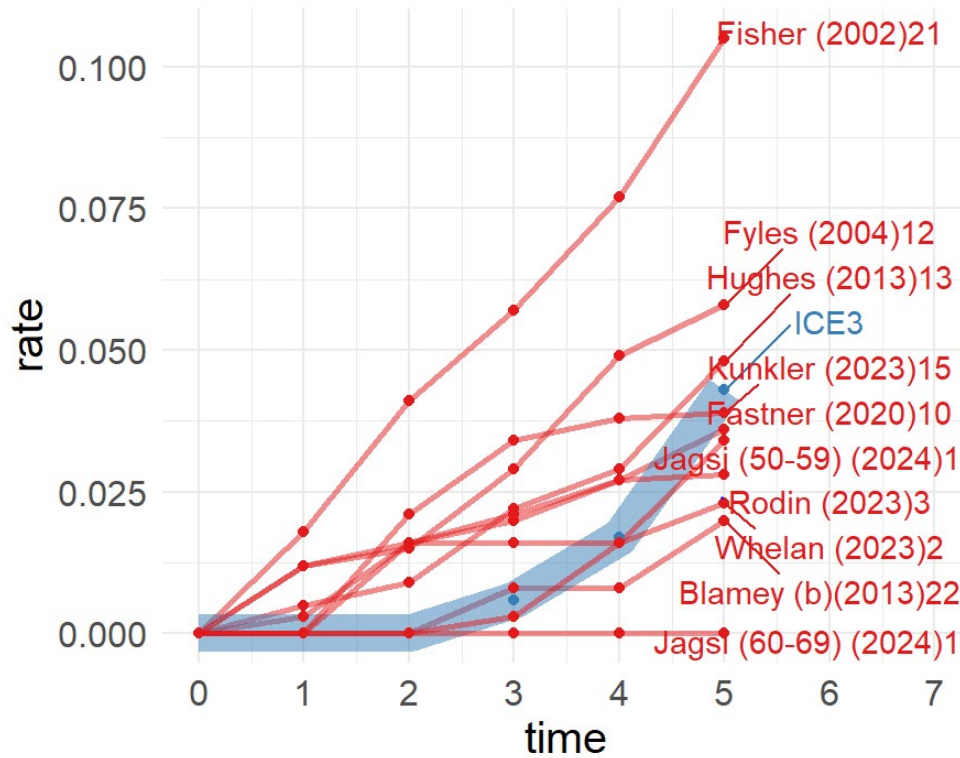
## Comparison to IceCure PRISMA Sensitivity (Lumpectomy with Endocrine Therapy)

As described in **Appendix C**, IceCure performed a sensitivity analysis of the PRISMA Systematic Review and Meta-analysis results to evaluate the 5-year IBTR rate excluding studies where <50% of treated patients received adjuvant endocrine therapy. As a result, Stenmark Tullberg (2021), Wickberg (2018), and Blamey (2013) treatment arm (a) were excluded from the sensitivity analysis. The 5-year IBTR rate calculated from the sensitivity analysis was compared to the 5-year IBTR rate for the ICE3 subpopulation (with endocrine therapy).

The results from the PRISMA Systematic Review and Meta-analysis sensitivity analysis represent the established and accepted outcomes of the existing standard of care treatment lumpectomy without radiotherapy and with endocrine therapy and constitute a valid reference rate for

comparison to the proposed indicated population (i.e., the ICE3 subpopulation (with endocrine therapy)). The PRISMA Sensitivity Analysis (Limited to Populations with Endocrine Therapy) resulted in an **estimated 5-year IBTR rate of 2.82% with a 95% CI from 1.62% to 4.83%**.

The reported IBTR rates from the subset of PRISMA-selected literature used in this sensitivity analysis and the ICE3 clinical study ICE3 subpopulation (with endocrine therapy) are summarized below.



**Figure 1. Ipsilateral Breast Tumor Recurrence (IBTR) Rate Up to 5-years for the ICE3 clinical study ICE3 Subpopulation (with Endocrine Therapy) and Each Study Included in the PRISMA Sensitivity Analysis (Limited to Populations with Endocrine Therapy)**

Both the ICE Subpopulation with Endocrine Therapy only and the PRISMA Sensitivity Analysis (Lumpectomy with Endocrine Therapy) populations evaluated patients who received adjuvant endocrine therapy only. A comparison between the populations can be seen below.

**Table 3: Comparison of ICE3 Subpopulation with Endocrine Therapy Only and PRISMA Sensitivity Analysis (Lumpectomy with Endocrine Therapy) Derived Comparator**

	5-Year IBTR	95% CI UB
ICE3 Subpopulation with Endocrine Therapy Only	3.7%	9.6%
PRISMA Sensitivity Analysis (Lumpectomy with Endocrine Therapy) Derived Comparator	2.82%	4.83%

## 1.1.2. Subpopulation Aligned with LUMINA Study

### Patient Selection

This subpopulation includes 56 patients from the ICE3 clinical study who for adjuvant treatment only received endocrine therapy. Additionally, these patients have a known Ki67 score of Ki67<14. This subpopulation excludes patients who are PR-. This subpopulation is aligned with the enrollment criteria of the LUMINA study.

### IBTR Rate

This subpopulation aligned with the LUMINA study demonstrated an estimated local IBTR rate of 2.17%.

*Please note: One patient returned late for their five-year visit (63 months post-treatment) and was observed to have recurred. Typically, Kaplan Meier survival analysis truncates the data contribution at the 5-year anniversary (Month 60 or Day 1825) and would not have included this event. The analysis shown in Table 12 conservatively includes this recurrence observed during the 5-year visit which occurred after the 5-year treatment anniversary.*

**Table 4: ICE3 Clinical Study Local IBTR: ICE Subpopulation Aligned with LUMINA Study (N=56)**

Time	N start*	Cumulative Recurrence	Survival Estimate†	Recurrence Estimate†	2-sided 95% CI LB	2-sided 95% CI UB
Operative	56	-	-	-	-	-
Month 6	56	0	100.0%	0.0%	0.0%	0.0%
Year 1	56	0	100.0%	0.0%	0.0%	0.0%
Year 2	55	0	100.0%	0.0%	0.0%	0.0%
Year 3	53	0	100.0%	0.0%	0.0%	0.0%
Year 4	50	0	100.0%	0.0%	0.0%	0.0%
Year 5	38	1	97.8%	2.2%	0.3%	14.5%

### Secondary Effectiveness Endpoint

The disease-free survival according to the ICE3 protocol definition was 82.2% and 97.7% according to the NCI definition, as shown below in **Table 13**. The overall survival and breast cancer survival were 92.5% and 98.0%, respectively.

**Table 5: ICE3 Clinical Study Summary of Secondary Effectiveness Endpoints - Survival Analysis - ICE3 Subpopulation Aligned with LUMINA Study (N=56)**

Secondary Effectiveness Endpoint (see outcome definition in Section 5.5)	Year 5 Survival Rate	2-sided 95% CI LB	2-sided 95% CI UB
<b>Disease Free Survival - Protocol Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	82.2%	68.5%	90.4%
<b>Disease Free Survival - NCI Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	97.7%	84.9%	99.7%
<b>Overall Survival Estimate</b>	92.5%	81.1%	97.1%

<b>Secondary Effectiveness Endpoint</b> (see outcome definition in Section 5.5)	<b>Year 5 Survival Rate</b>	<b>2-sided 95% CI LB</b>	<b>2-sided 95% CI UB</b>
<b>Breast Cancer Survival Estimate</b>	98.0%	86.9%	99.7%

Comparison to LUMINA Study

Both the patients in the ICE3 subpopulation aligned with the LUMINA study and the LUMINA study received adjuvant endocrine therapy. Both groups were PR-. The ICE3 subpopulation had a Ki67<14%, the LUMINA study has a Ki67≤13.25%.

As shown in the table below, the LUMINA study enrolled a patient population much larger compared to the ICE3 subpopulation. The relatively low sample size directly contributes to the high confidence interval, despite the fact that only one recurrence was noted in the ICE3 subpopulation.

**Table 6: Comparison of ICE3 Subpopulation Aligned with LUMINA Study and LUMINA Study**

	<b>5-Year IBTR</b>	<b>95% CI UB</b>
ICE3 Subpopulation Aligned with LUMINA Study (N=56)	2.2%	14.4%
LUMINA Study (N=500)	2.7%	4.1%

**1.1.3. Subpopulation Aligned with LUMINA and Nuclear Grade ≤ 2**

Patient Selection

This subpopulation includes 48 patients from the ICE3 clinical study who for adjuvant treatment only received endocrine therapy. These patients also have a known Ki67 score of Ki67<14 and a nuclear grade ≤ 2. This subpopulation excludes patients who are PR-. This subpopulation is aligned with the enrollment criteria of the LUMINA study and additionally has a nuclear grade ≤ 2.

IBTR Rate

This subpopulation aligned with the LUMINA study demonstrated an estimated local IBTR rate of 2.56%.

*Please note: One patient returned late for their five-year visit (63 months post-treatment) and was observed to have recurred. Typically, Kaplan Meier survival analysis truncates the data contribution at the 5-year anniversary (Month 60 or Day 1825) and would not have included this event. The analysis shown in Table 15 conservatively includes this recurrence observed during the 5-year visit which occurred after the 5-year treatment anniversary.*

**Table 7: ICE3 Clinical Study Summary of Secondary Effectiveness Endpoints - Survival Analysis - ICE3 Subpopulation Aligned with LUMINA Study and Nuclear Grade  $\leq 2$  (N=48)**

Time	N start*	Cumulative Recurrence	Survival Estimate†	Recurrence Estimate†	2-sided 95% CI LB	2-sided 95% CI UB
Operative	48	-	-	-	-	-
Month 6	48	0	100.0%	0.0%	0.0%	0.0%
Year 1	48	0	100.0%	0.0%	0.0%	0.0%
Year 2	47	0	100.0%	0.0%	0.0%	0.0%
Year 3	45	0	100.0%	0.0%	0.0%	0.0%
Year 4	42	0	100.0%	0.0%	0.0%	0.0%
Year 5	31	1	97.4%	2.6%	0.4%	16.8%

### Secondary Effectiveness Endpoint

The disease-free survival according to the ICE3 protocol definition was 90.1% and 96.6% according to the NCI definition, as shown below in **Table 5**. The overall survival and breast cancer survival were 93.3% and 97.7%, respectively.

**Table 8: Table 11: ICE3 Clinical Study Summary of Secondary Effectiveness Endpoints - Survival Analysis - ICE3 Subpopulation Aligned with LUMINA Study and Nuclear Grade  $\leq 2$  (N=48)**

Secondary Effectiveness Endpoint (see outcome definition in Section 5.5)	Year 5 Survival Rate	2-sided 95% CI	2-sided 95% CI UB
<b>Disease Free Survival - Protocol Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	90.1%	75.4%	96.2%
<b>Disease Free Survival - NCI Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	96.6%	77.9%	99.5%
<b>Overall Survival Estimate</b>	93.3%	80.8%	97.8%
<b>Breast Cancer Survival Estimate</b>	97.7%	84.9%	99.7%

### Comparison to LUMINA Study

Both the patients in the ICE3 subpopulation aligned with the LUMINA study and the LUMINA study received adjuvant endocrine therapy. Both groups were PR-. The ICE3 subpopulation had a KI67<14%, the LUMINA study has a Ki67 $\leq$ 13.25%. This subpopulation of ICE3 patients was confirmed to have a nuclear grade  $\leq 2$ .

As shown in the table below, the LUMINA study enrolled a patient population much larger compared to the ICE3 subpopulation. The relatively low sample size directly contributes to the high confidence interval, despite the fact that only one recurrence was noted in the ICE3 subpopulation.

**Table 9: Comparison of ICE3 Subpopulation Aligned with LUMINA Study and Nuclear Grade ≤ 2 and LUMINA Study**

	<b>5-Year IBTR</b>	<b>95% CI UB</b>
ICE3 Subpopulation aligned with LUMINA Study and Nuclear Grade ≤ 2 (N=48)	2.6%	16.8%
LUMINA Study (N=500)	2.7%	4.1%

## 1.2.Subpopulations Intended to Align with Proposed Indications

### 1.2.1. Subpopulation Aligned with ProSense™ Indications

#### Patient Selection

This subpopulation includes 147 patients from the ICE3 clinical study who are aligned with the proposed ProSense™ indications. This subpopulation excludes patients who are < 60 years of age and/ or without adjuvant endocrine therapy.

#### IBTR Rate

This subpopulation aligned with the ProSense™ indications demonstrated an estimated local IBTR rate of 3.08%.

*Please note: One patient returned late for their five-year visit (63 months post-treatment) and was observed to have recurred. Typically, Kaplan Meier survival analysis truncates the data contribution at the 5-year anniversary (Month 60 or Day 1825) and would not have included this event. The analysis shown in Table 4 conservatively includes this recurrence observed during the 5-year visit which occurred after the 5-year treatment anniversary.*

**Table 10: ICE3 Clinical Study Local IBTR: ICE Subpopulation Aligned with ProSense™ Indications (N=147)**

<b>Time</b>	<b>N start*</b>	<b>Cumulative Recurrence</b>	<b>Survival Estimate†</b>	<b>Recurrence Estimate†</b>	<b>2-sided 95% CI LB</b>	<b>2-sided 95% CI UB</b>
Operative	147	-	-	-	-	-
Month 6	146	0	100.0%	0.0%	0.0%	0.0%
Year 1	146	0	100.0%	0.0%	0.0%	0.0%
Year 2	142	0	100.0%	0.0%	0.0%	0.0%
Year 3	137	0	100.0%	0.0%	0.0%	0.0%
Year 4	130	2	98.5%	1.5%	0.4%	5.8%
Year 5	106	4	96.9%	3.1%	1.2%	8.0%

#### Secondary Effectiveness Endpoint

The disease-free survival according to the ICE3 protocol definition was 84.4% and 95.4% according to the NCI definition, as shown below in **Table 5**.

**Table 11: ICE3 Clinical Study Summary of Secondary Effectiveness Endpoints - Survival Analysis - ICE3 Subpopulation Aligned with ProSense™ Indications (N=147)**

Secondary Effectiveness Endpoint (see outcome definition in Section 5.5)	Year 5 Survival Rate	2-sided 95% CI LB	2-sided 95% CI UB
<b>Disease Free Survival - Protocol Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	84.4%	77.1%	89.6%
<b>Disease Free Survival - NCI Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	95.4%	90%	97.9%

Comparison to IceCure PRISMA Sensitivity (Lumpectomy with Endocrine Therapy)

Both the ICE3 subpopulation aligned with ProSense™ Indications and the PRISMA Sensitivity (Lumpectomy with Endocrine Therapy) includes patients who received endocrine therapy as adjuvant treatment.

**Table 12: Comparison of ICE3 Subpopulation Aligned with ProSense™ Indications and PRISMA Sensitivity Analysis (Lumpectomy with Endocrine Therapy) Derived Comparator**

	5-Year IBTR	95% CI UB
ICE3 Subpopulation Aligned with ProSense™ Indications	3.08%	8.0%
PRISMA Sensitivity Analysis (Lumpectomy with Endocrine Therapy) Derived Comparator	2.82%	4.83%

**1.2.2. Subpopulation Aligned with ProSense™ Indications and Nuclear Grade ≤ 2**

Patient Selection

This subpopulation includes 120 patients from the ICE3 clinical study who are aligned with the proposed ProSense™ indications and have a nuclear grade ≤ 2. These patients are ≥ 60 years of age and received adjuvant endocrine therapy. These patients may have also received other adjuvant treatments.

IBTR Rate

This subpopulation aligned with the proposed ProSense™ indications and with a nuclear grade ≤ 2 demonstrated an estimated local IBTR rate of 1.95%.

*Please note: One patient returned late for their five-year visit (63 months post-treatment) and was observed to have recurred. Typically, Kaplan Meier survival analysis truncates the data contribution at the 5-year anniversary (Month 60 or Day 1825) and would not have included this event. The analysis shown in Table 9 conservatively includes this recurrence observed during the 5-year visit which occurred after the 5-year treatment anniversary.*



**Table 13: ICE3 Clinical Study Local IBTR: ICE Subpopulation Aligned with ProSense™ Indications and Nuclear Grade ≤ 2 (N=120)**

Time	N start*	Cumulative Recurrence	Survival Estimate†	Recurrence Estimate†	2-sided 95% CI LB	2-sided 95% CI UB
Operative	120	-	-	-	-	-
Month 6	119	0	100.0%	0.0%	0.0%	0.0%
Year 1	119	0	100.0%	0.0%	0.0%	0.0%
Year 2	115	0	100.0%	0.0%	0.0%	0.0%
Year 3	111	0	100.0%	0.0%	0.0%	0.0%
Year 4	107	0	100.0%	0.0%	0.0%	0.0%
Year 5	87	2	98.1%	2.0%	0.5%	7.6%

### Secondary Effectiveness Endpoint

The disease-free survival according to the ICE3 protocol definition was 87.1% and 96.2% according to the NCI definition, as shown below in **Table 10**. The overall survival and breast cancer survival were 92.8% and 96.2%, respectively.

**Table 14: ICE3 Clinical Study Summary of Secondary Effectiveness Endpoints - Survival Analysis - ICE3 Subpopulation Aligned with ProSense™ Indications and Nuclear Grade ≤ 2 (N=120)**

Secondary Effectiveness Endpoint (see outcome definition in Section 5.5)	Year 5 Survival Rate	2-sided 95% CI LB	2-sided 95% CI UB
<b>Disease Free Survival - Protocol Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	87.1%	79.2%	92.2%
<b>Disease Free Survival - NCI Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	96.2%	90.0%	98.5%
<b>Overall Survival Estimate</b>	92.8%	86.1%	96.3%
<b>Breast Cancer Survival Estimate</b>	96.2%	90.2%	98.6%

### Comparison to IceCure PRISMA Sensitivity (Lumpectomy with Endocrine Therapy)

Both the ICE3 subpopulation aligned with ProSense™ indications and with nuclear grade ≤ 2 and the PRISMA Sensitivity (Lumpectomy with Endocrine Therapy) includes patients who received endocrine therapy as adjuvant treatment.

**Table 15: Comparison of ICE3 Subpopulation Aligned with ProSense™ Indications and PRISMA Sensitivity Analysis (Lumpectomy with Endocrine Therapy) Derived Comparator**

	5-Year IBTR	95% CI UB
ICE3 Subpopulation Aligned with ProSense™ Indications	1.95%	7.60%
PRISMA Sensitivity Analysis (Lumpectomy with Endocrine Therapy) Derived Comparator	2.82%	4.83%

## 1.3 Subpopulation Intended to Evaluate Impact of Adjuvant Radiotherapy

### 1.2.3. Subpopulation with Radiation Therapy

#### Patient Selection

This subpopulation includes 29 patients from the ICE3 clinical study who received radiation therapy as adjuvant treatment with or without endocrine therapy.

#### IBTR Rate

This subpopulation who received radiation therapy as adjuvant treatment demonstrated an estimated local IBTR rate of 0.00%

**Table 16: ICE3 Clinical Study Local IBTR: ICE Subpopulation Aligned with ProSense™ Indications (N=29)**

Time	N start*	Cumulative Recurrence	Survival Estimate†	Recurrence Estimate†	2-sided 95% CI LB	2-sided 95% CI UB
Operative	29	-	-	-	-	-
Month 6	29	0	100.0%	0.0%	0.0%	0.0%
Year 1	29	0	100.0%	0.0%	0.0%	0.0%
Year 2	28	0	100.0%	0.0%	0.0%	0.0%
Year 3	27	0	100.0%	0.0%	0.0%	0.0%
Year 4	26	0	100.0%	0.0%	0.0%	0.0%
Year 5	23	0	100.0%	0.0%	0.0%	0.0%

#### Secondary Effectiveness Endpoint

As shown below in Table 8, the 5-year rate of disease free survival was 88.7% according to the ICE3 protocol definition and 96.2% according to the NCI definition. The overall survival was 96.2%. Breast cancer survival in this cohort was 100% as no patient had local recurrence.

**Table 17: ICE3 Clinical Study Summary of Secondary Effectiveness Endpoints - Survival Analysis - ICE3 Subpopulation with Radiation Therapy (N=29)**

Secondary Effectiveness Endpoint (see outcome definition in Section 5.5)	Year 5 Survival Rate	2-sided 95% CI LB	2-sided 95% CI UB
<b>Disease Free Survival - Protocol Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	88.7%	69.0%%	96.2%
<b>Disease Free Survival - NCI Definition</b> <i>Includes events observed beyond 5 year anniversary</i>	96.2%	75.7%	99.4%
<b>Overall Survival Estimate</b>	92.4%	73.0%	98.1%
<b>Breast Cancer Survival Estimate</b>	100.0%	0.0%	0.0%