Statistical Analysis Plan (SAP)

A Prospective, Single-Arm Multi-Center Study of the HARMONIC® 1100 Shears and Generator G11 in Pediatric Surgical Procedures (General) and Adult Surgical Procedures (General, Gynecological, Urological, and Thoracic)

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The following individuals have reviewed this version of the Statistical Analysis Plan and are in agreement with the content:

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Revision History

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1.0	06JUL2023	First Version

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1 Introduction

This is the Statistical Analysis Plan (SAP) for the final analysis of data collected under Protocol ENG_2020_06 (Amendement 3, dated May 17, 2023). This SAP describes in detail the statistical methodology and statistical analyses for this protocol.

1.1 **Study Objectives**

The primary objective of this study is to demonstrate the acceptable performance and safety of the HARMONIC 1100 Shears and GEN11 devices when used per the instructions for use (IFU) in pediatric surgical procedures (general) or adult surgical procedures (general, gynecological, urological, and thoracic) where at least one single vessel is planned to be transected by the HARMONIC 1100 Shears.

The secondary objective of this study is to demonstrate the acceptable performance and safety of the HARMONIC 1100 Shears in the transection of > 5 to 7 mm diameter size vessels.

1.2 **Study Design**

This prospective, single-arm, multi-center study will collect clinical data in a post-market setting for the pediatric population (general surgical procedures) and adult population (general, gynecological, urological, and thoracic surgical procedures). Investigators will perform each procedure using the device in compliance with their standard surgical approach and the HARMONIC 1100 Shears and GEN11 IFUs.

Subjects will be consented and screened anytime during a period of 8 weeks prior to the date of surgery. Subjects will be considered enrolled when the HARMONIC 1100 Shears has been attempted to be used for at least one single vessel transection during pediatric surgical procedures (general) and adult surgical procedures (general, gynecological, urological, and thoracic). All subjects enrolled will be followed post-operatively through discharge and again at 28 days (± 14 days) post-surgery; therefore, from the surgery date to study exit, the duration will be approximately 6 weeks.

A minimum of 165 subjects enrolled (maximum of 328 subjects enrolled) will be included in the study from up to 15 surgery centers globally with the following procedure targets:

- A minimum of 33 subjects enrolled to a maximum of 40 subjects enrolled for Pediatric general procedure;
- A minimum of 33 subjects enrolled to a maximum of 119 subjects enrolled for Adults general procedures (Adult HPB, Adult lower GI and Adults gastric {gastrectomy [e.g., subtotal, total], esophagectomy, RYGB, sleeve gastrectomy, or BPD/DS});
- A minimum of 33 subjects enrolled to a maximum of 40 subjects enrolled for Adult gynecological (e.g., hysterectomy, oophorectomy, myomectomy or endometriosis resection);

- A minimum of 33 subjects enrolled to a maximum of 40 subjects enrolled for Adult urological (e.g., nephrectomy, cystectomy or prostatectomy); and
- A minimum of 33 subjects enrolled to a maximum of 89 subjects enrolled for Adult thoracic (lung resection [branches, excluding main pulmonary artery {PA}/pulmonary vein {PA}).

2 Treatment Assignment

This is a single-arm study where all enrolled subjects will have the HARMONIC 1100 Shears device utilized for transection of at least one vessel.

3 Randomization and Blinding Procedures

As this is a single-arm study, no randomization will occur, and no blinding procedures are required.

4 Interval Windows

Interval windows for the purpose of analysis in this study will not be defined outside of those already specified in the protocol for visit scheduling as the collection of data for the primary and secondary performance endpoints occurs intra-operatively. The final visit occurs approximately 4 weeks after surgery, thus no interval windows need to be defined given the absence of long-term follow-up in this study. The Schedule of Events specifies a window of 14 days around the scheduling of the 4-week follow-up visit, and any information entered in the eCRFs at this visit will correspond to the 4-week visit. There will be no assigning of observations to time points outside of the visit to which they are recorded in the eCRFs.

5 Levels of Significance

The hypotheses in this study will be evaluated using a one-sided significance level of 0.025. Estimation of all additional endpoints will be performed using two-sided 95% confidence intervals.

6 Analysis Sets

All Enrolled Set (AES): The all enrollment set contains all subjects who provide informed consent for this study . The AES will be used for subject disposition and all listings.

Full Analysis Set (FAS): The FAS will includeds all subjects in whom the HARMONIC 1100 Shears is utilized during the surgical procedure to transect at least one vessel and have assessment for the primary endpoint. The summary of all primary and secondary performance and safety endpoints will be performed by surgical population (pediatric and adult), surgical procedure group, procedure subgroup, and on the entire pooled set of subjects. The hypotheses described in Section 8.4.1 below will be evaluated on the pooled set of subjects and the > 5 to 7

mm diameter vessel population. The > 5 to 7 mm diameter vessel population will be based off the surgeon determination of the vessel size.

Safety Analysis Set (SAS): The SAS will include all subjects who signed informed consent and for whom HARMONIC 1100 Shears device is utilized during the surgical procedure. The safety endpoints will be summarized using SAS by surgical population (pediatric and adult), surgical procedure group, procedure subgroup, and on the entire pooled set of subjects.

7 Sample Size Justification

Primary performance analysis: A sample size of at least 165 subjects is required to have a minimum of 80% power for rejecting the null hypothesis that the rate of Grade 3 or lower transections is less than or equal to 87.5% when the expected rate of Grade 3 hemostatic transection is at least 94.0% using exact binomial test and a one-sided significance level of 0.025. This sample size estimation makes the assumption of one transection per subject which is likely very conservative as most subjects may have 2 or more transections during their procedure.Therefore, it is expected that greater than 165 transections will be observed and thus the study will be well powered for evaluating the performance goal hypothesis under the assumption of independence of observations. At the time of the study protocol amendement 3, it was found that on average there were 2 vessel transections per subject.

The assumption of independence and its impact on the overall study conclusion will be evaluated through sensitivity analyses. Given that the primary endpoint is being evaluated intra-operatively, it is not anticipated that there will be subject dropout prior to evaluating this endpoint, thus no adjustment or increase in subject enrollment is planned to account for dropout. A minimum of 33 subjects will be enrolled into each of the five individual procedure subgroups.

The performance goal of 87.5% was selected to represent a minimum acceptable level of clinical performance across all procedure groups. A recent energy study[1] with the same hemostasis endpoint in similar procedure groups showed point estimates of the primary endpoint ranging from 94.2% to 100.0%. The 95% confidence interval for the lowest performing group had a lower bound of 89.3%, thus selection of 87.5% represents a less than 2% region of indifference for maintaining average acceptable performance. In the recent study, average performance across all groups was 96.2% and selection of 87.5% as the performance goal corresponds to a noninferiority margin of 8.7% which is less than the commonly chosen value of 10%. Finally, consideration of the performance goal of 87.5% in the context of the relative reduction from the overall average performance of 96.2%, this can be interpreted as preserving at least 91.0% of the original average treatment effect (87.5/96.2*100% = 91.0%). Maintaining at least 91.0% of the expected effect of similar therapies represents a clinically meaningful metric for maintaining minimum acceptable performance and is actually considerably larger than the standard limits used for bioequivalence where the lower bound for relative therapeutic efficacy is set at 80%,

considerably lower than the 91.0% of therapeutic efficacy that is represented by a performance goal of 87.5% chosen for this trial.

Secondary performance analysis (Vessel size > 5 to7 mm): The hypothesis and evaluation of the hypothesis for the secondary performance endpoint will be similar to the primary performance endopoint. Therefore, a sample size of at least 165 vessels is required to have a minimum of 80% power for rejecting the null hypothesis. The number of vessels transected with a diameter size of > 5-7 mm will be continuously checked throughout the course of the study to achieve the required number of vessels.

Safety Analysis: From a safety perspective on the pooled analysis of 165 subjects and in consideration of rare AEs that may occur (e.g., bleeding requiring blood product transfusion), for an event that has an incidence rate of, for example, 1%, then in a sample of 165 subjects, the probability of observing at least 1 event is 81.0% under a binomial probability model. Thus, this sample size provides a high probability of observing rare events if they do occur, and provides reasonable assurance to conclude that the likelihood of such AEs is less than 2.3% if they do not occur based on the upper limit of an exact 95% confidence interval when 0 events out of 165 subjects are observed.

8 Analyses to be Conducted

8.1 General Conventions

Subject data will be summarized in tables and further details will be provided in listing. All eCRF data will be listed per subject for all subjects. Descriptive statistical analyses will be provided for pre-specified variables. Categorical variables will be summarized descriptively by frequencies and associated percentages. Continuous variables will be summarized descriptively by number of subjects, mean, standard deviation, median, minimum, and maximum.

Data recorded at the nominal visits will be presented in summary tables; however, unscheduled visits will not be included in summary tables. Listings will include both scheduled and unscheduled visits.

Analyses will be conducted using SAS software. During the course of programming of tables that are mocked up in this SAP, minor modifications may become necessary. Examples of these minor modifications include, but are not limited to, re-wording of a footnote, addition of a footnote, re-labeling of a column, or addition or removal of a column from a table or listing. In cases where modifications to tables or listings are not related to a change in statistical analysis methodology or conclusions that could be made on the originally proposed methodology, then no amendment of the SAP is necessary. Any final analyses that differ from what has been specified in this document will be identified within the final statistical output and documented within the clinical study report.

8.2 Disposition of Study Subjects

Subject disposition will be summarized in total, by surgical population (pediatric and adult), surgical procedure group, and procedure subgroups using counts and percentages. The number and percentage of subjects in the AES who failed screening, completed the study and discontinued from the the study will be tabulated along with the specific reasons for discontinuation.

8.3 Demographic, Baseline, and Surgical Characteristics

Summary statistics of subject demographics (age, gender, ethnicity, race and childbearing potential if female) and vital signs (height, weight, and body mass index) will be presented in total, and by surgical population (pediatric and adult), surgical procedure group, and procedure subgroups. Listings will be presented for subject's inclusion/exclusion criteria, demographic, vital sign, and background information.

Background information for pediatric general surgical procedure, and for hepatopancreatobiliary, lower GI, gastric, gynecological, urological, and thoracic adult procedures will be summarized separately. A listing will be provided for background information of each procedure group.

Medical history will be summarized by Medical Dictionary for Regulatory Activities (MedDRA) system organ class and preferred term in total, and by surgical population (pediatric and adult), surgical procedure group, and procedure subgroups. A listing will be presented for medical history. Prior and concomitant medications, surgical history and concomitant procedures will be provided in listing only.

Surgical characteristics including, at minimum, estimated blood loss, requirement for blood transfusion, and procedure duration will be summarized by surgical population (pediatric and adult), surgical procedure group, and procedure subgroups and in total

8.4 Primary and Secondary Endpoints and Associated Hypotheses

The study endpoints are representative of endpoints that are currently reported in the available literature for similar energy devices, and this will allow for comparisons with the results from this study.

8.4.1 Primary Endpoint and Associated Hypotheses

The primary performance endpoint in this study is the number and percentage of vessels where hemostasis (≤ Grade 3) is achieved using the HARMONIC 1100 Shears device. The hemostasis grading scale which was adapted from Siegel et al.[2] is defined as:

- Grade 1: no bleeding at transection site;
- Grade 2: minor bleeding at transection site, no intervention needed;
- Grade 3: minor bleeding at transection site, mild intervention needed, use of compression, basic energy devices (monopolar and/or bipolar device), and/or touch-

ups with HARMONIC 1100 Shears; or

- Grade 4: significant bleeding (e.g., pulsatile blood flow, venous pooling) requiring intervention such as extensive coagulation or use of additional hemostatic measures (e.g., hemoclips, staples, sutures, fibrin sealants, other advanced energy products).

The following hypothesis will be evaluated for the primary performance endpoint:

$$H_0: p \leq 87.5\%$$
$$H_1: p > 87.5\%$$

Where *p* is the percentage of transections achieving a Grade 3 or lower hemostasis rating and 87.5% is set as a performance goal for the lower bound of acceptable hemostasis. A two-sided 95% confidence interval will be calculated for *p* based on the sample proportion of transections where Grade 3 or lower hemostasis was achieved using exact Binomial methods proposed by Clopper-Pearson and the lower limit of this confidence interval will be compared to 87.5% to evaluate the above hypotheses. A p-value will be determined based on an exact binomial test. The null hypothesis will be rejected if the lower limit of the confidence interval is greater than 87.5%. .

The hypothesis and evaluation of hypothesis for the secondary performance endpoint for the proportion of vessels transected of diameter size > 5-7 mm with hemostasis grade 3 or lower will be similar to the primary peformace endpoint as described above.

The primary and secondary performance endpoint will be summarized at the vessel transection level as multiple vessel transections per subject are expected; that is, the denominator for the primary endpoint will be the total number of vessel transections and the numerator will be the number of vessel transections where hemostasis (≤ Grade 3) is achieved. The number and percentage of vessels where hemostasis is achieved will be summarized and an exact two-sided 95% confidence interval will be estimated for each surgical population (pediatric and adult), surgical procedure group, procedure subgroup, the > 5 to 7 mm diameter vessel population, and in total.

8.4.2 Secondary Endpoints and Associated Hypotheses

No formal hypotheses are specified for the secondary endpoints of this study.

Counts and percentages will be provided for the following secondary endpoints:

- Categories of 5-point scale (very dissatisfied, dissatisfied, neither satisfied or dissatisfied, satisfied, or very satisfied) for sealing and transection of lymphatic vessels completed by the HARMOINC 1100 Shears device:
- Categories of hemostasis grading scale (1-4) for all vessel transected
- Type, name, and number of additional hemostasis measures required to achieve hemostasis for Grade 4 vessel transections

8.4.3 Summary of additional key data

Summary statistics will be provided for the following endpoints:

- Procedure duration
- Use of any other energy device (monopolar, traditional bipolar, advanced bipolar, ultrasonic) in primary procedure (type, name, and number of device and reason for use), if applicable
- Surgeon questionnaire administered once per investigator
- Generator questionnaire after each procedure for each GEN11 used
- Surgical procedure conducted
- Hospital stay duration
- Name and number of vessels that were transected, surgeon determination of diameter size range (< 3 mm, 3 to 5 mm, and > 5 to 7 mm)
- Occurrence and location of cancer and occurrence of pre-surgical radiation/chemotherapy within 90 days prior to surgery, if applicable
- Occurrence of vessel skeletonization
- Presence of inflamed tissue, calcified tissues/vessels, atherosclerotic tissue, fibrotic tissue, or presence of adhesions intraoperatively
- Volume of estimated intra-operative blood loss
- Occurrence of blood transfusion, total required units of blood, and time point of transfusion
- Type of additional mild interventions (use of compression, monopolar, bipolar devices, and/or touch-ups with HARMONIC 1100 Shears) required to achieve hemostasis for Grade 3 vessel transections including number of times when HARMONIC 1100 Shears touch-ups were used
- Procedure-related AEs (Possible, probable or casual) and
- Protocol deviation classified as minor or major, rationale for deviation, type of protocol deviation, and outcome of protocol deviation in terms of subject's discontinuation of study

8.5 Safety Analyses

The primary safety endpoint in this study is the occurrence of device-related AEs. As per the study protocol, device-related AEs are those identified as having a relationship of possibly, probably, or causally. AEs unrelated to the study device are recorded as 'not related' on the CRF. Both device-related and procedure-related AEs reported during the study will be coded to MedDRA. All reported AEs will be summarized by MedDRA system organ class and preferred term by surgical population (pediatric and adult), surgical procedure group, procedure subgroup, and in total. Separate summaries will be provided for device-related and procedure-related AEs. Serious AEs will be summarized in a similar manner. In addition, these analyses will also be completed for the vessels in the > 5 to 7 mm diameter size vessel range.

Safety endpoints are as follows:

- All AEs
- All Serious AEs
- All AEs related to the study device
- Serious AEs related to the study device
- All AEs related to the study procedure
- Serious AEs related to the study procedure

Additional key safety outcomes identified in the CER #SCN070380 (ref. Table 40) that includes the bleeding/hemorrhaging and infection (including abscess) will be presented by surgical population (pediatric and adult), surgical procedure group, procedure subgroup, and in total. The observed event rates for these key safety coutomes from the current study will be presented along with the median and range event rates from the CER for descriptive comparison with no statistical test performed.

8.6 Plans for Interim Analysis

No interim analyses are planned for this study.

8.7 Handling of Missing Data

All summaries will be performed only on subjects undergoing the scheduled procedure and only observed data will be summarized. There will be no imputation of data for early terminated subjects or for missing data within the database.

8.8 Sensitivity Analyses

The analysis of the primary performance endpoint described above in Section 8.4.1 makes the assumption of independence of transections within a subject in estimation of variance for the confidence interval calculation. To account for the potential correlation among transections within a subject, a sensitivity analysis will be performed where the 95% confidence interval will be estimated using a generalized linear mixed model. The underlying distribution of the dependent variable in the model is logistic as the outcome variable is binary (i.e., hemostasis achieved or not achieved). A random intercept term as subject-level will be used in the generalized linear mixed model to account for the within subject correlation in achieving hemostasis. The distribution of the random intercept model is assumed to be Gaussian.
If the inference from the mixed model is not consistent with the inference from the primary endpoint analysis as described in Section 8.4.1 then the inference from the mixed model will be used to evaluate success.

Example SAS code:

*create a pseudo variable group so that the 95% CI for intercept can be estimated:

```
 data dat2;
    set dat1;
 group = “pseudo”;
run;
```

```
pro glimmix data= dat2;
class id group;
model outcome(Event ="0") = group / s dist=binary link=logit;
random intercept /subject = id;
lsmeans group/ cl ilink ;  * The results of interest (95% CI of the rate of hemostasis) are
presented on the last two columns;
run;
```

8.9 Subgroup Analysis

Subgroup analyses are planned to be performed for the subgroup of subjects who have a medical history of treatment for cancer (e.g. chemotherapy or radiation) and may be performed for additional groups pending the distributions of baseline demographic or clinical characteristics. These analyses will be exploratory and summary statistics for the procedure-related parameters will be provided for each subgroup.

8.10 Assessment of Site Homogeneity

No summaries or adjustments by study site are planned for this study.

9 Data Monitoring Committee (DMC)

No Data Monitoring Committee was planned or utilized during this study.

10 REFERENCES

1. Schilder J, Anderson D, Shah F. et al. Hemostatic efficacy of an advanced bipolar sealer in open gynecologic, thoracic, and colectomy procedure: a prospective cohort study. Int J Surg Open 2020;24:57-63.
2. Siegel JM, Cummings JF, Clymer JW. Reproducible, repeatable and clinically-relevant hemostasis scoring. J Adv Med Pharm Sci 2014;1:30-39.

Appendix: Table Shells and List of Listings to be Generated

Table shells are provided in a separate document for all summaries to be generated for this study. These shells are a guide to the general layout of data to be presented. Minor modifications can be made to suit existing programs or macros that are available. Additionally, a list of all listings to be created is provided corresponding to the eCRFs that are used in this study. All fields collected will be listed.