CLINICAL PROTOCOL COVER PAGE

A prospective, multi-centered, assessor-blinded clinical performance study to evaluate the sensitivity and specificity of the proposed cell/tissue histopathology image processor cCeLL – Ex vivo (Confocal Fluorescence Endomicroscopy) for intraoperative brain tumor diagnosis.

Sponsor Protocol No.:	cCeLL_BT_2022
Site Protocol No.:	22-VEHSV-01
Version:	1.3
Protocol Date:	6 March 2024
Sponsor:	VPIX Medical Inc.

Protocol Revisions

No.	Version No.	Date	Description of Changes
1	1.0	19 January 2022	N/A
2	1.1 CA	23 MAY 2023	This version is a Canadian specific version. • Addition of ISO 14155 compliant sections • Changes to sample size calculation and statistical methods • Refinement of eligibility criteria • Changes to some secondary endpoints • Addition of normal tissue and margin tissue • Removal of neurosurgeon assessment of images • Change of Canadian Primary Investigator and CRO
3	1.2	July 11, 2023	Number of specimens and basis for calculation, modification of secondary endpoints changed
4	1.3	March 6, 2024	• Correction to study summary: objective and secondary endpoints. • Updated the informed consent section (6.4) to indicate that patients will first be approached for the study by a clinical staff member not part of the research study, and that ICF will be obtained by a study team member not part of the circle of care. • Overall improved consistency of term and symbol use.

Table of Contents

Synopsis

Protocol Title	A prospective, multi-centered, assessor-blinded clinical performance study to evaluate the sensitivity and specificity of the proposed cell/tissue histopathology image processor cCeLL – Ex vivo (Confocal Fluorescence Endomicroscopy) for intraoperative brain tumor diagnosis.
Sponsor	VPIX Medical Inc.
Sites	Korea University Anam Hospital, Seoul, S. Korea Seoul National University Hospital, Seoul, S. Korea Samsung Medical Center, Seoul, S. Korea, Unity Health- St. Michael's Hospital, Toronto, ON, Canada
Coordinating Investigator	Shin-Hyuk Kang MD M.M.Sc Ph.D Korea University College of Medicine
Study Objective	The purpose of this clinical study is to confirm sensitivity and specificity of the cCeLL – Ex vivo in intraoperative brain tumor diagnosis as compared to analysis with frozen section.
Background	○ The existing workflow for intraoperative brain tumor diagnosis involves tissue biopsy, processing, analysis and confirmation by a pathologist using a frozen section, and relaying the result back to the surgeon. ○ The current intraoperative diagnosis based on frozen section analysis is time-, resource-, and labor-intensive. The process takes over 40 minutes on average, but may take longer if biopsied specimen is insufficient or requires repeated analysis. Thereby the development of an efficient device that allows intraoperative real-time diagnosis is enforced. ○ The "Digital Biopsy" imaging method, using micro-sized confocal laser endomicroscopy, is an emerging technology that enables intraoperative visualization of real-time high-resolution brain tumor tissue images. ○ Neoplastic brain tissue biopsied from participant will be visualized and images will be captured using cCeLL – Ex vivo. The corresponding biopsy samples will be examined with standard frozen-section analysis (H&E slide), to evaluate the sensitivity and specificity of cCeLL – Ex vivo in the diagnosis of brain tumors. This study will also be able to verify the potential of Confocal Endomiicroscopy to replace the current standard frozen-section analysis, in intraoperative brain tumor diagnosis.
Study Design	Prospective, multi-centered, assessor-blinded, clinical performance study
Sample Size	The sample size refers to the number of analyzable tissue samples collected from participants. There may be multiple samples collected from each participant. As it is expected that the 30% of tissue samples will be non-analyzable, the collection of tissue samples will be larger than the target sample size. The total samples to be collected will be up to 358 samples (286 cancer samples, 72 normal samples) - Korea University Anam Hospital: 51 samples (cancer: 42 samples, normal: 9 samples)

<table>
<tr><td></td><td>
- Seoul National University Hospital: 82 samples (cancer: 72 samples, normal: 10 samples)
- Samsung Medical Center: 82 samples (cancer: 72 samples, normal: 10 samples)
- St.Michael's Hospital, Unity Health: 143 samples collected from a total of 65 participants (cancer: 100 samples, normal: 43 samples)

After excluding the non-analyzable tissue samples, we expect to conclude the study with a total of 250 samples (200 samples for cancer, 50 samples for normal)

-Korea University Anam Hospital: 36 samples (cancer: 30 samples, normal: 6 samples)

-Seoul National University Hospital: 57 samples (cancer: 50 samples, normal: 7 samples)

-Samsung Medical Center: 57 samples (cancer: 50 samples, normal: 7 samples)

-St.Michael's Hospital, Unity Health: 100 samples collected from a total of 50 participants (cancer: 70 samples, normal: 30 samples)
</td></tr>
<tr><td>Basis for Calculating the Number of Acquired Images</td><td>
The data for agreement, sensitivity and specificity can be summarized as in the table below. For agreement, the terms agree and disagree are with respect to each other. For sensitivity and specificity, the terms agree and disagree are with respect to truth. So for sensitivity, truth is tumor and for specificity truth is not tumor.

Agreement Table

	cC		
FS Results	Agree	DisAgree	
Agree	a	b	a+b
Disagree	c	d	c+d
	a+c	b+d	n_T

The estimate of agreement is pAG = (a+d)/n_T. It is desired to test the hypothesis that the agreement is greater than 80%. This is compatible with the expected result that both the sensitivity and the specificity will be about 90%. Thus the hypothesis to be tested is:

$$H_0: \pi_{AG} = 80\% \text{ vs } H_A: \pi_{AG} > 80\%$$

Where π_{AG} is the percent agreement between Frozen Section and cCeLL. This will be tested utilizing the exact binomial test. Estimates of agreement from the pilot study indicate that the agreement will be greater than 86%. In this study, there will be 250 subjects available for analysis for the estimation and testing of agreement. For outcome results, if the agreement is 86%, with the sample size of 250, the probability the estimated agreement will be greater than 80% is greater than 90%.
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Sensitivity Table – This is only tabulated when the truth standard is positive for tumor.

Agreement Table with Tumor Truth

	cC		
FS Results	Agree w/Truth	DisAgree w/Truth	
Agree	e	F	e+f
Disagree	g	H	g+h
	e+g	f+h	n_1

The sensitivity of the Frozen Section is given by $p_{SenF} = (e+f)/n_1$ and the sensitivity of cCeLL is given by $p_{SenC}=(e+g)/n_1$. It is desired to demonstrate that the sensitivity of cCeLL is non inferior to that of Frozen section.

The non inferiority of the sensitivity of cCeLL result versus the sensitivity of the Frozen Section is given as:

$$H_0: \pi_{SenF} - \pi_{SenC} > \Delta \text{ versus } H_A: \pi_{SenF} - \pi_{SenC} \leq \Delta$$

Here π_{SenF} is the true sensitivity for Frozen Section and π_{SenC} is the true sensitivity for cCell. Where Δ is the non inferiority limit. In this study, it is proposed to utilize a Δ=0.05 (5%). If that Δ is used and the anticipated sensitivity is 90%, then with a sample size of 200 tumor positive samples the probability the difference between the estimated sensitivities is greater than 5% is greater than 90%.

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<tr><td>Eligibility Criteria</td><td>

Inclusion Criteria

The biological specimens (hereafter referred to as 'Tissue') and/or images to be used in this trial must be collected from patients who satisfy all of the following inclusion criteria

1. Male or female patients ≥19 years of age;
2. Patient is suspected to have a brain tumor and has been scheduled for neurosurgery with a potential tumor resection;
3. Patient is willing and able to provide informed consent and understand its content.

Exclusion Criteria

Tissue and/or images to be used in this trial must be collected from patients who do not meet any of the following exclusion criteria:

1. Patients has undergone several surgeries on the lesion of interest.

</td></tr>
<tr><td>Study Device</td><td>cCeLL - Ex vivo (Clearance No. 21-311)</td></tr>
<tr><td>Study Duration</td><td>18 months from the start date of the study</td></tr>
<tr><td>Study Procedure</td><td>1) Samples will be collected from consented participants who have satisfied the eligibility criteria.</td></tr>
</table>

2) Tissue will be removed as part of the standard neurosurgical procedure. Normal tissue obtained from inevitable resection during the course of surgery. There will be no deviation from normal surgical practice to aquire tissue samples.
3) Image recordings of sample tissue using cCeLL – Ex vivo are taken and the obtained data is stored (CC).
4) cCeLL – Ex vivo image data of the collected samples is sent to the central core.
5) The result of frozen section analysis (FS) will be collected from the pathology department of each site.
6) The result of permanent section analysis (PS) of corresponding paraffin-embedded tissue section (PS) will be collected from the pathology department of each site. PS will serve as a reference standard to the analysis result of CC and FS.
7) An interpreting pathologist will analyze given images (CC-H) and is not allowed to review the reference standards (PS) until all interpretations have been completed.
8) The cCeLL – Ex vivo image data will be anonymized such that only the unique reference numbers will be displayed in the images provided, and participant information including participant enrollment number and surgery information will be removed.
9) The types of the images obtained from this procedure are as follows:

cCeLL – Ex vivo will capture image data of a resected tissue, prior to frozen sectioning (FS) and permanent sectioning (PS). The biopsied samples can be used to evaluate the presence of tumors, and the tumor types.

Method of analysis	**Image data**	**Reference standard**	**Result of analysis by a pathologist**
cCeLL – Ex vivo	CC	PS	CC-H
Permanent section	PS	PS	
Frozen Section	FS	PS	

CC : Analysis of images obtained by cCeLL
FS : Frozen section analysis
PS : Permanent section analysis
H : Pathologist

10) Data will be analyzed by a statistician. Sensitivity and specificity will be calculated as follows:

	- The probability of interpretation results by the interpreting physician (1 pathologist) being positive given that the reference standards is positive - The probability of interpretation results by the interpreting physician (1 pathologist) being negative given that the reference standards is negative
Study Endpoints	**Primary Endpoint** The primary endpoints in this study are the agreement between cCeLL Ex vivo and frozen section, the sensitivity of cCeLL ex vivo and frozen section, and the specificity of cCeLL ex vivo and frozen section. These endpoints will be assessed by a single pathologist in the analysis of tumor tissue. **Secondary Endpoints** 1. Comparison of time taken in tissue scanning, visualization, and interpretation using cCeLL – Ex vivo to time taken in frozen section analysis. 2. Accuracy in determining the tumor type 3. Required image counts for adequate analysis and diagnosis using cCeLL – Ex vivo 4. Non-diagnostic Image 5. AUC value
Analysis Methods	**Primary Endpoints** Comparison of the sensitivity/specificity of cCeLL-Ex vivo reading results, and the sensitivity/specificity of the frozen section reading results in one pathological inquiry for brain tumor tissue • Sensitivity of cCeLL-Ex vivo reading result in 1 patient in pathological inquiry for brain tumor tissue: Probability that PS is positive if CC-H positive • Specificity of cCeLL-Ex vivo reading result in 1 patient in pathological inquiry for brain tumor tissue: Probability that PS is negative if CC-H is negative • Sensitivity of frozen section reading results for brain tumor tissue: Probability that FS is positive if PS is positive • Specificity of frozen section reading results from brain tumor tissue: Probability that PS is negative if FS is negative The ratio of the clinical sensitivity/specificity of a cCeLL Ex-vivo image and the sensitivity/specificity of the frozen section image of 1 patient in the pathological inquiry for the total reading results is presented, respectively. To identify a 95 % bilateral confidence interval for the difference between cCeLL- Ex vivo and the frozen section sensitivity/specificity. The statistical significance of the difference between cCeLL-Ex vivo and the frozen section sensitivity/specificity ratio is tested by the McNemar test. **Secondary Endpoints** 1. **Comparison of cCeLL-Ex vivo imaging and reading time with frozen section reading time** i. The cCeLL-Ex vivo imaging time is measured and recorded in minutes from the time the fluorescent dye is applied to the specimen for image acquisition

	ii. The cCeLL-Ex vivo reading time by the pathologist is measured and recorded in minutes from the time of capturing the cCeLL-Ex vivo image of each brain tumor to the time of the final reading. iii. The imaging and reading time for frozen section is measured in minutes from the time of resecting the specimen undergoing frozen section in the operating room to the time when the pathology department of each institution obtains a frozen section image and reports the findings. iv. Format: ___Hours ___Minutes 2. **Comparison of cCeLL accuracy of evaluating the tumor types via Ex-vivo imaging with the accuracy using frozen section** i. Evaluation of tumor types via cCeLL-Ex vivo images by 1 pathologist : Agreement between the result of CC-H and the result of PS ii. Evaluation of tumor types via frozen section: Agreement between the result of FS and the result of PS 3. **cCeLL – Time and number of images required to diagnose Ex vivo imaging** i. The number of images required for the pathologist to read the brain tumor cCeLL – Ex vivo image will be measured and averaged. ii. cCeLL – Ex vivo reading time uses that of secondary endpoint 1. iii. *Average number of images required for cCeLL – Ex vivo image reading of 1 brain tumor specimen: __ _ _ 4. **Non-diagnostic image** i. The total number of cCeLL Ex-vivo images among the images confirmed in the vivo reading, and the proportion of the number of images that cannot be read are stated as a percentage (%). ii. The number of images that cannot be read is recorded when the reader reads it. 5. **AUC value** i. Based on the results of each endpoint, the sensitivity and false positive rate (1-specificity) are used to draw the ROC (Receiver Operating Characteristic) Curve to evaluate the performance of the diagnosis to distinguish between positive and negative, and the accuracy of the diagnosis is evaluated through the AUC (Area Under the Curve) value obtained from the area below the ROC curve.

List of Abbreviations and Terms

ADE	Adverse Device Effect
AE	Adverse Event
AUC	Area under the curve
CC	Analysis of images obtained by cCeLL
eCRF	Electronic Case Report Form
DD	Device Deficiency
FAS	Full Analysis Set
FS	Frozen section analysis
GCP	Good Clinical Practice
ICF	Informed Consent Form
ISO	International Organization for Standardization
ITT	Intent-To-Treat
LOCF	Last Observation Carried Forward
MAST	Multiple Allergen Simultaneous Test
PBS	Phosphate Buffered Saline
PP	Per Protocol
PS	Permanent section analysis
REB	Research Ethics Board
SAE	Serious Adverse Event
SADE	Serious Adverse Device Effect
SOC	System Organ Class
SOP	Standard Operating Procedure
UADE	Unexpected Adverse Device Effect

1. Study Title

A prospective, multi-centered, assessor-blinded clinical performance study to assess the sensitivity and specificity of the proposed cell/tissue histopathology image processor cCeLL – Ex vivo (Confocal Fluorescence Endomicroscopy) in diagnostic yield during brain tumor surgery

2. Study Organization

2.1. Study Sites

Study Site	Location
Korea University Anam Hospital	73 Goryeodae-ro, Seongbuk-gu, Seoul, Republic of Korea
Seoul National University Hospital	101 Daehak-ro, Jongno-gu, Seoul, Republic of Korea
Samsung Medical Center	81 Irwon-ro, Gangnam-gu, Seoul, Republic of Korea
Unity Health- St. Michael's Hospital	36 Queen St E, Toronto, ON Canada **The Sponsor will obtain Health Canada authorization for an Investigational Testing application for this site..*

2.2. Sponsor

Company	CEO	Location	Phone
VPIX Medical Inc.	Kyungmin Hwang	2F, 774 Gyeryong-ro, Jung-gu, Daejeon, Republic of Korea	010-4366-3519

3. Background and Purpose of the Investigation

3.1 Study Objective

The purpose of this clinical study is to confirm sensitivity and specificity of the cCeLL – Ex vivo in intraoperative brain tumor diagnosis as compared to analysis with frozen section.

3.2 Background

Cancer is a leading cause of death, accounting for nearly 1 out of 6 death worldwide (Ferlay et al., 2020; de Martel et al., 2020; WHO, 2020).

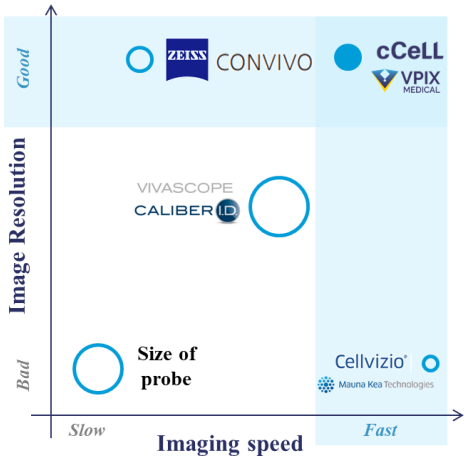
The existing workflow for intraoperative brain tumor diagnosis involves tissue biopsy, processing, analysis and confirmation by a pathologist using frozen section, and relaying the result back to the surgeon. The current intraoperative diagnosis based on frozen section analysis is time-, resource-, and labor-intensive. The process takes over 40 minutes on average but may

take longer if biopsied specimen is insufficient or requires repeated analysis. Thereby the development of an efficient device that allows intraoperative real-time diagnosis is enforced.

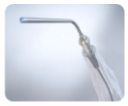
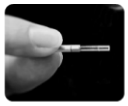
Confocal Laser Endomicroscopy is a promising diagnostic tool to replace current standard 'frozen section analysis' by allowing intraoperative real-time high-resolution visualization of tissue of interest. Confocal Fluorescence Endomicroscopy follows the general principles of fluorescence, which is the characteristic of fluorescence-stained specimen ('tissue') that absorb light of short-wavelength and emits light of long-wavelength. This medical device visualizes the intensity of light emitted from a specimen which varies according to the specimen's degree of light absorbance (White et al., 1987; Amos and White, 2003).

When the probe of the device, which contains optical fibers, is placed in contact with the specimen or close to within several hundred ㎛ of the specimen, the optical signal generated from the main body gets transmitted to the specimen through the probe and then absorbed by the specimen. The specimen then gets activated and emits fluorescent light, and this fluorescent light returns to the main body of the device through the optical fibers of the probe. The mechanism behind optical fibers is that light incident at a specific angle proceeds to total reflection due to the inside and outside of the optical fibers each being made with differing densities and refractive indices. The optical signal which returned to the main body through the optical fibers is detected as an electronic signal by the photodetector in the main body. The electric signal is then converted to a digital signal by the converter inside the main body of the device, and the digital signal is then processed by the PC through its software and exported as an image onto the monitor.

Several global medical device companies have developed promising digital-biopsy devices based on confocal microscopy for past few years and commercialized micro-probe-based optical fusion imaging devices that can be placed in contact with the patient's organs directly. However, technological barrier in micro-miniaturization of probe still exist as seen in marketed devices, which limits its use in various human organ systems›. These limitations are observed as the probe being too large for intraoperative use (Vivascope, USA) (Mavig GmbH, 2019), limited number of optic fiber bundles leading to inadequate resolution of images for pathologic diagnosis (Cellvizio, France) (Mauna Kea Technologies, 2021), or difficulty in compensating body movements for respiratory/circulatory activities due to limited laser scanning speed of the probe (Convivo, Germany) (Carl Zeiss Meditec AG, 2019). The proposed investigational device designed with an innovative optic probe and image-processing technology to enable detection and capture of high-resolution images whilst in operating room.





Device Name / Manufacturing Country	Caliber I.D. (U.S.A)	Mauna Kea Technologies (France)	Carl Zeiss (Germany)	VPIX Medical, Inc. (S.Korea)
	Vivascope	Cellvizio Series	Convivo	cCeLL™ Series
Target Organ System	Skin	Gastrointestinal (GI) Tract	Brain	Various Organ System
Price	$$	$$	$$$	$~$$
Micro-miniaturization	Poor	Advanced	Advanced	**Advanced**
Image Resolution	Intermediate	Poor	Advanced	**Advanced**
Image Scanning Speed	Intermediate	Advanced	Poor	**Advanced**
Image Size Adjustability	Available	Not available	Not available	**Available**

Anticipated Economic/Industrial Impact and Advancement – Establishment of Digital Histopathology System

① cCeLL – Ex vivo allows rapid scanning, capturing, and storing of high-resolution images of biopsy specimen, without undergoing lengthy standard procedure, as a complete Digital Histopathology System. Hospitals are suffering from the increased burden in storing pathology tissue slides, hence in process of converting originals into digital slides and storing into digital image repository.

② cCeLL – Ex vivo enables capturing of digital images without producing physical tissue slides, which is a labor-,time- and cost-intensive process, as well potentially reduces carbon emission. As the digital images created by cCeLL – Ex vivo are saved as electronic files without requiring utilization of separate viewer system, it allows efficient data management and accelerates conversion rate and smooth adaptation into digital histopathology system.

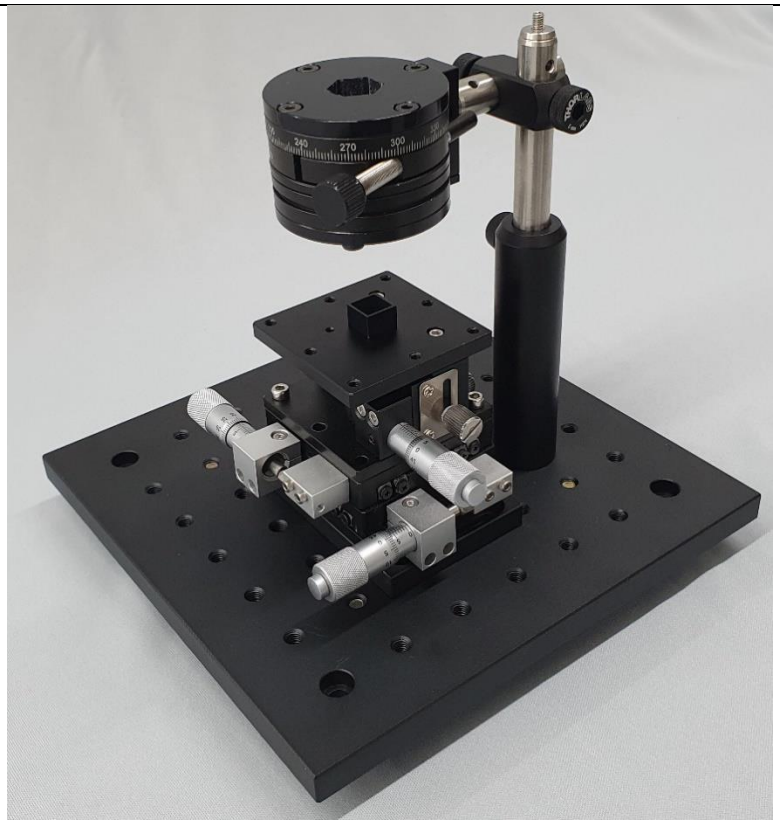
4. Investigational Device

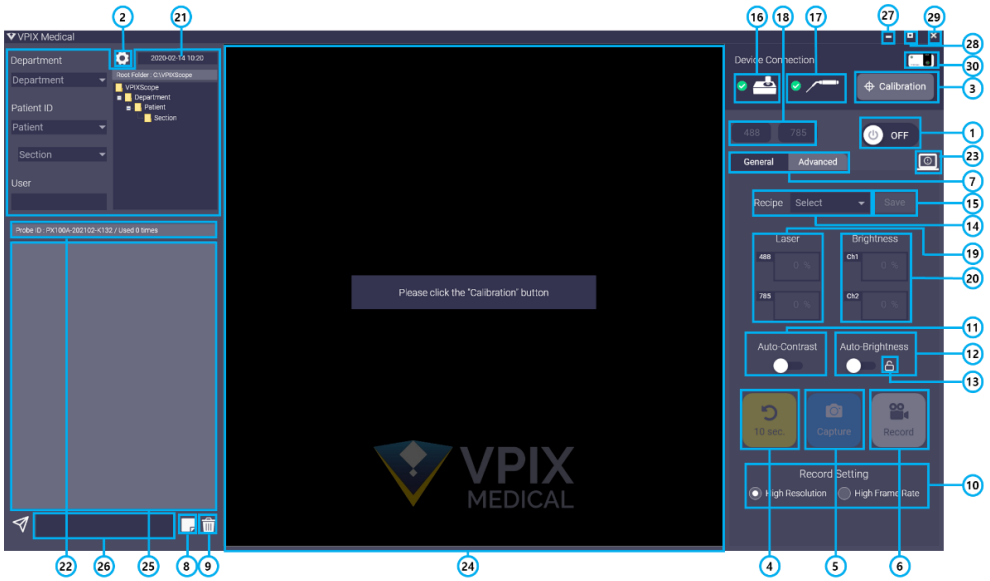
4.1 Device Standard

Item	Specification		
Electrical Specification	Maximum power consumption at the maximum output		300VA
	Electrical rating input		100-240VAC, 50/60Hz
	Protection degree for electric shock		Class 1 equipment
Performance specification	Laser	Output wavelength	1. CX100-2W2C488775: 480- 492 nm, 770- 785 nm 2. CX100-1W1C488: 480-492 nm 3. CX100-1W1C785: 770 -785 nm
		Laser Class	3R
		Output power	1. CX100-2W2C488775 a. 488 nm: max. 2.5 mW b. 775 nm: max. 3.5 mW 2. CX100-1W1C488: max. 2.5 mW 3. CX100-1W1C785: max. 3.5 mW
		Operation mode	Continuous mode
		Laser medium	Laser diode (semiconductor)
	Imaging	Field-of-View (FOV)	500μm(±10%) x 500μm(±10%)
		Frame count per second	2 - 10fps
Storage and Maintenance	In-Use	Temperature	15℃ - 35℃
		Humidity	30% - 75%
	Storage	Temperature	-10℃ - 50℃
		Humidity	10% - 90%

4.2 Appearance of the Device

Item	Description
Main device	
Probe	

Stage	 

Main screen (Software)	

4.3 Operating Mechanism

The investigational medical device visualizes cells of a biological specimen which has been resected from the body and to which dye has been applied by measuring the intensity of the fluorescence signal that is generated by shooting a laser onto the specimen. The device is capable of capturing images of regions several μm wide of a specimen at depth of several hundreds of μm, and visualizing them in 2D through its complementary software. Confocal Fluorescence Endomicroscopy follows the general principles of fluorescence. Fluorescence refers to a phenomenon in which a fluorescent object emits light of long wavelength by absorbing light of short wavelength. This device visualizes the intensity of light emitted from a specimen which varies according to the specimen's degree of light absorbance.

When its probe, which contains optical fibers, is placed in contact with the specimen or close to within several to several hundred μm of the specimen, the optical signal generated from the main body gets transmitted to the specimen through the probe and then absorbed by the specimen. The specimen then is activated and emits fluorescent light. This fluorescent light returns to the main body of the device through the optical fibers of the probe. The mechanism behind optical fibers is that light incident at a specific angle proceeds to total reflection due to the inside and outside of the optical fibers each being made with differing densities and refractive indices. The optical signal which returned to the main body through the optical fibers is detected as an electronic signal by the photodetector in the main body. The electric signal is then converted to a digital signal by the converter inside the main body of the device, and the digital signal is then processed by the PC through its software and exported as an image onto the monitor.

4.4 Intended Use

This investigational medical device is a diagnostic tool that aims to provide rapid and precise intraoperative assessment of biopsied neoplastic tissue specimen from a brain tumor. By enabling the fluorescence staining, visualizing, capturing and storing image data while in the operation room, standard time-consuming and labor-intensive preparation of specimen sectioning may not be necessary.

4.5 Target Health Condition

Benign/Malignant Brain Tumor

4.6 Device Accountability

4.6.1 Labeling

The investigational medical device will be labelled according to the requirements of ICH-GCP, ISO 14155, and all other applicable regulatory guidelines. The device label will contain the following information in required official languages (i.e. English and French in Canada; Korean in Korea):

a. Name of the device
b. Name and address of manufacturer
c. Identifier of the device
d. Package content
e. Expiry date
f. Specification (as necessary for proper use)
g. Directions for use
h. Warning and cautions
i. Contraindications
j. Storage condition

All investigational devices used in Canada will be labelled “Investigational Device” and “To be Used by Qualified Investigators Only” and “Instrument de recherche” and “Réservé uniquement à l’usage de chercheurs compétents” accordingly to Health Canada regulations.

4.6.2 Maintenance, Handling, and Record Keeping

The investigational medical device will be provided by Sponsor only after obtaining written approval/authorization from the REB and regulatory authorities. Access to investigational devices shall be controlled and the investigational devices shall be used only in the clinical investigation and according to the study plan. Device accountability will be maintained and will document the physical location of all investigational devices from shipment of investigational devices to the investigation sites until return or disposal. The Sponsor must document and retain all records related to device manufacture, shipping, receipt, return, or disposition. The investigator/investigator’s designee will acknowledge receipt upon reception.

Sponsor or Sponsor's designee must identify and document the SOP procedures for the return of investigational device upon study completion or in case of issue related to device.

The Sponsor or manufacturer must not provide or dispense any parts of investigational medical device to any other study site or investigator. The device must not be used other than for the intended use specified in the trial protocol.

4.6.3 Receiving, Storage, and Return

Device accountability will be maintained and will document the physical location of all investigational devices from shipment of investigational devices to the investigation sites until return or disposal. The principal investigator or an authorized designee shall keep records documenting the following:

a. name(s) of person(s) who received, used, returned, or disposed of the device;

b. the date of receipt, identification, and quantity of each investigational device (lot number);

c. the expiry date, if applicable;

d. the date or dates of use;

e. the date on which the investigational device was returned;

f. the date of return of unused, expired, or malfunctioning investigational devices, if applicable.

A site monitor will review the inventory and accountability log during site visits and at the completion of the study. If the study is completed, prematurely terminated, or suspended, the Sponsor shall promptly inform the investigators, the REB, the regulatory authorities of the reason for termination or suspension, as specified by the applicable regulatory requirements; and the investigational medical device will be return to the Sponsor or alternative disposition of used/unused products as per request by the Sponsor. The investigational medical device is intended for use by qualified investigators only.

5. Inclusion and Exclusion Criteria and Sample Size Determination

5.1 Inclusion Criteria

The biological specimens (hereafter referred to as 'tissue') and/or images to be used in this trial must be collected from patients who satisfy all of the following inclusion criteria:

1. Male or female patients ≥ 19 years of age.
2. Patient is suspected to have a brain tumor and has been scheduled for neurosurgery with a potential tumor resection.
3. Patient is willing and able to provide informed consent and understand its content.

5.2 Exclusion Criteria

Tissue and/or images to be used in this trial must be collected from patients who do not meet any of the following exclusion criteria:

1. Patient has undergone several surgeries on lesion of interest.

5.3 Tissue Eligibility

Eligible tissue will meet all of the following:

1. Tissue from an eligible and consented patient at a participating site
2. Any normal tissue obtained from inevitable resection during standard surgery

5.4 Sample Size

The sample size refers to the number of analyzable tissue samples collected from participants. There may be multiple samples collected from each participant.

There will be 250 analyzable tissue samples required (200 tumor positive and 50 normal) for this study.

5.4.1 Number of Specimens

A total of 250 analyzable samples (200 samples for cancer, 50 samples for normal) are required.

- Korea University Anam Hospital: 36 samples (cancer: 30 samples, normal: 6 samples)
- Seoul National University Hospital: 57 samples (cancer: 50 samples, normal: 7 samples)
- Samsung Medical Center: 57 samples (cancer: 50 samples, normal: 7 samples)
- St.Michael's Hospital, Unity Health: 100 samples collected from a total of 65 participants (cancer: 70 samples, normal: 30 samples)

With an expected dropout rate of 30%, the number of samples to be collected is 358 samples (286 cancer samples, 72 normal samples)

- Korea University Anam Hospital: 51 samples (cancer: 42 samples, normal: 9 samples)
- Seoul National University Hospital: 82 samples (cancer: 72 samples, normal: 10 samples)
- Samsung Medical Center: 82 samples (cancer: 72 samples, normal: 10 samples)
- St.Michael's Hospital, Unity Health: 143 samples collected from a total of 50 participants (cancer: 100 samples, normal: 43 samples)

5.5 Study Duration

It is expected that the study will run for about 18 months in total from the time the study sites are activated. Enrollment is expected to 8 months in Korea and up to 12 months in Canada. There will be additional time for data collection, data cleaning and analysis.

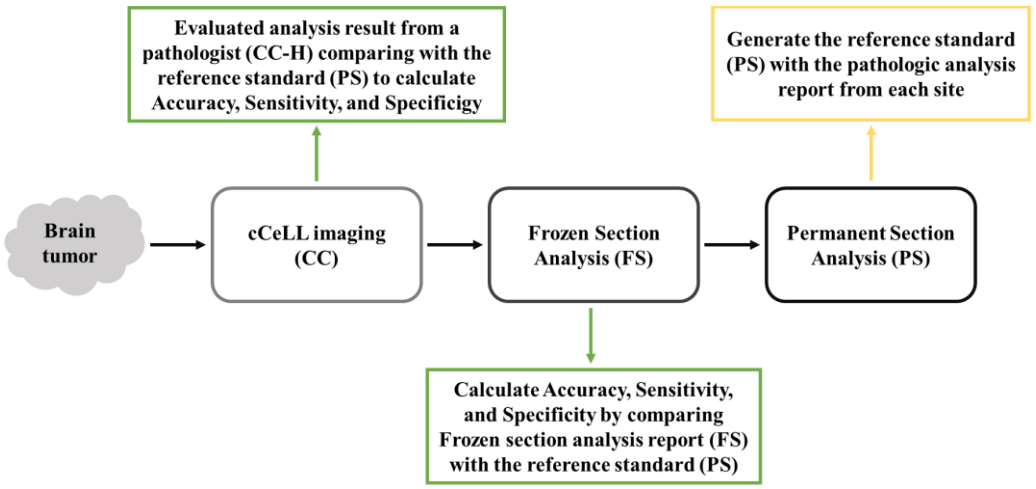
6. Study Procedures

6.1 Study Design

Prospective, multi-center, assessor-blinded clinical study. Samples will be paired and compared against the current standard of assessment.

6.2 Study Assessments and Procedures

There will be up to three (3) tissue sample types assessed for each participant: 1- center-of-tumor, 2- normal tissue (collected from inevitable standard resection), and 3- margin tissue. Depending on each participant's circumstance, the number of sample tissues harvested will differ from minimum of one to maximum of three. All tissue samples will be assessed with the investigative device and compared against frozen section analysis.



1. Samples will be collected from consented participants who have satisfied the eligibility criteria.
2. Tissues will be removed as part of the standard neurosurgical procedure. Normal tissues obtained from inevitable resection during the course of surgery.
3. Collected samples will be processed as follows:
    1. Wash the biopsied tissue with Phosphate Buffered Saline (PBS)
    2. Place the tissue on a glass slide
    3. Apply dye onto the tissue
    4. After 1 minute, wipe on gauze and then scan (i.e., cCeLL imaging) the tissue using Pixecton (probe) connected to cCeLL – Ex vivo
    5. Send the processed tissue to pathology to obtain images in the form for frozen sections analysis (FS)/permanent sections analysis (PS)
4. Image recordings of sample tissue using cCeLL – Ex vivo are taken and the obtained data is stored (CC).
5. cCeLL – Ex vivo image data of the collected samples is sent to the central core.
6. The result of frozen section analysis (FS) will be collected from the pathology department of each site.
7. The result of permanent section analysis (PS) of corresponding paraffin-embedded tissue section (PS) will be collected from the pathology department of each site. PS will serve as a reference standard to the analysis result of CC and FS.
8. An interpreting pathologist will analyze given images (CC) and is not allowed to review the reference standards (PS).
9. The cCeLL – Ex vivo image data will be de-identified such that only the unique reference numbers will be displayed in the images provided, and participant information including participant enrollment number and surgery information will be removed.
10. The types of the images obtained from this procedure are as follows:

cCeLL – Ex vivo will capture image data of a resected tissue, prior to frozen sectioning (FS) and permanent sectioning (PS). The sample is used to evaluate the presence or absence of tumors, and also to evaluate the tumor types.

Method of analysis	**Image data**	**Reference standard**	**Result of analysis by a pathologist 1**
cCeLL – Ex vivo	CC	PS	CC-H
Permanent section	PS	PS	
Frozen Section	FS	PS	

CC : Analysis of images obtained by cCeLL
FS : Frozen section analysis
PS : Permanent section analysis
H : Pathologist

6.3 Sample Image Number and Blinding

6.3.1 Participant Number Assignment

Participant No. will be assigned to participants satisfy the eligibility criteria and are included in the study.

The following rules will apply to generating Participant No:

Participant No.
R - __ __ - __ __ __
2 letter site acronym, 3-digit sequence (e.g., R-KO-001, R-KO-002)

The following rules will apply to generating Specimen No.:

Specimen No.
Participant No. - __ - __
1 letter specimen classification, 1-digit sequence (e.g., R-KO-001-C-1, R-KO-001-P-1, R-KO-001-N-1)

Each selected image will be assigned an enrollment number in a sequential manner. The investigator will register the images by entering such information as the participant number, image type, and order.

6.3.2 Reference Standard and Analysis Number Assignment

Reference standard and analysis No. will be assigned according to type as follows:

<Reference standard No.>

- Permanent section analysis (PS): Participant number – Specimen number – permanent section (e.g., R-KO-001-C-PS-01)

<Frozen section No.>

- Frozen section analysis (FS): Participant number – Specimen number – Frozen section (e.g., R-KO-001-C-FS-01)

<cCeLL image analysis No.>

- Pathologist 1 analysis of cCeLL image: Participant number – Specimen number – cCeLL – pathologist (e.g., R-KO-001-C-01-CC-H)

6.4 Informed Consent

Informed consent must be obtained prior to conducting any study-related assessments. A clinical staff member within the patient's circle of care (known to the patient), who is not part of the research team, will make first contact with potential patients to assess whether or not they are interested in learning more about the study. Then, they will contact a research study team member (not part of the patient circle of care) to discuss the study with the patient and obtain informed consent to participate in the study.

The research staff (not part of the patient's circle of care) will explain the clinical study to each potential participant, including all aspects of the clinical study that are relevant to the participant's decision to participate throughout the clinical study including, but not limited to, the following: purpose and nature of the study, study procedures, expected study duration, available alternative therapies, and the benefits and risks involved with study participation and the potential treatment. They shall avoid any coercion or undue improper influence on, or inducement of, the participant to participate and will not waive or appear to waive the participant's legal rights. Participants will be given a copy of the informed consent form and will be provided ample time to read and understand the document and the opportunity to ask questions. Participants will be informed of their right to withdraw from the study at any time without prejudice; consent forms will use local non-technical language and be provided in a language understandable to the participant. After this explanation, and before any study-specific procedures have been performed, the participant and the PI's qualified designee (a member of the study team outside the patient's circle of care), responsible for conducting the informed consent process will voluntarily sign and personally date the informed consent form. Prior to participation in the study, the participant will receive a copy of the signed and dated written informed consent and any other written information provided to the participant.

6.5 Demographics and Anthropometric Profile

Demographic information relating to the participant's age and sex will be recorded after enrollment.

6.6 Timing Data

Start and stop times and other similar data will be collected to determine the processing duration for each test from start to finish.

6.7 Assessment Data

Test results will be collected and compared from all three (3) modalities (cCell, FS, PS). Blinded assessments of cCell images will be documented from the site pathologist for each analyzable image.

7. Risks and Procedures to Minimize Risk

7.1 Anticipated Risks

The investigational medical device used in this prospective study is an ex-vivo diagnostic device which has no direct or indirect effect on the participant, and therefore there are no anticipated risks to be identified. Standard of care diagnostic images will be used to make clinical decisions in this study population.

7.2 Precautions of Use

7.2.1 General Precautions

1. To be used for ex-vivo diagnostic purposes only (for trained professionals only).
2. To be used by a trained professional only.
3. The user must be fully aware of the user instructions prior to use.
4. To be used with the parts and cables provided in accordance to the user instructions manual.
5. Do not block the vent of the device.
6. Not to be dissembled, repaired, or modified.
7. If device failure or abnormal behavior is suspected, stop using the device immediately and contact the head office.

7.2.2 Installation Precautions

1. Not to be used with cables other than the ones provided with the device.
2. Do not turn on the device until installation is complete.
3. Do not block the cooling fan of the device.
4. Never use with ungrounded wall outlet.
5. To be installed on flat surfaces only.

7.2.3 Precautions Before Device Use

1. Become fully aware of the device operation manual before use.
2. Inspect all parts before use.

7.2.4 Precautions During Device In-Use

1. Never directly stare at the laser emitting part of the device.
2. Do not use this device in places where combustible steam or explosive gases and chemicals are present or stored.
3. If issues not listed in the user instruction manual have occurred, do not attempt to resolve them on your own; instead contact the head office.
4. Keep this device and other electronic devices away from liquids.
5. Do not bend, twist or shock the probe

6. Be careful not to touch the laser emitting part of the probe with bare hand as this will contaminate the lens.

7.2.5 Laser Safety

1. Do not expose yourself to the laser of this device.
2. Never directly stare at the laser emitting end of the probe.

7.2.6 Maintenance Precautions

1. Back up data on a regular basis to avoid data loss.
2. Update the software on a regular basis.
3. Leave the device turned off during moving, washing, or maintenance.

8. Study Evaluation

8.1 Primary Endpoint

The primary endpoints in this study are the agreement between cCeLL Ex vivo and frozen section, the sensitivity of cCeLL ex vivo and frozen section, and the specificity of cCeLL ex vivo and frozen section. These endpoints will be assessed by a single pathologist in the analysis of tumor tissue.

8.2 Secondary Endpoints

1. Comparison of time taken in tissue scanning, visualization, and interpretation using cCeLL – Ex vivo to time taken in frozen section analysis.
2. Accuracy in determining the tumor type
3. Required image counts for adequate analysis and diagnosis using cCeLL – Ex vivo
4. Non-diagnostic Image
5. AUC value

8.3 Statistical Analysis and Interpretation

8.3.1 Methods and Sample Size

The data for agreement, sensitivity and specificity can be summarized as in the table below. For agreement, the terms agree and disagree are with respect to each other. For sensitivity and specificity, the terms agree and disagree are with respect to truth. So for sensitivity, truth is tumor and for specificity truth is not tumor. It should also be noted that since the total sample size was chosen to be 250, no power calculations were done; however, a probability calculation that the estimated parameter is greater than the null hypothesis was presented.

Agreement Table

	cC		
FS Results	Agree	DisAgree	
Agree	a	b	a+b
Disagree	c	d	c+d
	a+c	b+d	n_T

The estimate of agreement is $p_{AG} = (a+d)/n_T$. It is desired to test the hypothesis that the agreement is greater than 80%. This is compatible with the expected result that both the sensitivity and the specificity will be about 90%. Thus the hypothesis to be tested is that the agreement is 80% (H0: Null hypothesis) versus the alternative hypothesis that the agreement is greater than 80% (HA: Alternative hypothesis).

$$H_0: \pi_{AG} = 80\% \text{ vs } H_A: \pi_{AG} > 80\%$$

Where πAG is the percent agreement between Frozen Section and cCell. This will be tested utilizing the exact binomial test. Estimates of agreement from the pilot study indicate that the agreement will be greater than 86%. In this study, there will be 250 participants available for analysis for the estimation and testing of agreement. For outcome results, if the agreement is 86%, with the sample size of 250, the probability the estimated agreement will be greater than 80% is greater than 90%. That is, $P(p_{AG} > 80\%) > 90\%$.

Sensitivity Table – Sensitivity is only calculated when the truth standard is positive for tumor.

Agreement Table with Tumor Truth

	cC		
FS Results	Agree w/Truth	Disagree w/Truth	
Agree	e	f	e+f
Disagree	g	h	g+h
	e+g	f+h	n_1

The sensitivity of the Frozen Section is given by $p_{SenF} = (e+f)/n_1$ and the sensitivity of cCell is given by $p_{SenC}=(e+g)/n_1$. It is desired to demonstrate that the sensitivity of cCell is non inferior to that of Frozen section.

The non inferiority of the sensitivity of cCell result versus the sensitivity of the Frozen Section is given as:

$$H_0: \pi_{SenF} - \pi_{SenC} > \Delta \text{ versus } H_A: \pi_{SenF} - \pi_{SenC} \leq \Delta$$

Here π_{SenF} is the true sensitivity for Frozen Section and π_{SenC} is the true sensitivity for cCell. The parameter Δ is the non-inferiority limit. In this study, it is proposed to utilize a Δ=0.05 (5%). If that D is used and the anticipated sensitivity for both Frozen Section and cCell is 90%, then

with a sample size of 200 tumor positive samples the probability the difference between the estimated sensitivities is less than 5% is greater than 90%. That is $P((\pi_{SenF} - \pi_{SenC} \leq 5\%) > 90\%$.

Specificity Table – Specificity is only calculated when the truth standard is negative for tumor.

Agreement Table with Non-Tumor Truth

	cC		
FS Results	Agree w/Truth	Disagree w/Truth	
Agree	r	s	r+s
Disagree	t	u	t+u
	r+t	s+u	n_2

The specificity of the Frozen Section is given by $p_{SpeF} = (r+d)/n_2$ and the specificity of cCeLL is given by $p_{SpeC}=(r+d)/n_2$. It is desired to demonstrate that the specificity of cCeLL is non-inferior to that of Frozen section.

The non-inferiority of the specificity of cCell result versus the specificity of the Frozen Section is given as:

$$H_0: \pi_{SpeCF} - \pi_{SpeC} > \Delta \text{ versus } H_A: \pi_{SpeF} - \pi_{SpeC} \leq \Delta$$

Here π_{SpeF} is the true specificity for Frozen Section and π_{SpeC} is the true specificity for cCell. The parameter Δ is the non-inferiority limit. In this study, it is proposed to utilize a Δ=0.10 (10%). If that Δ is used and the anticipated specificity for both Frozen Section and cCell is 90%, then with a sample size of 50 tumor negative samples the probability the difference between the estimated specificities is less than 10% is greater than 80%. That is $P(\pi_{SpeF} - \pi_{SpeC} \leq 10\%) > 80\%$.

The sensitivity and the specificity hypotheses will be tested using McNemar's test by constructing an upper 97.5% confidence bound for the difference between the two methods. If the upper bound is less than Δ (5% for sensitivity and 10% for specificity) then the two images are said to be non-inferior. The upper confidence bound is given by:

$$p_{FS} - p_{cC} + z_{0.05} se(p_{FS} - p_{cCu})$$, where

$$se(p_{FS} - p_{cCu}) = \frac{\sqrt{(b+c)}}{n}$$

The standard error (se) for the difference depends on the number of discordant pairs (a and b). Where a and b correspond to f and g from the sensitivity table and to t and s from the specificity table. The estimates p_{FS} and p_{cC} correspond to the sensitivity of the Frozen Section,

p_{SenF} , and the sensitivity of cCell, p_{SenC} when sensitivity is being tested and they correspond to the specificity of the Frozen Section, p_{SenF}, and the specificity of cCell, p_{SenC}, when specificity is being tested.

The secondary endpoints will be summarized via descriptive statistics. No hypothesis testing will be performed for the secondary endpoints.

8.3.2 Analysis Population

The analyses described above will be performed on the intent to analyze population. That is, all samples in that are collected and are analyzable by frozen section will be utilized in the analysis regardless of whether they were successfully analyzed by both methods.

A secondary analysis will be done on the primary endpoints on the samples that were analyzable by both methods.

8.3.3 Demographic Baseline Data

Demographic data and other baseline characteristics will be summarized via descriptive statistics.

9. Operational Considerations

9.1 Regulatory, Ethical and Study Oversight

It is the Sponsor's responsibility to ensure that the study is conducted according to ISO 14155, the Declaration of Helsinki, other applicable regulatory requirements, the study protocol, and any conditions of approval imposed by the REB or Health Canada (HC). The Sponsor also will ensure proper clinical site monitoring.

The Sponsor has taken out an insurance policy for the total duration of the study. This insurance policy covers the participants in respect of the risks involved in this study according to this CIP. In the case of injury or disability deriving from participation in the study, the participant is requested to inform the treating physician responsible for the study without delay.

Prior to allowing the sites to start enrolling participants into the study, the Sponsor is responsible for selecting investigators, ensuring REB and HC approvals are obtained where applicable, and signing the Investigator Site Agreement with the investigators and/or hospitals. The study protocol (or amendment[s]), Informed Consent Form, and other applicable study related documents must be submitted to and approved by the REB and HC before enrolment of participants.

A signed copy of the REB approval letter addressed to the investigator must be submitted to the sponsor certifying study approval prior to participant enrolment. The sponsor is responsible for notifying HC of the intention to perform a clinical investigation under this protocol and will ensure an official approval is obtained prior to starting the clinical investigation. The investigation will not start before the required approvals or favorable opinions from the REB and HC have been obtained.

Any substantial amendments to the protocol will require REB and HC approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study participants.

9.2 Confidentiality and Privacy

All materials, information (oral or written) and unpublished documents provided to the participating physician (or any steps taken by the company on their behalf), including, but not limited to, the protocol, eCRFs, and IB are the sole property of the Sponsor.

These materials and information (both global and partial) may not be provided or disclosed by the participating physicians or any person on their team to unauthorized persons without the prior written consent of the Sponsor.

The participating physician considers all information received, obtained, or derived during the study to be confidential and will take all necessary steps to ensure that there is no breach of confidentiality, except for the information to be provided according to law.

The participant's personal data (age, sex, cCeLL images, date of surgery) and participating physician’s personal data which may be included in Sponsor database shall be treated in compliance with all local and international applicable laws and regulations. When archiving or processing personal data pertaining to the physician and/or to the participants, Sponsor shall take all appropriate measures to safeguard and prevent access to this data by any unauthorized third-party.

9.3 Sponsor Responsibilities

The sponsor has the overall responsibility for the conduct of the study, including assurance that the study satisfies international standards and the regulatory requirements of the relevant regulatory authorities.

The Sponsor will be responsible for:

- Selection of clinical investigators and sites: The Sponsor will select qualified investigators and facilities which have adequate study population to meet the requirements of the investigation.
- Training of investigators and site personnel and site monitoring: The training of the Investigator and clinical site personnel will be the responsibility of the Sponsor, or designee, and may be conducted during an investigator meeting, a site initiation visit, or other training sessions. Periodic monitoring visits will be conducted frequently enough to ensure that all clinical participant data are properly documented and that the study is properly conducted.
- Documentation: The Sponsor will collect, store, guard and ensure completion of all study relevant documents by the relevant parties.
    - Signed and dated eCRFs
    - Records of any Serious Adverse Events (SAEs) reported to the Sponsor during the clinical investigation

- Any statistical analyses and underlying supporting data
- Final report of the clinical investigation
- Submitting Reports
- Maintaining Records: The Sponsor will maintain copies of correspondence, data, SAEs and other records related to the clinical study according to requirements set forth by ISO 14155 and regulations.
- Monitoring: The Sponsor is responsible for monitoring the study to ensure compliance with ISO 14155 and regulatory requirements.

9.4 Investigator Responsibilities

- Protocol acceptance: Prior to starting enrolment of participants, the Investigator must read and understand this study protocol, and must sign and date the Protocol Signature page. The Site Agreement documents agreement to all conditions of the study protocol and agreement to conduct the study accordingly. This study will be conducted in accordance the Declaration of Helsinki and other applicable regulatory requirements and standards, and any conditions of approval imposed by the REB or regulatory authorities.
- Required documents: The following documents must be submitted to the Sponsor, or designee prior to participant enrolment:
    1. Signed Protocol Signature Page
    2. Recent (≤ 2 years old) signed and dated English Curriculum Vitae (CV) of the Principal Investigator and co-investigators of the clinical site. The CV should clearly show the Investigator/co-investigators' qualifications and experience.
    3. Copy of the written confirmation of the REB regarding approval of the protocol including version number and date, ICF (including version and date) and other adjunctive participant material.
    4. List of voting REB members
    5. Signed Clinical Trial Agreement
- REB approvals and notifications: According to the local requirements, the Investigator must have all necessary approvals, including written approval from the REB of the clinical site or other accepted REB prior to enrolling participants in the study. A copy of the written approval must be provided to the Sponsor.
    - Serious Adverse Event (SAE) reports as well as annual and final reports will be submitted to the REB as required.
- Obtaining informed consent: A member of the study team, who is not part of the patient's circle of care, will obtain informed consent in accordance with the procedure described in this study protocol and the requirements of the REB.

- Medical care of participants
- Reporting requirements
- Audits / Inspections: In the event that audits/inspections are initiated by the Sponsor (or its designee) or national/international regulatory authorities, the Investigator must allow access to the original medical records and must provide all requested information. In the event that audits are initiated by regulatory authorities, the Investigator will immediately notify the Sponsor.

9.5 Clinical Monitoring

The Sponsor will be responsible for ensuring that adequate monitoring at each site is completed to ensure protection of the rights of participants, the safety of participants, and the quality and integrity of the data collected and submitted. Each clinical site will be monitored according to the study monitoring plan to ensure to verify that:

- The rights and well-being of the participants are protected
- The reported study data are accurate, complete and verifiable from source documents
- The conduct of the study is in compliance with the currently approved CIP/ amendment(s), GCP, the applicable regulatory requirement(s) and applicable requirements of the REB
- There is adequate participant enrollment
- Investigational device accountability (if applicable) is maintained

Monitoring visits will be performed remotely or onsite by qualified Sponsor representatives and will include, but not limited to, the following:

- Verification of the protocol adherence
- Source documentation verification and accuracy of the eCRFs
- Verification that informed consent was obtained for all participants participating in the study in accordance with requirements described in the study protocol
- The safety status of all participants (i.e. (S)AEs/(S)ADEs/DD)
- Verification of completeness of the Site File
- Verification of accuracy of all study logs such as the Delegation of Authority Log, etc.
- Compliance with applicable regulations
- Identification and action to resolve any issues or problems with the study.

At the end of each visit the Monitor will meet with the PI and study site personnel to discuss any relevant findings. Failure by the PI/site staff to address adequately issues of non-compliance may cause the Sponsor to put the Investigator or Site on probation or to withdraw the Investigator/Site from the study. The frequency of monitoring will be based upon

considerations such as enrollment levels, study duration, compliance, or suspected data inconsistency requiring further investigation.

9.6 Source Documentation

Regulations (Health Canada MDR; ISO 14155) require that Investigators maintain information in the participant's medical records that corroborate data collected in the eCRF. In order to comply with these regulatory requirements, at minimum, the following is a list of information that should be maintained and made available as required by monitors and/or regulatory inspectors:

1. Medical history/physical condition relevant to the inclusion/exclusion criteria
2. Dated and signed notes on the day of entry into the study, protocol number, clinical site participant number assigned and a statement that informed consent was obtained and is on file in the participant medical chart
3. Adverse events reported and categorized as Serious Adverse Device Effects (SADEs) their resolution, including supporting documents such as discharge summaries and lab results

The Investigator and the head of the medical institution (where applicable) agree to allow the Clinical Monitor direct access to all relevant documents.

9.7 Data Handling

All eCRF information, study records, reports, and source documents that support the eCRF must be retained in the files of the responsible Investigator according to national requirements following notification by the Sponsor or designee that all investigations have been completed, and will further be retained in accordance with local and international guidelines as identified in the Investigator Site Agreement. This documentation must be accessible upon request by international regulatory authorities or the Sponsor (or designee). The Sponsor or designee must approve archiving or transfer of the documentation for relocation purpose of premises, in writing, prior to the actual file transfer. The Investigator must notify the Sponsor, in writing, of transfer location, duration, and the procedure for accessing study documentation. The Investigator must contact the Sponsor, or designee, in writing at least 30 days before the transfer location, duration, procedure for accessing study documentation or destruction of any records and reports pertaining to the study to ensure they no longer need to be retained.

If the Investigator retires, relocates, or for other reasons withdraws from assuming primary responsibility for keeping the study records, custody per written notice must be submitted to the Sponsor, or designee, indicating the name and address of the person accepting primary responsibility. The REB must be notified in writing of the name and address of the new custodian.

Data will be retained by the Sponsor for 15 years after the end of the study; or, where the investigational device is subsequently placed on the market, for 15 years after the last device has been placed on the market. Clinical data are to be retained by the study site in conformance with regional medical practice requirements.

All clinical study information will be recorded, handled and stored in a way that allows its accurate reporting, interpretation and verification. Data are to be submitted promptly via eCRF after collection. Missing or unclear data will be corrected as necessary throughout the study. The Sponsor will request further documentation such as physician a procedure notes when complications or malfunctions are observed and reported. During monitoring visits, the findings from the review of source documents will be discussed with the Investigator.

All documents shall be maintained in a secure location, protected from unauthorized access or damage, and they shall be archived as per local requirements.

9.8 Protocol Deviations

A protocol deviation is defined as any divergence from the study protocol. PIs and site support staff must not deviate from the protocol. Any deviations from clinical protocol requirements will be considered protocol deviations and need to be reported to the Sponsor within 24 hours. The Sponsor will not make any exceptions to the protocol and will not provide waivers to participants with any protocol deviations.

Deviations from the protocol that may affect the rights, safety or well-being of human participants, or the scientific integrity of the study may not proceed without prior approval of the Sponsor and the REB. However, under emergency circumstances, deviations from the protocol to protect the rights, safety or well-being of human participants may proceed without prior approval of the Sponsor, the REB and the regulatory authorities as required. Such emergency deviations shall be documented by the Investigator and reported to the Sponsor within 24 hours.

The Sponsor may disqualify an Investigator if, without having obtained prior written approval by the Sponsor, there is a deviation from this protocol in a manner that has affected or could affect the rights, safety or well-being of human participants, or the scientific integrity of the study.

The Sponsor will determine the effect of the protocol deviation on the scientific value of the clinical study and participant safety and determine if additional reports or actions are required. Additional action may include site re-training, removal of the devices, and/or site termination.

The Investigator is responsible for promptly reporting protocol deviations to the REB per the REB's policies, and to the Sponsor.

9.9 Quality Assurance and Quality Control

The Sponsor will implement and maintain quality systems defined by written procedures to ensure that trials are conducted and data are generated, documented and reported in compliance with this Clinical Investigational Plan, Good Clinical Practices (ISO 14155), and the applicable regulatory requirements. Included in these procedures will be quality control measures to ensure that all data are reliable and have been processed correctly at each stage of handling.

The PI will be responsible for ensuring that all personnel involved in the study are trained on such procedures. The Sponsor will secure agreement with all parties to ensure direct access to

the site, and to source data/documents and reports, for the purpose of monitoring and auditing by the Sponsor, REB review and for regulatory inspection. Such agreements will be verified as part of site initiation.

9.10 Publication Policy

The study will be registered in a publicly accessible database prior to participant inclusion, and the results of the study will be made publicly available upon conclusion in accordance with the Declaration of Helsinki. At the conclusion of the study, a report will be prepared for presentation and for publication. Authorship is based on the ICMJE (International Committee of Medical Journal Editors) guidelines.

All parties should adhere to accepted guidelines for ethical reporting. Negative and inconclusive as well as positive results must be published or otherwise made publicly available. Sources of funding, institutional affiliations and conflicts of interest must be declared in the publication.

9.11 Financial Disclosure

The Sponsor will reimburse the site for participation in the study in accordance with the executed Clinical Study Agreement. No further compensation will be provided to the site or investigators. Additional details are provided in the Clinical Study Agreement.

Investigators will provide the Sponsor with sufficient, accurate financial information as requested to allow the Sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities. Investigators are responsible for providing information on financial interests during the course of the study and for one year after completion of the study.

10.Safety Oversight

The Sponsor will be responsible for providing safety oversight and reviewing the information about the investigational product and reported safety events. This oversight includes reviewing safety information and providing applicable recommendations. This data and safety review facilitate early detection of safety signals and continued appropriateness of the research and protection of human study participants.

10.1 Definition and assessment of safety related events

Information on safety related events will be collected while the device is implanted (i.e., from implant up to and including explant) by the investigators or their designate. Classification of events is based on ISO 14155:2020. [1]

- Adverse Event (AE): any untoward medical occurrence, unintended disease or injury, or untoward clinical signs (including abnormal laboratory findings) in

[1] ISO 14155, 2020: Clinical investigation of medical devices for human subjects — Good clinical practice.

participants, users or other persons, whether or not related to the investigational device. This definition includes events related to the procedures involved.

- Serious Adverse Events (SAE): An adverse event that led to death or led to serious deterioration in the health of the participant, that either resulted in:
    - Death
    - A life-threatening illness of injury, or
    - A permanent impairment of a body structure of a body function, or
    - In-patient or prolonged hospitalization, or
    - Medical or surgical intervention to prevent life-threatening illness or injury or permanent impairment to a body structure or a body function, or
    - Fetal distress, fetal death or a congenital abnormality of birth defect.

Planned hospitalization for a pre-existing condition, or a procedure required by the clinical investigation plan without serious deterioration in health, is not considered a serious adverse event.

- Adverse Device Effect (ADE): An adverse event related to the use of an investigational device. This definition includes adverse events resulting from insufficient or inadequate instructions for use, deployment, implantation, installation or operation, or any malfunction of the investigational device. This definition includes any event resulting from use error or from intentional misuse of the investigational medical device.
- Serious Adverse Device Effect (SADE): Adverse device effect that has resulted in any of the consequences characteristic of a serious adverse event.

For the purposes of this study, only (S)ADEs will be collected and reported. Since this is an ex vivo procedure, it is expected that the incidence of (S)ADEs will be low to none.

10.2 Device Deficiency (DD)

A DD is defined as any inadequacy of an investigational medical device with respect to its identity, quality, durability, reliability, safety, or performance. Device deficiencies include malfunctions, use errors, or inadequacy in information supplied by the manufacturer.

All DDs must be reported to the Sponsor within 24 hours.

10.3 Reporting of Safety Related Events

Reporting of serious incidents to the regulatory authorities will be performed based on Health Canada regulations:

- Incident means any malfunction or deterioration in the characteristics and/or performance of the study device, including use-error due to ergonomic features,

any inadequacy in the information supplied by the manufacturer and any undesirable side-effect.

- Serious incident means any incident that directly or indirectly led, might have led or might lead to any of the following:
    - Death of a participant, user or other person,
    - Temporary or permanent serious deterioration of the participant's, user's or other person's state of health,
    - Serious public health threat.

Safety related events, which require preventive or corrective measurements intended to protect participants, may have to be reported to the local REBs. The Sponsor will report any (S)ADEs and DD(s) to Health Canada as required and within stipulated timelines.

11. References

Amos WB, White JG. How the confocal laser scanning microscope entered biological research. Biol Cell. 2003;95(6):335-342. doi:10.1016/s0248-4900(03)00078-9

BIS Research Inc. Medical market data. Updated 2022. https://blog.bisresearch.com/bis-research-market-insights-quick-bytes

Carl Zeiss Meditec AG. Convivo: Putting in vivo cellular imaging at your fingertips. 2019:Brochure. Available at: https://www.zeiss.com/content/dam/Meditec/ref_master/products/convivo/documents/convivo_brochure.pdf

Chand P, Amit S, Gupta R, Agarwal A. Errors, limitations, and pitfalls in the diagnosis of central and peripheral nervous system lesions in intraoperative cytology and frozen sections. *J Cytol*. 2016;33(2):93-97. doi:10.4103/0970-9371.182530

de Martel C, Georges D, Bray F, Ferlay J, Clifford GM. Global burden of cancer attributable to infections in 2018: a worldwide incidence analysis. Lancet Glob Health. 2020;8(2):e180-e190.

Ferlay J, Ervik M, Lam F, Colombet M, Mery L, Piñeros M, et al. Global Cancer Observatory: Cancer Today. Lyon: International Agency for Research on Cancer; 2020 (https://gco.iarc.fr/today, accessed February 2021).

IndustryARC. Oncology Market - Forecast (2020 - 2025). 2020.

Mat Zin AA, Zulkarnain S. Diagnostic Accuracy of Cytology Smear and Frozen Section in Glioma. *Asian Pac J Cancer Prev*. 2019;20(2):321-325. Published 2019 Feb 26. doi:10.31557/APJCP.2019.20.2.321

Mauna Kea Technologies. Cellvizio: Real-time in vivo cellular imaging platform. 2021:Brochure. Available at: http://www.cellvizio.net/uploads/media/media_pdf/0001/03/MKT-132-EN-Cellvizio%20Web%20Brochure.pdf

MAVIG GmbH. Vivascope: Confocal laser scanning microscopy - In vivo histology for cellular level skin analyses in cosmetic research and dermopharmacy. 2019:Brochure. Available at: https://www.vivascope.de/wp-content/uploads/2019/06/Cosmeticbrochure.pdf

Samal S, Kalra R, Sharma J, et al. Comparison between crush/squash cytology and frozen section preparation in intraoperative diagnosis of central nervous system lesions. *Oncol J India.* 2018;1:25–30.

Tofte K, Berger C, Torp SH, et al. The diagnostic properties of frozen sections in suspected intracranial tumors: A study of 578 consecutive cases. *Surg Neurol Int.* 2014;5:5.

Tofte K, Berger C, Torp SH, et al. The diagnostic properties of frozen sections in suspected intracranial tumors: A study of 578 consecutive cases. *Surg Neurol Int.* 2014;5:5.

White JG, Amos WB, Fordham M. An evaluation of confocal versus conventional imaging of biological structures by fluorescence light microscopy. *J Cell Biol*. 1987;105(1):41-48. doi:10.1083/jcb.105.1.41

WHO. Assessing national capacity for the prevention and control of noncommunicable diseases: report of the 2019 global survey. Geneva: World Health Organization; 2020.