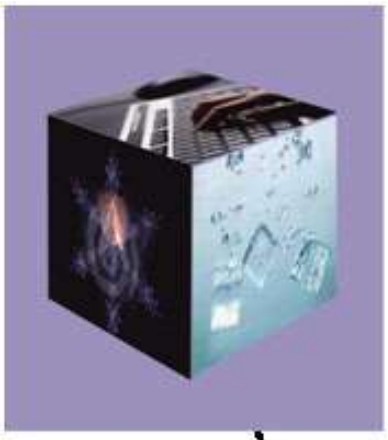
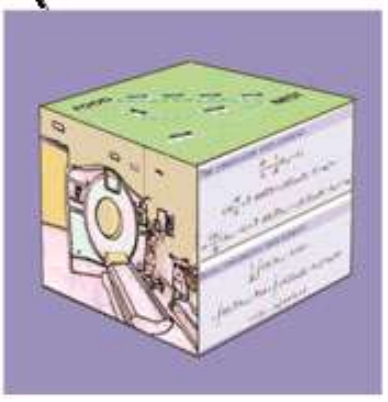


Prof. Massimo Alessandro Vercelloni
Prof. Leopoldo Avalle

FROM STUDY TO REALIZATION

EVALUATIONS ON THE EVOLUTION OF RESEARCH ON CRYOSURGERY



SURGICAL CONSIDERATIONS REGARDING THE POTENTIAL CHARACTERISTICS OF AN INTEGRATED INSTRUMENT FOR OPTIMAL CRYOSURGERY INTERVENTION.

EXAMINATION OF ALL POSSIBLE RELATIONSHIPS AND CONSEQUENCES RELATED TO THE NATURE AND POTENTIAL CHARACTERISTICS OF THE CRYOSURGICAL SYSTEM PATENT.

BIOPHYSICAL CONSIDERATIONS THAT UNDERLIE THE PATENT DESIGN OF AN INSTRUMENT FOR OPTIMAL CRYOSURGERY INTERVENTION.

Prof. Massimo Alessandro Vercelloni - Prof. Leopoldo Avalle

INTRODUCTION.

With this report we are going to evaluate and consider all the technical parameters that led to the formulation of the patent inherent to the project by name:

"Closed Cycle Multifunctional Cryosurgical System" (SCP CC).

which with the use of specific equipment allows the cryosurgical ablation of injured anatomical parts,

This project has been patented and has received Italian and European approval. (It is being evaluated in other countries of the world).

The applications and concessions expressed above are represented by the following numbers:

National patent (Italy)

Question: 102015000083117 of 14/12/2015

Concession: 102015000083117 of 20/8/2018

European Patent

Question: 16836090.7 of 12/12/2016

Priority claimed: No 102015000083117 14/12/2015

GENERAL CONSIDERATIONS.

One of the negative consequences to be generally attributed to surgery on tumour exeresis is the occurrence of local recurrences or distant metastases. Extremely important is the staging of TNM disease. The absence of lymph node involvement allows radical surgical exeresis with consequential follow-up.

The removal of neoplastic tissue sometimes determines the risk of dissemination of intraoperative tumor cells, being free to move, which can cause the onset of local recurrences or distant metastases.

This situation can have negative consequences taking into account that the patient has reduced immune defenses.

In this regard, cryosurgery is a useful indication for the treatment of advanced disease and distant metastases allowing debulking to obtain synergistic responses with chemotherapy.

It is, therefore, useful and necessary to study a set of instruments that can partially solve (or mitigate) these problems.

Among the various perspectives, low-impact surgery using cryosurgery we think that it can represent an accessory choice of great therapeutic interest.

Gelification allows to destroy the DNA of neoplastic cells preventing their replication and consequently the possibility of spreading the neoplasm in the patient undergoing such therapeutic treatment.

This effect, according to our considerations, is one of the reasons that should lead surgeons to use this technique when indicated.

We believe that the design of technologically advanced instruments that allow to perform interventions on various tumor pathologies that today are inoperable by traditional means, may be the first step towards the use of this surgical method of great impact for patients with truly considerable therapeutic results.

The application of these low temperatures must take place with particular amounts of cryogenic energy appropriate to the size and type of tumor tissue to be treated.

From a careful investigation, however, this approach hides some critical points that we will describe below that must be considered and overcome. Neoplastic tissue is indoctrinated in healthy tissue and the "cold front" cannot interrupt itself, in a natural way, when it comes into contact with adjacent normal tissue.

There are no physical or biological techniques to stop the advancing cryogenic front.

Cold, like heat, is transmitted in biological tissue in compliance with complex laws that are difficult to detect and govern. [1]

One way to ensure the integrity of healthy organs is to place hot needles (probes brought to body temperature and kept at the same thermal value) in appropriate positions to protect them.

The result of these probes is to stop the cryogenic front where the destructive effect due to low temperatures is no longer required but harmful.

We could call these needles "sentinels" or more appropriately **''heat shields**" for their protective function.

It is not easy to predict the cryogenic advancement fronts that are generated by the combination of the cold effect of **cryods** and the thermal effect of sentries. Just as it is not as easy to determine their optimal insertion position given the complexity and variability of the thermal curves that evolve in the patient's body.

This configuration due to the lowering of the temperature in the biological tissue can be "**seen**" using a mathematical tool that engineers use in thermotechnical studies.

The patent we are about to describe its characteristics uses this elegant and powerful mathematical tool.

It is a mathematical physics methodology that we will describe later in this report.

We are referring to the formulation of differential equations to partial derivatives which are the mathematical method to represent the evolution of heat in bodies. [1] [8]

It is the most advanced mathematical technique that can provide designers dealing with these topics of thermal transmission.

Let us note, however, that we consider the problem of transmission of low temperatures, which are quite different from the hot fronts characteristic of the iron and steel industry.

There is, moreover, another very important parameter to evaluate: the medium by which the cold fluid is transmitted is not the classic homogeneous solid body (like a steel bar or other conforming and well known material whose structure does not change during the process) but a biological tissue composed of complex cellular elements permeated with water.

A medium that changes state (from liquid to solid as a whole) during the transmission process.

It is actually almost another physical world.

This problem has been studied and realized in previous works dealing with cryosurgical probes from a mathematical point of view. [1] [3] [4] [5] [6].

Heat transfer in biological tissues, and in particular around a cryosurgery probe, is characterized by a dense capillary network with low blood perfusion; it is modelled by the classic Pennes equation [8] obtained from studies by the biophysicist Stefan (see Stefan's direct problem) which is mathematically expressed by the following formula:

$$C\frac{\partial T}{\partial t} = \nabla \cdot (k\nabla T) + \dot{w}_b C_b (T_b - T) + \dot{q}_{met}$$

where C is the specific volumetric heat of the tissue, T the temperature of the tissue, t the time, k the thermal conductivity of the tissue, wb the rate of blood perfusion, Cb the specific volumetric heat of the blood, Tb the temperature of the blood and qmet the source of volumetric heat given by cell metabolism.

It is a very complex mathematical expression and very difficult to solve (even and just for an expert mathematician).

In this case a technique called Finite Difference Numerical Solution is helpful. The use of the computer and the appropriate numerical techniques allows to overcome and solve in an elegant and almost perfect way the difficulties of calculation expressed above.

This differential equation subjected to repeated and rigorous experimental tests on biological tissues has given excellent results by simulating biophysical reality in space and time.

In other words, it has been adherent and almost perfectly consistent with the progress in the biological tissues of the cryogenic front.

The researchers used this mathematical expression bringing small differences to the biophysical values that characterize the strictly biological nature of tissues.

Let us also note that from the scientific literature and medical-surgical information it has been found that cooling biological tissues causes, as already expressed above, the death of cells. This phenomenon, however, is not as global as it might seem.

When thawing, a significant number of cells survive the cryogenic impact.

To make the "cold" effect lethal, it is necessary to cool the tissues as quickly as possible. While thawing must occur as slowly as possible. The destructive effect increases if, the cooling and thawing cycle occurs at least twice.

Thermal cycles are defined and can be automatic, i.e. made by the equipment or appropriately guided by the surgeon who manipulates the control structures of cryogenic sources.

Increasingly refined laboratory research aimed at understanding the cryogenic mechanisms of cells has led to the determination that in an ablation process not only the achievement of low temperatures but also the **time** with which the tissues were frozen played a very important role.

It was observed that not only the time had **to be as fast** as possible but also the thawing had **to be as slow** as possible to achieve maximum tissue destruction.

This topic of fundamental importance for tumor ablation was studied by Prof. Chua of the University of Singapore and its publication has become a basic reference for designers and surgeons working in this field.

Chua et al. [2] used a finite volume method to solve the transient biological heat equation using single or multiple probe geometry; the simulated results showed good agreement with experimental data obtained from in vivo clinical studies.

The calibrated model was also used to analyze the effects of several freeze-thaw cycles on tumor cell damage.

This study provided a fundamental basis for the design of an optimized cryosurgery protocol that incorporates thermal effects and the extent of cell destruction within the tumor.

In summary, we can summarize: **from thermal reduction to fast freezing** and slow thawing to the concept of **thermal cycle** to achieve maximum cell destruction.

This last notion is at the basis of the design motivations that make up the patent we are going to describe.

At this point, the difference in treatment to which the tissues on which the cryogenic probes act is spontaneous and evident.

Those closest to the cryodes will be affected by the effects of thermal cycles while those furthest away will be less and less affected by cryogenic aggression as they move away from the source needle.

With regard to these biological considerations, the Chua cycle will produce three different effects depending on the distance from the base of the cryode, namely: **zone of maximum destructive effect, zone of intermediate destructive effect and zone of minimum destructive effect.**

This technical information is already known in advance because it depends exclusively on the thermal conditions of the cryode and the cryogenic characteristics of the equipment, and the system designers have taken it into account.

For these reasons we speak of **maximum destruction volume combined** with each cryode. [2] [4]

This first synthetic analysis shows that the patient and cryosurgical intervention approach must necessarily be preceded by a preparatory phase in order to "inform the team preparing for the intervention in the strictest possible way".

The information should indicate the surgical methods and topological positions indicating where and at what depth to insert the probes (both cold and hot).

For these reasons it is more than evident that the patent considers two fundamental phases for its cryogenic realization:

[virtual cryosurgical operation **(VCO)]** (virtual because it takes place without the presence of the patient).

[steered cryosurgical operation **(SCO)**] (real intervention on the patient).

Let us reconsider analytically the patent proposal and the two operative phases.

VIRTUAL CRIO-SURGICAL INTERVENTION *(virtual cryosurgical operation (VCO))*

We would like to point out that both interventions are interrelated and represent grounds for patent claims.

We will describe, therefore, with more detail the operation of these functional apparatus both from a logical-mathematical and cryosurgical point of view.

VCO is only a softweristic component in which the relationship with the surgeon is realized, who, for obvious reasons, must be a true expert of human anatomy.

The virtual cryosurgical operation (VCO).

This intervention has been defined virtual by the fact that there is no physical presence of the patient while the cryosurgical operation is simulated (as previously mentioned).

This is the first logical elaboration process that must be carried out for the successful outcome of the subsequent real operation.

In the patent process a very first information is required regarding the "language" to be used in the following phases of interface with the physician.

The user can create his own particular glossary using the system previously organized for such operations.

In it are read the data of the X-rays and pathological information of the patient, the results of the consequent elaborations will be the input for the consequent real intervention process that will follow.

Unfortunately it is not possible to represent a neoplastic mass with a simple mathematical formula or a regular geometric figure.

In substance it is a "real" model and it is therefore necessary to take into account three fundamental variables:

1) the **accessibility** of the part to be treated,

2. the characteristics of the '**mass**(es)' to be cooled,

3. possible **cold damage** to peri- injurious tissues.

For these reasons the presence of the surgeon who "dialogues" and acts with the system is as indispensable as it is fundamental.

The operational steps are as follows:

a) If the medical investigation detects the presence of a pathology for which it is decided to resort to cryosurgery, in a first operation (extraneous to virtual surgery) the patient must undergo a 3D CT scan. The images and any other diagnostic form will be the input of the system that characterizes the virtual intervention.
b) The data previously collected will be read and processed by the specific software that characterizes the virtual intervention.
c) In this phase the dimensions of the neoplasms will be determined (after a predetermination of the physical origin of the axes and the value of their dimensional scale, shape, volume and position on the respective Cartesian axes).

d) This preliminary operation is very important because it will determine the size, number and position of the neoplasm within the healthy organ.
e) From the previous point the system is able to calculate the position, geometric dimensions and, consequently, the total mass of the tumour and any metastases that must be destroyed by cryosurgery.
f) At this stage of the process, the system must read and compare the physical data of the probes with the total volume of neoplastic tissue to be destroyed.

Each cryod, previously stored with its dimensions and with the fundamental parameter that has been defined **volume of maximum destruction** mentioned above, is represented with different colours by a cylinder with a small diameter and a discrete length.
At one end of this needle is displayed in an ovoid image whose dimensions are proportional to the volume of maximum security (destruction) indicated above.
For obvious reasons of scale, the system adopts a single dimensional ratio to facilitate the ongoing investigation.
The X-ray image is presented in three dimensions and, at the cryosurgeon's pleasure, can rotate on its own axes.
The simulated cryode recalled by the operator can move in any direction at will.

On the screen will also appear some numbering that will indicate the values in percentage of the tissues reached by the cold.

For the user's convenience, the calculation system will offer the percentages of neoplasms reached by the cryogenic front and will also provide the values of healthy tissues affected by the cryogenic front.

These numerical parameters will indicate to the operator the efficiency of the insertion of the cryods (always in percentage values).

The physician will thus be able to virtually insert the maximum number of cryods in the structure on which he operates.

Once the maximum number is reached, the system will inform this event and suggest a different operative strategy.

This function is fundamental for the correct calculation of the maximum number of cryods that will have to operate in the imminent intervention phase.

The positions and depths of each needle (cryod) will therefore be quantified, thus allowing an overall optimization of the cryogenic set.

We point out that every graphic representation of the virtual system is not stable and that the cryosurgeon can, by rotating and moving the positions of the cryodes (and of the heat shields), by consulting the percentage indicators tend to values close to 100% which is the optimum for each intervention.

g) Now the process is able to determine the cryogenic isothermal curves. These curves will be the thermal result of the effects of cold needles, the patient's biological condition and sentinel probes (heat shields).

At predetermined times, information about the geometry of the isothermal curves and their value will be stored on a magnetic medium.

When, through the use of technical image, the boundaries of the neoplasm to be treated will be reconstructed and any physiological indications coming from the medical staff will be

considered, the problem of defining the free parameters of a cryosurgical operation becomes exclusively related to the physical dynamics of heat propagation inside the tissue. [3][10]

The definition of the most appropriate configuration of parameters preceding each cryosurgical intervention is called cryosurgical planning and is generally entrusted to automatic systems based on complex mathematical and computational tools.

In conclusion, we can say that virtual intervention (VCO) plays a very important role in the global cryosurgical management.

It defines the total number of cryods to be used and sentinel probes (heat shields).

But not only that, their "topological" position is determined and the global impact on sick and healthy tissues is calculated.

This last operation could, in later times, be carried out by particular robots that, under the control of the cryosurgeon, place the probes in the patient.

In addition, the various "cryogenic fronts" are calculated and displayed in graphic form, which, as a result of the effects of the hot and cold probes, occur at "set times" and are useful for the control (automatic or not) of the advancement of cold in the tissues.

The management activity (allow us this term) is not to be underestimated because the positioning exercise of the various probes has a highly "formative" role improving the efficiency and sensitivity of the surgical room staff[3].

THE STEERED CRYOSURGICAL OPERATION (*steered cryosurgical operation* (SCO)).

Let us remember that the most important event that was the fulcrum around which cryosurgery began to act was the discovery of the use of ultrasound (US) in the detection of cold fronts in prostate surgery. This real time ultrasound (high frequency sound waves) allows modest temperature variations to effectively alter the speed of sound transmission through any medium. The images due to ultrasound monitoring of the liver are very eloquent and reveal the ease of understanding and consequently the usefulness that the surgeon can draw from them in the operating room. [7]

Other types of surgery, such as kidney, lung and brain surgery, have their own unique surgical accomplishment.

The proposed system is able to pre-dispose itself and adapt to each phase that international protocols require, giving the surgeon the advantages of cryosurgery without its local complications.

This surgery must in practice "execute" the orders and information that are stored in the magnetic medium resulting from the virtual surgery operation (VCO).

It is natural that in order to make a comparison it becomes necessary to be able to follow the real cryogenic front that follows during this cryosurgery phase and verify it with the one that, at certain times, has been created in the virtual surgery.

As a premise to the functions that the unit must possess, there is the possibility (obviously protected) to change some parameters that characterize the "Chua Cycle". [2]

We will remember that, summarizing what has just been said, we can say that the maximum destructive effectiveness of a cryogenic instrument consists in operating on a certain tissue by freezing it very quickly and then slowly heating it and performing at least two/three cycles (cooling - heating).

For these reasons the modification of the cycles is a very delicate operation and should not be performed except by a surgeon with extensive operational experience.

This patent has the terms that characterize it as **multifunctional** and **closed cycle**.

The first term means that the system has been designed to perform every kind of cryosurgical intervention on every organ.

The second term means that the cooling is carried out by specific components of the system without resorting to external supplies (as already indicated).

In reality the Functional Unit is composed of two parts that are not necessarily joined together. One (refrigerator) generates cold even in an environment that does not necessarily have to be the operating room, while the other **(operating instrument)**, with all its sockets for the various cryods, resides in the operating room.

The cold is stored in special mobile tanks containing an incongelable fluid (at least up to the classic -100°C more than useful for cryosurgery) which will be transferred between the two parts (refrigerator / functional unit).

Ethyl alcohol has been chosen as the real cryogenic "carrier" that carries the cold to the tips of the operating probes. Ethyl alcohol is chosen for its non-toxicity and its low freezing point (-114°C); it is however advisable that the temperature of the cold chamber does not drop below -100°C to avoid the formation of brines and pumping problems through the circuits.

The number of cylinders is such that at least two (or more) cryosurgical devices can be supplied.

One cylinder is more than sufficient to supply the intervention time.

The designers foresee the installation of two in order to respect the maximum power supply safety. [9]

The phases into which the SCO intervention can be divided are the following:

a) Preparation of the patient for surgery and its positioning on the surgical bed in the vicinity of the low temperature generator instrument located in an adjacent room.
This may coincide with the reading of the magnetic support obtained from the previous phases.
b) The information contained in the magnetic support generated in the virtual surgery allows the cryosurgical staff to review the radiological images and to prepare the instruments to be used in the following phase.
c) Given the calculated insertion positions of the cryods, the staff introduces the probes according to the coordinates imposed by the study previously carried out (in the future, as already expressed above, this operation can be done by a robotic system assisted by surgeons).
d) Once the probes have been arranged, the cryosurgeon starts cooling them. With this command you have the effects to follow.
e) Start of the internal clock. This is a very important tool to perform cold front checks. As we have said above the system of the cryogenic fronts will be compared, at specific times, with the one created previously in the realization of the virtual intervention.
f) With the activation of the system's internal timer, the echo sound monitoring is turned on to detect the progress of the cold front (US). As we have just said, this front will have to be compared, at predetermined times, with the one theoretically calculated. This is a phase that, at first, will be done by the cryosurgeon who will compare the two curves. Then particular software will perform this operation and the final task of assistance will be delegated to the doctor.
g) Control of cryosurgical operations in compliance with international protocols.

Before performing the SCO cryosurgery intervention, the medical staff that has the "computer documents" that are a consequence of the virtual intervention, can review at will all the phases of intervention that it deems appropriate, making the corrections that it deems most suitable for the success of the intervention itself, using the software in possession of the operating unit.

In these computer supports are recorded the information that generated the virtual simulation (besides the modifications made by the cryosurgical staff).

The medical team, therefore, inserted the support with the data related to the project (VCO) previously processed, displays the preliminary information on the display and prepares the patient for the intervention.

Following these indications, supported if necessary by an ultrasound unit, ***the cryodes and the sentinel probes (shield) must be placed in the exact positions that the processing of the virtual intervention recommends.***

These operations may, in the future, be performed by special robots (Robotic Surgery).

Once these operations have been completed, the surgeons will start cooling the probes and thus start ***the cryosurgery pilot surgery (SCO)***.

The cryosurgery unit must therefore be equipped with a computerized process system that can allow the reading of diagnostic images and preliminary medical measurements that are part of the project (VCO).

It is therefore really important to use "3D imaging" tools whose figures, read and interpreted by a specific software, can be processed to contribute to the exact location of the targets. This operation, for a correct execution, must be able to recognize the structures to be hit, simulate the insertion of cold needles (cryodes) in the right position and intervene by lowering the temperature of the latter. [11]

No less important is the possibility to check the cold front to verify the safety of healthy organs. [12] [13]

The insertion of the hot probes (heat shields or sentinels) must be well calibrated and studied with the utmost care (obviously in the VCO phase).

The cryosurgical apparatus in question must have the following characteristics:

Functional:

1. The machine must be able to perform a wide range of cryosurgery specific interventions: insertion and use of cryosondes for renal, prostate or liver, lung, brain or on the treatment of ubiquitous metastases, for skin surgery (dermatological cryosurgery), cryogenic fluid intake to feed the internal probes of the "cryo-baloon" technique.
2. The apparatus must be able to carry out the specific "thermal cycles" of cryosurgery operations; leaving the choice to the surgical staff to intervene manually or automatically (as they wish).
3. There must be the possibility to insert cycles at will of the cryosurgical team (only under strict supervision and control of the system).
4. The computer structure of the equipment must be able to allow the virtual cryosurgery intervention (VCO)". This intervention must be carried out in harmony with the cryosurgeon using the peculiar logical-physical-mathematical structure that will interact with the experience of the physician. This procedure, after three-dimensional radiological measurements (imaging activities), will determine the useful number of cryo-probes to be used in each specific case in compliance with the laws of thermal diffusion in biological tissues. For this purpose a mathematical-informatics method will also be used that will optimize the use and calculate the ideal positions of insertion of probes and sentinels in the structure on which they are to be operated. [9]
5. The storage phase of the data calculated with the activation of the virtual intervention will be carried out by the system. The processing will take into account the images of the tissues to be operated, the number of cryosondes to be used and the number of protective probes (sentinels or shields), the position of each needle both hot and cold, their penetration depth and, last but not least, the geometries of the various cryo-genic isothermal fronts which will be determined by the combined effect of the probes in the tissues at predetermined times.
   It should be stated that the point of origin of the axes used will be specified; a fundamental point for any topological and geometric reference. For obvious reasons of memory occupation these thermal fronts will be detected at predetermined time intervals (as mentioned above).
   In this way the thermal situations of the simulated intervention will be photographed at regu-

lar moments with images and graphs that will be useful in the consequent comparison with those generated in the real piloted intervention (SCO).

6. Once the intervention conditions have been identified, the surgical staff will be able to move on to the real intervention, i.e. with the presence of the patient (Real piloted intervention (SCO)).
   The insertion of the probes, both hot and cold, can also take place automatically using a "surgical robot" which, in compliance with what was done and elaborated in the previous phase, will insert these needles in the required position respecting the depths calculated in the virtual intervention phase.
   Any positioning errors must be contained within the tolerances that medical biology and engineering allow.
   The cryosurgical staff, assisted by a 3D ultrasound instrument, will supervise these operations and will give the appropriate orders that will allow the continuation of the real intervention operations.
7. When the cryosurgical staff will start the activation of the probes, the intervention will proceed as foreseen in the previous phase. The thermal fronts will be followed by an ultrasonic control and, within the established timeframe, the system will compare and verify the real trend of the isothermal lines. In this way, the isothermal lines realised in the virtual intervention phase will be compared with the real cryogenic front. If any differences are detected between the previously calculated trend and that detected by the ultrasonic structure, the appropriate modifications will be made in order to correct these variations, informing in any case the surgical staff who can intervene with the means available to the structure.
8. There must be a particular button that, activated, will allow **the quick release of the cryods**. **[3]**
   It is a mandatory function that allows rapid heating by acting on internal valves that will release the cryogenic tips by delivering a heated fluid. Surgeons can then, if necessary, remove the probes from the patient without waiting for the normal warm-up time.

Constructive:

The system must have the maximum compactness compatible with the operational functions described above. We summarize in the following points how much globally must be respected:

1) No gas must be released into the atmosphere;

2) Continuity and constancy of operation must be respected;

3) Electricity consumption must be limited;

4) Only one electrical outlet and one (if any) of cooling water must be required;

5) The management must be fully computerised (i.e. managed by an electronic control system).

6) The equipment must comply with all regulations and safety requirements for electrocution risks for both the patient and the surgical operators.

Operational functional diagram.

The above can be summarized in the following images:

- Patient symptomatology: medical diagnosis - detection of radiological images (Rx , Ultrasound, CT chest-abdomen, RNM).
- Virtual intervention with calculation of the number of cryodes and sentinels and their disposition. Previous X-rays are widely used.
- Real intervention (with patient), position of cold and hot needles (as elaborated in the previous phase). Detection of the cryo-genic front trend using ultrasound instruments. [7][9]

THE FOLLOWING TABLE FURTHER CLARIFIES THE PROCEDURES SET OUT ON THE PREVIOUS PAGES.

SURGICAL

CRYO OPERATION

DESIGN (VCO)

EXECUTION INTERVENTION (SCO)

Multifunctional unit

It provides for the monitoring and management of cryosurgery.

The unit is programmed in order to carry out the various operations in compliance with the Chua cycles, in manual or automatic mode.

The biological and functional parameters of the patient are also considered and controlled.

The temperature and position of the cryods are realized in the values determined during the previous medical operating phase (V.C.O.).

All phases of the intervention can be controlled with a 3D ultrasound or radiographic device.

Cryo contentainer

Cryo container Housing

Crio generator

It generates the accumulation of frigories, cooling a particular fluid that will activate the multifunctional unit for cryosurgical intervention through the cryo container inserted in the multifunctional unit.

Bibliografy.

[1] Sun F., Wang G.X., Kelly K.M., Aguilar G., 2005, *Numerical Analysis of Tissue Freezing in Cutaneous Cryosurgery Using Liquid Nitrogen Spray*, 5th International Symposium on Multiphase Flow, Heat Mass Transfer and Energy Conversion, Xi'an, China

[2] Chua K.J., Chou S.K., Ho J.C., 2007, *An Analytical Study on the Thermal Effects of Cryosurgery on Selective Cell Destruction*, Journal of Biomechanics, Vol. 40, 100-116

[3] Avalle L. et al., 2016, *Cryo-electric – Multidisciplinary Centre for Studies and Research on the Application of Cold in the Health Sector*, available at http://www.cryo-electric.it/eng/pag1.html

[4] Larrey D.J., 1817, *Mémoires de Chirurgie Militaires et Campagnes*, Paris

[5] Korpan N.N., 2001, *Atlas of Cryosurgery*, 314-321, Springer, Wien

[6] Campbell-White A., 1889, *Liquid Air in Medicine*, The Medical Record, New York

[7] Onik G.M., 1989, *Transperineal Prostatic Cryo-surgery under Transrectal Ultrasound Guidance*, Seminars in Interventional Radio, 6, 90–96

[8] Rabin Y., Shitzer A. 1998, *Numerical Solution of the Multidimensional Freezing Problem During Cryosurgery*, Journal of Biomechanical Engineering, Vol. 120, 32-37

[9] Etheridge M.L., Choi J., Ramadhyani S., Bischof J.C., 2013, *Methods for Characterizing Convective Cryoprobe Heat Transfer in Ultrasound Gel Phantoms*, Journal of Biomechanical Engineering, Vol. 135, 1-10

[10] Giorgi G., Avalle L., Brignone M., Piana M., Caviglia G., 2011, *An Optimization Approach to Multiprobe Cryosurgery Planning*, Computer Methods in Biomechanics and Biomedical Engineering, 1-14

[11] Baglietto S., Piana M., Caviglia G., Massone A.M., 2013, *A Procedure Based on Images for the Optimal Design in a Cryosurgery Operation*, Università degli Studi di Genova

[12] Çengel Y.A., Boles M.A., 2014, *Thermodynamics: An Engineering Approach*, 8th edition

[13] Haaf S., Henrici H., 2002, *Refrigeration Technology*, Ullmann's Encyclopedia of Industrial Chemistry

Written on 01/02/2020