# UNIVERSITÀ POLITECNICA DELLE MARCHE

FACOLTÀ DI INGEGNERIA

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Corso di Laurea magistrale Biomedical Engineering

# 3T PET/MR equipment for the administration of sensory stimuli and motor task in research and diagnostics


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A.A. 2021 / 2022

## Abstract

During physical and/or mental activity several brain areas are involved. To detect these activations the use of specific imaging techniques and well-designed tasks are essential in clinical and diagnostic settings. The largest section of the human brain is the cerebral cortex, which is linked to higher-order cognitive processes. Magnetic resonance is a non-invasive technique that uses a strong magnetic field and radiofrequency waves to create detailed images of organs and tissues. In neurological clinics and research, functional magnetic resonance imaging (fMRI) is used to assess small regional blood flow changes within the cortex, detected during a scan procedure which also involves task performance.
This thesis aims to investigate the setup of stimulation protocols for functional studies based on the accessory instrumentation equipment recently installed in the Marche University Hospital.
The equipment reviewed is produced by a Norwegian company called NordicNeuroLab. This company provides several modular solutions for both clinical and research fMRI needs, thanks to many hardware and software tools. NordicAktiva software thanks to an intuitive interface allows to select different paradigms and manage the stimuli delivery. For this project, three paradigms were applied to a healthy young subject during an fMRI procedure, with a 3T Signa PET/MR scanner. The first paradigm was a General Motor Left task (open and close left hand), the second was an Auditory task (with classic music) and the third was a Visual Checkerboard task (with goggles mounted on the scanner). Each task lasts 5m 12s, with 5 alternating 30s-cycles of resting and active states. Functional and T1-weighted images were acquired and then preprocessed and analysed using BrainVoyager software. For pre-processing, brain extraction, motion correction, slice time correction, temporal filtering, co-registration, and normalization steps were performed. In the results for the motor task strong activation of contralateral motor cortex was present, as expected. In the auditory task, the music stimulation shows a bilateral activity in the primary auditory cortex. The visual stimulation with the flickering checkerboard exhibits strong activity within the occipital part of the cortex as well as in the posterior parietal area. To improve stimuli delivery protocol, the use of software like nordicAktiva is game-changing, however, setting up the trial duration in the main code is a quite time-demanding procedure. A big limitation in this analysis is the involvement of just one healthy subject. A larger group of subjects could be essential for brain mapping statistical study. Also, it could be better to include in the investigation neuropathological patients since they have difficulty staying motionless in the scanner and even performing some tasks, leading to incomplete or incorrect data.
In conclusion, the employment of protocols for stimuli delivery during fMRI investigation is essential for brain mapping in clinical and diagnostics settings. Researchers must precisely plan their methodology and use well-suited paradigms, considering several features related to the psychophysical status of the subject.

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# 1. Introduction

The brain is a fascinating organ that regulates body processes and interprets information from the external world. Numerous things governed by the brain include intelligence, creativity, emotion, and memory [1].

Understanding the brain's functioning will help to discern the principles behind human behaviour and actions. All healthy human brains share the same fundamental structures and functions, even though each one is unique as an individual. People differ particularly in how their brain cells communicate, and both heredity and interactions with the environment have a great impact on this aspect. The brain changes as a result of experience, both positive and negative, strengthening and weakening existing connections, while also forming new ones [2].

When a person does both some physical and/or mental activity, different areas of the brain are involved. The activation of these areas depends on several cognitive and physiological conditions, therefore the use of specific imaging techniques and well-designed tasks to detect them is a fundamental procedure in clinical and/or diagnostic settings.

## 1.1 Brain structure and activation areas

The brain structure is composed of three main parts: forebrain, midbrain, and hindbrain.

In the forebrain, there is the cerebrum, which is the largest section of the human brain, and it is linked to higher-order mental processes like cognition and action.

The grey surface, the cerebral cortex, is made up of nerve cells, while beneath the surface, white nerve fibers transmit messages to nerve cells in different regions of the brain and body.

The cerebral cortex is divided into four "lobes" (Figure 1):

- Frontal Lobe: it is located just below the forehead and is related to the brain's capacity for thought, organization, planning, speech, movement, facial expressions, serial tasking, problem-solving, inhibition control, spontaneity, initiating and self-regulating behaviours, attention, memory, and emotional control.

- Parietal Lobe: situated in the upper back of the brain, it is responsible for the control of sophisticated actions and senses like touch, body awareness, and spatial orientation. This lobe is essential for the integration of sensory data from multiple body areas, understanding relationships between numbers, manipulating objects, visual-spatial processing, language understanding,

construction capacity, bodily placement and movement, left-right distinction, and self-awareness/insight.

- Occipital Lobe: it is situated in the posterior part of the brain, and it is involved in processing visual information such as visual attention, visual recognition, spatial analysis (movement in a three-dimensional environment), and visual perception of body language, including postures, expressions, and gestures.

- Temporal Lobe: located close to the ears, it is involved in processing verbal memory, visual memory, language production (including fluency and word-finding), general knowledge, and autobiographical memories. It is also associated with comprehending spoken language, verbal memory, and visual memory.

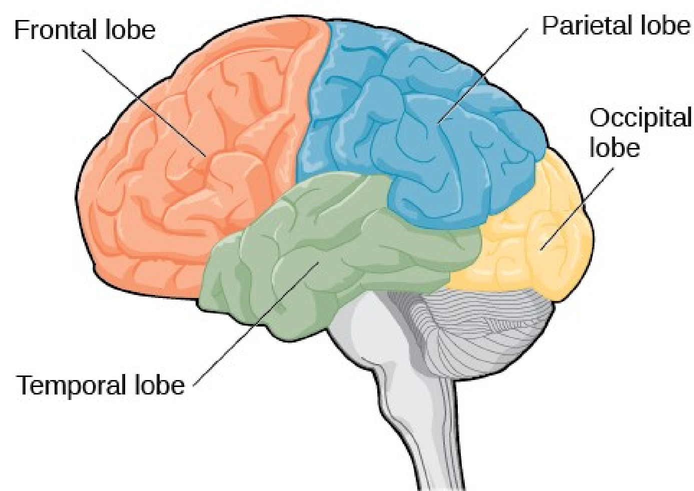


*Figure 1: lobes division in the cerebral cortex.*

The cerebrum is divided into right and left hemispheres. Each of them controls the opposite side of the body, but they don't share all the functions. In fact, speech, comprehension, mathematics, and writing are generally under the authority of the left hemisphere (called “dominant”), whereas creative, spatial, artistic, and musical abilities are all governed by the right hemisphere. Moreover, in 92% of persons, preferred hand function and language are dominated by the left hemisphere.

The *midbrain*, the smallest part of the brain, is situated close to the centre of the brain, above the hindbrain, and under the cerebral cortex. The midbrain's principal function is to work like a relay station for our visual and auditory processes. In this area is located the limbic system, (also known as the “emotional brain”) composed of the thalamus, which transports sensory data from other

areas of the brain to the cerebral cortex, and the hypothalamus, which controls numerous hormonal and somatic activities. (Figure 2).

The *hindbrain* is divided into the cerebellum ("small brain") and the brain stem.
The cerebellum is similar to the cerebrum thanks to its two hemispheres and highly folded surface, and it is related to the control and synchronization of movement, posture, and balance.
The brain stem (Figure 2), mainly composed of pons and medulla, is responsible for vital life processes like breathing, heartbeat, and blood pressure [3].

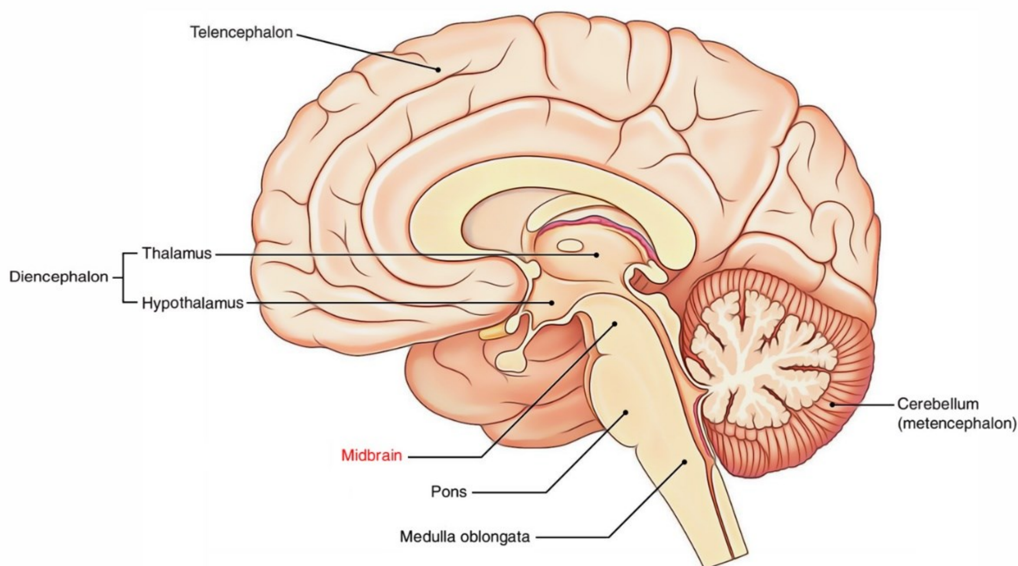


*Figure 2: forebrain, midbrain and hindbrain are shown in the picture. In particular are pointed out the principal components of the limbic system and the brain stem.*

### 1.1.1 *Brodmann areas*

Different regions of the cerebral cortex are involved in various cognitive and behavioural processes.
The *Brodmann areas* (Figure 3), named after Korbinian Brodmann, are a method of mapping the cortex and its distinctive functions. The brain cortex can be split into 52 sequentially numbered sections using this system. These areas, discriminated by microscopic anatomy through shapes, types of cells, and their connections, were initially based on the cytoarchitectural organization of neurons in the cerebral cortex.

The most important Brodmann areas are:

- Brodmann areas 1, 2, and 3 – *Primary somatosensory cortex*: it is in charge of processing physical sensations. These sensations, such as touch, pain, temperature and the localization of touch, are picked up by receptors located all over the body. This area is also fundamental for skilled and coordinated motions and motor learning.
- Brodmann area 4 – *Primary motor cortex*: The primary motor cortex is responsible for initiating and coordinating motor movements. On the contralateral side of the body (the opposite side of the body with respect to the brain's hemisphere control), each region of the motor cortex corresponds precisely to a different body part.
- Brodmann areas 17, 18 and 19 – *Primary and secondary visual cortex*: located in the occipital lobe, these areas are essential for processing visual stimuli. The primary visual cortex (area 17), which is the most extensively researched in terms of visual function, is highly specialized for processing both static and moving things as well as pattern recognition.
- Brodmann area 22, 39 and 40 - *Wernicke's area*: Brodmann 22 is a part of the Wernicke's area and is implicated in sophisticated language and auditory processing. Wernicke's area is responsible for speech fluency, or more specifically, it allows the completion of coherent sentences out of a string of words.
- Brodmann areas 44 and 45 – *Broca's area*: Broca's area, which is in the frontal lobes, is crucial for language development. This area can put pieces of language together, choose information from a range of sources, and assist in the production of data about the motor movements necessary for spoken and written language. Broca's area can also help in regulating syntactic processing mechanisms and in building complicated sentences and speech patterns [4].

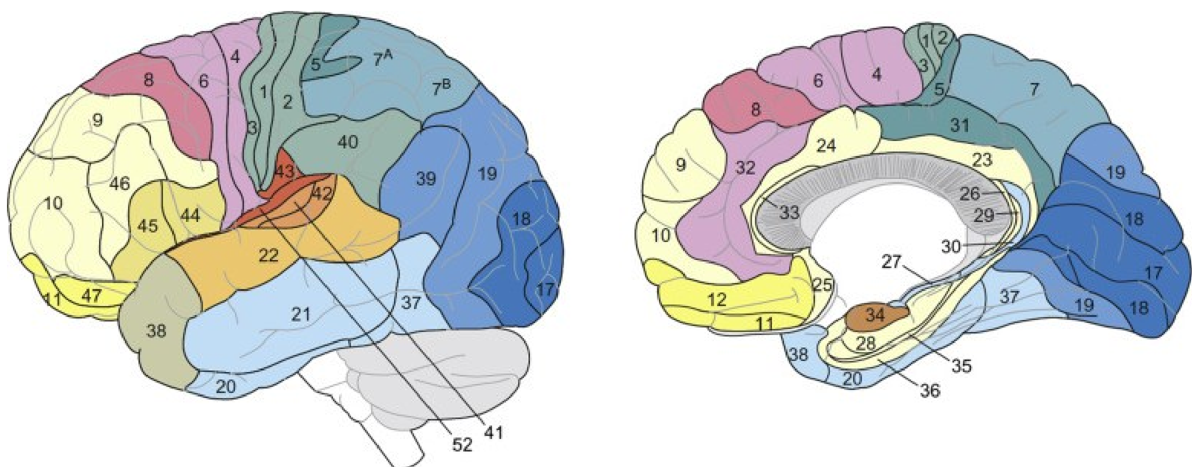


*Figure 3: Brodmann areas in the cerebral cortex; on the left lateral surface, on the right medial surface of left hemisphere [4].*

## 1.2 Magnetic resonance: structural and functional imaging for investigating brain activation

Magnetic resonance imaging (MRI) is a non-invasive medical imaging technique that, through a magnetic field and computer-generated radio waves, creates detailed images of human body organs and tissues.

The magnetic field in an MRI machine, using powerful magnets, causes the body's water molecules to momentarily rearrange, and these aligned atoms emit tiny signals used to make cross-sectional MRI images (slices).

Different types of images can be produced by altering the order in which radio frequency (RF) pulses are applied and collected. The interval between succeeding pulse sequences delivered to the same slice is known as the repetition time (TR). The time between the RF pulse being delivered and the echo signal being received is known as the time to echo (TE).

Two different relaxation times—T1 and T2—can be used to characterize tissue. T1-weighted images are produced by using short TE and TR times, and the T1 properties of tissue determine the contrast and brightness of these images. On the other hand, longer TE and TR periods are used to create T2-weighted images, in which the T2 characteristics of the tissue dominantly control the contrast and brightness.

Since MRI does not require x-rays or other radiation, it is particularly well suited when frequent imaging is required for diagnosis or therapy, especially in the brain.

### 1.2.1 *Functional Magnetic Resonance Imaging*

Functional MRI (fMRI) is employed in clinical settings, such as neurosurgery, for intraoperative monitoring of brain areas (awake surgery) and preoperative planning. This enables the programming of the surgical strategy to reduce the risks of postoperative deficit. Moreover, fMRI is frequently used in the domains of cognitive neuroscience and neurological research to examine the many cognitive functions (language, attention, memory) in healthy and pathological subjects. When compared to other techniques like Positron Emission Tomography (PET), Electroencephalography (EEG) and Magnetoencephalography (MEG), this technology offers significant advantages. First, it is non-invasive (no use of ionizing radiation as PET), safe, and has high spatial resolution (down to millimeters). Although, low temporal resolution (seconds) is one of its drawbacks [5].

Functional MRI is used to assess small regional blood flow changes in the cortex that occur when a patient performs precise tasks in a high-field MRI scanner. Such tasks, thoroughly described to subjects before the procedure, typically last three to six minutes, and may demand repetitive movement of a part of the body or the accomplishment of defined language or visual tasks. Following a detailed examinations of the scan's raw image data, "activation maps" are created using analysis of each task's outcomes (or "paradigm") and strict image quality control analysis.

Delivering periods of stimulus presentation, exactly coordinated with the scanner, can be done in a series of blocks or epochs that takes place at predetermined intervals (usually every 30 s). A "block paradigm" is a term used to describe this type of configuration.
An "event-related paradigm" is another option, in which the stimuli are provided during scanning at random or pseudorandom time points. While event-related paradigms deliver stimuli more realistically, block paradigms have more statistical power for detecting the active brain [8].

There are two main fMRI techniques: the *blood-oxygen-level dependent* (BOLD) technique and the dynamic or exogenous technique. The BOLD technique is preferred since it does not require intravenous contrast media.
The BOLD method makes use of the reduced signal intensity during scanning procedure caused by the conversion of diamagnetic oxyhemoglobin to paramagnetic deoxyhemoglobin that occurs with brain activation [6]. Since oxygenated hemoglobin is diamagnetic, whereas deoxygenated hemoglobin is paramagnetic, this latter results in local proton dipole dephasing and loss of T2* (“observed" or "effective" T2) MRI signal.
Brain regions that are metabolically active as a result of a stimulus or cognitive effort receive substantially more blood than non-active regions due to cerebrovascular autoregulation.
A regional increase in cerebral blood flow caused by active neurons overcompensates for the localized increase in oxygen demand. Therefore, an increase in the T2* signal occurs from a local increase in the oxyhemoglobin to deoxyhemoglobin ratio in metabolically active brain.
A T2* sensitive scanning approach, such as an echo planar-gradient echo (EPI/GRE) pulse sequence, can be used to detect these signal variations (which range from roughly 2% on a 1.5 Tesla MRI scanner to 12% on a 7 Tesla MRI scanner). Repeatedly scanning the brain with an EPI-GRE pulse sequence is the most used method for obtaining an fMRI scan.

## 1.3 Aim of the study

The main purpose of this thesis is to investigate the development of the accessory instrumentation of the 3T PET/RM equipment recently installed in the Marche University Hospital. This instrumentation is used for the administration of sensory stimuli and for the recording of motor responses, both during research protocols and clinical investigations. In particular, the goal is to set up stimulation protocols to be administered during functional studies.

To achieve this aim the following objectives have been accomplished:

1. Study of the manuals relating to the accessory components (hardware and software) for the management and administration of brain stimuli using well-designed paradigms.
2. Analysis of functional and structural brain images performed using the predesigned stimulation tasks.

# 2. NordicNeuroLab integrated products and software

NordicNeuroLab (NNL) is a Norwegian company provider of integrated products and services for fMRI, used to evaluate diseases and injuries that affect brain function. Modern post-processing and visualization software for BOLD fMRI, Diffusion/DTI, and Perfusion DSC/DCE analysis, as well as fMRI hardware for audio and visual stimulation, eye-tracking, and patient response collection, are all included in the product line (Figure 4).

The ambition of this company is to commercialize top neuroimaging technologies while making them simple to use. As a result, NNL has developed comprehensive fMRI systems that contain all the components needed to perform an fMRI scan, also providing modular solutions for both clinical and research needs.

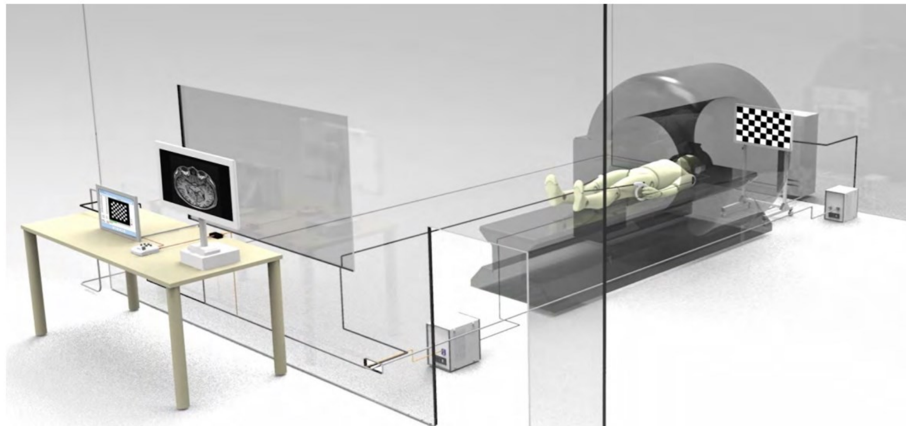

*Figure 4: Examination and scan room with a NNL system overview.*

## 2.1 NordicNeuroLab Hardware components

The hardware package of NNL includes items made specifically for fMRI, providing research and modular solutions.
The devices include standardized interface adapters for audio and visual input and are interoperable with MRI scanners from all major manufacturers.
The fMRI hardware system was designed, developed, and assembled using a Quality Management system certified ISO 13485.

### 2.1.1 *Inroom Viewing device*

The Inroom Viewing device is an LCD screen suitable for 1,5T, 3T, and 7T scanning environments (Figure 5). An optimal choice to provide images or video inside the MRI scan room and a simple alternative to standard projectors or goggle-based image delivery systems, due to its slim design, 40” display with high definition, and excellent picture quality.

The monitor can be easily placed anywhere in the MRI room by using its height-adjustable movable foot stand and thanks to a front-facing camera it can provide continuous patient monitoring during the exam.

By enabling the operating staff to remain in the examination room throughout procedures, the screen improves clinical efficiency by allowing uninterrupted patient care and quick response times.

The principal use for the Inroom Viewing device is to display instructions, words, pictures, videos, and symbols during an fMRI scan procedure; however, it can also provide the patient a break from the exam and calm their tension.

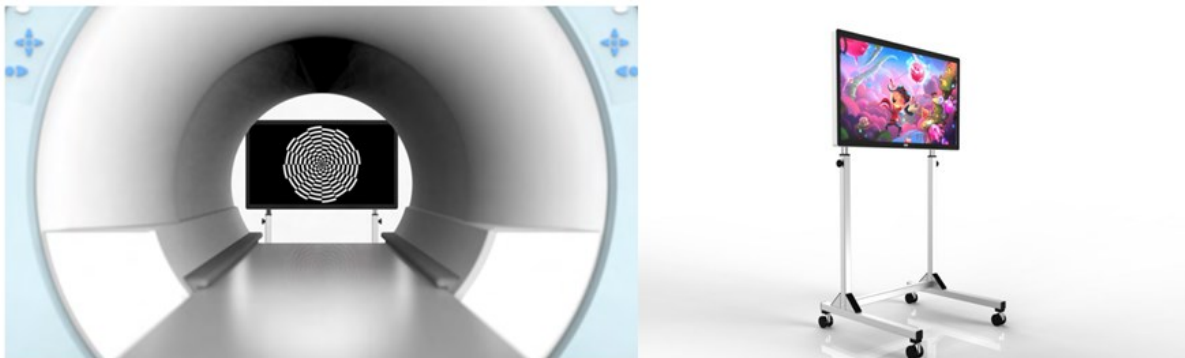

*Figure 5: Inroom Viewing device, respectively, show in full, on the right, and placed in front of the MRI scan tube, on the left.*

### 2.1.2 *VisualSystem HD*

NNL VisualSystem HD is a powerful and adaptable option for providing visual stimuli within the MR scanner (Figure 6). High quality graphics or text are presented to the patient using clear images and vibrant colours. This tool represents new opportunities and challenges for fMRI research and therapeutic settings, immersing the participant or patient in a new environment. Due to the proximity to the eyes, there are no outside distractions present during stimulus presentation, and people are less likely to feel uncomfortable due to the confined environment of the scanner.

The VisualSystem HD has provided clinicians the capacity to follow and record participants' gaze and pupils in real-time, as well as to assess their level of alertness and overall task involvement.

With coil-specific adapters, this component is simple to attach and features a design that fits a variety of head coils. The movable arm enables quick and accurate positioning at the desired viewing angle. The built-in diopter correction and pupil distance fine-tuning are simple to control and tailor to each patient, whether an adult or a young one.

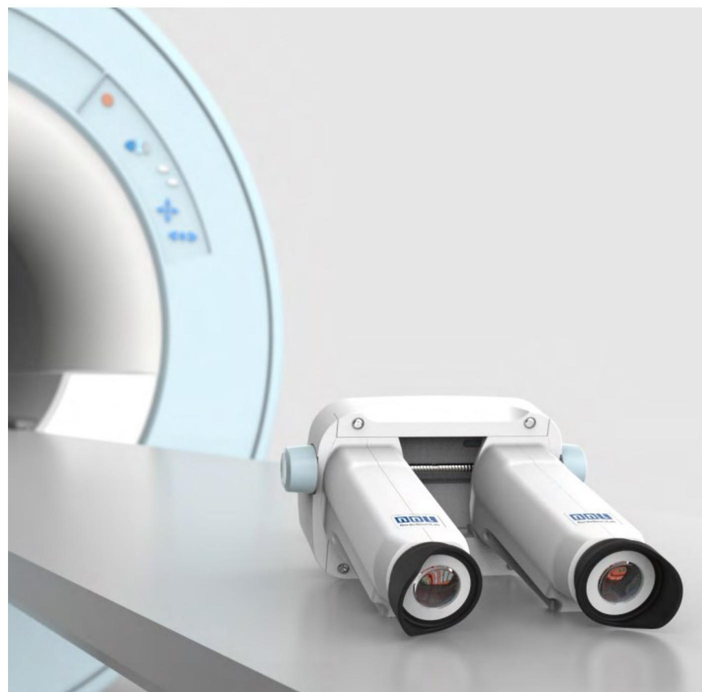

*Figure 6: VisualSystem HD device.*

### 2.1.3 *Audio System*

NNL company provides headphones (Figure 7) characterized by an excellent sound that reproduces even the smallest details with remarkable precision, due to the state-of-the-art electrostatic transducers. During the presentation of audio stimuli, the communication with the patient is uninterrupted, thanks to the device's significant reduction in scanner noise.

Specifically made for fMRI, the improved sound quality and higher noise attenuation provide more precise audio stimulation, resulting in a more significant BOLD response than conventional pneumatic audio systems. The Communication Console allows for one-way patient communication and complete audio setting flexibility through simple and intuitive controls.

*Figure 7: a clinician is placing the NNL headphones on a patient before the scan procedure.*

### 2.1.4 ***SyncBox***

Synchronizing stimulus presentation with MR data acquisition is one of the issues in fMRI. For the validity of the results, timing information must be accurate and verified. Thus, the SyncBox allows to choose how the trigger pulse from the scanner is communicated to the software, presenting the stimuli through a versatile and user-friendly options panel (Figure 8).

This tool, which works with the top software programs, offers a straightforward method for precise control over stimulus presentation and easy accessibility to timing information for data analysis.

The trigger signals generated by the scanner during an MRI sequence can be mimicked by the SyncBox, decreasing the need for testing in an expensive scanning environment by allowing the user to construct and test the whole experimental paradigm in the office.

The device interacts with a range of external tools and is scanner independent, enabling synchronization of data from various hardware sources and accurate time stamp logging.

*Figure 8: SyncBox device.*

There are two "choose modes" on SyncBox's first panel.

By selecting "Synchronization" the device works in synchronization mode, therefore it waits to receive trigger pulses from the scanner. In this way SyncBox will sends the PC activation pulses according to the settings in the menu.

Instead, by selecting "Simulation", the device works simulating the trigger signals according to the specified settings. In simulation mode, the SyncBox can be completely controlled by the PC. Using a serial interface, all settings, starts, and stops can be set from the software.

In the main menu (Figure 9), in line with the acquisition settings, it is necessary to enter the following features:

- Volumes: number of volumes (series of consecutive scans that together sweep out a volume of space) that the SyncBox should expect during a single session.
- Slices: number of slices that compose a single volume (note: if the scanner sends only one trigger signal on each volume, always set this value to 1).
- Pulse length: the time the scanner takes to purchase a single portion.
- TR-time: time between the start of the acquisition of a single volume and the start of the acquisition of the next one. This value can be set only during simulation mode.
- Trigg on slice: the SyncBox will send a signal to the PC on the specified portion number (pressing the right button to select the portion number).
- Trigg on volume: the SyncBox sends a signal to the pc on the specified volumes.

To run synchronization/simulation mode it is necessary to push START SESSION. Instead, to check on PC communication it is necessary to push "send trigger pulse to PC".

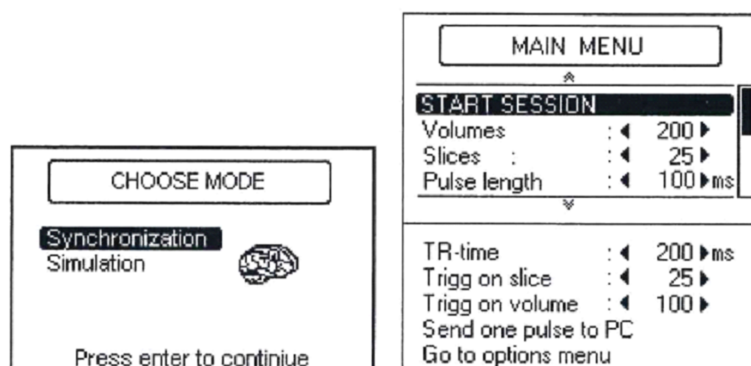


*Figure 9: on the left is shown the panel for the "Choose Mode" in which it is possible to select "Synchronization" or "Simulation" mode; on the right is shown the "Main Menu" in which it is possible to set all the acquisition settings.*

## 2.2 NordicAktiva

With the nordicAktiva software, a single technician can manage stimulus presentation and image acquisition simultaneously. The user is guided step-by-step through the process of presenting stimuli during image acquisition using an intuitive interface and clear, comprehensive instructions that are available in several languages.

During the MRI examination, nordicAktiva displays the paradigm to the patient and runs seamlessly with the fMRI hardware. Automatic MRI acquisition and paradigm presentation synchronization allow the acquisition of reliable fMRI data (Figure 10).

This software gives the option to either use pre-defined paradigms, adapt them based on preferences, or just create a library specifically designed for subjects. Additionally, it supports a variety of audio and video files and paradigms in multiple languages.

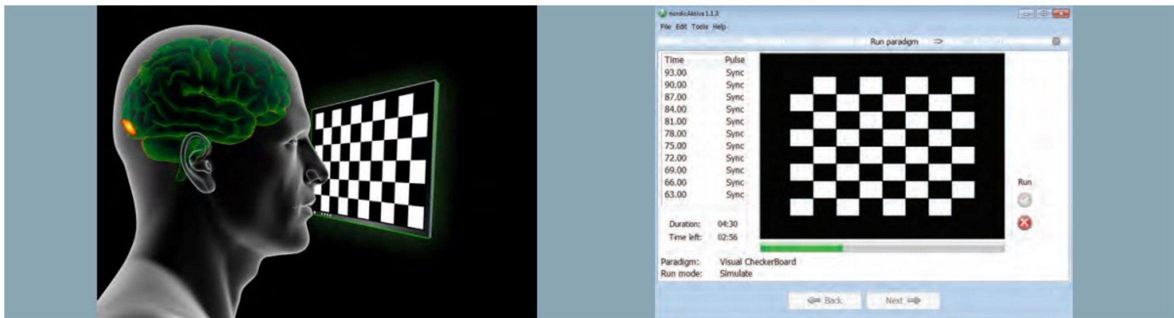


*Figure 10: left panel, example of visual task displayed thanks to nordicAktiva and correlated brain activation on the patient; right panel, screen on the nordicAktiva program when the visual task is run, with the features of a visual stimulation paradigm during the fMRI scan.*

### 2.2.1 *NordicAktiva paradigms*

A paradigm is a temporal sequence of stimuli aimed to elicit BOLD responses from the subjects. Detailed paradigms with stimuli or events are utilized during an fMRI examination to trigger a subject's hemodynamic response or brain activation [7].

The principal paradigms categories involve motor, visual, auditory, and language stimulation. Each of them has clinical relevance, for example, language task paradigms have a significant role in presurgical language assessment in the treatment of patients with brain lesions (or tumours), as well as the visual one for patients with epilepsy.

The following tasks are based on the recommendations provided by the American Society of Functional Neuroradiology, but it is important to keep in mind that field strength, coil design,

distribution systems, other ambient conditions, and post-processing techniques all have an impact on particular paradigm timing parameters [8]. To minimize the widespread variability in clinical fMRI, using standard paradigms is just the first essential step in practice convergence [9].

### 2.2.2 *Auditory Tones*

**Paradigm description:** The Auditory Tones paradigm implies the patient listens to a series of tones presented randomly and click a button with either their right or left hand when they hear the odd (high) tone. This task is alternating with rest intervals.

There is one timing option:

- 30 seconds each; 1 rest plus 4 cycles, 4 minutes 30 seconds.

**Instructions:** A series of tones will be randomly generated, and the patient will hear them throughout the task. A low tone (standard tone) will be presented more frequently than a high tone (odd tone). When the high tone is heard, the patient's assignment is to use either his/her right or left hand to hit a button.
Every auditory stimulation will be followed by a resting interval, during which the patient should remain still and rest while concentrating on the fixation cross in the centre of the screen. There will be multiple repetitions of the auditory stimulation task followed by rest periods, however, the scan will start and end with rest periods. The response to stimuli needs to be quick and accurate as possible.

### 2.2.3 *Breath Hold*

**Paradigm description:** The Breath Hold paradigm requires the patient to hold his/her breath for 20 seconds (4 seconds inhale + 16 seconds hold). This task alternates with rest intervals while the patient is told to breathe normally.

There is one timing option:

- normal breathing for 40 seconds followed by a 20 second breath hold, 4 cycles plus breathing normally, 4 minutes 30 seconds.

**Instructions:** during the task, the patient will be instructed to hold his/her breath for 16 seconds after inhaling for 4 seconds. The countdown is visible on the screen so that the patient will know for how long he/she must hold the breath.
After each breath hold period, there will be a 40 second rest in which the patient should lie still, breathe normally, and concentrate on the text in the centre of the screen.
There will be multiple repetitions of the breath hold task followed by normal breathing, however, the scan will begin and end with a period of normal breathing.

### 2.2.4 *General Motor Left*

**Paradigm description:** a flashing green dot on the left side of the screen indicates the left user-defined motor task. The task is alternating with rest intervals (Figure 11).

There are two time options available:

- 20 seconds each, 1 rest period plus 4 cycles of activity and rest, 3 minutes.
- 30 seconds each, 1 rest period plus 4 cycles of activity and rest, 4 minutes 30 seconds.

**Instructions:** the patient will see a flashing green dot on the left side of the screen during the General Motor Left task, which requires performing a left-hand motor task that the operator defines. After each motor task session, there will be a rest period where the patient should keep still and concentrate on the fixation cross in the centre of the screen.
There will be multiple repetitions of the task followed by rests, however, the scan will start and end with rest periods.

*Figure 11: on the left, it is pictured an example of what the patient sees during a Left Motor Task; the flashing green dot on the left part of the screen gives the signal to do the left motor task. On the right, it is shown what is displayed during the rest interval.*

Examples of motor tasks:

- Finger tapping, the challenge is to quickly tap the left-hand fingers (right-hand fingers in the General Motor Right task) onto the thumb. If the patient is incapable of touching each finger separately, he/she can either tap all the fingers simultaneously or even clench the fingers into a fist.
- Toe movement, in which the patient has to flex and extend his/her left toes (right toes in the General Motor Right task) as fast as possible.

### 2.2.5 *General Motor Left Right*

**Paradigm description:** A flashing green dot on the left or right side of the screen signals a user-defined right and left-side motor task (Figure 12).

Two time options available:

- 20 seconds each, 1 rest period plus 3 cycles of activity and rest, 3 minutes 20 seconds.
- 30 seconds each, 1 rest period plus 3 cycles of activity and rest, 5 minutes.

**Instructions:** A flashing green dot will appear on the left or right side of the screen when the patient is performing the General Motor Left Right task. Depending on whether the dot is on the right or left side of the screen, the assignment is to execute a right or left motor task specified by the operator. There will be rests between motor task periods where the patient should be still and focus on the fixation cross in the centre of the screen.

There will be multiple repetitions of the task followed by rests, however, the scan will start and end with rest periods.

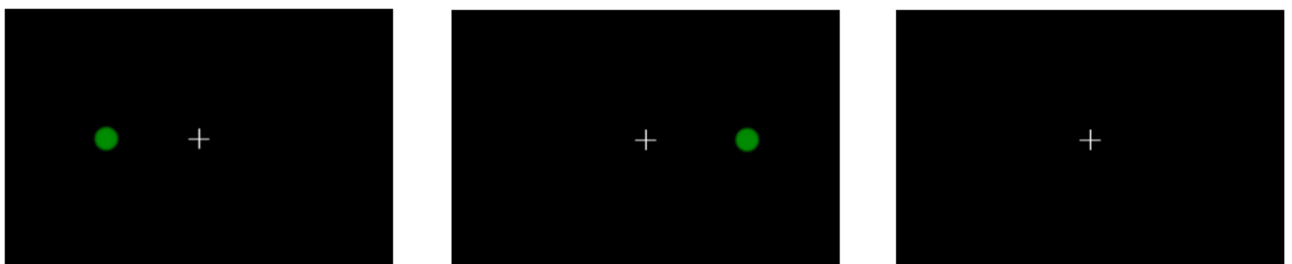

*Figure 12: the left and central pictures represent what is shown during the General Motor Task Left Right; the flashing green dot alternates on the left and right part of the screen gives the signal to do a left or right motor task. On the right, it is shown what is displayed during the rest interval.*

### 2.2.6 *General Motor Right*

**Paradigm description:** a flashing green dot on the right side of the screen indicates the right user-defined motor task. The task is alternating with rest intervals (Figure 13).

There are two time options available:

- 20 seconds each, 1 rest period plus 4 cycles of activity and rest, 3 minutes.
- 30 seconds each, 1 rest period plus 4 cycles of activity and rest, 4 minutes 30 seconds.

**Instructions:** the patient will see a flashing green dot on the right side of the screen during the General Motor Right task, which requires them to perform a right-hand motor task that the operator defines. After each motor task session, there will be a rest period where the patient should keep still and concentrate on the fixation cross in the centre of the screen.
There will be multiple repetitions of the task followed by rests, however, the scan will start and end with rest periods.

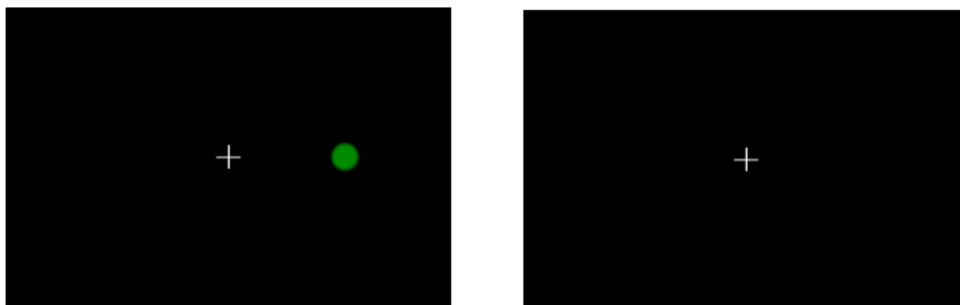

*Figure 13: on the left, it is pictured an example of what the patient sees during a Right Motor Task; the flashing green dot on the right part of the screen gives the signal to do the right motor task. On the right, it is presented what is displayed during the rest interval.*

### 2.2.7 *General Motor Start Stop*

**Paradigm description:** Displaying a visual START or STOP signal indicates the START and STOP of user-defined tasks (Figure 14).

Two timing alternatives are available:

- 20 seconds each, 1 rest period plus 4 cycles of activity and rest, 3 minutes.
- 30 seconds each, 1 rest period plus 4 cycles of activity and rest, 4 minutes 30 seconds.

**Instructions:** A START sign will be displayed in the centre of the screen while the General Start Stop task is performed. When the START sign appears, the patient's job is to execute a motor task established by the operator. After each motor task period, there will be a rest period signed by a STOP symbol. During this time, the patients should remain still and keep their attention on the STOP symbol in the centre of the screen.

There will be multiple iterations of the START and STOP periods, but the scan will start and end with a rest period.

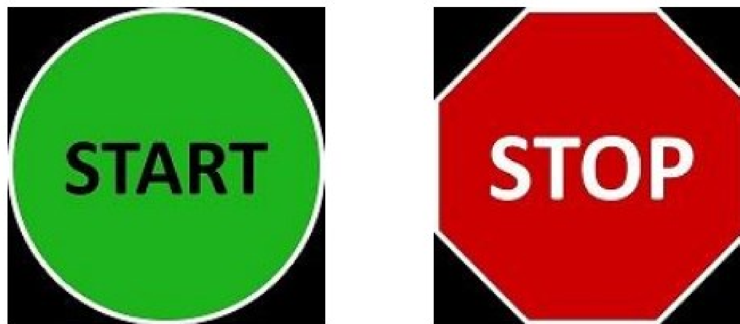


*Figure 14: on the left picture it is shown the START sign which indicates the patient to execute a designed motor task; on the right, instead, it is shown the STOP sign which indicates the end of the task and so the beginning of the rest period.*

Examples of motor tasks:

- <u>Tongue movement</u>, when the START sign appears, the patient has to move the tongue along the back of his/her upper teeth (a very slight movement with the lips closed) and stop the task when the STOP sign appears.
- <u>Toe movement</u>, which consists of simultaneously flexing and extending both right and left toes quickly when the START sign appears and stopping the activity when the STOP sign appears.

### 2.2.8 *Language Sentence Completion*

**Paradigm description:** the Sentence Completion paradigm involves a task in which the patient is instructed to silently complete blanks in sentences displayed on a screen. The task is alternating with rest intervals in which are shown groups of scrambled letters (Figure 15).

There are two time options available:

- 20 seconds each, 1 rest period plus 6 cycles of activity and rest, 4 minutes 20 seconds.
- 30 seconds each, 1 rest period plus 4 cycles of activity and rest, 4 minutes 30 seconds.

**Instructions:** during the task, the patient will view sentences that end in a blank space, so the goal is to silently complete the sentence without moving the head or the lips. Each sentence completion session will include several different sentences, and after each session, there is a rest period where the patient will see scrambled letters to look like text.
There will be multiple repetitions of the task followed by rests, however, the scan will start and end with rest periods.

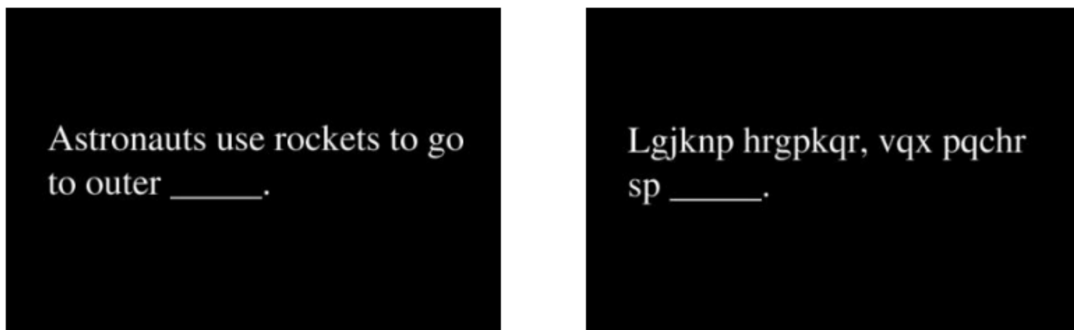


*Figure 15: on the left, there is an example of a sentence presented during the Language Sentence Completion task; the patient during the fMRI scan will silently read the sentence and complete the blank space without saying the word out loud. On the right, there is the screen displayed during the rest interval in which is presented a nonsense text.*

### 2.2.9 *Language Silent Word Generation*

**Paradigm description:** the Silent Word Generation paradigm asks the patient to create as many words as they can using the letters that are randomly shown on the screen. The patient will see nonsense symbols during the rest interval in between these tasks (Figure 16).

Two timing alternatives are available:

- 20 seconds each, 1 rest period plus 6 cycles of activity and rest, 4 minutes 20 seconds.
- 30 seconds each, 1 rest period plus 4 cycles of activity and rest, 4 minutes 30 seconds.

**Instructions:** the patient will be shown letters one at a time appearing on the screen for a few seconds, so the task is to silently think as many words as they can that begin with the given letter, without moving the head or mouth. During each word generating period, different types of letters will be given. Every active session alternates with a rest period in which random symbols are presented in the centre of the screen, and the patient rests focusing on the symbol without moving the head. The scan will begin and end with a rest period.

*Figure 16: left, the image represents the screen during a Silent Word Generation task, in which the patient sees the letter on the centre of the screen and thinks of as many words as they can that begin with the given letter; right, the image displays the random symbol presented on the screen during rest.*

### 2.2.10 *Language Rhyming*

**Paradigm description:** the Rhyming paradigm asks the patient to determine whether two words on the screen rhyme (optionally, if they do, the patient can voluntarily click a button on the Response Grips) (Figure 17). This task is followed by one in which the patient is shown pairs of symbol strings and asked whether they match or not (optionally, if they do, clicking a button on the Response Grips).

Two time options are available:

- 20 seconds each, 1 control plus 6 cycles, 4 minutes 20 seconds.
- 30 seconds each, 1 control plus 4 cycles, 4 minutes 30 seconds.

**Instructions:** during the task pairs of words will be displayed on the screen for a few seconds, and it is up to the patient to determine whether the words rhyme (if they do, the patient can press the button with the left hand). Each block of rhyming words will provide a variety of word pairs.

Every rhyming period will be followed by a symbol-matching period where the patient will see pairs of symbol strings in the centre of the screen, and he/she should decide whether the symbol strings match (if they do, the patient can press the button with the left hand). Each symbol-matching session will provide different pairs.
Rhyming and symbol matching will alternate multiple times, but the scan will start and end with symbol matching.

*Figure 17: on the left, it is shown an example of pairs of words displayed during the Language Rhyming task; the patient during the exam has to determine whether the words rhyme. On the right, it is shown an example of symbol strings displayed during the Symbol-matching period which follows the rhyming one; the patient has to determine whether the symbol strings match.*

### 2.2.11 *Language Object Naming*

**Paradigm description:** the Object Naming paradigm involves asking the patient to silently name various random object drawings/pictures displayed on the screen. If the image depicts a living object, the patient can be directed to press a button with their left hand to enhance more complex semantic processing. The task alternates with a rest period in which confused images of the same objects are shown (Figure 18).

There are two timing alternatives:

- 20 seconds each, 1 rest period plus 6 cycles of activity and rest, 4 minutes 20 seconds.
- 30 seconds each, 1 rest period plus 4 cycles of activity and rest, 4 minutes 30 seconds.

**Instructions:** the patient will see one black and white image at a time on the screen for a few seconds throughout the task. The goal is to silently name the object in the photo without moving the head or lips. In each object-naming period, different pictures will be shown. Optional, it is possible to press a button with the left hand if the image depicts an alive object. After each session, there will be a rest during which a scrambled image will appear in the centre of the screen. The patient should stay calm throughout the rest phase, keeping the attention on the scrambled image.
The scan will begin and end with a rest period.

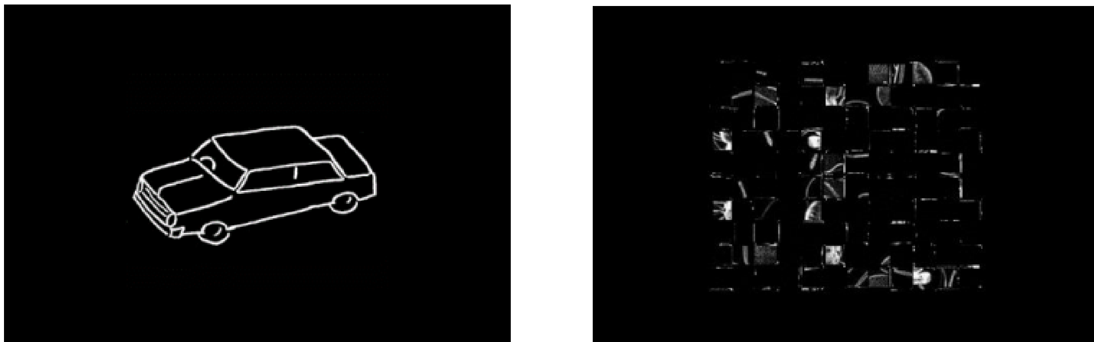

*Figure 18: left, example of objects displayed on the screen during the Object Naming task; the patient has to silently name the objects during the exam. Right, example of a scrambled image that appears during the rest interval.*

### 2.2.12 *Language Antonym Generation*

**Paradigm description:** the Antonym Generation paradigm consists of asking the patient to think silently to words opposites of those displayed on the screen. A rest period during which the patient is shown random sequences of symbols alternates with this task (Figure 19).

Two time options are available:

- 20 seconds each, 1 rest period plus 6 cycles of activity and rest, 4 minutes 20 seconds.
- 30 seconds each, 1 rest period plus 4 cycles of activity and rest, 4 minutes 30 seconds.

**Instructions:** during the task, one word at a time will stay on the screen for a few seconds, so the job is to silently consider the term that denotes the opposite of the word displayed without making any facial or mouth movements. After each antonym generating period, there will be a rest session where the patient will see a series of symbols in the centre of the screen. The patient looks at the succession of symbols during the rest, as though reading without moving their head.
There will be multiple repetitions of the task followed by rests, however, the scan will start and end with rest periods.

*Figure 19: on the left, it is pictured an example of a word presented during the Antonym Generation task, in which the patient has to silently think of a term that indicates the opposite of the displayed word. On the right, there is an example of symbol series presented during rest.*

### 2.2.13 *Language Synonyms*

**Paradigm description:** in the Synonyms paradigm is asked to determine if two word pairs displayed on the screen are synonyms (if they are, optionally, the patient can press a button on the Response Grips with the left hand). This task is followed by one in which the patient is shown pairs of symbol strings and asked to determine if they match (Figure 20).

Two timing alternatives are available:

- 20 seconds each, 1 control plus 6 cycles, 4 minutes 20 seconds.
- 30 seconds each, 1 control plus 4 cycles, 4 minutes 30 seconds.

**Instructions:** when performing the Synonym task, pairs of words will be displayed on the screen for a few seconds, so the patient has to determine whether the words are synonyms, i.e. if their meanings are similar (if they are, answer by pressing a button with your left hand). Each synonym period will have different word pairings.

A symbol-matching task time will come after each synonym phase, during which the patient will view pairs of symbol strings in the centre of the screen and determine if the symbol strings match. If they do, he/she must click a button with his/her left hand to indicate the response. Each block of symbol matching will have several different pairs. The synonym and symbol-matching task will switch off multiple times, but symbol-matching will start and terminate the scan.

*Figure 20: left, example of pairs of words displayed during the Synonyms task, so that the patient has to determine whether the words are synonyms. Right, example of pairs of symbol strings that are shown during the symbol-matching task which follows the synonyms one.*

### 2.2.14 *Language Tones Naming*

**Paradigm description:** the Tones Naming paradigm consists of a responsive naming task during which the patient silently answers questions. This task is followed by a control session where a series of random tones are presented.

There is one timing option:

- 20 seconds each, 1 rest period plus 6 cycles of activity and rest, 4 minutes 20 seconds.

**Instructions:** the patient will hear a simple sentence in the form of a question during the task (for example: "What colour is the ocean?"), so the job consists in silently considering the response laying very still, and without making any lip or head movements. After each response naming period, there will be a rest interval during which the patient will hear a series of random tones. For a few seconds, a low tone (standard tone) will be introduced more frequently than a high tone (odd tone). The patient should be completely still while keeping their eyes on the cross in the centre of the screen and listening for the high tone (no response needed).
The responsive naming task and auditory tones period will alternate several times; however, the scan will begin and end with a rest period.

### 2.2.15 *Language Verb Generation*

**Paradigm description:** in the Verb Generation paradigm random nouns are displayed on the screen, and the patient is asked to think of as many verbs related to that noun as possible. A rest interval during which the patient is shown a string of symbols alternates with this task (Figure 21).

Two timing alternatives are available:

- 20 seconds each, 1 rest period plus 6 cycles of activity and rest, 4 minutes 20 seconds.
- 30 seconds each, 1 rest period plus 4 cycles of activity and rest, 4 minutes 30 seconds.

**Instructions:** one word at a time, for a few seconds, will be displayed on the screen so that the patient's task is to silently list as many verbs as they can, without moving their head or lips, related to the given word. Each verb generation period will include several different words. Following the task session, there is a rest interval in which a series of symbols appear in the centre of the

screen. During the rest, the patient should look calm and still at the symbols without moving their head.
The scan will begin and end with a rest period.

*Figure 21: on the left, it is presented an example of a word that is displayed during the Verb Generation task, in which the patient has to silently list as many verbs as they can relate to the given word. The right picture is presented as an example of what is displayed during a rest period.*

### 2.2.16 *Language Passive Story Listening*

**Paradigm description:** in the assigned task, the patient listens to passages from the story "The Cat in the Hat". This period is followed by a rest interval where the patient hears the same extracts read backward.

One timing alternative is available:

- 20 seconds each, 1 rest period plus 6 cycles of activity and rest, 4 minutes 20 seconds.

**Instructions:** the patient will hear extracts from a book during the ask and pay close attention to the story, but no response is required. Every story listening period, which lasts several seconds, is followed by a reversed speech interval where the patient should remain calm, rest, and keep their eyes on the fixation cross in the centre of the screen.
The scan will begin and end with a reversed speech period.

### 2.2.17 *Visual Checkerboard*

**Paradigm description:** the Visual Checkerboard paradigm involves having the patient concentrate on a flickering black-and-white checkerboard (8 Hz). The task alternates with rest intervals during which the patient focuses on the fixation cross. The scanning personnel must know before the scan starts if the patient has ever experienced epileptic seizures when exposed to flashing lights or patterns (Figure 22).

One timing alternative is available:

- 30 seconds each, 1 rest period plus 4 cycles of activity and rest, 4 minutes 30 seconds.

**Instructions:** the patient will see a flickering checkerboard on the screen for several seconds while performing the Visual Checkerboard task, and he/she is asked to stare at it as long as it remains on the screen without moving the head. The patients should keep still and keep their attention on the fixation cross in the centre of the screen during the resting intervals that come after each flashing checkerboard period.
The scan will begin and end with a rest interval.

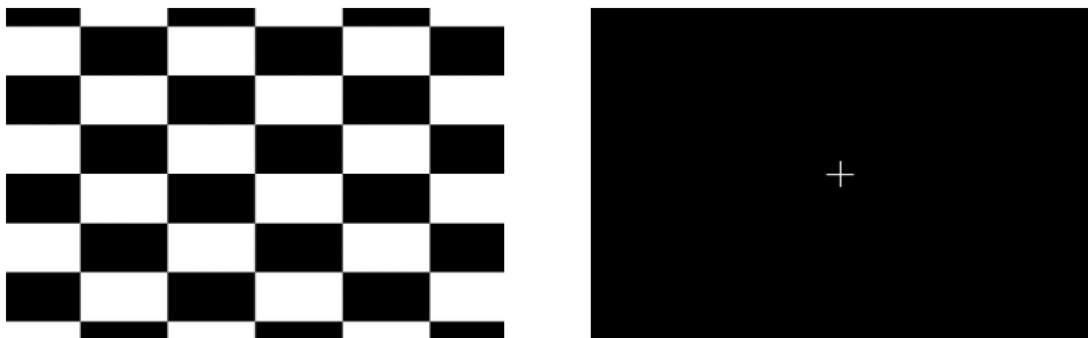

*Figure 22: The left picture is presented as an example of a flickering checkerboard displayed during the Visual Checkerboard task. On the right, there is a picture representing what is displayed during rest phase.*

# 3. Experimental part

The present section explains the MRI acquisition parameters and the pre-processing steps used to analyse the fMRI data obtained from the subject involved in this investigation during auditory, motor and visual stimulation. The research for this thesis was carried out in conjunction with the Department of "Scienze Cliniche Specialistiche ed Odontostomatologiche" of Università Politecnica delle Marche and the Riuniti Hospital of Ancona.

## 3.1 Participant

For this study, data were acquired from one right-handed healthy subject S1. MRI acquisitions periods range between November 2022-January 2023. Table 1 reports the specifics of the clinical subject.

*Table 1: patient clinical information*

| Case | Gender | Age at testing | Weight (Kg) | Pacemaker wearer | Metal prosthesis wearer | State of pregnancy |
|---|---|---|---|---|---|---|
| S1 | F | 25 | 50 | NO | NO | None |

## 3.2 MRI data acquisition

Data sessions were constituted of three different trials:

- General Motor Left task: the personnel instructed the subject during the scan procedure to open and close the left hand.
- Auditory task: during the procedure the subject listened to classic music, that was reproduced in the scan machine thanks to the integrated speaker.
- Visual checkerboard task: thanks to the VisualSystemHD mounted in the scan machine, the subject saw during the fMRI acquisition the visual paradigm displayed through the nordicAktiva software.

Each task has 5m of duration, composed of 5 cycles of 60s each, in which there is the alternation of 30s-resting state and 30s-stimulation period.
For the visual checkerboard task, the time session in the main file of the paradigm code was modified. The software default time was 4 minutes 30 seconds, but it was changed to 5 minutes by varying the "ShowTime" variable. Also, it was added an initial phase at rest by using a "waiting" block.

During the resting state fMRI acquisition, the subject was instructed to lie down and stay calm as possible, to keep the eyes open and relax.
BOLD fMRI data and T1-weighted structural images were collected utilizing a 3T Signa PET/MR scanner.

The functional images were acquired through a gradient-echo EPI sequence with the following settings:

- TE: 83 ms
- TR: 3000 ms
- Flip angle: 90°
- FOV: 64 x 64 mm
- Number of volumes: 98 (100 acquired but 2 skipped)
- Number of axial slices: 36
- Slice thickness: 4 mm (0 mm gap thick)
- Voxel resolution: 3.75 x 3.75 x 4.0 mm
- fMRI duration: 300 s (5 min and 12 s but the first 12 s were cut out).

A T1-weighted structural image was collected through an MPRAGE sequence for anatomical localization with 3D data. The parameters used were:

- TE: 3.2 ms
- TR: 8.5 ms
- FOV: 256 x 256 mm
- Number of sagittal slices: 257
- Slice thickness: 1 mm (0 mm gap thick)
- Voxel resolution: 0.5 x 1.0 x 1.0 mm

## 3.3 Pre-processing

After data collection, BrainVoyager, a specialized, all-inclusive, powerful, cross-platform neuroimaging software, was used to process the structural and functional MRI images.

Several pre-processing steps are commonly applied to raw data in fMRI to remove artifacts and noise components: brain extraction, motion correction, slice time correction, temporal filtering, and registration.

### 3.3.1 *Brain extraction*

To distinguish between brain and other tissue, the T1-weighted structural data underwent the first pre-processing step, the brain extraction. Since both brain and non-brain tissue can be detected in structural images, the non-brain tissue areas need to be removed before the registration phase is carried out, to improve the robustness of the process.

This step is an automatic process with four steps: background cleaning, brain extraction, white matter detection, and estimation of the bias field within white matter voxels [10].

In BrainVoyager, is possible to do it, enhancing the anatomical data set's quality, using the automatic intensity inhomogeneity correction tool (auto-IIHC) (Figure 23).

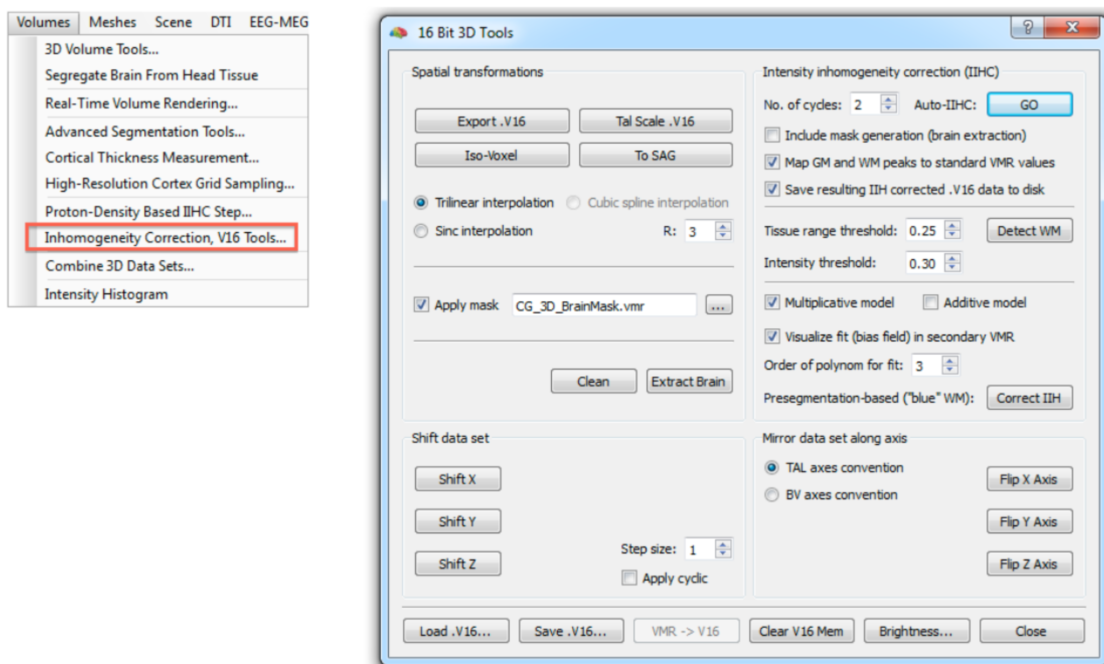


*Figure 23: Intensity Inhomogeneity correction tool to brain extraction in BrainVoyager.*

### 3.3.2 *Motion correction*

Small movements of the subject during the scan procedure can lead to important artifacts, bringing an impairment in the quality of the fMRI data. For this reason, motion correction or realignment is the main pre-processing procedure.
Because of the head movements, subsequent slices may not align correctly, resulting in incorrect anatomical placements between voxels in future images. Different head immobilization systems exist, yet they cannot totally stop movements, especially unintentional ones [11].

Motion correction works by choosing one functional volume from a run (or another one from the same scanning session) as a reference to which all other functional volumes are aligned. The majority of algorithms use six parameters to represent head movements: three for translation (displacement) and three for rotation. The x, y, and z axes, as well as rotation about these, can be used to represent any spatial displacement of rigid bodies, making these parameters adequate for describing rigid body motion.

In BrainVoyager, to run this step "3D motion correction" was selected from the FMR Data Preprocessing menu, with values left as default (Figure 24).

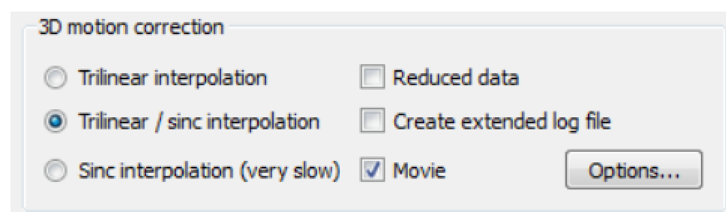


*Figure 24: options available to control 3D motion correction in BrainVoyager.*

### 3.3.3 *Slice time correction*

A crucial aspect of pre-processing is the correct management of differences in time when individual slices are acquired. The concern about different slice scanning times results from the fact that a functional volume (such as the entire brain) is typically covered by a sequence of successively measured 2D slices, and not covered at once.
Slices can be recorded either sequentially, in ascending or descending order or interleaved (the odd slice numbers are recorded first and followed by the even slice numbers).

To correct the problem of slice acquisition delays, the sequences of individual slices are "shifted" in time to match a reference time point, such as the first or middle slice of a functional volume. When implementing the data-shifting method to correct timing differences, the data must be re-sampled at time points between the measured data points. Considering measurable data points "in the neighbourhood" (from time points measured close by), it is possible to approximate the values at the unmeasured time points.

In BrainVoyager, three interpolation methods are available: linear, cubic spline and windowed sinc interpolation (Figure 25).
In the "FMR Data Preprocessing" was selected Slice time correction and the cubic spline as interpolation function, with ascending order. The cubic spline was chosen because it avoids smoothing the data by using more points in the neighbourhood, leading to a very accurate resampling.

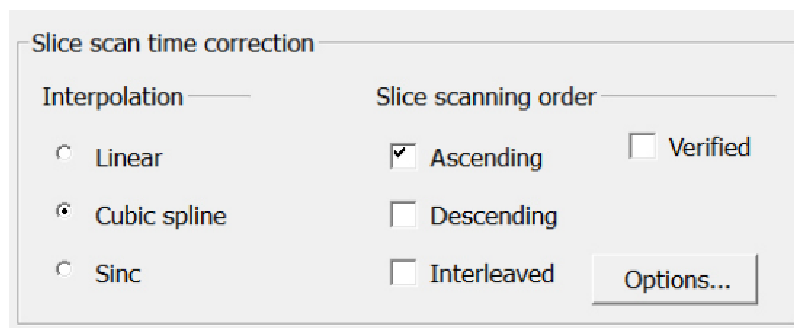


*Figure 25: BrainVoyager window for slice scan time correction settings.*

### 3.3.4 *Temporal filtering*

Low-frequency drifts are frequently visible in the voxel time courses of fMRI data, and they are assumed to be the result of both physiological and physical (scanner-related) noise.
The suppression of these undesirable signal components from each voxel's time series is one of the most crucial pre-processing steps, and it should always be done due to the significant impacts on fMRI data analysis. Nevertheless, this temporal filtering step is also one of the most dangerous since, if implemented incorrectly, condition-related signal changes may also be removed, losing important information.
In principle, fMRI data is high-pass filtered, so the lowest frequencies, ideally lower than the low frequency fluctuations representative of the BOLD signal, are removed from the data.

Cut-off frequency, expressed in Hertz, or cut-off period, expressed in seconds, are two features to represent the quantity of temporal filtering that has to be applied, and they reflect on the quality of the data. Higher cut-off frequencies (0.001 Hz) can be applied to obtain high quality datasets and remove less noise while retaining more data, whereas lower cut-off frequencies (0.01 Hz) are frequently used with low quality datasets to eliminate more noise [12].

In BrainVoyager it was applied a high-pass filter with a cut-off of 0,015 Hz (Figure 26).

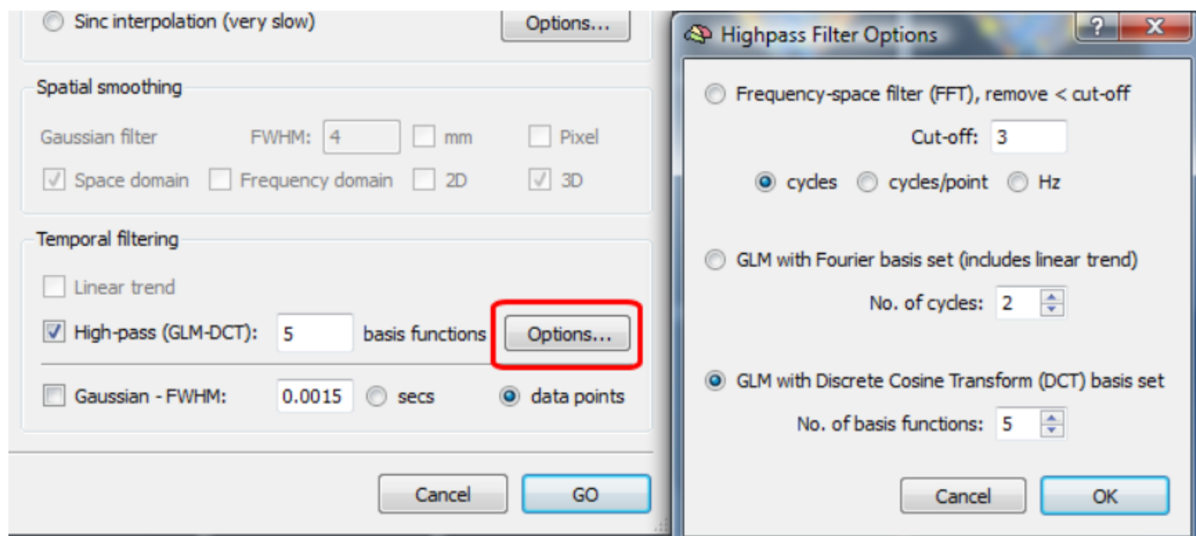


*Figure 26: BrainVoyager Highpass Filter Options in which it is possible to set the cut-off frequency (in cycles) and the type of filtering.*

### 3.3.5 *Registration*

Since the same subject's functional images (collected while performing a task or at rest) and T1-weighted structural images were both recorded during fMRI trials, the registration step is essential to establish a voxel-anatomical relationship between them.

The functional image, derived from the BOLD signal, has a lower spatial resolution than the structural image. It is very important to align the functional image with the T1-weighted structural image of the subject to define and properly recognize the regions of interest within the functional image, interpret the results, and understand in which brain regions the voxels are activated during the scan procedure.

The subject's functional and structural images are both present in the native space where the data were collected, but it is necessary to align them with a standard space. This procedure leads to results more easily compared and integrated when a comparison between several subjects is performed, because the human brain varies from one person to another in terms of both size and shape.

All acquisitions can be aligned to a particular template, that is an image representing the atlas (a reference for locating the activation and interpreting the outcome) and that provides a target for the alignment of individual images [6]. One of the most common atlases is the Talairach atlas.

This atlas was created by Jean Talairach, and it is modelled on the post-mortem brain of a 60-year-old woman. Anatomical landmarks such as the anterior commissure (AC), posterior commissure (PC), midline sagittal plane, and the external boundaries of the brain at each edge provide the basis for his "three-dimensional proportionate grid" (Figure 27). These markers allow us to define the origin (zero-point) as the point where the AC joins the midline sagittal plane in three dimensions. Following this, the axial plane is defined as the plane parallel to the AC/PC line that is orthogonal to the midline sagittal plane, and the coronal plane is defined as the plane that is orthogonal to the sagittal and axial planes. Additionally, the space has a boundary box that indicates its extent in each direction and is defined by the brain's most extreme regions in each direction [13].

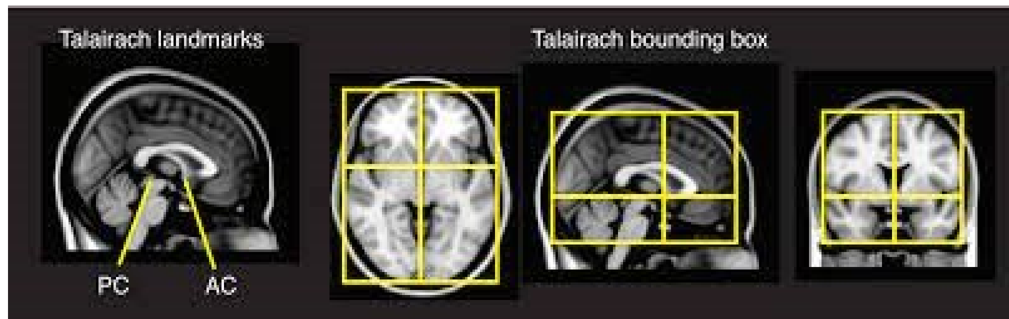


*Figure 27: on the left it is pointed out the Talairach landmarks AC and PC. The Talairach space boundary box is identified on the right [5].*

The main two steps of registration are the *co-registration* and the *normalization*.
The first step is the co-registration, in which the pre-processed functional images are realigned on the appropriate T1-weighted anatomical image (-weighted). The aim is to superimpose the data from the functional images onto an image that allows for the differentiation of anatomical regions. In BrainVoyager this is carried out by opening the "3D Volume Tools" in the co-registration section and aligning the functional (FMR) and structural images (VMR), by running both the Initial Alignment and Fine-Tuning Alignment.
The subject's images (both functional and anatomical) are registered to the Talairach space in the second stage, which is referred to as normalization.

# 4. Results

The results of the analysis discussed in chapter 3 are presented in this chapter.

## 4.1 General Motor Left task

Figure 28 shows the pre-processed functional 3D image, acquired during the General Motor Left task activity.

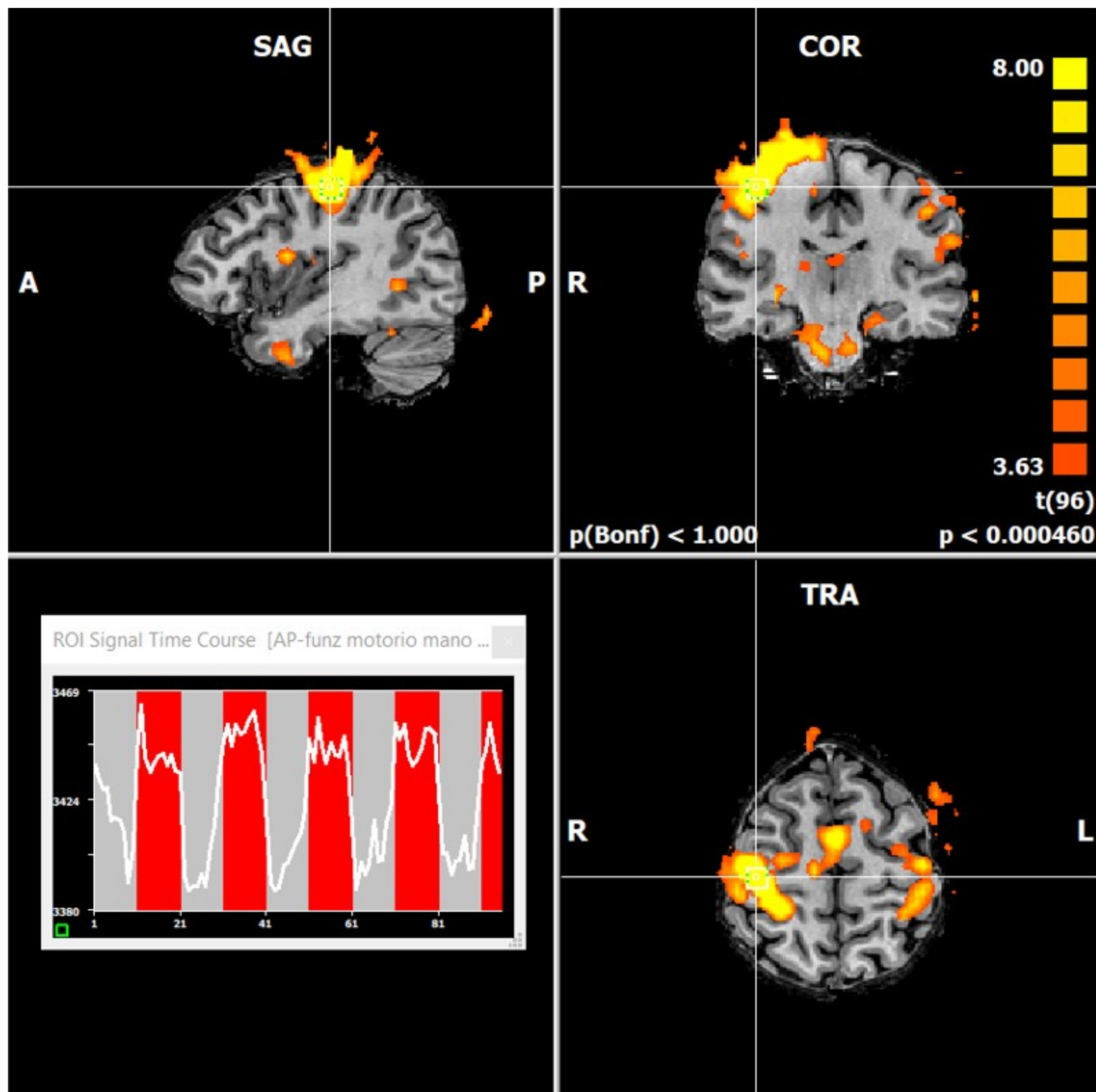


*Figure 28: functional 3D image of the general motor left task activation represented in the three main planes (sagittal, coronal and transverse).*
*In the bottom left box, it is represented the Time Course window, which shows the time course of regions-of-interest (ROIs). This signal represents the average time course from all voxels in the corresponding ROI. The x-axis of the window displays time while the y-axis displays the fMRI signal strength (raw values).*

In figure 29 it is presented the results of the inhomogeneity correction using auto-IIHC.

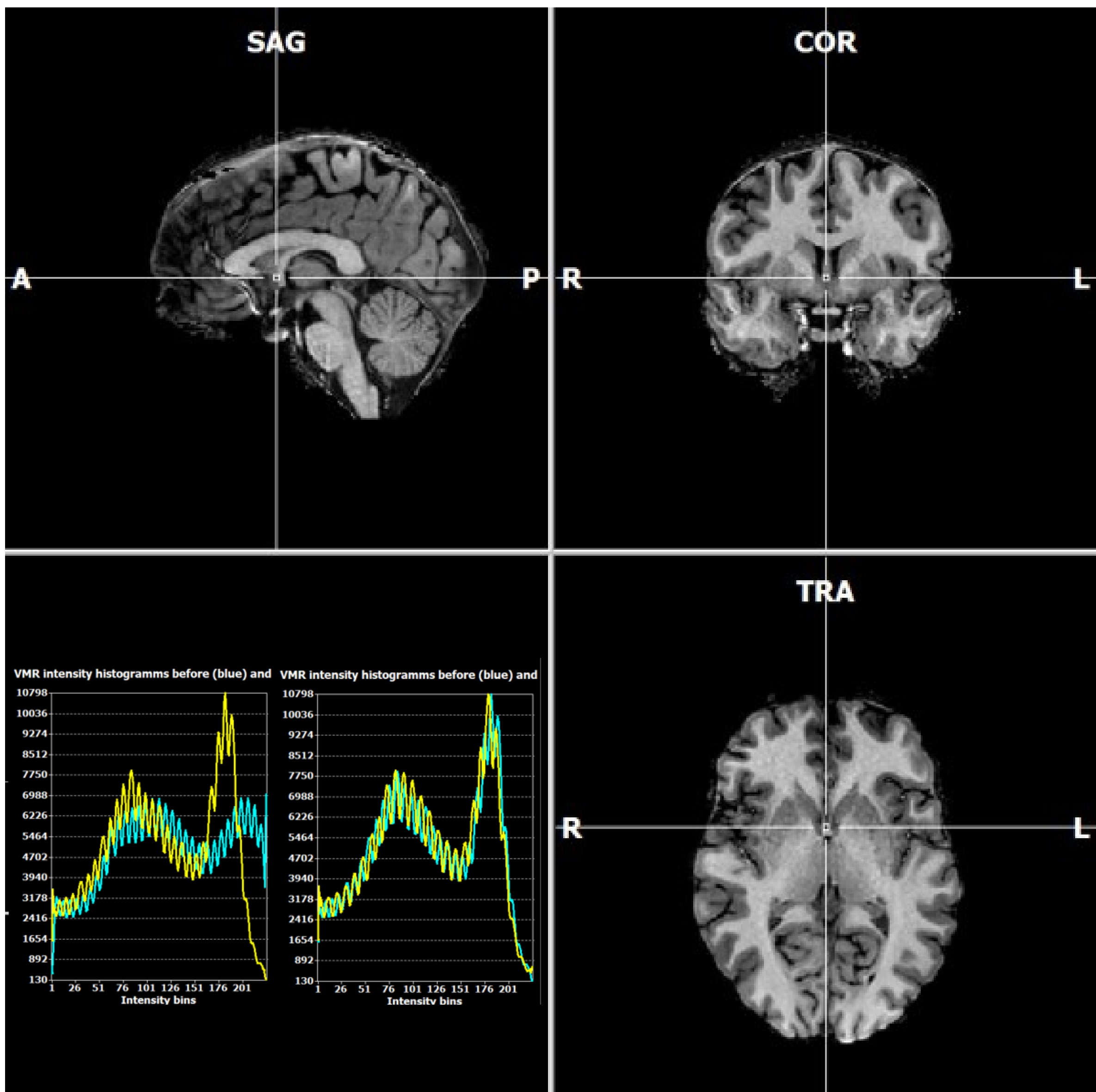


*Figure 29: 3D anatomical images of the brain after the brain extraction step in sagittal, coronal, and transverse planes. The panel on the left below shows the VMR (3D anatomical file) Intensity histograms. Each histogram shows the result before (blue line) and after (yellow line) application of one cycle of correction. The plot on the left side displays the first iteration, instead on the right side the second iteration, in which further improvement is visible.*

Figure 30 and 31 shows the brain images in the three main planes after the co-registration and normalization steps.

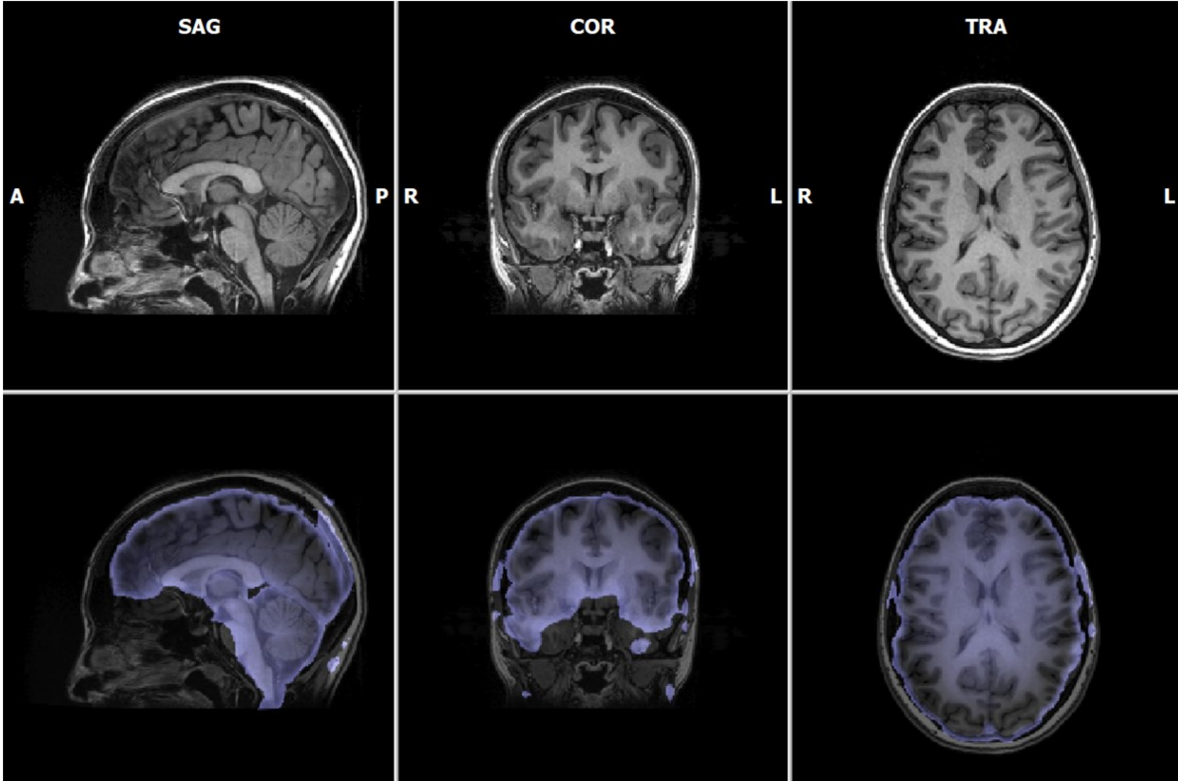


*Figure 30: Coregistration of functional pre-processed file (transparent) in structural IIHC image.*

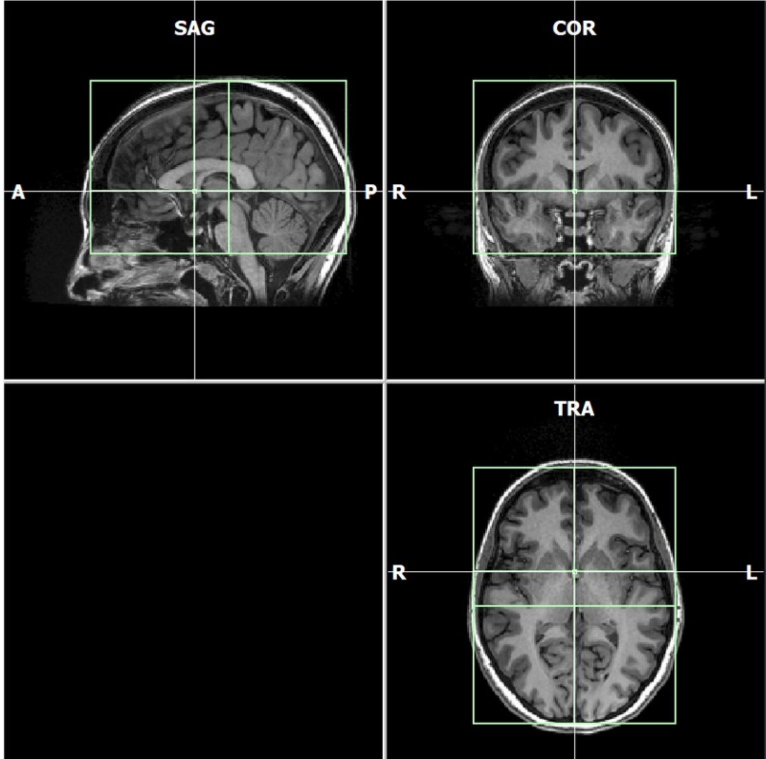


*Figure 31: Normalization in the Talairach space.*

## 4.2 Auditory task

Figure 32 shows the functional images of the 36 slices, instead Figure 33 shows the pre-processed VTC image (Talairach + 3D structural + functional image), both acquired during the auditory task activity.

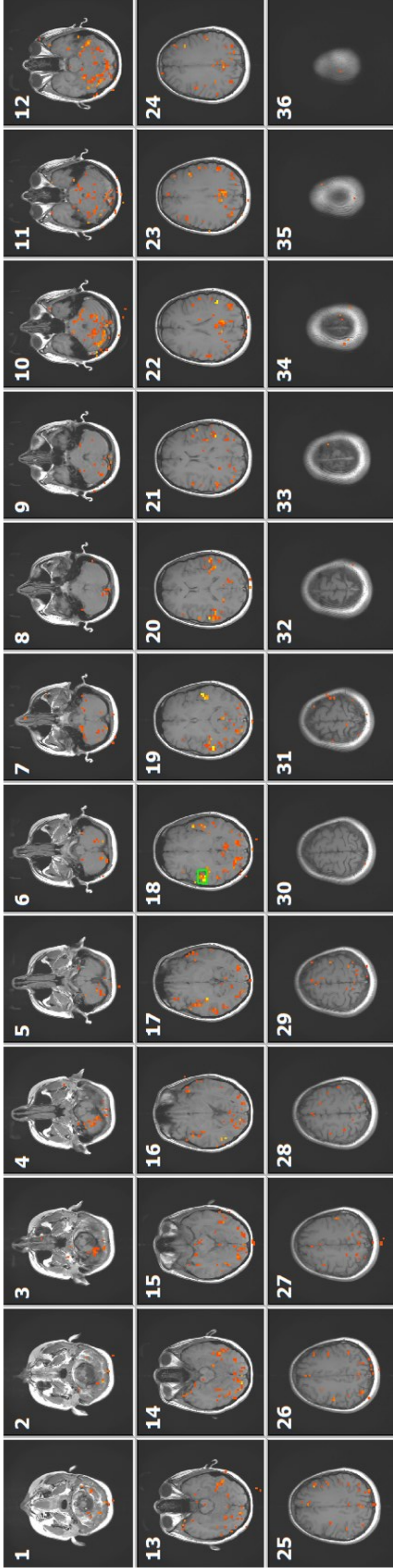

*Figure 32: Functional auditory task images of all 36 slices.*

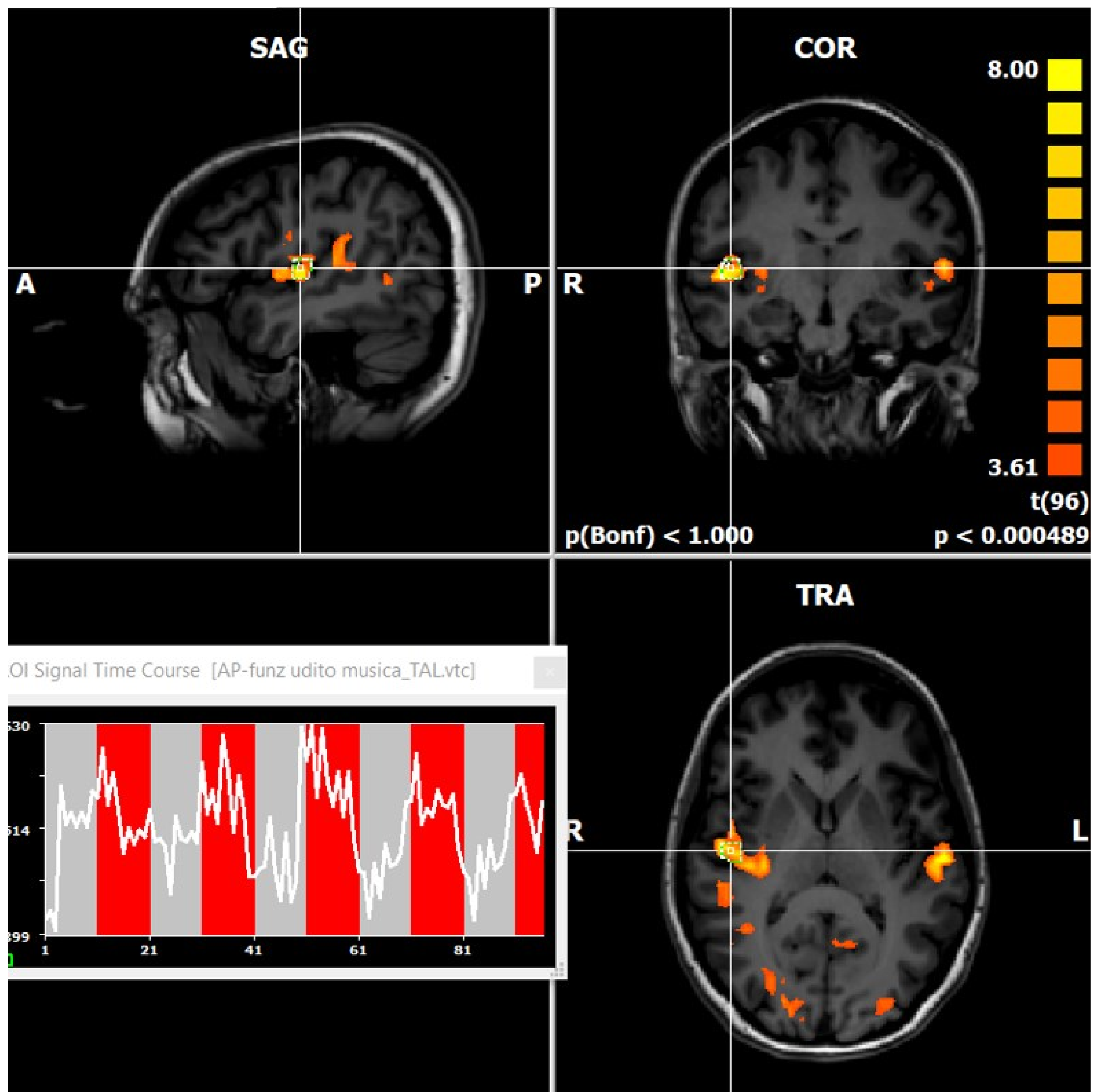


*Figure 33: VTC image of the auditory task activation represented in the three main planes (sagittal, coronal and transverse). In the box below on the left it is represented the Time Course window, which shows the time course ROIs.*

## 4.3 Visual checkerboard task

Figure 34 shows the functional images of the 36 slices, instead figure 35 shows the pre-processed VTC image, both acquired during the visual checkerboard task activity.

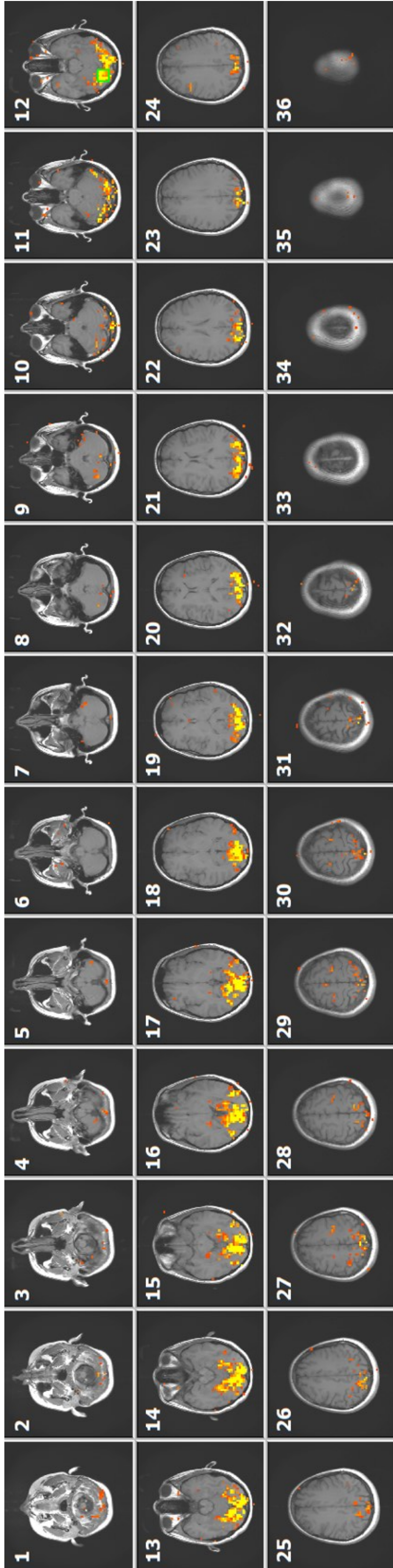

*Figure 34: Functional visual checkerboard task images of all 36 slices.*

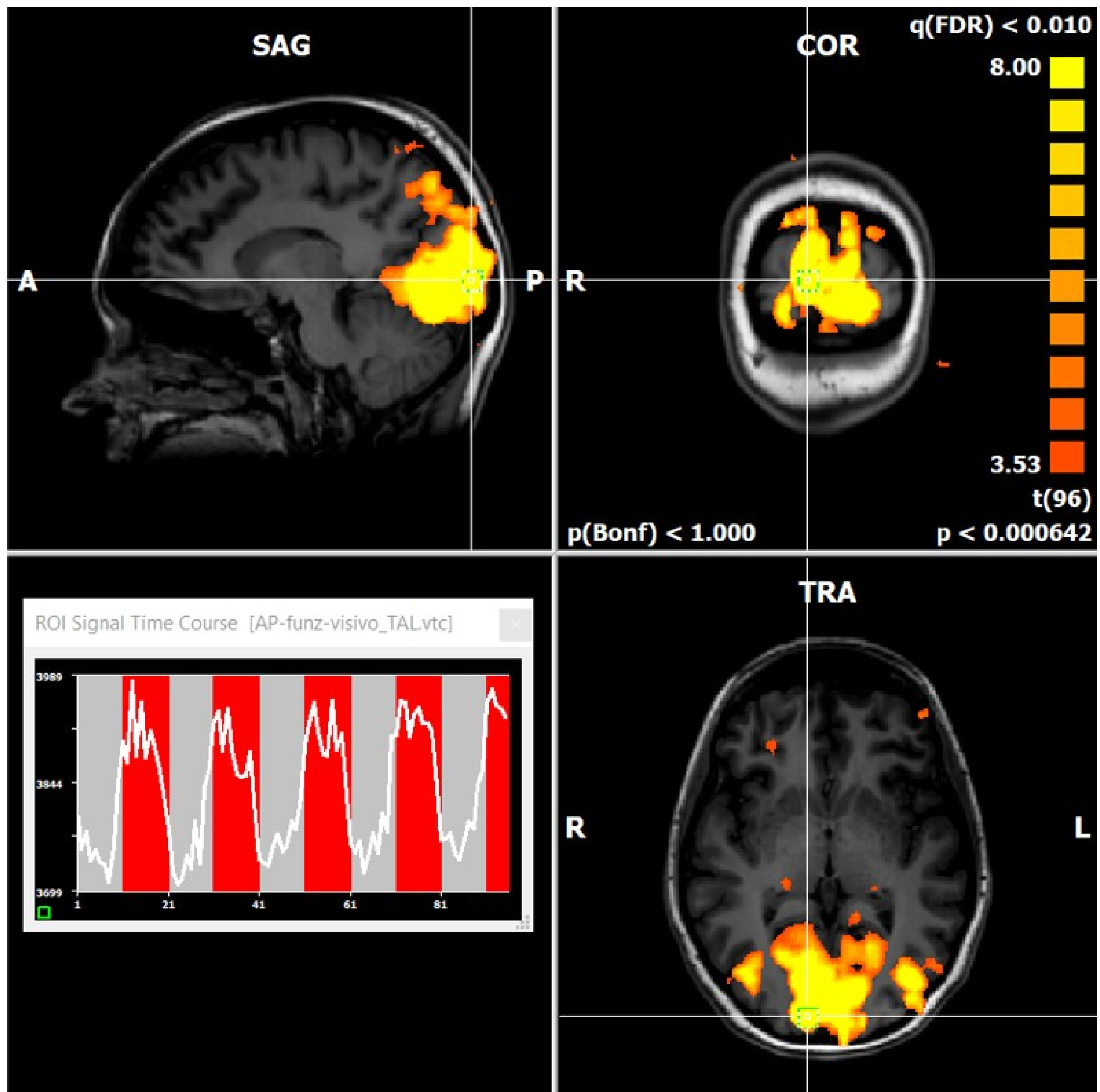


*Figure 35: VTC image (Talairach + 3D structural + functional image) of the visual checkerboard task activation represented in the three main planes (sagittal, coronal and transverse).*
*In the box below on the left it is represented the Time Course window, which shows the time course ROIs.*

# 5. Discussion

The main purpose of this thesis is to analyse stimulation protocols to be administered during functional magnetic resonance studies. Stimulation protocols may be one of the most important steps to make functional MRI became a routine diagnostic neuroimaging procedure, to be used for preoperative planning.

Three types of responses to designed tasks are provided in the results: motor, auditory and visual. Unilateral consecutive finger tapping (General Motor Left task) causes significant activity in the cortex around the central sulcus, which is the expected somatotopic position. Strong supplementary motor area (SMA) activation is detected, along with main primary sensory-motor activation and contralateral activation (of the right hemisphere for a left task). It is possible to observe secondary somatosensory and premotor cortical activity as well.
Sequential finger tapping frequently causes also ipsilateral activation (on the same side of the task) within the anterolateral precentral gyrus.
In many studies, it is reported that the contralateral activation becomes more relevant for a sequential finger tapping on the dominant compared to the nondominant hand [14].
It can be difficult to understand fMRI results by just considering segmented task components related to a specific stimulation activity. For instance, a normal motor performance relates to sensory feedback from the haptic, proprioceptive, and visual senses. Both the motor task region and these sensory regions may become more activated thanks to this feedback. It can be challenging to account for imperceptible environmental factors like force or visual feedback during motor activity, even though activation areas are incredibly sensitive to their effect [15].

Visual stimulation by flickering checkerboard causes strong activity in the primary visual cortex, as expected. Significant activity is also present in the secondary visual cortex, as well as in the posterior parietal area.
Functional MRI has provided a valuable tool in validating cortical retinotopy (visual input mapping from the retina to visual cortex) and quantitatively determining the cortical extension of central foveal vision (responsible for sharp central vision), obtaining results that would otherwise have required invasive procedures [16, 17]. Stimuli presentation with goggles is frequently used, with respect to a standard monitor, since it enables different types of stimulations (monocular, several combinations of hemifields, and quadrants), and allows to investigate of better cortical visual retinotopy. Additionally, since the patient can only look within the goggles, it helps them concentrate better [18].

Auditory stimulation using classic music causes robust bilaterally activity in the primary auditory cortex, as well as in the secondary auditory area. It is also notable a left and right activation of the lateral premotor cortex.
Recent neuroimaging research has demonstrated that listening to music passively stimulates brain areas that are part of the actual motor system [19].
A hypothesis of motor activity during music perception known as the ASAP (Action Simulation for Auditory Prediction) was advanced by Patel and Iversen [20]. According to this idea, the motor planning system uses the same neural mechanisms that simulate bodily movements to synchronize neural activity with the musical beat.
An additional investigation could involve the different activation in musicians and non-musicians. Musicians show plasticity-induced changes in perceptual and motor abilities as well as altered structural and functional neural connectivity. For instance, musicians passively listening to music should activate the supplementary motor cortex as well as the cerebellum, which has a larger volume in musicians [21].
Running further research that directly contrasts various types of music-listening tasks is another interesting direction to consider.

*Technical considerations and limits*

In the present study, the designed paradigm duration was 5 minutes with 5 cycles of 60s each, since several clinical studies use this as blocked design fMRI time. To set up this duration in the nordicAktiva software, it is necessary to search for the main file code in several folders, and for each paradigm is necessary to change the active and resting block duration variables. Standard paradigm timing in the software is referred to the American Society of Functional Neuroradiology (ASFN) recommendations, for which generally the task duration changes based on the type of paradigm.
To improve stimuli delivery protocol, the use of software like nordicAktiva is game-changing, however, the timing set up in code blocks is a time-demanding procedure, which may be improved using designed manipulation software.

A significant limitation of this thesis is the investigation and analysis of only one healthy subject. The involvement of a larger group of subjects, or even pathological patients, could be essential

also for a statistical study which could improve the setup of stimulation protocols for clinical applications such as presurgical mapping.

The wide variety of tasks that have been suggested and utilized to optimize fMRI brain mapping can be explained by the complexity of mapping motor, language, visual, and other cognitive functions [22].
An active task baseline, as opposed to a resting condition, can more precisely identify important brain regions by removing background signals connected to the task but not specifically related to the function of interest. Cognitive tasks can coactivate other activities like attention and visual processing, so a suited active baseline condition can enable to account for signal variations [15].

Functional MRI has a high sensitivity to motion because of its prolonged acquisition times. Patients with brain injuries and neurological abnormalities may have more difficulty maintaining their head motionless inside the scanner than healthy control volunteers [23]. In addition, children are more likely to experience motion artifacts, with roughly one-third of fMRI tests performed on them providing non-diagnostic results [24]. For these reasons, task selection, well suited for the type of subjects involved in the clinical analysis, may lead to a reduction in motion artifacts. For instance, lip pursing and toe wriggling are tiny motions that could be utilized to map the motor cortex with the least amount of non-task-related muscle activation.

Moreover, according to resting-state fMRI studies, neurocognitive and neuropsychological testing, neurosurgical patients perform less effectively than healthy subjects in attention, language memory, and executive function [25]. These deficiencies restrict their capacity to comprehend and complete the tasks adequately, which leads to incomplete or incorrect data because task performance is a crucial factor in determining the accuracy of task-based fMRI. Preoperative analysis may be improved by modifying the study design to take into account the capabilities, cooperation, and cognitive function of each patient. If a patient is unable to do several tasks in one session, separate study periods could be more tolerable [15].

The outcomes also depend on the right pre-processing method. A debatable but important method that can boost the signal-to-noise ratio (SNR) is spatial smoothing. The sensitivity of detecting activation in specific brain regions is increased by smoothing. This procedure is delicate and must be managed well since in some cases it could improve the detection of active areas, and in others it could make it worse. It depends on the size of the cortical area that is expected to be activated: for a small area, which requires detailed inspection, the spatial smoothing may complicate the analysis, instead for a bigger area may be more efficient to be used.

Additionally, it improves the SNR of images with lower resolution to reduce the need for time-consuming high-resolution image acquisition [26]. However, excessive smoothing can suppress the activation signal whereas it can reduce the spatial resolution.

*Conclusion*

In conclusion, the employment of specific protocols for stimuli delivery during fMRI investigation is essential for brain mapping in clinical and diagnostics settings. To acquire the most meaningful data for brain activation, researchers must precisely plan their methodology and use well-suited paradigms, taking into account several features related to the psychophysical status of the subject.

Since blocked design (repeated actions of a task during about 30s blocks) may skip transiently activated regions it will be a good choice in the future to combine blocked and event-related tasks (transient BOLD response related to a single task), having the advantage of monitoring both prolonged and transitory functional activity.

Specific tools such as the VisualSystem HD (goggles) and the Syncbox, used in combination with the nordicAktiva software, may represent a resolute implementation for stimuli delivery and management. Nevertheless, some technical adjustments related to the correct positioning of these instruments and improvements in the paradigms design need to be done.

To sum up, subjects must be considered under ideal circumstances tailored to their unique cognitive and physiological states, to properly detect cerebrovascular changes related to significant neuronal activity. These alterations may be accentuated or reduced by the cognitive, psychological, and physiological conditions of the subject, and these challenges may be overcome by protocol design.

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# Ringraziamenti

Vorrei dedicare questo spazio a chi ha contribuito direttamente ed indirettamente alla produzione di questa tesi, risultato finale del mio percorso universitario.

Un ringraziamento particolare va alla mia relatrice Prof. Burattini Laura per aver permesso la realizzazione di questo elaborato.

Un sentito grazie va alle mie correlatrici Prof. Mara Fabri e Dott. Marcantoni Ilaria per la loro infinita disponibilità e per avermi seguito nel processo di scrittura dell'elaborato.

Un sentito grazie va anche al Prof. Gabriele Polonara per avermi dato la possibilità di svolgere il tirocinio, che ha portato alla stesura di questa tesi, presso il suo dipartimento.

Ringrazio anche le Dott.sse Mojgan Ghoushi e Nathalie Herbert, i Dott.ri Marco Valenti, Luca Reversi e Francesco Mariotti, e i tecnici Simone Marinelli e Alessio Canari del dipartimento di Neuroradiologia, per il tempo e le risorse impiegate.