# Trabajo Fin de Grado de Psicología

Neural Correlates of a Whistled Language: an fMRI-task

Correlatos Neurales de un Lenguaje Silbado: una tarea de fMRI

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#### Abstract

Are whistled languages and normal spoken languages treated the same by our brain? Whistled speech appears in different non-connected regions of the world, due to a communicative necessity across long distances. In the current study, we seek to identify similarities and differences in neuronal activity between both whistled and normal spoken types of language. Blood oxygen level dependent (BOLD) response in a whistled speech have been first studied by Carreiras, López, Rivero & Corina in 2005. Their experiment concluded that the same language areas were activated in a passive-listening to *Silbo* and Spanish speech task. However, this conclusion is undermined by several methodological problems. Here, we attempted to address the same question as Carreiras et al. (2005), but solving some of their problems. We found similar activation in the left temporal lobe for Silbo and Spanish, concretely a higher activation in the superior temporal gyrus, widely considered a key area for language comprehension. This suggests that there are common language areas underlying *Silbo* and Spanish.

*Keywords*: Whistled speech, *Silbo gomero*, Lateralization, functional Magnetic Resonance Imaging (fMRI).

#### Resumen

¿Trata nuestro cerebro de igual forma un lenguaje hablado y un lenguaje silbado? Los lenguajes silbados han surgido en diferentes regiones del mundo aparentemente no conectadas entre sí, debido a la necesidad de comunicación a través de largas distancias. En este estudio, buscamos identificar similitudes y diferencias entre estos dos tipos de lenguaje, utilizando el español hablado y el Silbo gomero. Carreiras, López, Rivero & Corina, en 2015, estudiaron el nivel de oxígeno en sangre debido a la actividad neuronal (BOLD) en respuesta a la comprensión de estos dos lenguajes, concluyendo que ambos activaban áreas comunes de lenguaje en el cerebro de silbadores. Nosotros hemos encontrado una activación similar en el lóbulo temporal izquierdo, concretamente el giro temporal superior, ampliamente considerado un área clave para la comprensión del lenguaje. Esto nos sugiere que hay áreas comunes en el procesamiento del lenguaje silbado y español hablado.

Palabras clave: Lenguaje silbado, Silbo gomero, lateralización, Resonancia Magnética Funcional (fMRI).

Neural Correlates of a Whistled language: an fMRI-task

This work presents a task designed to examine the similarities and differences in neural processing of a whistled language, with respect to a common speech processing. Other experiments have followed this objective (Carreiras et al., 2005), but due to methodological inconsistencies and different results, there is not a reliable answer. However, this question is not the only purpose of the work. The technique we used was fMRI, and therefore, another important goal was to understand and broaden knowledge about the technique. Thus, before we present our study and details, we will commence with a brief introduction to fMRI development history and performance.

### **Magnetic Resonance Imaging**

Magnetic Resonance Imaging (MRI) is based on the intrinsic angular momentum of particles (spin), and it was suggested by Wolfgang Pauli in 1924. In the 1920s, Isaac Rabi and Otto Stern discovered that these particles, in a magnetic field, would absorb radio frequency energy. They proved it with particles traveling in a vacuum. Later, Felix Bloch from the Stanford University and Edward Purcell from Harvard University, reach the same effect but with particles staying in liquids and solids, what is called nuclear magnetic resonance (NMR). Due to this, they won the Nobel Prize in Physics in 1952. Within the 1950s and the 1960s, this phenomenon was demonstrated in organic tissues. In 1973, Paul Lauterbur showed that NMR could be used to create an image by causing the resonance frequency of nuclei to vary linearly with their spatial location. Peter Masfield created the foundation for snapshot MRI. In 2004, Paul Lauterbur and Peter Masfield shared the Nobel Prize in Medicine. Richard Ernst improved the technique sensibility (its resolution) by increasing the radio frequency. Finally, Ogawa showed in 1990 that

MRI water signals can be sensitized to brain oxygenation, and this effect is used in fMRI, as we will see later (Pekar, 2006).

Nowadays, fMRI is constantly gaining in popularity, as: First, it does not use ionizing radiation and is a safe method. Second, discriminates between white and grey matter with a high precision. And third, can be used to detect changes in blood oxygenation related to neural activity (Ward, 2015).

The following is a summary of the events required to obtain a brain image using this technique. The magnetic resonance uses the selective energy absorption of electrons, protons, or atomic nuclei which have an odd number of protons or neutrons (Junqué & Barroso, 2009). These particles can absorb radio frequency electromagnetic energy from a potent magnetic field. Hydrogen is composed by a proton, a neutron and an electron, and is chosen due to its abundance in the human body. These atoms are found in hemoglobin, which can have diamagnetic behavior (oxyhemoglobin) or paramagnetic behavior (deoxyhemoglobin). In paramagnetic behavior, hydrogen protons are constantly spinning around an axis, and this movement of a positive charge causes a little magnetic field. Furthermore, paramagnetic behavior involves an odd number of particles that are not compensated, and this allows a particle to align with a strong magnetic field (Junqué & Barroso, 2009). This phenomenon is what MRI utilizes.

Once the person is correctly positioned inside the MRI machine and it starts working, the first thing that occurs is that a magnetic field is applied. This magnetic field would be constant during the scan with a strength measured in Tesla (T). The scanner we utilized in this work had 3T, and usually scanners have between 1.5 and 3T (Ward, 2015), although 7T scanners have been used in research. The stronger the magnetic field is, more atoms will be aligned to it in a longitudinal magnetization (Figure 1). Protons align in two ways: parallel and antiparallel to the

external magnetic field (Schild, 1990): parallel alignments are given when the protons are in a low energy state which align with the north pole, and antiparallel alignments when they are in a high energy state and align with the south pole (as shown in figure x).

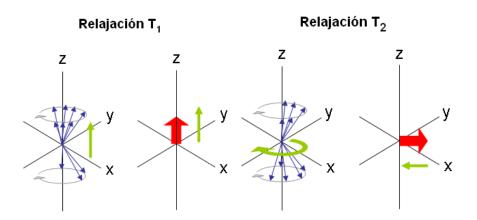


Figure 1: T1 relaxation to the left, shows a longitudinal magnetization, whereas T2 relaxation to the right, shows a transverse magnetization. Source: http://uptmkleberramirez.blogspot.com.es

Protons preserve the alignment while they move in a particular way, called precession movement. They move on the basis of their axis but drawing a circle with the top of it (Figure 2). When protons are aligned, a radio frequency pulse is sent by the machine and exchanges energy with them. Some protons absorb enough energy to pass from a low energy state to a high energy state, and it leads to a reduction on the longitudinal magnetization (Figure 1). At the same time, these protons have a coordinate precession movement in step, what is called *in phase* (Schild, 1990), and it makes a transversal magnetization.

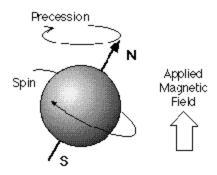


Figure 2: Precession movement of a proton that is aligned with a magnetic field. Source: https://physics.stackexchange.com

The precession speed is measured by the precession frequency or times that the proton precess per second, and this frequency depends on the strength of the magnetic field to which the subject is exposed. Precession frequency can be calculated by using the Larmor formula:  $\Theta_0 = \gamma \times B_0$ , where:  $\gamma$  is a constant depending on the atoms used,  $B_0$  the magnetic field strength and  $\Theta_0$  correspond to the frequency (Schild, 1990). 1T equals 10.000 gauss, and using this formula, T = 42.58 MHz, T = 85.16 MHz and T = 127.74 MHz, when using hydrogen.



Figure 3: A head coil can be a transmitter, a receiver, or both. As a receiver coil, obtains the energy from the current produced by the transverse magnetization of the protons (Klomp et al., 2011). Source: http://mrinstruments.com

Finally, transverse magnetization induces current, that is received by the antenna, or head coil (Figure 3). After removing radiofrequency signal, protons suffer relaxation and go back to their initial position. Depending on the moment in which we collect the signal, we will obtain different kind of images. In T2 images, transverse magnetization is lost and no energy is transferred (T2 relaxation); and for T1 images, protons in the high energy state go back to low energy state relaxation, longitudinal magnetization, and there is an energy transfer to sensor from the MRI machine (T1 relaxation), (Shild, 1990).

### **Functional Magnetic Resonance Imaging**

Haemoglobin is important in functional Magnetic Resonance Imaging. Previously, we explained the two different behaviours for the haemoglobin: deoxyhaemoglobin and oxyhaemoglobin. Besides, functional imaging measures the changes in deoxyhaemoglobin concentration, due to its magnetic properties, in what is called blood oxygen-level dependent (BOLD) response. This technique is based on the cellular and metabolic processes that take place during the neuronal activity in what is known as hemodynamic response function (HRF).

The HRF is described in three steps (Figure 4): first, cells need extra oxygen and energy to perform their functions, and blood provides these metabolites to them while they are active in metabolic activity. Blood liberates oxygen from oxyhaemoglobin molecules located the nearest to these regions, and consequently what remains is deoxyhaemoglobin. This increase in deoxyhaemoglobin leads to an *initial dip* of the BOLD signal. The second step constitutes a reaction to the loss of oxygen in blood flow to the region involved, which increases considerably (*overcompensation*), and is measured in fMRI. Finally, before returning to the original oxygen levels, there is another dip caused by a venous relaxation, an *undershoot* (Ward, 2015). Images obtained in fMRI are called T2\* images.

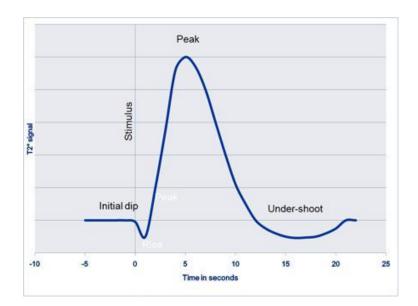


Figure 4: Model HRF.

Source:

https://radiopaedia.org

In summary, fMRI is a technique that measures brain activity in an indirect way, by detecting differences in BOLD response. It is based on biological properties of this activity (metabolic processes and its effects in blood changes) and physical properties of a very abundant particle in our bodies, hydrogen. It is an innocuous technique, and MRI that allows researchers and clinical experts to obtain various types of images (T1, T2 and T2\*), with complementary information.

## **Experimental development in language**

Investigation in language has been crucial for scientific and clinical reasons. It contributes to our knowledge about a very important and special ability of humans, and there are yet many things to elucidate in this matter. Neural correlates of language processing have been object of interest since early times, but first relevant findings were identified through postmortem analysis of clinical patient with injured brains, as we will explain latter. Studies in this field have been of relevance for delimitation of impairments, such as aphasias, and to obtain necessary information for application of their correct treatment or interventions.

Moreover, other investigations indicate that language related areas are not exclusively used for spoken language. They are also involved in other type of languages, such as sign languages or symbols relevant to social communication. It is evidenced in a study carried out by Bellugy, Poizner & Klima (1989) with deaf from birth individuals with acquired left hemisphere injuries affecting to areas highly implicated in language processing. These participants presented considerable deficits in sign production and comprehension, compared to non-injured deaf from birth participants. This result shows that areas involved in spoken language are not exclusive to it but also implied in sign language.

Thus, the question we addressed here was whether areas involved in normal language processing would be also associated to a whistled language. Some research has been made in this regard, but there are divergent conclusions.

Whistled language is an unusual type of language, composed by whistles, and used in many regions of the world as a language replacement. People living in these regions switch to whistled languages when they want to communicate through long distances and across mountains.

Our study is based on whistled language of La Gomera, also called *Silbo Gomero*, and it is based on the spoken Spanish from this island. It is composed of five different vowels, as Spanish, but two of them are strongly overlapped (Meyer, 2008), and four consonants (Trujillo,1978). Also, rules of tonic accent are mostly respected in Silbo (Meyer, 2008). Both whistled vowels and consonants vary in a high to low pitch dimension and the melodic line can be continuous or interrupted (Carreiras et al., 2005).

Here below, we expose a brief revision of cross-time language investigation and we will end up commenting the advances in the language concern using fMRI, and particularly, results in whistled languages.

# Language and the brain

The neural correlates of cognitive aspects such as language were, at first, studied in clinical patients with injured brains. In those times, patients' behavior and linguistic responses were examined while alive, and an autopsic study would be realized, if possible, when deceased. This technique was carried out by Wernicke, Broca, Lichtheim and Déjerine within XIXth and half of XXth centuries and has revealed basic anatomical structures involved in language, that we recognize nowadays. From then until the 80's, in vivo investigation techniques, such as radioisotopes exploration, was developed, and later to this, computerized axial tomography (CAT). Finally, neuroimaging techniques are providing a new kind of in vivo information about injured and non-injured brains and provide information about the anatomical progression of injuries; and functional imaging techniques, as fMRI and positron emission tomography (PET), have allowed us to observe the brain activity produced while patients or participants perform a linguistic task (Junqué & Barroso, 2009).

## Classical background

Paul Broca discovered a key area for language in the third left frontal circumvolution (1861, 1865). His first patient, Leborgne, had lost his ability to produce speech, while understanding was preserved. He was able to answer Broca's questions by gestures and presented right arm and leg paralysis. The post mortem examination indicated an extended damage in the left hemisphere caused by an infection. Broca estimated that the infection started in the third left frontal circumvolution (Brodmann area, BA, 44/45), causing the articulatory

disability, and after so, it extended to other areas. He concluded that this area was necessary for speech articulation (Purves et al., 2012).

In 1874, Karl Wernicke showed his work with different patients. These patients presented a noticeable alteration in their speech understanding. Their speech was fluent and prosodic, but they had a difficulty to find the correct words, and an alteration in repetition and naming (with visual clues). This issue is associated to left posterior superior temporal region, posterior to the primary auditory cortex (BA 22). Wernicke proposed this area as a midpoint for word's auditory images and considered it indispensable for speech understanding (Junqué & Barroso, 2009).

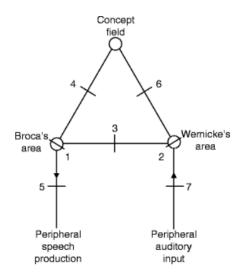


Figure 4: The Wernicke-Lichtheim model. Source:

http://michelegaldi.wixsite.com

Wernicke also established a connectionist model of spoken language production, afterwards developed by Lichtheim in 1885. The Wernicke-Lichtheim model (Figure 4) proposed that language production is due to Broca's and Wernicke's areas activation, a concepts area, and the connections between these three areas. The network connecting the concepts area and Broca's area would be used for intentional speech production, and on the other hand, the network connecting Wernicke's and Broca's areas is involved in speech that does not

require access to the subject's concepts, such as repetition. The left angular gyrus (AB 39) was conceptualized by Déjerine, as a visual-verbal zone necessary to reading and writing. And later, Norman Geschwind (1965, 1972) would consider Broca's area as an articulatory codification rules store, that provides instructions about the necessary muscles and their coordination. For him, Wernicke's area plays a significant role in oral speech recognition based in patterns, and for this reason it is central for understanding. The arcuate fasciculus communicates Wernicke's with Broca's area in a posterior-anterior connection, and the angular gyrus transforms visual into acoustic information to provide Wernicke's area with the last one. Finally, he proposed that the system of structures described until now in this section, is communicated with other associative cortical areas (Junqué & Barroso, 2009).

## Recent approaches

In a recent review, Price (2000) concludes that activation in Wernicke's area activates words' auditory representations. Also, the left frontal operculum/anterior insula (Broca's area) activation corresponds with the words' motor representations; and the areas peripherical to the Sylvian fissure would be equivalent to concepts area described by Wernicke.

Nowadays, many cognitive abilities are hypothesized to occur in multiple, parallel processing streams. Some studies point a dissociation between anterior and posterior streams in speech perception. The anterior stream constitutes a 'what' pathway associated with the processing of speech meaning and the identity of the speaker, and which includes anterior belt and para-belt regions, and anterior temporal and ventrolateral frontal regions (Scott and Johnsrude, 2003). The posterior stream would constitute both a 'where' and a 'how' pathway (Ward, 2015), and includes posterior belt and para-belt, superior and posterior temporal and dorsolateral frontal regions, and the inferior parietal lobe, including the angular gyrus and the

arcuate fasciculus. This stream is involved in sound localization and speech gestures representation (Scott and Johnsrude, 2003; Ward, 2015).

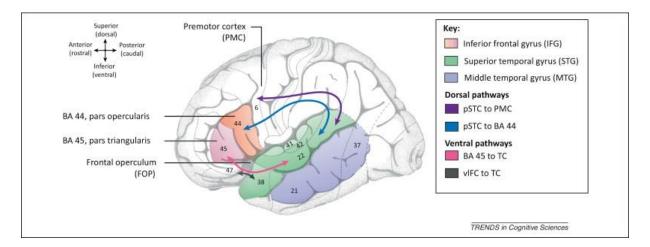


Figure 5: Pathway in pink indicates the 'what' route; in blue and purple dorsal pathways. Source: http://www.cell.com

For the anterior pathway, more intelligible words, show more anterior activity in the temporal lobe, measured with fMRI (Scott and Wise, 2004). And an involved area in speech gestures perception is the superior temporal sulcus (STS), while representations in the premotor cortex are perceptual and motoric (Kohler et al., 2002).

Regarding the lateralization of language, Engelien et al. (1995) and Tzourio et al., (1997) reported a lateralization to the right when the subjects attended to nonverbal stimuli. Binder et al. (1995) showed in an fMRI study that language areas were strongly lateralized on the left, whereas the tone decision task activated the right auditory cortex to a higher extent. Moreover, in 2003, Woermann et al. compared determination of language dominance, in 100 patients with different localization-related epilepsies, using fMRI with results of the Wada test. They concluded that 91% of the patients were presented the same lateralization measuring with both techniques, and that this result shows the reduced necessity of the Wada test due to the precision of fMRI in the language domain.

Furthermore, planum temporale is a region that is often activated in speech tasks. It is assumed to be involved in the processing of auditory and non-auditory information. This region is located posterior to the primary auditory cortex, in BA 22 (Ward, 2015), as Wernicke's area.

These newer results do not contradict the explained conclusions from classical studies, but adds more concrete information, and as we saw here fMRI is a useful tool employed in many language studies.

### Research in Whistled languages and the brain

The first study analyzing the neural activity produced by a whistled language was carried out by Carreiras et al. (2005). They examined areas related to comprehension of whistled language, concretely Silbo Gomero, with two different comprehension tasks. First, participants listened passively to Silbo and Spanish speech sentences, and they used as baseline the digitally reversed version of Silbo sentences. Second, they were asked to monitor series of Silbo and Spanish randomized words. They found common activation areas in the left superior temporal gyrus and right superior midtemporal region, in both conditions, but only for experienced whistlers. Their explained activations were thresholded Z > 3.09, and P < 0.05 uncorrected. Thus, they concluded that Silbo comprehension activated areas related to normal speech comprehension.

However, there are several problems with the methodology of the experiment that undermine this conclusion: First, analysis was done with just two subjects in the first task, and three in the monitoring task, which is not enough for a concluding analysis. Second, baselines to the auditory stimuli in the first task were reversed *Silbo* for both, the Spanish and the *Silbo* condition. Spanish sentences should have been contrasted with a same-length reversed Spanish condition. Third, different orators and whistlers were alternated, and this could be problematic

because right temporal regions have been found that are associated with speaker changes (Rauschecker & Scott, 2009). Thus, observed right temporal activities may be related to speaker change and not language change (Spanish vs. *Silbo*). Fourth, in the first task, participants were asked to remember what they had been listening to, and this contaminates the comprehension activity with remembering processes. In the second task, they monitored a target item they had seen before the scanning session and within this task, they were also asked to remember some of the experiment content.

We consider that due to these problems, conclusions may be premature. In addition, a more recent experiment shows non-lateralization of whistled syllables recognition in dichotic listening to Turkish whistled language (Güntürkün, Güntürkün and Hahn, 2015). They used 31 subjects for their experiment, and the dichotic listening task compared lateralization of whistled and vocal syllables recognition. Their results indicate non-lateralization for whistled dichotic listening and a clear lateralization for vocal dichotic listening.

Therefore, we repeat the passive listening to *Silbo Gomero* and Spanish sentences task, but making some important changes. We analyse data from nine participants; contrast Spanish speech condition with its digitally reversed version, instead of reversed *Silbo*; used a single orator and whistler; and removed the remembering task. We expected that Spanish would activate the common language areas (Broca, Wernicke, etc) and that *Silbo* would also activate these same areas.

#### Methods

### **Data acquisition**

Functional MR images were acquired using a 3T MRI system (Signa Excite, General Electric Milwaukee, WI, EEUU). Subjects used headphones adapted to the fMRI machine. For functional imaging in sentences-task, 440 volumes were acquired. We gathered 36 axial slices, with a slice thickness of 3.7mm each and slice gaps of 0.3 mm, using an interleaved acquisition. The parameters of the functional sequence were: a 64×64 matrix; Field of view (FOV), 256×256mm; repetition time of the pulse sequence (TR), 2.00 seconds; echo time, 22.5ms; and a flip angle of 75° (optimum flip angle for our TR is between 73°-77°). Voxels measured 4×4×4mm, 64mm³.

Data collected during the experiment were functional images (T2\*), a structural image (T1) and a diffusion tensor imaging (DTI) for each subject.

#### Stimuli

The analysed data in the present work corresponds to the results given in a fMRI task extracted from a three-fMRI-tasks experiment. These tasks or runs were separated by brief time gaps, in which the subjects were given no auditory or visual stimuli.

First task consisted of a naming task in Whistled or Spanish language, in which a wide list of visual inputs was used (drawings in black and white). These inputs were divided in ten blocks of five images each one and the language instruction were alternated. We collected 164 volumes. (5.47 minutes).

Second task was a word listening task and alternated 24 blocks which were assembled in six blocks with five words each one. It alternated Spanish language, reversed Spanish, *Silbo Gomero* and reversed *Silbo gomero*. We collected 344 volumes in this task. (11.3 minutes).

The last task, the one we use in the present work, was a passive-listening task to Silbo and Spanish sentences. It alternated 24 blocks that were assembled in six groups. Within each of these six blocks, subjects listened to four blocks of five sentences each one. The first five sentences were spoken in Spanish language, the following five were in reversed Spanish (so it was intelligible) after a short pause. Then, another pause, and five sentences in *Silbo gomero*. Eventually, another pause, and five more sentences in reversed *Silbo Gomero*. We collected 439 volumes. (14.63 minutes).

Whistled sentences and words lasted more than the same ones in spoken speech. The reverse auditory stimuli were given as baseline to counteract these mentioned length differences between stimuli of the same task, and the effect of the background noise produced by the machine. We present length differences in sentences in table x.

Table x
Stimuli timing

Condition	Mean	Standard Deviation
Silbo	23.5725	2.2451
Silbo reversed	23.5729	2.4662
Spanish	12.7839	0.8685
Spanish reversed	12.7848	0.9434

*Note:* Average and standard deviation of the stimulus extent.

#### **Participants**

Nine whistler participants (8 men and one woman), aged 12-60 years, with an average of 23.67 and a standard deviation, 15.30. All of them were right handed and were selected by a whistled language teacher.

Participants signed their consent and awareness about safety aspects before the commencement of the experiment in the MRI machine. They could abandon the experiment in

any moment if they considered it necessary. Also, participants were informed of the tasks the experiment required and the necessity of staying motionless while the experiment was running.

About the motion concern, whistled language usually requires arms and fingers to be produced (Figure 2), but due to the motionless requirement to our subjects, they were asked to whistle in what they call *Silbo fino*, only using their mouths, as in common speech.

### Preprocessing and registration

Image preprocessing and analysis has been done using FSL, Oxford University, United Kingdom, based on Unix. No image was removed before the analysis. First, we used the BET program (brain extraction) to perform a brain/non-brain segmentation and remove non-brain tissues. This step is important for the later registration step. We did not perform any slice timing correction.

Motion correction was performed with MCFLIRT. In this programme, motion is estimated utilizing a chosen image from the middle of the time series, as model. Then, the program compares each of the other images to this model. It can realign the slices, taking the parameters obtained from the comparison of each image to the image taken as reference, and reslice each moved image to best fit with the model. We have chosen the middle image because is the closest one to the other images in the time series. This motion correction technique uses a rigid-body, what means that the program does not make changes on the brain shape.

Otherwise, it makes changes on the brain position and orientation (Poldrack, Mumford & Nichols, 2011). Moreover, we executed a highpass temporal filtering to remove low frequency artefacts. Also, a spatial smoothing FWHM (full width half max) of 8mm. We increased the smoothing from the standard used (5mm), because we were looking for large areas. Although it

removes small effects. This smoothing is executed on each volume separately and is performed to reduce the mismatch across individuals (Poldrack, Mumford & Nichols, 2011).

Both T1 and T2\* images were registered to standard with 12 degrees of freedom (DOF). 12 DOF represent an affine transformation composed by the following linear transformations: three rotations, three translations and three scalings. This step is important due to the anatomical differences across individuals. Thus, brains must be aligned, and we employed a widely utilized template which has been developed by Montreal Neurologic Institute (MNI), MNI152 (Poldrack, Mumford & Nichols, 2011).

### **Statistical analysis**

Our purpose was to illustrate the common activation areas in response to listening to Silbo and Spanish speech sentences. First, we contrasted Silbo and Spanish conditions with their reversed versions. We did this in two phases: a first-level analysis (the contrast for each subject), and a higher-level analysis using a mixed-effects model, in which we consider the cross-subject variance. Once we finished these contrasts, we did a conjunction using both (*Silbo > Silbo reversed* and *Spanish > Spanish reversed*) to distinguish the shared areas, which was the main purpose of the work. Additionally, we contrasted *Silbo - Silbo reversed > Spanish - Spanish reversed* and *Spanish - Spanish reversed > Silbo - Silbo reversed*. We explain better these analysis in the paragraphs below.

#### First-level Analysis

Data from each participant was analysed individually. To detect variation, we used the general linear model (GLM), in which BOLD response is the dependent variable and the expected BOLD timecourses are the independent variables (Poldrack, Mumford & Nichols, 2011).

With respect to the predicted BOLD response, a double-gamma HRF model was used. Double-gamma HRF model considers the overcompensation and the undershoot. Then, this model was compared with the obtained signals. The threshold was Z>2.3 for each voxel.

# **High-level Analyses**

In the high-level analysis, we compared all the subjects of our sample. We used a mixed effects model, considering the cross-subject variance.

The Z values we expose in the section below, show the consistency of the real signal with the expected one.

## Conjunction

The conjunction established the common activation based on the minimum significant value comparing voxels in both contrasts. This function was carried out with the easythresh\_conj script (Nichols, Brett, Andersson, Wager & Poline (2005).

#### Results

# Conjunction

We found common significant BOLD responses for both Spanish and Silbo conditions in the left temporal gyrus. The results reported from this conjunction are thresholded at P < 0.1 and not corrected. Maximum activation, Z=2.95, under these circumstances was obtained within the left superior temporal gyrus, posterior division, and its anterior division was also detected. Left middle temporal gyrus and the left temporal lobe were also activated areas in this cluster (Table 1). Activated areas shown in a standard space can be seen in Figure 6.

Contrasts Silbo > Silbo reversed and Spanish > Spanish reversed did not indicate any significant BOLD response, and due to these non-corrected contrasts were employed.

Table 1.

Conjunction Silbo – Silbo reversed and Spanish – Spanish reversed

Location	Cluster size (voxels)	Max Z	X	y	Z
Left Superior Temporal Gyrus, posterior division	3129	2.95	74	47	35
Left Middle Temporal Gyrus, anterior division		2.82	70	57	25
Left Middle Temporal Gyrus, posterior division		2.8	72	58	25
Left Superior Temporal Gyrus, anterior division		2.8	72	59	30
Left Temporal Pole		2.67	70	73	26

*Note:* The maximum voxel activation in the cluster indicated in bold letters. Locations below are other significant activations in the same cluster. Locations in reference to Harvard – Oxford Cortical Structural Atlas (Lateralized).

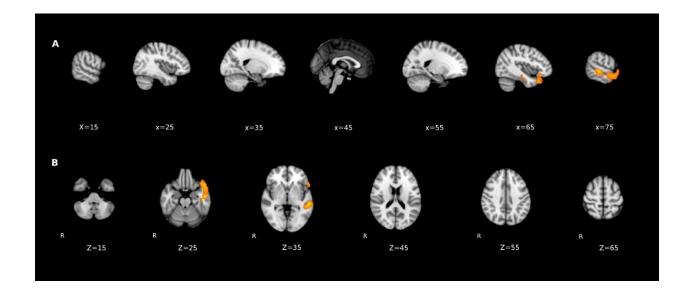


Figure 6. BOLD response to the conjunction Silbo – Silbo reversed and Spanish – Spanish reversed.

# Contrast Silbo – Silbo reversed > Spanish – Spanish reversed

Silbo sentences contrasted to Spanish sentences and vice versa did not show any significant activated areas when the analysis was corrected. Uncorrected analysis of Silbo – Silbo reversed > Spanish – Spanish reversed contrast indicated an activation, Z=3.06, in a voxel located in the left postcentral gyrus. The biggest cluster found (390 voxels), covers part of the left inferior frontal gyrus, in the pars opercularis and the left precentral gyrus. Some other activated regions are exposed in Table 2.

Table 2.

Contrast Silbo – Silbo reversed > Spanish – Spanish reversed

Location	Cluster size	Max z	X	у	Z
Inferior Frontal Gyrus, pars opercularis (LH)	390	2.87	68	69	41
Precentral Gyrus (LH)		2.77	71	66	40
Postcentral Gyrus (LH)	312	3.06	<b>75</b>	<b>58</b>	49
Cingulate Gyrus, anterior division (RH)	230	2.81	43	77	51
Paracingulate Gyrus (LH)		2.71	48	77	52
Insular Cortex (RH)	101	2.67	<b>29</b>	<b>72</b>	31
Frontal Operculum Cortex (RH)		2.48	24	71	37

Frontal Orbital Cortex (LH)	88	2.8	62	73	30
Frontal Pole (RH)	<b>76</b>	2.69	34	94	31
Postcentral Gyrus (LH)	47	2.81	49	43	64
Superior Temporal Gyrus, posterior division	29	2.42	80	<b>55</b>	38
(LH)					
Superior Frontal gyrus (LH)	23	2.43	48	<b>70</b>	64
Precentral Gyrus (RH)	10	2.48	16	62	45

*Note:* Harvard – Oxford Cortical Structural Atlas (Lateralized). Bold written data corresponds to the maximum voxel activation in a cluster.

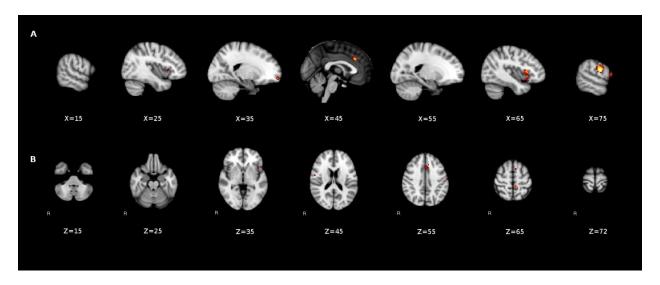


Figure 7. BOLD response in the non-corrected contrast 'Silbo' - 'Silbo' reversed > Spanish - Spanish reversed.

# Contrast Spanish – Spanish reversed > Silbo – Silbo reversed

Furthermore, Spanish speech contrasted to Silbo sentences did not show any significant result either. Another non-corrected contrast showed activation in many areas from the right hemisphere, such as the lateral occipital cortex, Z=3.14 in a cluster of 392 voxels; the right temporal pole; right fusiform cortex; and some other areas represented in Table 3 and Figure 8.

Table 3.

Contrast Spanish – Spanish reversed > 'Silbo' - 'Silbo' reversed

Location	Cluster size	Max z	X	у	Z
Lateral Occipital Cortex, superior division	392	3.14	21	26	50
(RH)					
Cingulate Gyrus, posterior division (RH)	289	2.72	41	42	<b>50</b>
Precuneus Cortex (RH)		2.54	39	35	41
Precuneus Cortex (LH)		2.51	46	34	41
Lateral Occipital Cortex, inferior division (LH)	127	2.67	68	<b>30</b>	43
Lateral Occipital Cortex, superior division (LH)		2.61	70	28	45
Left Middle Temporal Gyrus, temporo occipital		2.6	68	32	41
part					
Right Temporal Pole	95	2.8	20	69	<b>17</b>
Right Middle Temporal Gyrus, anterior	87	2.69	<b>17</b>	63	<b>26</b>
division					
Cerebellum Left Crus II	87	2.9	<b>62</b>	25	10
Cerebellum Right VI	60	2.59	<b>26</b>	<b>37</b>	22
Cerebellum Right Crus I		2.58	23	39	21
Right Temporal Occipital Fusiform Cortex		2.53	25	39	25
Right Angular Gyrus	29	2.6	23	<b>37</b>	43
Left Hippocampus	28	2.45	<b>56</b>	<b>58</b>	25
Cerebellum Right VIIb	17	2.48	26	38	11
Cerebellum Right Crus II		2.44	26	37	13
Right Superior Frontal Gyrus	12	2.58	38	73	61

*Note:* Cerebellar Atlas in MNI152 space after normalization with FLIRT, and Harvard – Oxford Cortical Structural Atlas (Lateralized). Bold written data corresponds to the maximum voxel activation in a cluster.

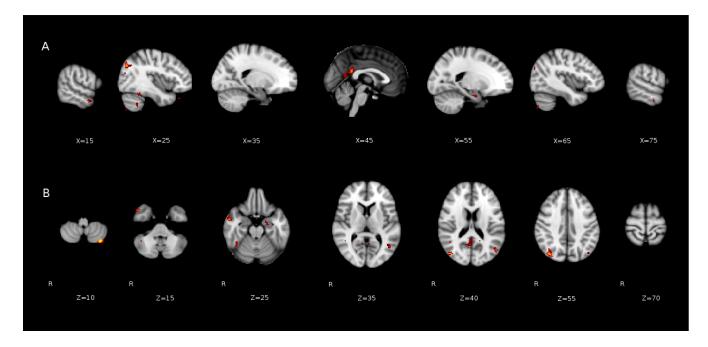


Figure 8. BOLD response without correction in Spanish – Spanish reversed > Silbo – Silbo reversed contrast. A row corresponds with sagital brain slices, and B row with axial ones.

# **Discussion**

In this experiment, nine whistlers performed a passive-listening task, to Silbo and Spanish sentences. The aim of this study was to know whether Silbo and normal speech are treated the same by our brain.

A non-corrected analysis of our data shows common activated areas in the left temporal lobe. Concretely, the highest activation was found in the left superior temporal gyrus, posterior division. Also, activation in its anterior portion, the left middle temporal gyrus, anterior and posterior division, and the left temporal pole. Left inferior frontal gyrus, pars opercularis, was activated in Silbo – Silbo reversed and not activated in Spanish – Spanish reversed. Left middle temporal gyrus, temporo-occipital part was activated in Spanish – Spanish reversed and not in Silbo – Silbo reversed.

The common activated areas in the temporal lobe might imply that there are common anatomical regions in language comprehension for both whistled and Spanish languages. The superior temporal gyrus (STG) in its posterior division, matches with the Wernicke's area and is related to language comprehension and the visual representation of words (Junqué & Barroso, 2009). The left temporal cortex, out of the classical language areas, works as an intermediate of conceptual and linguistic systems, thus, it allows the access to lexical pertinent forms to the activated concepts (Junqué & Barroso, 2009).

Also, the activation shows a ventral-anterior pattern, which implies the activity of the neural circuit involved in speech meaning processing and the identity of the speaker, the 'what' pathway (Scott and Johnsrude, 2003).

Our results have similarities with these found in Carreiras et al. (2005), but we do not find any significant activation in the right temporal lobe. It might be due to the effect of different and alternated speakers and whistlers that we have mentioned in the introduction. A more significant activation may be found in the right temporal lobe when the speaker changes, independently from the vocalization they produce, *Silbo* or Spanish (Rauschecker & Scott, 2009).

Moreover, Güntürkün, Güntürkün and Hahn (2015), studied lateralization of syllables processing, and both the experiment from Carreiras et al., 2005 and our experiment utilized sentences. The non-lateralization concluded in the first experiment could be due to the lack of semantic processing of their inputs. Even though, they show a lateralization in the same task for vocalised syllables.

Additional contrasts, shows differences in other areas activation that have not been considered as key areas for language in our review.

### **Conclusions**

Corrected analysis did not show any significant result but tendencies, and it is probably due to the little sample we used. An experiment with a larger sample would be necessary for making more consistent conclusion. Another interesting comparison would include non-whistler participants.

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#### References

- Bellugi, U., Poizner, H. & Klima E. S. (1989) Language modality, and the brain. *Trends Neuroscience*. 12: 380-388.
- Binder, J. R., Rao, S. M., Hammeke, T. A., Frost, J. A., Bandettini, P. A., Jesmanowicz, A., & Hyde, J. S. (1995). Lateralized human brain language systems demonstrated by task subtraction functional magnetic resonance imaging. *Archives of Neurology*, *52*(6), 593-601. doi:10.1001/archneur.1995.00540300067015
- Carreiras, M., Lopez, J., Rivero, F., & Corina, D. (2005). Linguistic perception: Neural processing of a whistled language. *Nature*, *433*(7021), 31-32. doi:10.1038/433031a
- Cowan, G. M. (1975). Mazateco whistle speech. Bibliotheca Phonetica, (11), 108-117.
- Engelien, A., Sibersweig, D., Stern, E., Huber, W., Frith, C., & Frackowiak, R. S. J. (1995). The functional anatomy of recovery from auditory agnosia. *Brain*, *118*(6), 1395-1409. doi: 10.1093/brain/118.6.1395
- Guentuerkuen, O., Guentuerkuen, M., & Hahn, C. (2015). Whistled turkish alters language asymmetries. *Current Biology*, 25(16), R706-R708.
- Junqué, C. & Barroso, J. (2009). Manual de Neuropsicología. Madrid: Editorial Síntesis.
- Klomp, D. Koning, W., Hoogduin, H., Raaijmakers, A., Petridou, N., Van den Berg & N.,

  Luijten, P. (2011). *Practical design of RF transmit and receive arrays*. Proceedings of

  European Society of Magnetic Resonance in Medicine and Biology (ESMRMB)

  Congress, Leipzig.
- Kohler, E., Keysers, C., Umilta, M. A., Fogassi, L., Gallese, V., & Rizzolatti, G. (2002). Hearing sounds, understanding actions: action representation in mirror neurons.

  Science, 297(5582), 846-848.

- Meyer, J. (2008). Typology and acoustic strategies of whistled languages: Phonetic comparison and perceptual cues of whistled vowels. *Journal of the International Phonetic*Association, 38(1), 69-94. doi:10.1017/S0025100308003277
- Nichols, T., Brett, M., Andersson, J., Wager, T. & Poline, J.B. (2005). Valid Conjunction Inference with the Minimum Statistic. *NeuroImage*. 25 (3), 653-660. doi:10.1016/j.neuroimage.2004.12.005 Preprint
- Pekar, J. J. (2006). A brief introduction to functional MRI. IEEE Engineering in Medicine and Biology Magazine, 25(2), 24-26.
- Poldrack, R. A., Mumford, J. A., & Nichols, T. E. (2011). Handbook of functional MRI data analysis. Cambridge University Press.
- Price, C. J. (2000). The anatomy of language: contributions from functional neuroimaging. *Journal of anatomy*, 197(3), 335-359. doi:10.1046/j.1469-7580.2000.19730335.x
- Purves, D., Augustine, G.J., Fitzpatrick, D., Hall, W.C., LaMantia, A.S. & White, L.E. (2012).

  Neuroscience. Sinauer Associates. Fifth Edition.
- Rauschecker, J. P., & Scott, S. K. (2009). Maps and streams in the auditory cortex: nonhuman primates illuminate human speech processing. Nature neuroscience, 12(6), 718-724. doi:10.1038/nn.2331
- Schild, H.H. (1990). fMRI made easy. Berlin/Bergkamen: Schering.
- Scott, S. K., & Johnsrude, I. S. (2003). The neuroanatomical and functional organization of speech perception. *Trends in Neurosciences*, 26(2), 100-107. doi:10.1016/S0166-2236(02)00037-1

- Scott, S. K., & Wise, R. J. (2004). The functional neuroanatomy of prelexical processing in speech perception. *Cognition*, 92(1), 13-45. doi:10.1016/S0010027703002269
- Trujillo, R. (1978). *El Silbo Gomero: Análisis Lingüístico*. (Canaria-Andrés Bello, Tenerife, 1978).
- Tzourio, N., El Massioui, F., Crivello, F., Joliot, M., Renault, B., & Mazoyer, B. (1997).

  Functional anatomy of human auditory attention studied with PET. *Neuroimage*, *5*(1), 63-77. doi:10.1006/nimg.1996.0252
- Uppenkamp, S., Johnsrude, I. S., Norris, D., Marslen-Wilson, W., & Patterson, R. D. (2006).

  Locating the initial stages of speech-sound processing in human temporal cortex.

  Neuroimage, 31(3), 1284-1296. doi:10.1016/j.neuroimage.2006.01.004
- Ward, J. (2015). The Student's Guide to Cognitive Neuroscience. Psychology Press.
- Woermann, F. G., Jokeit, H., Luerding, R., Freitag, H., Schulz, R., Guertler, S., ... & Ebner, A. (2003). Language lateralization by Wada test and fMRI in 100 patients with epilepsy. *Neurology*, *61*(5), 699-701.