Report Koen Nelissen, Research Project BB/11/08.

The role of the visual and motor systems in action understanding.

1. Introduction

The ability to understand what others do is a prerequisite for communication and for functioning in a social environment. Although the brain seems to cope with extracting this type of information from the visual scene naturally and effortlessly, it is only when deficits in this ability become evident (as is the case in individuals with autism-spectrum disorders), that one begins to appreciate the importance of gaining insights in the neuronal substrates underlying these abilities. Cognitive neurosciences investigating the topic of action understanding received a major boost about 20 years ago, when one of the still most hotly debated types of visuo-motor neurons was discovered in the premotor cortex of the monkey (di Pellegrino et al., 1992). These mirror neurons fire during active motor acts carried out by the animal (grasping a piece of apple), but also during observation of other individuals performing similar motor acts (Gallese et al., 1996) and it has been suggested they play a role in different aspects of social cognition in humans and non-human primates, ranging from action and intention understanding, empathy, language, theory-of-mind skills and imitation learning. Twenty years after their initial discovery, our knowledge of both the organization and function of the mirror neuron system in far from complete. An investigation of the monkey mirror system with functional MRI will be essential to understand the apparent discrepancies between the organization and role of the mirror neuron system in both human and non-human primate species.

A better understanding of the organization and function of the mirror neuron system is timely and of significant importance, given recent proposals that a dysfunctional human mirror system constitutes the _basis of autism_, a still highly controversial view. Furthermore, recently action observation has received a lot of attention because of its potential use in promoting _motor recovery in people affected by stroke_. This non-invasive action observation therapy relies on the ability to stimulate the motor cortex through the visual component of the mirror neuron system. Therefore, a more in-depth understanding of how visual information has access to the motor cortex will be important for optimizing current stroke recovery therapies using action observation.

2. Aims of the project

***Aim 1**: To map the monkey brain regions activated during execution and observation of prehension motor acts using awake fMRI with high spatial resolution (1mm isotropic). Monkeys will be trained to both passively observe videos showing grasping actions done by others as well as actively perform grasping movements in the MR scanner, while brain activity will be measured.*

***Aim 2**: To investigate the role of the monkeys` mirror neuron system in understanding goal-directed actions (grasping), using both fMRI adaptation and action categorization, followed by reversible lesioning of the different mirror neuron and action observation responsive sectors.*

***Aim3**: To investigate the presence of mirror responses in the somatosensory cortex of the monkey, outside the classical grasping-related mirror system.*

2. Methods and results

2.1 Methods and Results Aim 1:

An important aspect of mapping the mirror neuron system using functional MRI, involves the development of MR-compatible motor tasks, allowing investigation of brain responses during the execution of skilled manual motor tasks.

We developed two different MR-compatible carousels (Fig.1) in order to study the different components of skilled grasping motor actions including reaching, grasping and lifting in non-human primates.

Monkeys were seated in an MR-compatible chair (Fig.1A,B; 1) while a rotating carousel (Fig.1A,B; 2) containing several objects (Fig.1A,B; 4) was placed in front of the chair. In one setup (Fig.1A), monkeys performed grasping movements in the dark, while fixation on a translucent screen in front of them (Fig.1A; 7). In the second setup, monkeys performed the grasping action in the light, while directly fixation the objects in front of them (Fig.1B; 4). Fig 1 C-F shows a more detailed view of the rotating carousels containing different graspable objects.
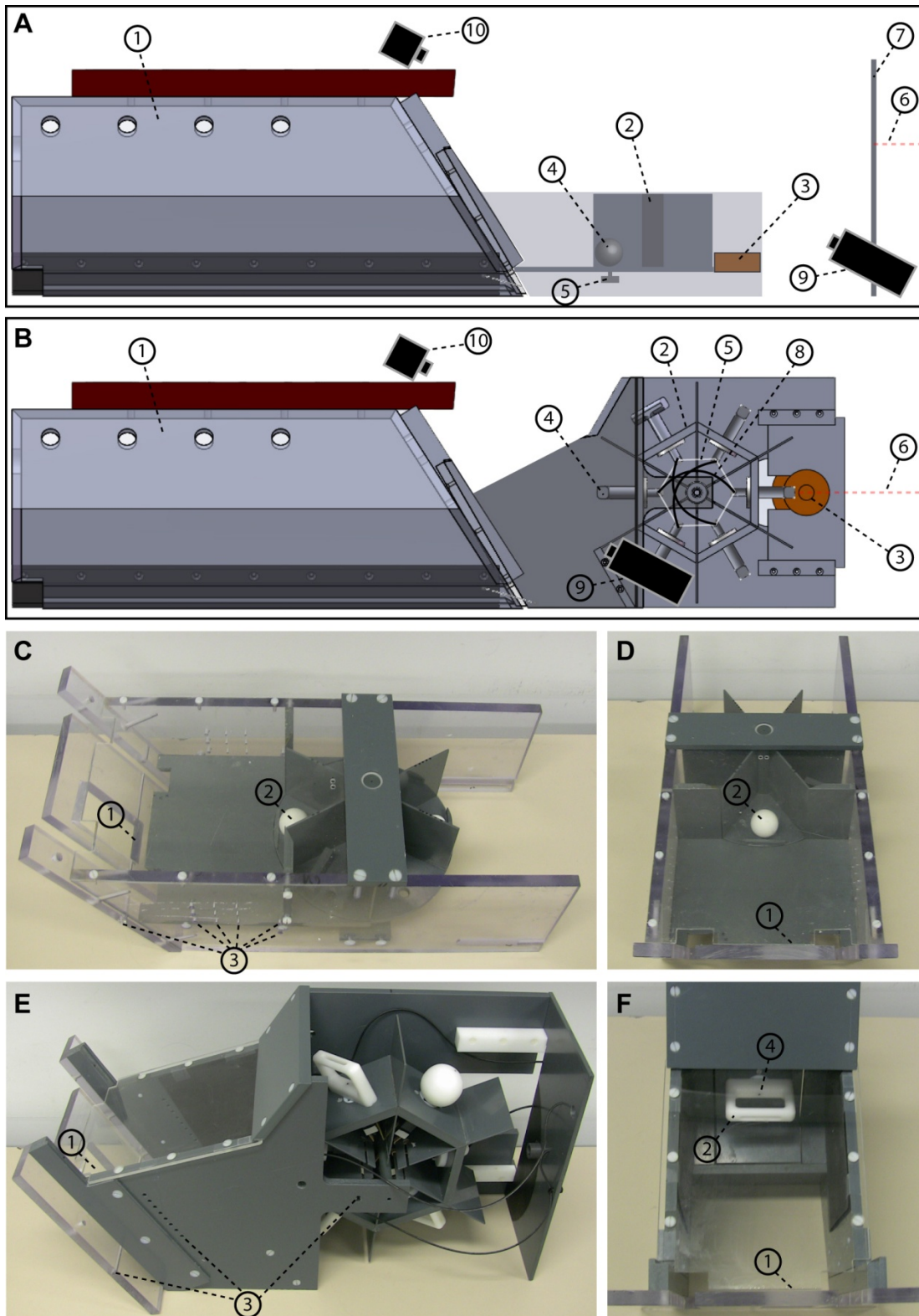
Figure 1: MR-compatible grasping setups.

In order to map the brain regions involved in grasping observation and execution, monkey were trained to alternate between active motor tasks and passive observation tasks. The initial motor tasks were only performed in the dark (to exclude brain responses during active

grasping tasks due to vision of the own hand) and monkeys were trained to respond to different colored fixation targets presented on the screen in front of them. Figure 2 shows an overview of a typical reach-and-grasp trial. Monkeys were trained to fixate a green fixation point on the translucent screen in front of them (Fig.1A; 6) while keeping their hand in the start position. When the fixation point turned to blue, they were required to reach forward and grasp the object in front of them in order to receive a liquid juice reward. During reach-only trials, monkeys were cued with exactly the same visual cues, but instead of an object, an empty slot on the rotating disk was positioned in front of them (Figure 3). During this motor task, when the cue turned blue, they were required to reach forward and place their open hand on the disk in order to receive a juice reward. During baseline 'rest' blocks, monkeys were rewarded for fixating a red cue while keeping their hand at the start position (Fig.2, left side).
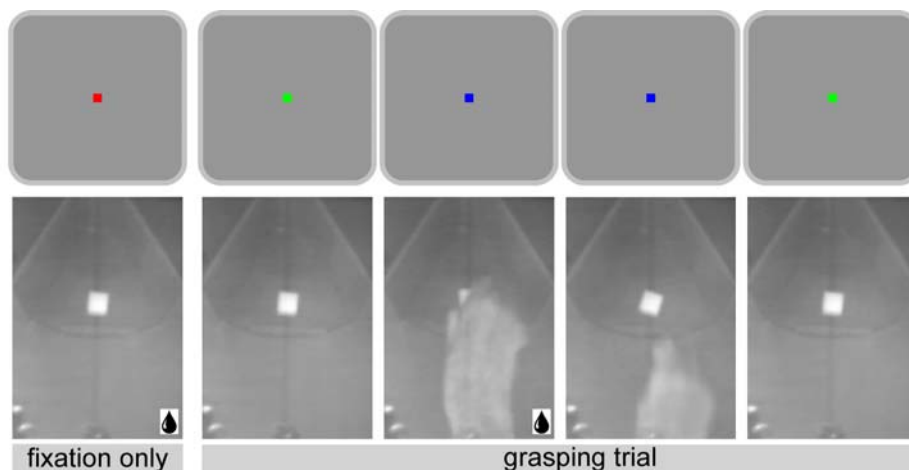

Figure 2: Fixation only and grasping motor trial

Figure 3 shows an overview of the actual fMRI grasping mirror neuron system experiment. Monkeys performed the different visual (reaching and grasping observation) and motor (reaching and grasping execution) tasks in consecutive blocks of 30 seconds. During 'rest' blocks, monkeys fixated on a small red fixation spot presented on the screen in front of them as indicated in Figure 2. These 'rest' blocks served as a baseline condition. In alternation blocks, monkeys either performed the reach-and-grasp or reach-only motor tasks, or passively fixated to videos showing persons performing similar reach-and-grasp (grasp observation) or reach-only (reach observation) actions.

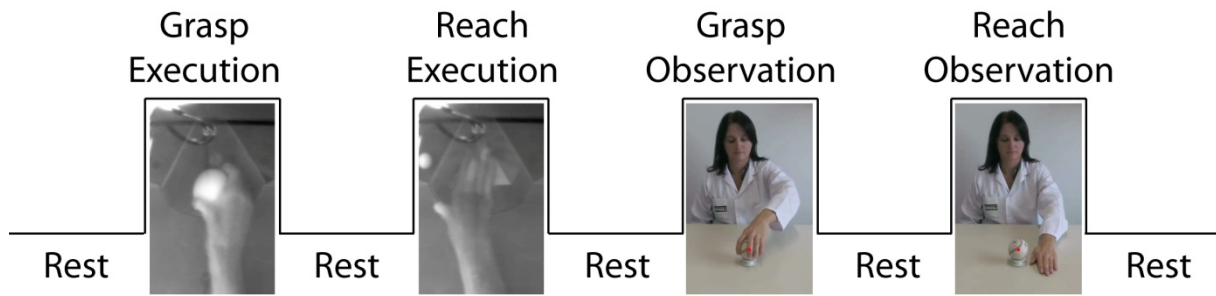| Grasp Execution | Reach Execution | Grasp Observation | Reach Observation |

Rest | Rest | Rest | Rest | Rest

Figure 3. Different experimental conditions used in the mirror system mapping fMRI scans.

Previous electrophysiology studies in monkeys have demonstrated grasping-related mirror neurons in parietal (areas PF/PFG and AIP), premotor (F5) and motor (F1) cortex. Figure 4A demonstrates the location of these regions on a lateral view of a monkey hemisphere.
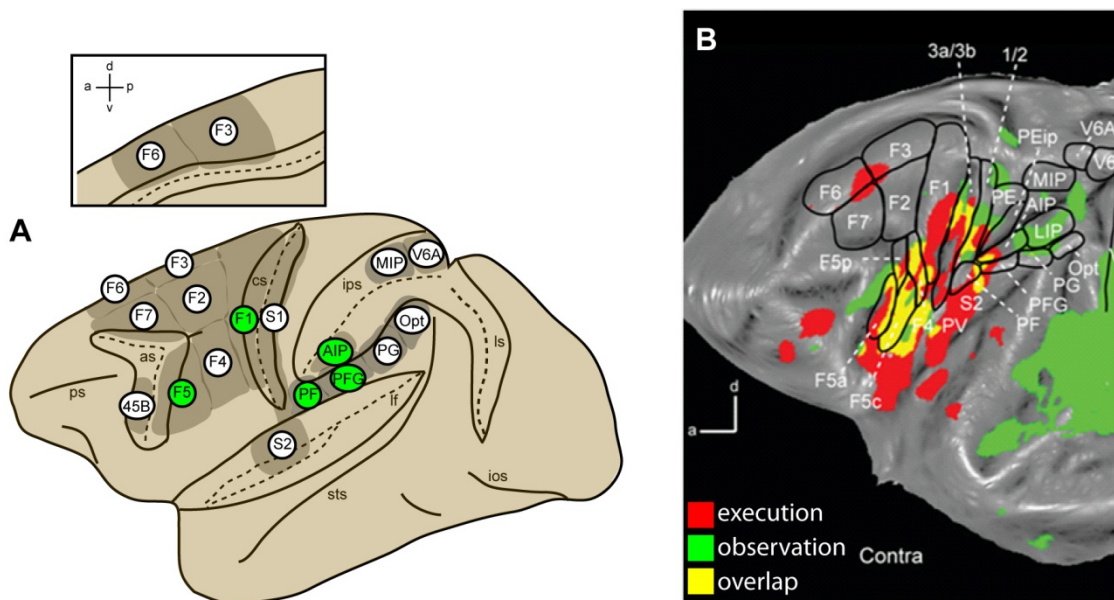

Figure 4. Location of grasping mirror neurons in the monkey (A) and overlapping grasping execution/observation fMRI responses in the monkey brain (B).

The preliminary results of the whole brain grasping mirror system mapping experiments are shown in figure 4B. A flattened representation of the left brain hemisphere is shown with areas active during grasping motor actions color-coded in red, areas active during grasping observation in green and areas responding both during grasping execution and observation (putative grasping mirror regions) in yellow. Brain regions responding to grasping execution and observation included portions of parietal areas PF/PFG and AIP, in addition to several

premotor (F4 and F5) and motor (F1) sectors. Finally, clear overlap was also observed in primary and secondary somatosensory cortex. This overlap between grasping execution and grasping observation responses in somatosensory cortex is in line with recent observations in the human and suggest a more extended mirror system in the monkey than previously assumed based upon single cell studies. These fMRI mapping studies can guide future electrophysiology recordings to investigate the mirror responses at the single cell level.

2.2 Methods and Results Aim 2:

Previous electrophysiology and imaging studies in monkeys looking into the possible neural correlate of action understanding, have shown brain responses during passive action observation in superior temporal sulcus, parietal, premotor and frontal regions. However, from these studies it is difficult to draw conclusions on the monkeys' ability to understand the goal of the observed action since these experiments required only passive fixation. In addition, these studies do not allow making strong inferences about a regions' role in action understanding, since they did not demonstrate a causal role of activity in these regions and the monkeys' ability to understand actions done by others.

Therefore we developed an active goal categorization task to investigate rhesus monkeys' ability to categorize videos of hand-object actions according to the goal depicted in the action (grasping vs not-grasping).

Two rhesus monkeys were trained to categorize hand action videos according to the goal of the action depicted (grasping versus not-grasping). In the initial training, after a 3 sec video presentation, the video disappeared and was followed by presentation of a peripheral target point (Figure 5A). Monkeys were required to make either a leftward or rightward saccade, depending on the goal depicted in the action video (grasping or not-grasping, respectively).

In the final test, 2 peripheral targets appeared simultaneously and monkey were rewarded for making a saccade to the left or right target when a grasping action or a non-grasping action, resp. was shown (Fig.5B) During several baseline trials, monkeys fixated a fixation point for 3 seconds after which the two peripheral fixation points were shown (monkeys were rewarded for making a saccade to either one of the two peripheral targets).

After performance in the categorization task was proficient (more than 90% correct trials), monkeys were tested on their ability to generalize to other untrained grasping or not-grasping actions. During these generalization tests, untrained hand action videos depicting

either grasping or not-grasping actions were shown and monkeys were rewarded for making a saccade to either side, in order to excluded learning effects (Fig 5C).
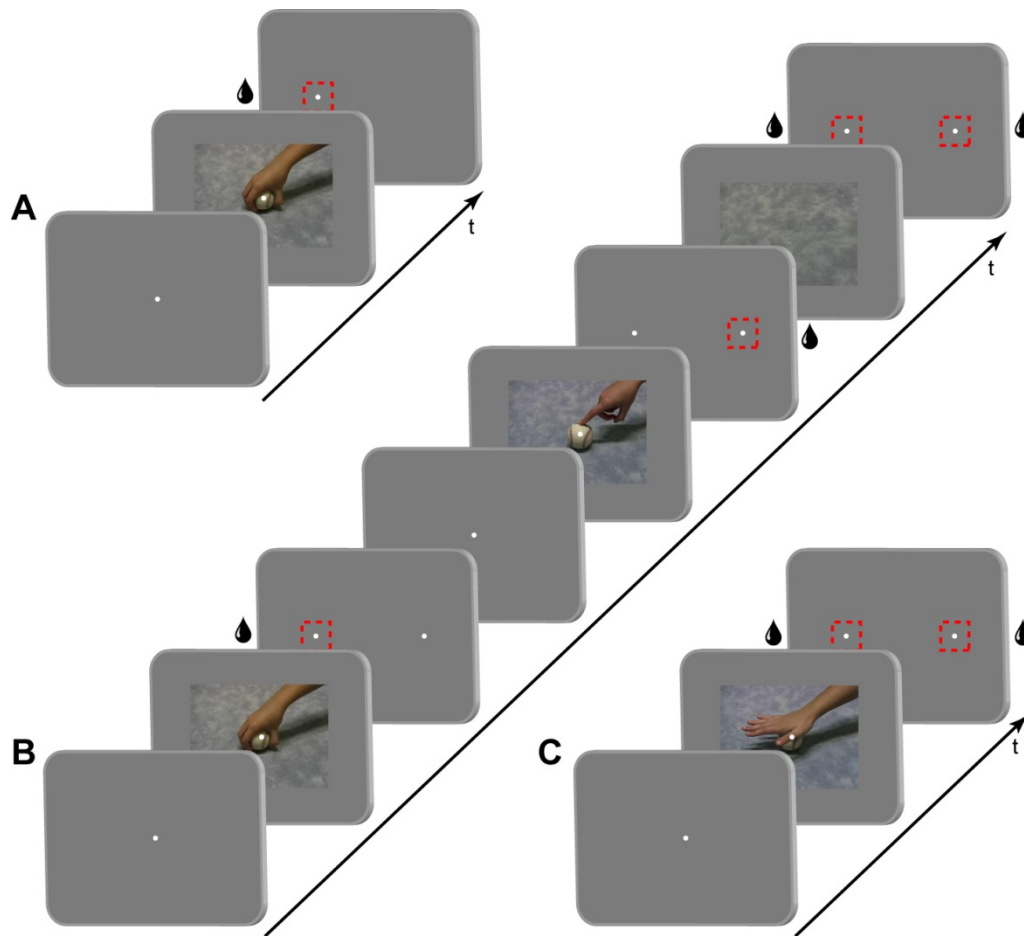

Figure 5: Action categorization experiment and generalization test.

These generalization tests showed that monkeys were proficient at correctly categorizing both new untrained grasping actions with different grip types, objects or biological effectors (Fig. 6A,B; 1-4), as well as new not-grasping actions including mimicked grasps and open hands touching the object (Fig. 6A,B; 5-8). It should be noted however that this generalization to untrained videos did not work for all tested videos. Monkeys failed to categorize correctly an artificial robotic arm performing a grasp (Fig.6A,B; 9) or a closed fist touching an object (Fig. 6A,B; 10).

These results demonstrate the feasibility to use cognitive more demanding active action recognition tasks in rhesus monkeys to investigate the neuronal correlates of action understanding.
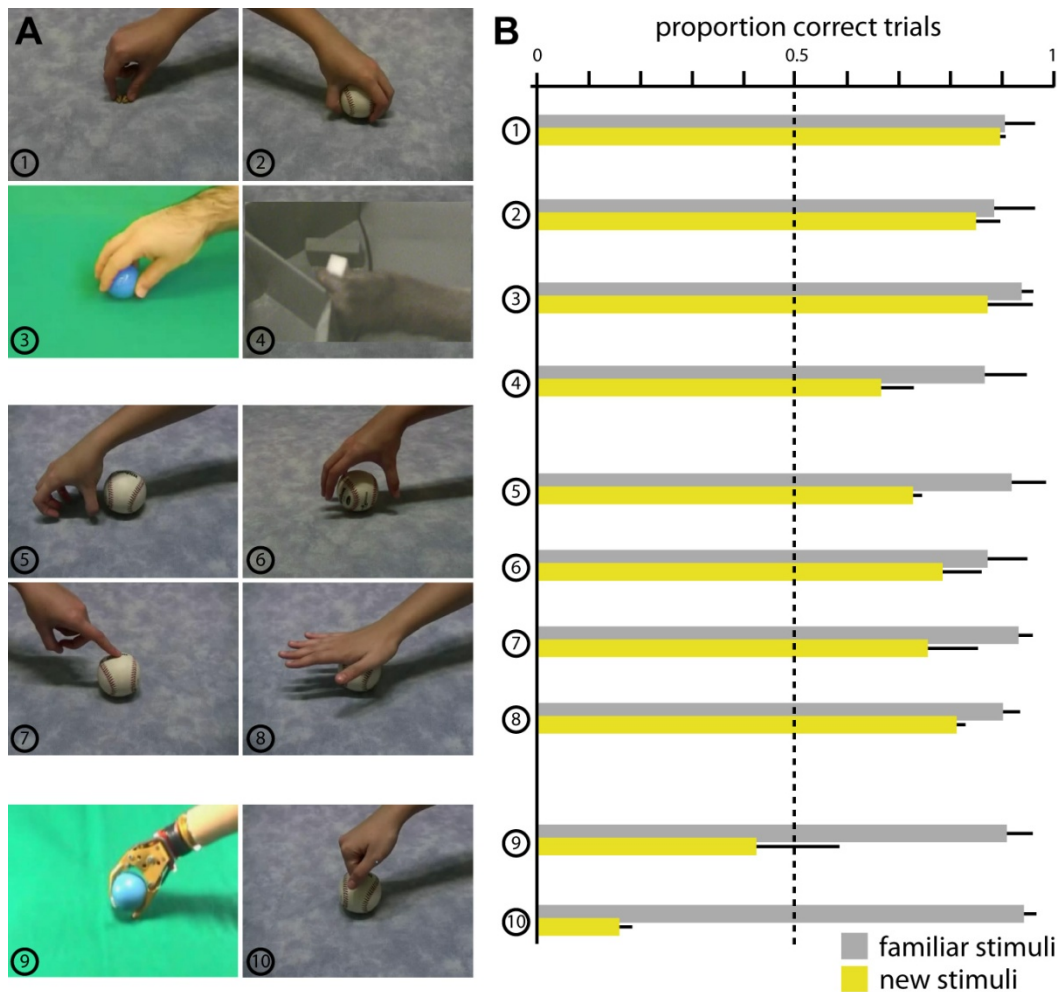
Figure 6: Result of action generalization tests.

In order to investigate the brain regions activated during this active action categorization task, the two monkeys were also scanned while performing this goal categorization task. Preliminary results of these experiments suggest that active action categorization (grasping vs non-grasping) activates both visual and motor regions (Fig. 7B). Interestingly, grasping categorization (versus controls) yielded significant MR signal increases in several brain sectors (Fig. 7B,C) that also respond during active grasping motor tasks (Fig. 7A), including parietal area AIP and premotor F5, in addition to portions of somatosensory cortex S2.
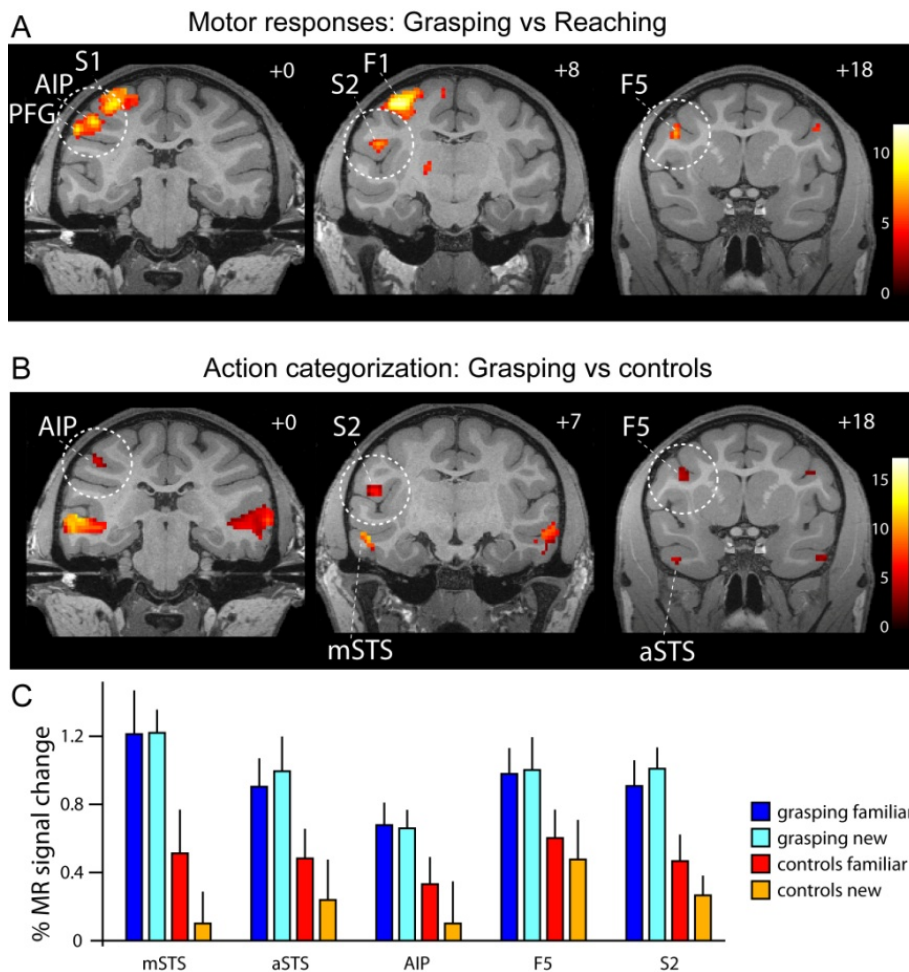
Figure 7: Brain responses during grasping execution (A) and active action categorization (B,C)

During future experiments, I will try to disentangle the specific contribution of these visual and motor sectors to goal understanding by assessing the causal behavioral effects of selective inactivation of these sectors while the animals perform this active action categorization task.

In parallel, I will also perform similar behavioral and functional imaging experiments in humans, to investigate the role of the mirror system in goal understanding.

Methods and Results Aim3*: To investigate the presence of mirror responses in the somatosensory cortex of the monkey, outside the classical grasping-related mirror system.*

Both the mirror system mapping (Aim 1) and action categorization (Aim 2) experiments showed that passive grasping observation also yields responses in somatosensory cortex in the non-human primate. This suggests that the sensory events might also be mirrored in

additional brain regions outside the motor system. These observations are in line with data obtained in humans using functional imaging, showing overlapping responses in the subjects' brain both when the subject is being touched and when he observes a person being touched. It has been proposed that these mirror responses in somatosensory cortices play a role in empathy in humans and a possible similar neural correlate in non-human primates has so far not been investigated.

In order to map the hand field of the monkey somatosensory cortex, I performed several anaesthetized fMRI experiments during which the fingers or face of the subject were being stimulated with air puffs. These yielded significant responses in portions of both primary and secondary somatosensory cortex. In future experiments, I will investigate the overlap between these somatosensory responsive regions and the fMRI responses obtained during observation of videos displaying both active and passive touch.

3. Added value of the project for the research program and host unit

The topic of action recognition in the human and non-human primate brain using fMRI have been part of several previous national projects in our lab (GOA 2005/18, excellence financing EF/05/14 and IUAP), with the electrophysiology counterpart being studied by Prof. R. Vogels. The execution of grasping motor acts, which complements the visual studies, has been studied with fMRI during my FWO postdoc fellowships (2006 – 1012) and my Belspo Return Fellowship (2012 - 2014), as well as with electrophysiology by the group of Prof. P. Janssen in the EF project and another European project (Neuroprobes IST 027017). The topics of action recognition and execution are also part of the current IAP and PF financing. Currently, comparative studies on the processing of facial gestures in monkeys and humans are being performed in collaboration with the group of Psychiatry (Prof. M. Vandenbulcke). In addition, the topic of action recognition is studied in parallel with fMRI in the human brain in collaboration with Dr. J. Jastorff (Psychiatry, KU Leuven) and Dr. R. Peeters (Radiology, UZ Leuven). Finally, we continue to have a strong collaboration with the anatomist group of Parma University to study the organization of the grasping motor and mirror system by combining fMRI data with hodological data (Prof. G. Luppino and Prof. S. Rozzi).

4. Perspectives of integration of researcher into the Belgian scientific milieu

Opinion of Wim Vanduffel (promoter and head of lab for Neuro- & Psychophysiology): Dr. Koen Nelissen is one of the world leaders in functional neuroimaging of non-human primates. He is specialized in the mirror neuron system and was the first one to perform fMRI experiments with monkeys performing complicated motor tasks inside the scanner. At the KU Leuven, Dr. Nelissen is the driving force in this research domain in which he already made substantial contributions as evidenced by several highly cited landmark papers. Recently, Dr. Nelissen developed a research plan for the next 5 years that contains a healthy mixture of high-risk high gain and solid incremental research projects. In this context, his research fits perfectly with the line of research of the Laboratory of Neuro-and Psychophysiology. Dr. Nelissen was recently awarded with a BOF-ZAP tenure track position at the lab for Neuro- and Psychophysiology at KU Leuven, starting October 1th 2014. I'm convinced that he will outperform most of his peers and will have a brilliant academic future.

5. Publications since start of project

Peer-reviewed papers
1. Arsenault JT, **Nelissen K**, Jarraya B, Vanduffel W. Dopaminergic reward signals selectively decrease fMRI activity in primate visual cortex. Neuron, 2013; 1174-1186. Impact factor (IF): 14.76
2. Zhu Q*, **Nelissen K***, Van den Stock J*, De Winter FL, Pauwels K, de Gelde B, Vanduffel W, Vandenbulcke M. Neuroimage, 2012; 66C: 402 − 411. Dissimilar processing of emotional facial expressions in human and monkey temporal cortex. **First authorship shared.** IF: 5.90
3. **Nelissen K***, Jarraya B*, Arsenault J, Rosen B, Wald L, Mandeville J, Marota J, Vanduffel W. Neural correlates of the formation and retention of cocaine-induced stimulus-reward associations. Biological Psychiatry, 2012; 72: 422-428. **First authorship shared**. IF: 8.67

Abstracts
1. **Nelissen K**, Vanduffel W. Goal understanding in non-human primates: active action categorization tasks. Annual meeting of Vision Sciences Society. Naples, Florida, 10-15 May 2013, Abstract No. 26.415.
2. **Nelissen K**, Vanduffel W. Goal understanding in non-human primates: active action categorization tasks. 10th meeting of the Belgian Society for Neuroscience, May 31th 2013, Campus Jette, Brussels.
3. **Nelissen K**, De Maziere P, Vanduffel W. Basal ganglia fMRI responses during skilled manual motor tasks in the non-human primate. Society for Neuroscience. New Orleans, 13-17 October 2012, Abstract No. 380.16/VV16.

4. Orban GA, Peeters R, Vanduffel W, Rizzolatti G, **Nelissen K**. Using parallel fMRI in human and nonhuman primates to locate premotor area F5c in humans. Society for Neuroscience. New Orleans, 13-17 October 2012, Abstract No. 13.10.

5. Mantini D, Zhu Q, **Nelissen K**, Van den Stock J, De Winter FL, Vandenbulcke M, Vanduffel W. Inter-species activity correlations between face-selective brain areas in humans and macaques. Society for Neuroscience. New Orleans, 13-17 October 2012, Abstract No. 13.06.