# **PART TWO** | Development of Molecular Imaging strategies

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# Chapter 7

## MR-based molecular imaging of the brain: the next frontier

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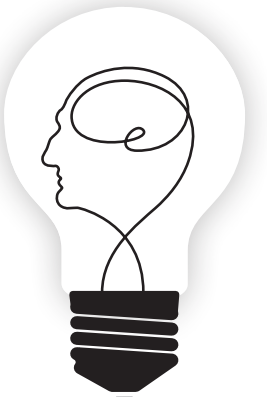

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## Abstract

In the foreseeable future the molecular imaging field could greatly assist neuroradiologists. Reporter molecules provide information on specific molecular or cellular events that could not only aid diagnosis, but potentially differentiate between different stages of disorders and treatments. In order to accomplish this, reporter molecules literally need to pass a barrier, the blood-brain barrier, which is designed to repel non-essential molecules from the brain. Although this is not a trivial task, several transport systems could be tricked into guiding molecules into the brain.

The non-invasive nature in conjunction with a wide availability makes MRI particularly suitable for longitudinal neurological imaging studies. This review explains the principles of MR contrast, delineates different types of reporter molecules and describes strategies to transport reporters into the brain. It also discusses recent advances in MR hardware, pulse sequences, the development of (targeted) reporter probes and future directions of the MR neuroimaging field.

## Introduction

Anatomical images have always been the center of gravity in the daily work of radiologists. They provide the basis of many diagnoses supplemented by physiological magnetic resonance data or metabolic profiling if necessary. Despite the sophistication of these techniques and the wealth of information that can be obtained, the diagnostic information often remains non-specific and evidence regarding the nature of the underlying disease commonly remains circumstantial. In contrast to generic contrast agents used in the clinic, the 'molecular imaging' (MI) field uses reporter molecules tailored for *in vivo* detection of specific molecular or cellular events. Formally, MI encompasses techniques that directly or indirectly monitor and record the spatiotemporal distribution of molecular or cellular processes for biochemical, biologic, diagnostic, or therapeutic applications.[1,2] (**Table 7.1**) The technique is widely used in pre-clinical research and is on the verge of entering the clinical arena. It enables radiologists to add molecular or cellular information to their array of diagnostic tools, which will have a tremendous effect on the diagnosis of neurological disorders, where invasive diagnostic techniques like biopsies can rarely be used.[3,4] Radiologists may even be able to detect "pre-disease" or "pre-symptomatic states" when molecular and cellular changes arise before they lead to anatomical or functional disturbances. Following an early diagnosis, MI could closely monitor the effectiveness of therapeutic interventions.

MR imaging is already the imaging technique of choice for neuroradiologists and MR imaging systems are widely available. In principle one could collect anatomical and physiological information and report the location of reporter molecules in a patient during a single MR imaging session. For these reasons this review focuses on MR-based MI of the brain, explaining the principles of MR reporter molecules, describing strategies to target them to the brain and reviewing the state of the art in CNS MI.

## Brain targeting

An important feature of the brain that sets it apart from other organs in terms of MI is the presence of the blood-brain barrier (BBB), a selective barrier to the CNS that impedes the influx of most compounds from blood to brain. (**Figure 7.1A**) It permits the passage of metabolic compounds and ions to maintain neuronal function, while shielding off possible harmful compounds. The effective barrier results from the selective permeability of tight junctions between endothelial cells, although the underlying layers and cell types also exhibit great influence on its function and permeability. The endothelial cells in the cerebral vasculature differ from normal endothelial cells in having low pinocytotic activity, abundant mitochondria, fewer fenestrations and specialized junctions to adjacent cells; all these features play a role in the impermeability of the BBB.[5]

The BBB is generally regarded as a bottleneck for MI imaging of the brain, as it severely hinders the delivery of reporter probes to the brain. Designing reporter molecules such that they may cross the barrier is not trivial, although a good understanding of BBB physiology has resulted in several delivery strategies.[6] With paracellular transport sealed off, transmembrane transport

**Table 7.1 Definitions**

| | |
|---|---|
| Molecular imaging | *In vivo* imaging of the spatiotemporal distribution of molecular or cellular processes. |
| Cellular imaging | *In vivo* imaging of the spatiotemporal distribution of cellular processes. |
| Contrast agent, label | Chemical functional group that allows visualization by an appropriate imaging technique. For example, Gd chelates or iron oxides are MR contrast agents, 18F atoms are PET contrast agents, and fluorophores are optical contrast agents. |
| Nanoparticle | Molecules in the 10-1000 nanometer range that serve as an imaging platform. Examples include SPIO particles, liposomes, dendrimers and quantum dots. |
| Reporter probe, Reporter molecule | A molecule or nanoparticle that is used to image particular biological processes. The molecule or nanoparticle is a composite of a contrast agent and targeting moiety. |
| Reporter cell | A cell that contains a contrast agent. |
| Reporter gene | A gene that encodes for a protein that (directly or indirectly) is easy to assay. Reporter genes are linked to genes of interest to study expression levels. |

through endothelial cells is the only way to gain access to the brain. Several strategies to transport MI probes across the BBB can be followed, exploiting different endogenous transport systems: passive diffusion, carrier-mediated transport, receptor-mediated endocytosis and adsorptive endocytosis.[7-10] (**Figure 7.1B**)

Small molecules such as oxygen and carbon dioxide readily diffuse into the brain, but the BBB is quite restrictive for other compounds. Nevertheless, small non-charged lipophilic compounds may be engineered to passively enter the brain.[11] However, lipophilic compounds have major drawbacks, including enhanced uptake and retention by peripheral tissues, complicating the compound biodistribution and pharmacokinetics.[12] In practical terms, lipophilic compounds are generally not soluble in aqueous solutions. Organic solvents are usually excluded from *in vivo* experiments for toxicity reasons, while others that are in clinical use, such as DMSO, reduce the integrity of the BBB, which clearly is not desirable for non-invasive imaging purposes.[13]

Metabolites such as glucose, amino acids, nucleosides and neurotransmitters are transported into the brain by carrier-mediated transport through proteins in the plasma membrane of endothelial cells that catalyze bidirectional transport (blood to brain and *vice versa*).[14] These pumps operate on both sides of the cell to maintain a nutritional balance. Reporter molecules that mimic the structure of a nutrient could trick the transport system to gaining entrance to the brain, although efflux pumps on the luminal (blood) side may impede this effort.[15] Positron emission tomography (PET) has extensively employed this strategy, e.g. by using fluorodeoxyglucose as a glucose analog.[16]

Larger reporter probes may target internalizing receptors, resulting in receptor-mediated endocytosis: following complex formation of the probe and the receptor, the complex is internalized, transported to the abluminal (brain) side of endothelial cells and released into the brain. The insulin receptor and transferrin receptor are well-known examples.[8] This transport mechanism is suitable for the translocation of macromolecules and nanoparticles and is therefore particularly interesting for MI.[7] A prerequisite for this mechanism is a receptor-binding ligand, such as a molecular mimic of an endogenous ligand or an antibody against the receptor of interest. Evidently, the ligand should be conjugated to a contrast agent or an MR imaging-detectable nanoparticle to be able to visualize it using MR imaging.

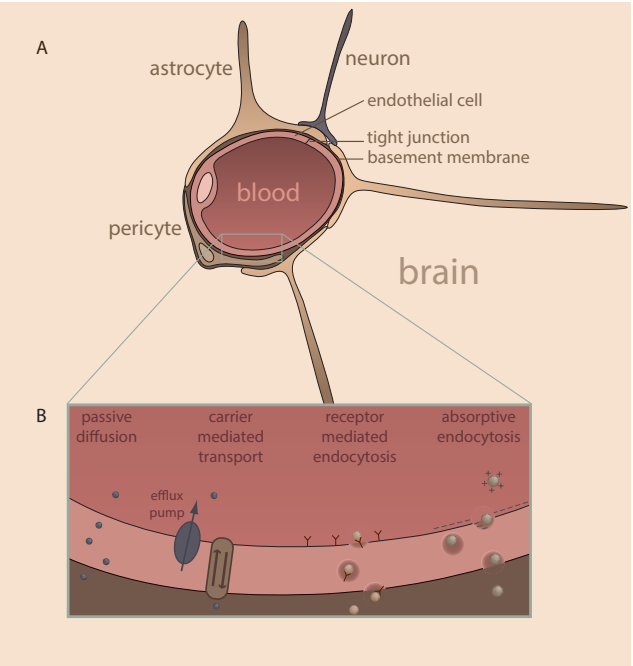


**Figure 7.1 Blood-brain barrier**
(**A**) Schematic representation of the blood-brain barrier(BBB) and (**B**) transport mechanisms for BBB passage (see text for details).

Compounds may also be internalized by non-specific interactions with the cell surface. Cationized albumin, for example, interacts with the anionic cell surface of the endothelial cells, and is then internalized via adsorptive endocytosis.[8] This is a general internalization mechanism that is not specific for endothelial cells, so with regard to MI this strategy is not very useful, as reporter probes would be internalized by a range of cell types, resulting in a high non-specific background signal.

It is possible to disrupt the BBB temporarily to gain access to the brain, e.g. through osmotic pressure (mannitol). A method that appears relatively safe is the injection of microbubbles into the bloodstream followed by focused local ultrasound exposure causing cavitation, leading to BBB disruption.[17] Such methods may be used for preclinical studies, but will not be suitable for clinical use of targeted probes, except perhaps for image-guided drug delivery.

## Generating MR Contrast

Whatever the object of interest is, the reporter system used to visualize it should contain an MR imaging-visible contrast agent. An overview of these agents is provided here below and in **Table 7.2**. Iron oxide particles (SPIO, CLIO and MION) are composed of iron oxide crystals with polymer coatings and are often biodegradable. They are synthesized in different forms and sizes, ranging from 300 nm to 1.6 μm in diameter, and each size category exhibits different pharmacodynamical behavior and relaxation effects.[18] All iron oxide particles possess relatively large (negative) magnetic susceptibilities, thereby reducing $T_2$ and $T_2$* relaxation times, resulting

**Table 7.2 MR contrast**

| MR Contrast agent | Examples | Predominant effect | MR sensitivity | Notes |
|---|---|---|---|---|
| $T_1$ agent | $Gd^{3+}$, $Mn^{2+}$ chelates | $T_1$ | ++ | Gd is chelated for toxicity reasons |
| $T_2$ agent | SPIO, USPIO MION, CLIO | $T_2$ | ++++ | Large range of sizes |
| CEST; paraCEST | Amino acids, sugars; Eu-DOTA | saturation transfer | ++ | Pre-clinical stage |
| Heteronuclei | $^{19}F$, $^{31}P$ | "Hot spot" imaging | + | Negligible background signal |

in signal voids or hypointense regions in $T_2$- or $T_2$*-weighted MR images. The fact that SPIOs cause 'negative contrast' could make them difficult to distinguish from imaging artifacts or other susceptibility sources. To overcome this disadvantage, imaging sequences are being developed that produce positive contrast using SPIOs, although the generated contrast is still non-specific and cannot be exclusively attributed to the presence of iron oxide particles.[19] Some SPIOs are approved by the FDA, making them popular choices for cellular and molecular imaging, although their relatively large size makes them less useful for targets that are not easily accessible, as in neurological applications.

Paramagnetic agents (e.g. Gd) have small positive magnetic susceptibilities that cause a modest decrease of relaxation times, particularly T1, resulting in 'positive contrast' on $T_1$-weighted images.[20] The positive contrast effect of paramagnetic contrast agents is much weaker than the negative contrast effect of SPIOs. Sensitivity could be enhanced by conjugating multiple Gd-containing chelates to a single probe, such as a dendrimer or protein, but the design of large complexes is always a trade-off between sensitivity and molecular weight. The larger (and 'brighter') contrast agents are synthesized, the more difficulties they will encounter to cross the BBB. A special class of paramagnetic contrast agents is encompassed by 'smart' or 'responsive' contrast agents. Their relaxation properties significantly change in response to local physiological changes, e.g. in pH, temperature or enzyme activity.[21,22] Paramagnetic agents are small and generally easy to conjugate to probes of interest, which makes them suitable for neuro-imaging, although they remain relatively insensitive.

Chemical Exchange Saturation Transfer (CEST) is contrast mechanism that has been developed over the past decade and is based on magnetization transfer through proton exchange.[23] CEST agents contain exchangeable protons with a resonance frequency different from bulk water. Upon selective irradiation of this frequency, magnetization is transferred to bulk water through chemical exchange, causing a decrease in the signal intensity of bulk water. The contrast can thus be switched 'on' (by presaturation of the CEST protons) or 'off' (no presaturation). The efficacy of CEST agents depends on the exchange rate and frequency difference between the exchangeable 'CEST protons' and bulk water and can be improved by the incorporation of lanthanide ions (PARACEST).[24] CEST probes can be engineered to work at different excitation frequencies, which potentially allows us to study interactions of cells labeled with different CEST agents.[25]

Contrast agents described thus far are detected indirectly, in the sense that they affect the relaxation properties of water protons. Their effect is registered in the final proton image. A more direct approach is heteronuclear MR imaging. In principle any nucleus with a non-zero magnetic spin and a large enough 'magnetic sensitivity' could be used. Fluorine ($^{19}$F) is a popular choice in the MI field, as it exhibits favorable MR characteristics with a spin-½, a magnetic sensitivity close to that of proton and 100% natural abundance. Moreover, there are hardly any endogenous fluorine-containing molecules, so fluorine-based reporter molecules do not suffer from background signal. Compared with proton imaging, however, the fluorine signal will be rather low; fluorine probes accumulate at most in the millimolar range, whereas the water concentration *in vivo* is roughly 40 mol/l. The design of fluorine-based probes is very variable, ranging from low-MW molecules with covalently bound fluorine atoms to perfluorocarbon molecules measuring hundreds of nanometers.[26-28] Low-MW fluorine probes may be designed to accumulate in the brain, where they could take full advantage of the 'hot spot imaging' principle if they locally agglomerate to sufficiently high concentrations.

## Reporter Systems

MR-based MI is used to detect a wide range of biological events, which can be classified in three categories (with some overlap between them). Firstly, the presence of specific *molecules*, such as receptors, can be imaged (see "*Detecting molecules*"). Secondly, specific *cells*, such as stem cells after transplantation (see "*Detecting cells*"), can be traced, and thirdly, the expression of specific *genes* (see "*Reporter genes*") can be visualized.

**Detecting molecules**

Molecular reporter probes come in many shapes and sizes, but their principal components are a targeting molecule and a contrast agent. The probe should be targeting a unique hallmark of the biological process of interest. Popular target choices are receptors, enzymes and cytokines, which are often expressed to much higher levels in pathologies. Once a target is chosen, one needs to select a complementary ligand that binds the target with high affinity and specificity. Natural ligands, such as receptor agonists, can be used, or specific ligands, such as antibodies or peptides, can be developed.

Reporter probes are synthesized in a wide range of sizes and can be divided in low-MW probes and large MW probes or nanoparticles. (**Figure 7.2**) Low-MW probes are based on drugs, metabolites or inhibitors, in which single atoms or small functional groups are substituted with a contrast moiety. For neurological MI applications, low-MW probes most likely find their way to the brain via passive diffusion or carrier-mediated transport. A beautiful example is the small fluorine-containing reporter probe developed by Higuchi *et al*.[27] This molecule targets amyloid plaques, one of the hallmarks of Alzheimer's disease (AD) and was intravenously injected into transgenic AD mice. (**Figure 7.3A-C**) Using a combination of $^{19}$F and $^{1}$H MRI, Higuchi showed the presence of this probe in the brain of these mice, presumably bound to amyloid plaques.

High-MW reporter probes (with MWs over 5 kDa and diameters ranging from a few to hundreds of nm) are based on proteins or nanoparticles to which contrast agents and ligands are

conjugated. Some nanoparticles, such as SPIOs or perfluorocarbon particles, generate contrast themselves (see section "*Generating MR contrast*"), while others (such as liposomes and dendrimers) simply supply a crosslinking platform. There is a trend in the field to construct

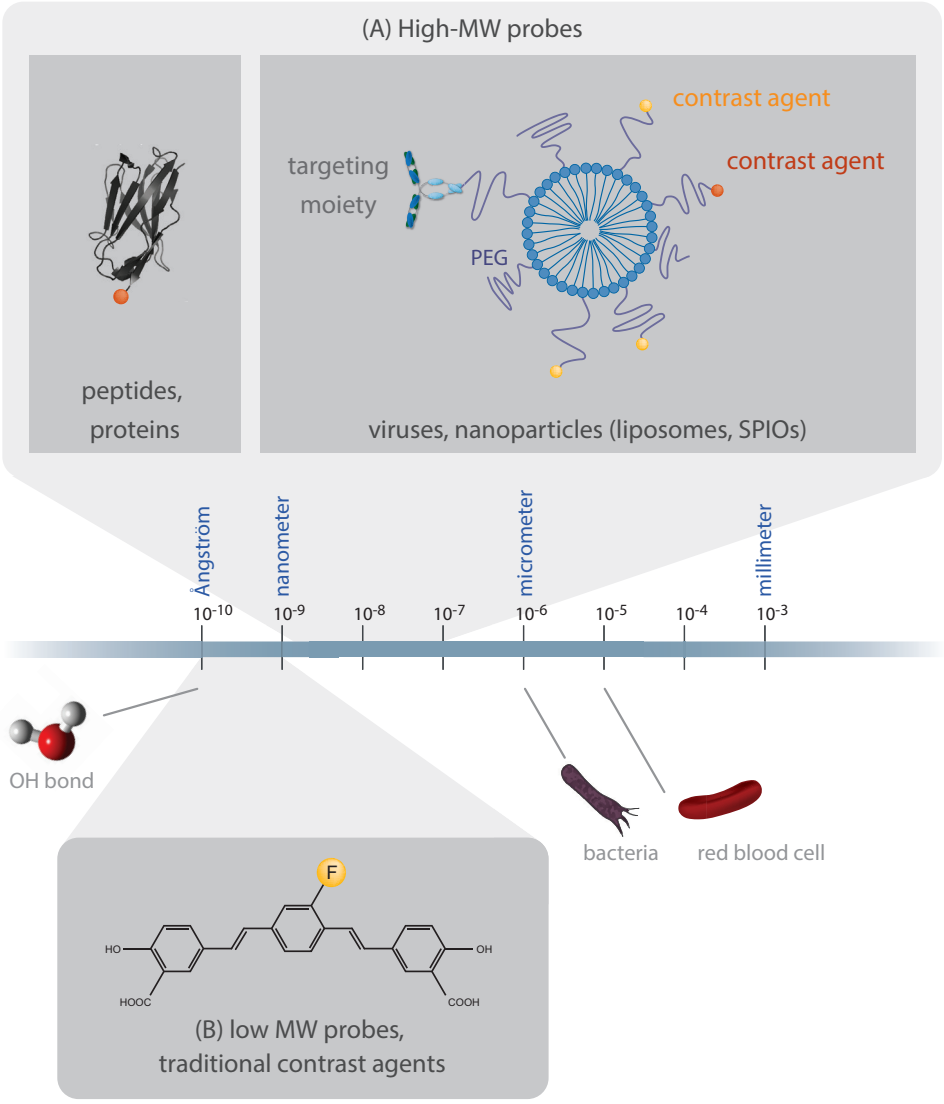


**Figure 7.2 Reporter probes and their orders of magnitude**
The actual size of reporter molecules ranges from a several Ångstroms ($10^{-10}$m) to hundreds of nanometers. (**A**) High-MW biomolecules (not drawn to scale), such as peptides or antibodies, that have target affinity could directly be labeled with a contrast agent. Reporter molecules based on liposomes or SPIO particles, tens to hundreds of nanometers in size, are conjugated to targeting moieties and contrast agents. (**B**) Low-MW reporter probes are small molecules that contain contrast-generating labels. They fall in the low nanometer range. The figure shows reporter probe FSB that is able to cross the blood brain barrier and bind to amyloid plaques. It is labeled with $^{19}$F for MR imaging.[27]

nanoparticles that contain multiple types of contrast agents, which can be visualized by different imaging modalities.[29] This concept, referred to as multimodality imaging, combines the strengths of different imaging techniques, such as sensitive PET tracers with high resolution MR images. For example, Kircher *et al*. set out to aid neurosurgeons visualizing brain tumor margins.[30] They synthesized a long-circulating multimodality reporter molecule, composed of an iron-oxide nanoparticle and a near-infrared fluorochrome and injected this into rats bearing gliosarcoma that expressed green fluorescent protein. MR imaging confirmed that microglia had sequestered the nanoparticle and displayed the tumor as hypointense regions in $T_2$-weighted images. Subsequent optical imaging allowed discrimination of tumors from brain tissue, which ultimately could be used by surgeons during surgery.

**Detecting cells**

Instead of visualizing reporter molecules, it is also possible to label intact cells, allowing cell tracking and gaining knowledge on cell behavior, such as migration patterns of immune cells following immunotherapy or stem cell survival following transplantation.

Evidently, cells should remain viable and functional after the labeling procedure. Cellular contrast agents should ideally remain within the desired cell type and not dilute with cell division to enable reliable longitudinal studies.[31,32] SPIO particles are by far the most used MR contrast agents used for MR cell tracking. However, a particular confound is that the MR image cannot distinguish viable cells from non-viable cells. Additionally, due to susceptibility effects, hypointense regions in MR images are significantly larger than the actual cluster of labeled cells, which could lead to misinterpretations of the iron source. MR imaging-based cell tracking does not exclusively lean on SPIO particles; Gd-based approaches benefiting from positive contrast have also been demonstrated, although substantial amounts of lanthanide-based contrast agents are required to affect MR images, which increases toxicity issues.[33] Phosphorous and fluorine imaging have the advantage of a low background and can often be performed in a quantitative manner, but they too suffer from sensitivity issues. Finally, CEST imaging is emerging as a very useful technique, especially in reporter gene imaging.[34]

Cell labeling can be performed *in vitro*, after which the labeled cells are implanted or the contrast agent may be injected systemically, which is then taken up by phagocytotic cells.[35] Many cell types readily take up contrast agents via phagocytosis, but non-phagocytic cells (including stem cells) can be labeled via transfection agents, fluid phase pinocytosis, encapsulation, receptor-mediated uptake or magneto-electroporation, amongst others.[31,32] Cells that have been labeled for CNS application include neural stem cells, oligodendrocyte precursors and macrophages.[36-38] Tracking neuronal stem cells empowers us to study the best way to administer the cells (intravenous *versus* intraparenchymal transplantation), estimate the dose (how many cells to use), assess the survival rate, evaluate their migration patterns, check if they find their target site and home, and optimize the therapeutic time window.[32,39] To illustrate this, Hoehn *et al*. transplanted SPIO-labeled stem cells into the contralateral hemisphere of a mouse displaying ischaemic stroke.[37] $T_2$*-weighted MR images showed the migration of these cells across the brain from the implantation site to the edge of an ischaemic lesion. (**Figure 7.3D-F**) This example demonstrates the possibility to follow cells in real time, which may be used to study the therapeutic potential of stem cells in the future.

**Reporter genes**

The previous two sections focused on reporting specific molecules or cells; this section addresses imaging gene function, which in contrast to cell tracking, exclusively reports on viable cells. Reporter gene imaging is a technique in which gene products (i.e. reporter proteins) are imaged *in vivo*.[40,41] Essentially, a reporter gene is transcribed to mRNA, which in turn is translated into a reporter protein (which are far more abundant in the cell than DNA or RNA). A good reporter protein must be easy to assay and must not normally be expressed in the cells of interest or, when encoding for endogenous proteins, must be expressed at much higher levels than normal. The gene of interest is unlikely to encode an MR-visible protein, although the protein of interest may interact with exogenous reporter molecules.[42] Often, the gene of interest is teamed up with a reporter gene. These genes can be engineered such that they are both driven by the same promotor. Upon activation of this promotor (which can be conditional or tissue-specific, for example), the expression of both genes is simultaneously enhanced; imaging the reporter protein thus 'reports' on the expression of the gene of interest. The reporter protein may produce endogenous contrast or may be imaged via exogenous reporter molecules.[42]

Reporter gene imaging is currently in a preclinical stage, but its potential is enormous. One could monitor gene expression, track cells in normal and abnormal development, map dynamic protein interactions and check cell transplantation therapy. Taking this one step further, one could follow the effects of gene therapy, in which cells are genetically modified to produce a therapeutic effect.[40,42] Reporter gene imaging is commonly used in the nuclear and optical imaging field, with green fluorescent protein[43] and luciferase[44] as prominent examples, but MR imaging starts claiming a place on stage as well, by virtue of its non-invasive nature and whole-body coverage.

Needless to say, for MR reporter gene imaging, the reporter protein needs to be MR detectable and common strategies are outlined in recent reviews.[40,42,45] Reporter genes may encode for artificial proteins, which are detectable by CEST imaging (**Figure 7.3G-I**)[34], but most MR reporter systems are based on the accumulation of iron, circumventing the administration of exogenous contrast agents. When reporter genes overexpress proteins such as ferritin[46,47], the transferrin receptor[48] or MagA[49] at high levels, they cause a local build-up of iron, which leads to enhanced negative contrast in $T_2$- or $T_2$*-weighted images. This principle has been demonstrated by Genove *et al*.[47], who visualized gene expression using genetically modified replication-defective adenovirae carrying the genes for the light-chain or heavy-chain subunits of ferritin. They injected the adenovirae into the striatum of a living mouse, which lead to the local overexpression of ferritin and the accumulation of endogenous iron. This resulted in enhanced negative contrast in $T_2$-weighted MR images, which was visible for weeks.

The field of MR reporter gene imaging is developing at a steady pace, but before it becomes a mainstream clinical tool it has to overcome several issues. The efficiency of gene transfer is currently very modest, which in combination with low MR sensitivity, makes MR reporter imaging a challenge. Reporter gene imaging (in combination with gene therapy) often uses viral based vectors to package genes, and a delivery device to pass them to the target organ. In this respect the method is not strictly non-invasive. Current applications of reporter gene imaging are therefore mostly confined to small animal models, although clinical applications are anticipated in the future, most likely as a tool to visualize gene therapy.

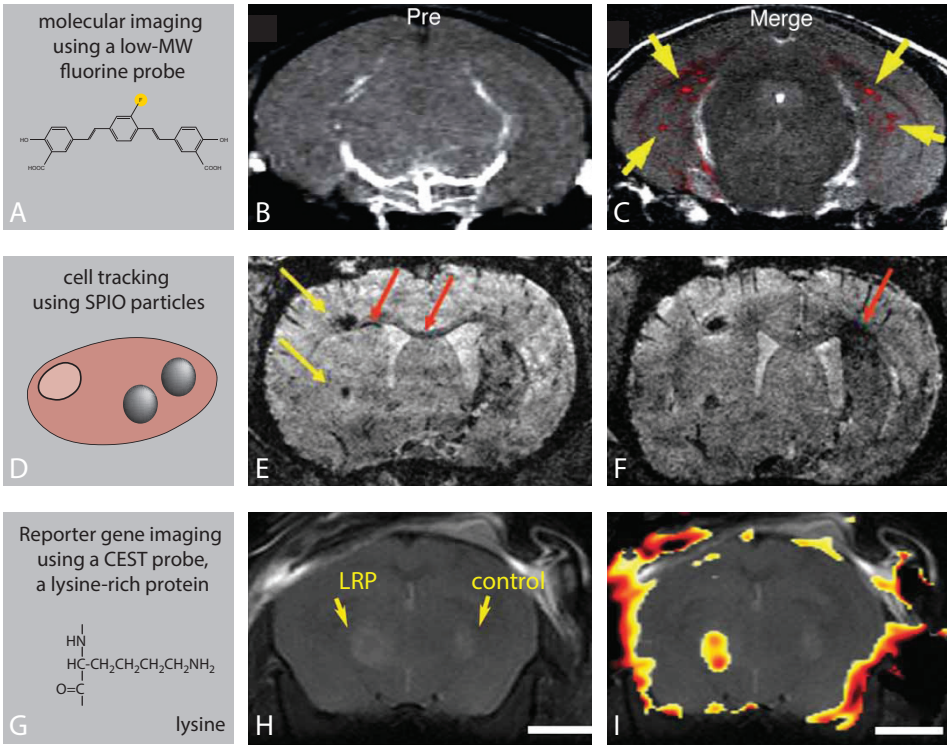


**Figure 7.3** ***In vivo*** **examples of molecular neuroimaging using MRI**
(**A-C**) MR images of transgenic mice displaying symptoms of AD. (**A**) A fluorine containing reporter probe with affinity for senile plaques was intravenously injected and imaged by $^{19}F$ and $^{1}H$ MRI. (**B**) Proton MR image before injection of the reporter probe; (**C**) merged proton and fluorine image after injection of the reporter probe, indicating the anatomical locations of the reporter molecule in the brain.[27] (**D-F**) Cellular imaging of migrating stem cells in a mouse model of ischaemic stroke. (**E**) Stem cells were labeled with ultra-small SPIO particles and transplanted into the contralateral hemisphere. (**F**) They migrate from the implantation sites along the corpus callosum towards the ischaemic border zone.[37] (**G-I**) *In vivo* reporter gene imaging using CEST reporter proteins. The left hemisphere of a mouse brain was injected with glioma cells expressing LRP (lysine rich protein, a CEST agent); the right hemisphere was injected with control tumor cells. (H) Anatomical MR image of the brain; (I) CEST signal intensity-difference map superimposed on the anatomical image, indicating the LRP-expressing tumorxenograft.[34]

## Current developments

The field of molecular imaging is developing in several directions. Imaging modalities perform better, molecular biologists construct ingenious reporter genes and transgenic mice, while chemists improve contrast agents and create multimodal reporter molecules.[20,50]
There is a continuous effort to increase the field strengths of human scanners, and these high-field magnets are making their way into the clinic. The most commonly used clinical scanners operate at field strengths of 1.5 Tesla, while 3 Tesla is regarded as high-field, but ultra-high field magnets at 7 Tesla are also used for patient studies. As the MR signal is proportional to the magnetic field strength, using high-field scanners improves the sensitivity and enhances the signal to noise ratio, improving anatomical and functional imaging.[51] A higher magnetic field

also increases the spectral resolution, which is an additional benefit for $^{1}$H-MR spectroscopy and CEST-based imaging. Unfortunately high-field magnets do not come without expense: increased magnetic susceptibility effects, field inhomogeneity and energy deposition pose worries and technical challenges, but these are partly overcome by using optimized coils, fast and parallel imaging and refocusing flip angles.[52,53]

With regard to molecular imaging, high-field scanners are able to detect lower concentrations of reporter molecules, which decreases toxicity issues. This is particularly the case for iron oxide-based particles, heteronuclear contrast agents and CEST agents. On the other hand, the current lanthanide-based contrast agents do not perform as well at high fields, and their relaxation behavior should be significantly improved.[20] Regardless of the field strength there is a continuous effort to tailor and improve pulse sequences, such as ultrafast imaging sequences and creating positive contrast for SPIO particles.[19]

## Outlook

The road to MR-based MI has been a long one. MR-based contrast agents are inherently far less sensitive than radiotracers, which makes them more likely to fail due to sensitivity or toxicity problems. Nevertheless, the advantage of providing anatomical, physiological and molecular or cellular information during a single examination is so great that research in this area is booming.

In the near future we anticipate that fluorine-based reporter molecules in conjunction with MR imaging at high(er) fields holds great promise, combining "hot spot imaging" and sensitivity. In contrast to fluorine-based radiotracers, MR reporter probes are relatively long-lived and could be imaged at a regular basis while avoiding excessive ionizing radiation. While BBB research is in fifth gear, MI imaging of the brain is already in use for pathologies with a (possible) compromised BBB, such as cerebrovascular diseases, inflammatory conditions and gliomas. Without the need to cross the BBB, this approach could take full advantage of current developments in the field.

Also, alternative therapies for neurological disorders, such as gene therapy and stem cell transplantation, are active areas of research and we expect that MI will play an essential role in the development of these therapies, for example by tracing labeled stem cells or by developing a common platform for treatment and diagnostics. Examples for this common approach are already known from cancer research, where liposomal particles have been used that target tumor sites, contain anti-cancer drugs and MRI-visible contrast agents to monitor the treatment response.

Many radiologists may regard MI as science fiction, but they should be aware that this field develops at a fast pace. Although currently most research is applied in animal models, the first radiological studies in humans have already been performed. It is generally believed that MI based on radiological techniques will be introduced in the clinical arena within the next few years. However, the clinical implementation of MR-based MI will only occur when academic radiologists are actively involved in research programs in which they master data collection and interpretation of MI images. This could be achieved by providing MI courses to residents,

organizing postgraduate courses for radiologists and by creating MI fellowships. Radiologists could also assist (bio)chemists in the development of reporter probes to advance clinical applications. Seeing that MI is on the brink of leaping to the clinic, radiologists should prepare themselves now to join that jump in the near future.

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# **PART TWO** | Development of Molecular Imaging strategies

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