times brighter than those from current synchrotron sources, and will be capable of producing high-resolution image data rapidly before the sample is destroyed. According to Kornberg, the objective is to match the resolution of current x-ray diffraction analyses without the need to prepare crystals.

Another significant development lies in image analysis algorithms. Work on the proteome and cell structure involves the identification of common basic features within entities that show considerable individual variation. In constructing a three-dimensional map of a particular human cell, for example, an image analysis algorithm must cope with high levels of individual sample variation. Two cells that are fundamentally the same may vary not just in shape, but also in the location and number of organelles. Improved algorithms are needed to 'separate the wheat from the chaff' and identify the underlying structural similarities while ignoring individual variations. Such processing is highly computationally intensive, and recent approaches exploit parallel computing techniques to break up such problems into smaller components that can be executed simultaneously (Fernandez *et al.*, 2004).

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The study of larger structures also relies on the integration and combination of several imaging or microscopy techniques. Optical microscopy may be used to resolve larger details of the whole cell and is unmatched in revealing real-time processes *in vivo*. Electron microscope tomography could be used to probe local structures and derive the broad geometrical shape of proteins down to around 50-nm resolution. X-ray crystallography or NMR might be used to derive the detailed structure of individual protein complexes.

"I think the move to integration is very important for structural biology," commented Matthias Wilmanns, head of the EMBL outstation in Hamburg (Germany).

Wilmanns and his team have used a variety of techniques to determine the structure of the giant muscle protein titin, which, at over 3 million Daltons, is the largest polypeptide yet discovered. "The titin molecule is too large to get high-resolution images, so we have had to use integrative approaches," explained Wilmanns. Indeed, there is growing cooperation between the various disciplines of microscopy and imaging, including computational biologists and crystallization experts, to tackle broader problems, such as unravelling functional relationships within and between cells.

Cooperation at the technical level is also important to maintain innovation and apply emerging technologies. Europe has been particularly strong in creating networks that link researchers in optical microscopy with manufacturers, as evidenced by Carl Zeiss's acquisition of EMBL's SPIM technology. It has also spawned several related initiatives, notably the European Light Microscopy Initiative and the EU-funded European Advanced Light Microscopy Network.

In this way, microscopy follows the evolution of genomics, proteomics and systems biology, in which closer cooperation, integration and interdisciplinary research have created an enormous momentum that now

produces huge amounts of data on genes, proteins and their interactions in the cell. But it will be the application and combination of improved microscopy and imaging technologies that enable scientists to link this information into a larger and more detailed picture of the cell.

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When volunteers are not healthy

Dealing with incidental findings in basic research challenges researchers and study participants alike

In November 2000, sports photographer Mickey Pflieger was standing on the sidelines of an NFL football game when one of the players knocked him down during an errant play. Pflieger was unconscious for several minutes and was taken to hospital, where a brain scan revealed that, although he had suffered no ill effects from the accident, he did have a large brain tumour. The chance discovery of his tumour, an 'incidental finding' in medical terms, allowed Pflieger to undergo the appropriate treatment earlier and may have improved his chances of long-term survival.

Although the circumstances are rarely as dramatic as being knocked out by a 250-pound football player, incidental findings are fairly common during medical and scientific studies on human volunteers, and they pose a dilemma for both researchers and their subjects. When Pflieger received his diagnosis he was already a patient, and doctors guided him to the appropriate therapy. In the research setting, however, scientists, who are often not experienced clinicians, face a complex mixture of risks and responsibilities when deciding when and how to hand

over a subject with a potential medical problem to the healthcare system.

Although virtually any study involving a human subject could reveal an incidental finding, researchers in neuroimaging and genetics in particular have turned their attention to this issue in recent years. Because of technological advances that make it possible to examine the human brain and the human genome in far more detail than ever before, these research areas are rapidly expanding. With that expansion comes the likelihood that more incidental findings can and will be detected.

So far, assessments of the frequency and types of incidental findings in neuroimaging have focused on magnetic resonance imaging (MRI) studies (Illes *et al*, 2004b; Kim *et al*, 2002; Katzman *et al*, 1999). The rate of abnormalities was in the range of 2–8% and comprised mostly benign findings, such as chronic sinusitis, that did not require any clinical follow-up. More serious conditions included blood vessel malformations (Fig 1), cysts and tumours.

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Genetics researchers have to grapple not only with true incidental findings, such as the discovery of a discrepancy in paternity during a family study, said Flavia Facio, a genetic counsellor at the US National Human Genome Research Institute (NHGRI; Bethesda, MD, USA), but also with sequence variations, called variants of unknown clinical significance, whose effects are simply not known. Today, most people who participate in genetic studies have symptoms or a family history of the disease under consideration, so unlike participants in neuroimaging studies, they already suspect that they have a medical problem. Facio predicts, however, that as genetics research advances, researchers will begin to

Fig 1 | An MRI image taken during a research study shows an arteriovenous malformation (circled in red) in the brain of a 25-year-old volunteer. Image courtesy of Matthew Kirschen, Stanford University, CA, USA.

recruit more reportedly healthy volunteers. Thus, geneticists face some of the same ethical questions as neuroimagers in the arena of incidental findings.

Incidental findings can be a particular problem in brain research because the scientists running the protocols are usually not medical doctors and are not qualified to draw clinical conclusions from their data, said Judy Illes of Stanford University's Center for Biomedical Ethics (CA, USA), who studies the ethics of incidental findings in neuroimaging. In addition, the data are frequently not of clinically interpretable quality. Thus, a suspicious finding is just that—a suspicion—unless the researchers consult with medical experts. Some neuroimaging groups therefore include neurologists and radiologists on their research teams, just in case they unexpectedly need a medical opinion on one of their scans. When Stanford student Matthew Kirschen discovered a golf-ball-sized structure in the brain of one of his volunteers, he called in his principal investigator, the director of the imaging center, the manager of the MRI magnet and a Stanford neurologist. The group was able to assess the problem quickly, and within hours had helped the subject obtain appropriate medical care. All of the people

Kirschen consulted were officially part of the research team. “We do that on purpose,” said Kirschen, “so that if something like this happens, we have the ability to share the information with [the team] or get advice from them without having to go outside of the confidentiality or privacy domains.” Lining up a team of medical experts may be relatively straightforward when the research imaging facility is affiliated with a large hospital, but the task can be more daunting for researchers who work in a purely academic setting.

Before genetics researchers discuss a finding of potential clinical significance with a subject, they will generally verify it with an independent laboratory carrying Clinical Laboratory Improvement Amendment (CLIA) certification. If the CLIA laboratory confirms the researchers' results, they can feel confident that they are giving their subjects accurate information,

according to Vivian Ota Wang, a Programme Director at the NHGRI. However, CLIA laboratories do not always provide a clinical interpretation of the result, Facio stressed. That is still left to the judgement of the researchers, who, given the increasingly widespread use of genetic techniques by scientists from diverse backgrounds, may not have formal training in genetics.

Researchers also face the difficult problem of communicating a suspected problem to a study participant. The responsibility of informing the subject may, by default, fall to the principal investigator as leader of the group; however, he or she may not always be the most appropriate person. In a study on the expectations of participants in neuroimaging, Kirschen, Illes and colleagues found that a majority of subjects would prefer to learn about an incidental finding from a physician on the

Genetics researchers have to grapple not only with true incidental findings ... but also with sequence variations ... whose effects are simply not known

research team. "Part or most of the motivation for that is that they want somebody who can answer their questions," commented Kirschen. Consequently, consent forms for neuroimaging studies at Stanford University ask for contact information for participants' primary care physicians, so that they can be consulted if necessary (Illes *et al.*, 2004b). The speed with which suspicious results are reviewed and shared with subjects is also important, especially in a field such as neuroimaging in which some conditions require urgent medical attention (Illes *et al.*, 2004b). Another important aspect of communication is assessing the subject's understanding of and reaction to the finding. Even though she discusses the possibility of variants of unknown clinical significance during pre-test counselling, Facio finds that "it's not unusual for people who receive these results to be disappointed or even surprised."

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The question of how carefully to screen subjects for abnormalities is still open in the neuroimaging community. At one end of the spectrum, the US National Institutes of Health (NIH; Bethesda, MD, USA) gives a full clinical brain scan to everyone who volunteers for its neuroimaging studies (Check, 2005). This practice offers participants maximal reassurance that any problems will be discovered, and minimizes the chances that researchers will miss an abnormality. However, it is also extremely expensive in both time and money and some researchers feel very strongly that the increased costs are not worth it. "In the present financial climate [giving every participant a clinical scan] means that half as much research gets done," said Alan Evans, a neuroimaging researcher from McGill University (Montreal, QC, Canada). "And that has, in my mind, a much greater impact on society and the health of society than the very, very low incidence of incidental findings which are actually correct and an even lower incidence where there is something you could have done." In

addition, doubling the scanning time may exceed the patience of some volunteers, particularly in paediatric studies, Evans added. Other researchers have adopted a less comprehensive, but still expensive, policy in which a radiologist reads every research scan, according to a recent survey (Illes *et al.*, 2004a). Evans' group, and the majority of those surveyed, take a moderate approach, which Evans described as a 'Good Samaritan' policy. They inform participants upfront that their scans are not of clinical quality and are not being interpreted by medically qualified personnel. However, if they do notice something suspicious, they will consult with a radiologist for confirmation and inform the subject of the results. At the other end of the spectrum, about 18% of neuroimaging researchers who were not required by their institutions to discuss volunteers' brain scans with a radiologist said they never did.

A final issue that troubles researchers is legal liability—the risk that they will be sued if they misdiagnose or miss a serious medical condition in one of their subjects. Beyond the impact on the individual researchers, Evans worries that a lawsuit might prompt the affected institution to disband its clinical research entirely, and hopes that the NIH will formulate some guidelines to help researchers protect themselves from lawsuits. So far, the problem is hypothetical. Neither Wang nor Illes knows of any legal action arising from an incidental finding in genetics or neuroimaging. However, "We don't need a legal event to be proactive. Our goal is to offer some ethical guidelines that protect our subjects, the investigators, and the research," Illes said.

For the research subject, an incidental finding may at first appear to be a blessing; in reality however, it can impose a substantial psychological and financial burden. Clinically evaluating the finding often requires extensive follow-up tests. Moreover, said Illes, in the USA "where one has to purchase healthcare and isn't guaranteed health insurance by any means", a finding may create additional costs for the patient and even affect a subject's future insurability. In an anonymous letter to *Nature*, one person described how he was denied disability insurance because of a brain tumour that was discovered during a research scan (Anonymous, 2005). The anxiety is particularly onerous when the medical implications of the

finding are not clear. For example, a woman who learns that she has a sequence variation of unknown clinical significance in her *BRCA1* gene, which is implicated in hereditary breast and ovarian cancer, may end up undergoing more frequent clinical breast exams or other screening, even though her particular variation may not increase her risk of breast cancer at all. In cases where the woman is already deeply concerned about breast cancer, perhaps because of a family history of the disease, such a result can "insert even more ambiguity and more anxiety, and...feeling of loss of control," said Facio. In fact, some consider the potential negative impact of an incidental finding to be a risk that needs to be explicitly discussed during the informed consent process before participating in a research study (Grossman & Bernat, 2004). Facio has known people who declined to participate in a study because they do not wish to deal with the consequences of an ambiguous result.

To address some of these issues, Illes and others organized a workshop in January 2005 on the 'Detection and Disclosure of Incidental Findings in Neuroimaging Research'. About 50 neuroimaging and genetics researchers and representatives of the ethical and legal communities met for two days for debate and discussion in Bethesda, MD, USA. Despite some areas of disagreement, the group ultimately adopted several recommendations. Among other things, they concluded that researchers should anticipate the possibility of incidental findings and decide beforehand how to handle them; that someone capable of rendering a medical opinion on a scan should be available to the research team; that a qualified member of the research team should promptly inform the subject of any clinically significant findings; that the possibility of incidental findings should be discussed with subjects during the informed consent process; and that guidelines on incidental findings should outline the responsibilities of researchers while not impeding the progress of research. Even though the focus was on neuroimaging, geneticist Wang felt the discussion was broadly applicable because it allowed people from different backgrounds "to cross-fertilize a little bit better and really learn from the lessons of other professions". Kirschen called it "absolutely astounding" to watch the

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diverse assembly debate the issues and said that it did an “excellent job” of settling on broadly applicable consensus points.

Evans was more critical of the meeting. He is concerned that the debate will lead to unnecessary bureaucratic regulations that will have a “chilling effect” on clinical research. “It’s already a tough enough business to do clinical research...[If you] raise it to a further level of paperwork complexity and bureaucratic obstructionism, people will just say ‘forget it’.” The meeting’s organizers are acutely aware of the need to develop guidelines that will protect researchers as well as their subjects. Co-organizer Emmeline Edwards, Deputy Director for Extramural Research at the

US National Institute of Neurological Disorders and Stroke (Bethesda, MD, USA), cites the desire to strike the right balance as the main reason why some aspects of the issue are still under discussion. “We certainly do not want to hamper neuroimaging research,” she said, “but we also want to raise awareness of the responsibility of the researcher...to manage the finding.”

On the basis of the conclusions of the workshop, Illes and her colleagues at Stanford have developed a pathway for managing incidental findings as well as some suggested language for informed consent forms. The group is also continuing its efforts to inform and learn from the research community. Kirschen hopes that their work will resonate beyond the neuroimaging community, noting that the aspects of incidental findings that they are debating “will also be relevant in other aspects of research and medicine as we push technology forward”.

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