Giovanni Paganelli:

nuclear medicine's enthusiastic ambassador

➜ Marc Beishon

Radioisotopes can be used to locate tumours and to deliver targeted therapeutic radiation internally. Sounds ideal for cancer, yet nuclear medicine remains an underdeveloped field, held back by irrational fears of all things nuclear, false perceptions of its potential, and bureaucratic and cost barriers to accessing agents. Giovanni Paganelli is a believer, and he is spreading the word.

It is a given that a complex field such as cancer spawns an increasing number of sub-specialities – and that there will inevitably be debates about which are most deserving of resources. Those working in fields that do not command much support can nevertheless spend many years quietly building a solid scientific and clinical reputation against the odds to emerge eventually with very important work – which arguably now applies to nuclear medicine, and in particular its application in therapeutic oncology.

Giovanni Paganelli, director of nuclear medicine at the European Institute of Oncology in Milan, describes what the specialty means for cancer. "It is the use of radioisotopes to both locate cancer cells for diagnosis and to deliver energy to the target cells to destroy them. I liken my field to an aircraft – to make it take off we need both diagnostic and therapeutic wings."

The problem for nuclear oncology, he adds, is that too many professionals – including oncologists – see mainly the diagnostic side of the subject, where practitioners often come from radiology, and not from internal medicine (as Paganelli himself does). This perception is reinforced by the fact that nuclear medicine departments are often located in the darkest bowels of hospitals – where large and possibly dangerous equipment often lurks – divorcing specialists from the multidisciplinary discussions taking place on the floors above.

Together with a public suspicion of anything 'nuclear' in some countries, the technical and bureaucratic difficulties of sourcing and preparing radioisotopes, the cost of equipment and agents, and to date fairly narrow progress in truly therapeutic applications, nuclear oncology has faced tough challenges. And as Paganelli notes, the specialists themselves are partly to blame. "We tend to spend most of our time talking to other colleagues in nuclear medicine – and not getting our messages across to medical oncologists and surgeons at the right meetings," he says.

"If you had talked to me 10 years ago I would have been quite depressed about our prospects. But in the last few years there has been much more progress." The arrival of PET (positron emission tomography) and SPECT (single photon emission computed tomography) as widespread techniques has triggered new awareness in the medical profession of the use of radioisotopes, says Paganelli, although again this is largely because of diagnostic potential. "What excites

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me – and what I consider true nuclear medicine – is the growing use of targeted diagnosis and therapy, and it is one of the few areas in oncology where we are really doing translational medicine from bench to bedside."

It is a bold claim, but Paganelli has a 20-year track record in researching the targeting potential of radioisotopes in combination with other agents, and while he concedes that clinical applications are currently limited, no one who visits his department in Milan could fail to be infected with his enthusiasm – and his persuasive arguments about the tremendous potential for tackling some of the most difficult oncology problems. Work on high-grade brain tumours, lymphoma and neuroendocrine treatments are in train, while a sequence of clinical research projects to aid breast-conserving surgery is a landmark at the Institute.

Paganelli always intended to become a doctor – "It was my dream as a boy to do research and understand why people get ill" – although he was later to discover that science does indeed follow Edison's famous maxim: 1% inspiration, 99% perspiration. He chose to pursue internal medicine while at the University of Bologna, and further selected geriatrics as a speciality – the growing 'market' of an aging population being a draw. He recognises a logical and philosophical link between aging and cancer – "After we have reproduced, maybe the DNA doesn't care

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whether it's damaged or not" – but the move to nuclear medicine came about by chance when he was given an oncology research grant. He quickly embarked on his targeting research.

Leaving a permanent assistant's post in the nuclear medicine department at the Bufalini hospital in Cesena, near Bologna, Paganelli took up a fellowship to do basic research at London's Hammersmith Hospital. His Italian boss was none too pleased – but the opportunity to join a group working on new radioimmunotherapy targeting methods using monoclonal antibodies, under well-known medical oncologist Agamemnon Epenetos, was too good to pass up.

"The field started as a branch of pathophysiology," explains Paganelli, "and cardiology was the most important application 10–15 years ago. That's changed dramatically now. I estimate that about 80% of nuclear medicine is now focused on oncology, although heart and other organ functioning are still crucial areas." All told, there are now more than 100 procedures in regular use in nuclear medicine departments. However, as he adds, one of the first examples of nuclear medicine – and a truly targeted therapy and an oncology application to boot – was the use in 1946 of radioactive iodine to treat thyroid cancer, making use of the unique ability of the gland's cells to absorb iodine and so localise the killing effect of radiation. "A protein traps the iodine like a Trojan Horse," says Paganelli.

The first commercial radiopharmaceutical, based on iodine-131, went on sale in 1950; the scintillation (gamma) camera came out in 1958; cyclotrons to produce medical radioisotopes were introduced in the early 1960s (as was the forerunner of PET); and heart,

The nucleus. Paganelli with core members of his 40-strong department of nuclear medicine, including chemists, physicists, technicians, physicians, nurses and admin staff, grouped alongside one of their PET-CT scanners

lung and other organs became standard scanning targets for nuclear medicine, which was recognised as a speciality in the US, in 1971. But it was a demonstration by David Goldenberg, in 1973, of the targeting of tumour antigens by radiolabelled antibodies that set in train Paganelli's arrival at Hammersmith in the 1980s.

By then, melanoma patients had been treated with iodine-131-labelled monoclonal antibodies. But as Paganelli points out, while an excellent idea, this targeting had limitations – not least that the radiolabelled antibody went everywhere in the body. "The idea I had was to add a pretargeting stage. We first target the cancer cells with a non-radioactive antibody, which clears from the rest of the body. Then we deliver a second, radiolabelled molecule that is attracted by the antibody, and which is also cleared rapidly from the body." The aim is to deliver a more effective dose to the target, while minimising side-effects, and has been a plank of Paganelli's subsequent research and clinical practice; indeed it was the subject of a patent filed jointly with the Italian ministry of research in 1991.

The sheer amount of work – that perspiration — and multidisciplinary understanding is a feature of such research, says Paganelli. "Nuclear medicine is one of the few branches of medicine where you have to be on top of maths, physics and chemistry as well as the biology. If you want to know how to apply approaches such as pretargeting – the amount of agents, the timing of doses, the molecular operation and so on – you need to assemble a huge amount of information." His more than 40-strong department in Milan – comprising physicists, chemists and technicians as well as medical doctors – is testimony to the need for a multidisciplinary approach.

Paganelli first went to work in general nuclear medicine at the San Rafael hospital in Milan, with a licence to continue his research, before being asked by Umberto Veronesi to set up a fledgling department at the European Institute of Oncology in 1994. It has become the first and leading centre in Italy to carry out therapeutic targeting work, and his closest collaborators are now chemist Marco Chinol and physicist Marta Cremonesi, both based in his unit.

He had a window of opportunity at the start, as there were no regular patients for a few months, and he set about developing the pretargeting technique with high-grade brain tumours, which have a very poor outlook in the vast majority of cases. "I started with glioblastoma because it is an orphan disease with a suitable marker [the protein tenascin] and we could only have a few months to see if it worked or not – and if it did, it may then also work more easily in cancers such as lymphoma. We used the avidin biotin system and published some interesting results." His closest colleague on this work then was Antonio Siccardi, professor of biology and genetics at the University of Milan.

Paganelli describes avidin and biotin as a "fantastic natural system" (it's been described as nature's gift to molecular biology). Avidin – found in the egg whites of birds and in bird and reptile tissues – is a protein that binds to the much smaller biotin molecule (also known as vitamin H) with tremendous affinity. In his early work with brain tumour patients with grade III and IV astrocytoma, he employed a three-stage process – first injecting a monoclonal antibody tagged with biotin that binds to the tumour, then avidin, which binds to the antibody, and finally the radionuclide – in this case yttrium-90-labelled biotin. Although the number of patients was small, and the work took several years, a quarter (12 patients) showed a reduction in tumour size and three people had complete remission.

Now PAGRIT – pretargeted antibody guided radioimmunotherapy (and a trademark) – has entered the oncologist's lexicon, and is suitable for several other applications, notably in lymphoma, which is a cancer with good radiosensitivity and which also expresses tenascin. It also has potential in other cancers, according to Paganelli. A form of pretargeting is also now playing a role in his most important work to date, on breast cancer.

The complex nature of this work is obvious – and a further potential barrier is the availability of reagents.

The avidin biotin system has been described as nature's gift to molecular biology

"High costs should be defrayed by more competition and more centralised facilities to prepare reagents"

"When I started the work on glioblastoma there was a company that supplied the antibody for human injection – you need a lot of resources to prepare these agents, and now we also have to comply with European Good Manufacturing Practice regulation. But my supplier was sold to another company that was not interested in continuing production." It is only recently – after a gap of some 15 years – that he has found another company to step in (Sigma Tau, based in Rome), such that he is now in the registration process for the PAGRIT model for brain tumours with EMEA along with a phase I/II trial. He adds that there is – encouragingly – much more interest from commercial quarters in agents for nuclear medicine generally, from both smaller biotech firms and larger companies (one US commentator predicts a therapeutic market worth $1.9 billion by 2012, from just $71 million in 2005).

Two radioimmunotherapies of note for non-Hodgkin's lymphoma are now on the market – Bexxar (tositumomab and iodine-131 tositumomab) and Zevalin (90-ibritumomab tiuxetan). They hit the spotlight in the US recently when Medicare, the healthcare programme for older people, baulked at the very high reimbursement costs set by their makers – as much as $30,000 (€20,600) for one treatment.

These treatments are nothing new to Paganelli – he has been working with non-commercial versions of most nuclear agents for some time. Having prepared 90-ibritumomab tiuxetan himself at Milan, Paganelli comments that the costs could certainly be much lower, and argues that the high costs should be defrayed by more competition among biotech firms and more centralised facilities to prepare reagents for sharing among oncology centres.

"There is no central radiopharmacy in Europe for preparing monoclonal antibodies or peptides for therapeutic purposes," he says, adding that he is in discussion with colleagues to set up just such a facility at European level, and also at local level for the various clinics in and around Milan.

The short half-life of many medical radioisotopes does mean that supplies must be constantly on tap, and while medical cyclotrons can produce some isotopes, especially for glucose used in PET, others such as the widely used technetium-99 and iodine-131 are mainly produced in nuclear reactors (sometimes as longer lived 'parent' isotopes that are then used to generate 'daughters' locally). In Italy, nuclear reactors have been rejected by a referendum, so the country will always be dependent on outside supplies.

That nuclear medicine is dependent on a reliable commercial supply of radioisotopes was brought home very recently – a reactor in Canada that supplied a large proportion of the worldwide market was off-line longer than expected last December, leading some medical centres in North America to postpone procedures and to scramble around for alternative supplies.

Paganelli's most well-known work, in breast cancer, came in response to Veronesi's drive to cut unnecessary surgery. "In 1995 he asked me if I had anything that could avoid axillary dissection of lymph nodes, perhaps using PET. I said we could look at using blue dye to identify sentinel nodes, which was also being done with melanoma. Looking further, I realised we could optimise the approach, as blue dye can miss a lot of lymph nodes." The result was the first protocol for sentinel node lymphoscintigraphy in breast cancer, identifying the node using a radioactive marker injected into the tumour to see the extent of tumour spread. "I sent it to Veronesi, who was cautious, but we started to optimise the amount of radioactivity and size of particle we were injecting and we published in 1997.

"While I was doing this I realised that, after injecting the material into the tumour, sometimes it did not move from the cancer site and we were missing 30% of the nodes," he continues. Paganelli and colleagues found that injecting near the tumour instead found the sentinel node with much greater precision. "After now carrying out more than 12,000 sentinel node lymphoscintigraphies, we have missed the sentinel node in only 99 cases, which is a

Nuclear family. With his wife, Stefania, who also works at the Institute

sensitivity of more than 99%, and the technique is now routine for breast cancer." It is notable, though, that Europe differs in practice from the US. "The molecule we use is not authorised by the Food and Drug Administration, and is larger than the one used in America – and they see more lymph nodes as their molecule is not so easily trapped by the sentinel node. So our method is more precise."

This sparked off another innovation. Challenged in the coffee bar by a surgeon to solve with his 'high-tech methods' the growing problem of locating non-palpable lesions more precisely, Paganelli's immediate and not entirely serious response was, "Simple – inject a drop of radioactive material into the centre of the lesion and use a gamma probe to locate and remove it." But the idea became a study protocol within a few days and proved very effective – and has become known as ROLL (radioguided occult lesion localisation).

Now 'on a roll' with the work, Paganelli and his team have taken the elegant step of combining it with the pretargeting approach to add another option for eliminating residual cancer after a tumour has been removed in breast conserving surgery. Postoperative, partial breast irradiation using external beam technology is the standard treatment, but usually requires travelling to and from hospital for daily sessions over a period of six weeks. A technique now in phase II trial is to shorten this therapy with a radioisotope treatment, using the tried and tested avidin biotin system.

"The surgeon injects avidin into the tumour bed during the operation – no special skill is needed – then the day after, or when the patient has recovered, she receives an injection of radioactive biotin in the nuclear medicine department. It's very simple, very cheap and can be done anywhere, and I think we may be able to replace external beam radiotherapy altogether. There are many places where linear accelerators for radiotherapy are not available or not covered

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Along with Veronesi, he knows only too well the struggle to get new therapeutic approaches accepted

by insurance – as many as half of women in the US still have a mastectomy instead of breast conserving treatment. I think this is the best idea I've had in my career so far."

The next step will be to add an antibody specific to breast cancer with the avidin, but this is not ready for trial yet. The technique without the antibody is known as IART – intraoperative avidination for radionuclide therapy – and Paganelli says, after successful completion of the present trial, it should go to a multicentre study this summer. There are several other techniques to accelerate breast irradiation currently in trial, but none that are as simple to apply as IART (see *Clin Cancer Res* 13:5646s–5651s), and he would like to extend the idea to other cancers where conventional radiotherapy can have major side-effects, such as in the head and neck.

It is another chapter in the now very lengthy story of breast conserving treatment, and Paganelli, along with Veronesi of course, knows only too well the struggle to get new therapeutic approaches accepted – and bureaucracy in Italy is a particularly tough nut to crack. He also anticipates running up against vested interests. "I'm sure the makers of linacs will not be so happy – but in fact if you cut the number of applications for them you can treat more patients."

One other notable area of therapeutic work at Milan is the treatment of neuroendocrine tumours with peptide receptor radionuclide therapy – Paganelli and colleagues have built on work pioneered in Rotterdam and Basel on this technique. "These are rare tumours, but not as rare as you think – and we can show a benefit in more than 70% of patients, with 20% in complete remission. It's attracted commercial interest and we are looking at peptides for other cancers." He says the peptide treatment has expanded around Italy, with about 50 patients seen each week. In Milan, quite a few come from abroad for this and other treatments such as for brain tumours.

All this is well known in the relatively small circle of nuclear oncologists, a profession that seems to have a number of entry points and patchy representation globally. Paganelli says there are more in Europe with a background in internal medicine, compared with the US, where he says nuclear medicine tends to be more a branch of radiology. The Netherlands and Germany are among the stronger countries, he adds, while pockets of excellence exist in several places – a case in point is in Nantes, France, where a high-intensity cyclotron is being built, and which is hosting a conference at the end of March: Nuclear Medicine Tomorrow (see www.arronax-nantes.fr).

The European Association of Nuclear Medicine – appropriately sited in Vienna, home to the International Atomic Energy Agency – is an active organisation, he notes. It established a school for continuing education in 1997 and is forging closer links with the European Society of Radiology as the crossover between imaging techniques becomes more pronounced (although this has courted opposition from some quarters, not least because nuclear medicine appears to be stronger in those countries where it has been allowed to flourish as a physician-led discipline). Paganelli is just keen to spread messages about nuclear oncology. "I'm more likely to attend meetings with surgeons and medical oncologists now than the nuclear medicine events," he says. He's also taken up teaching posts at the universities of Milan and Bologna, and is pleased to report that in Italy there are now at least 10 centres routinely doing work introduced at Milan.

Paganelli spends much time trying to convince companies to invest in the work. He also registered more patents recently – not for personal gain, "but because they are necessary to convince firms of commercial value."

Elsewhere there have been gloomy reports on prospects – in the US, federal cuts in 2006 led to "many important scientific projects related to nuclear medicine being abandoned", according to the Society of Nuclear Medicine. In the UK in 2003 nuclear medicine was said to be "close to collapse" – the country had just four PET scanners at the time, according to the *British Medical Journal*, and there

Enthusiast. Fishing is Paganelli's great passion

were concerns about a depleted workforce. Certainly a PET/CT set-up is costly – about €1.5 million – but as Paganelli notes, it should be justified as a front-line diagnostic tool and not shunted to the end of a queue of other techniques used for investigation.

Part of the debate about funding also revolves around the use of very costly external beam technologies. As Paganelli points out, there are different radioisotopes he can use internally that deliver not only beta particles (i.e. electrons/positrons) but also alpha particles comprising protons and neutrons (i.e. a type of hadrontherapy, and he's in a good place to monitor progress in the external use of ion beams, as the TERA Foundation, the Italian hadrontherapy project, is based in Milan). By and large, internal radionuclide approaches tend also to be safe and well-tolerated, he says, especially with pretargeting, while it is a myth that all such treatments need bunker-like facilities to be administered.

While the radionuclides show promise in a fairly limited number of treatments so far – bulky, solid tumours have been less amenable to the targeted approach – Paganelli is in no doubt that, given the right backing, they should remain at the forefront of translational research, including a growing use in studying the action of targeted drugs.

Apart from medicine and his family – four daughters and his wife Stefania, who works in administration at the institute – Paganelli's biggest passion is that ultimate in targeting, fly fishing (medicine is a hobby compared with this, apparently).

Nuclear medicine does, perhaps, need more high-profile leaders, but Paganelli is content to keep developing his base in Milan and doing some teaching, albeit with ongoing skirmishes with the Italian authorities. Heading a society, or following in the footsteps of that most famous Italian nuclear scientist, Enrico Fermi, to the US as so many other Italians have done, is not for him. "I'm happy to serve my country – but all the time trying to kill bureaucracy with evidence," he says, adding that he will be content if in 10 years' time targeted radionuclide therapy is mainstream in cancer centres. "Whenever you propose something new, people say it is not true. Then they say it doesn't work. And when you show it works, they say it's not new. Nobody will give you anything – you have to fight with great enthusiasm and work equally with your heart and brain."

And that is just what he is doing, forging new paths in search of innovative ways to put nuclear medicine to the service of cancer patients. Fourteen years after being diagnosed with a terminal brain tumour, his first patient at the Institute, now alive and well, and the thousands of women who have safely retained their healthy lymph nodes, are among the many who have reason to be glad of this spirit of innovation and enthusiasm.

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