How Cancer Genomics is Transforming Cancer Care

Professor Sanjay Popat FRCP PhD

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Understanding our genes is the fundamental basis to understanding cancer and how to treat it. Most of the cells in the human body have a central structure called a nucleus. Within this nucleus, are our chromosomes; we have 46 of these to be precise, that's 22 pairs, numbered 1-22, one of which we inherit from our mother and one of which we inherit from our father. In addition, we have another set called X and Y that determine if we are male or female. The chromosomes are tightly coiled into strands of DNA: the basic building blocks of life. And it's our DNA that is the blueprint for everything that the cell does: it is a sequence of letters (nucleic acids), and that precise sequence of letters determines how that individual cell functions.

So how does this miracle occur? Well, there's a lot of complex biology, but from the sequence of nucleotides of DNA, a modified copy of this sequence called mRNA is created. Here, every 3 letters or bases is important as each three letters encodes an amino acid, a part of a protein called - the machinery of a cell. The RNA is subsequently read, and a string of amino acids is made which then ends up being a protein. So, you can see how the DNA sequence turns into a protein which then does the functions of a cell.

In recent years we have witnessed three major events that have allowed us to harness this biology to transform the way we treat cancer.

The first of these was publication of the first draft of the human genome in 2001. It was amazing that prior to then, our actual understanding of the DNA sequence of all 23 pairs of chromosomes in human beings was relatively limited. The reason that this publication was so fundamentally important was that it allowed us to understand what normal genetic sequence is, and alongside it, all of its normal variations. And by understanding what's normal, only then can we begin to understand what is abnormal and especially when this comes to cancer. At the same time, TIME magazine published an influential article on how it predicted that we would use this understanding of genetic sequence to help predict which drugs we took for what condition: a prophecy which is now in routine clinical care for cancer.

Understanding the human genome allowed us to better understand the cancer genome. We knew that if a sperm cell or an egg had a mutation- a change in the nucleotide sequence or a different spelling letter in the DNA- then this mutation would be present in every single cell of all of us. However, what we have identified is that if the same mutation does not occur within the egg or sperm but occurs later on in development, within a tissue such as the skin or the lung or the colon then that genetic abnormality is only present within that particular group of cells these are called somatic mutations. And this is exactly what we have identified occurs in cancer. Specifically, mutations otherwise known as spelling mistakes within the DNA sequence, occurring within particular cells of a particular organ, not present in any other part of the body.

Now when we look at the sequence of DNA, we actually find multiple different abnormalities that occur in nature. The commonest type that we see are called mutations. This is, as I explained before, when the spelling mistake of the DNA sequence changes, and this results in an abnormal protein. There are different

classifications of mutations classified as transitions or transitions, missense mutations, or insertions or deletions.

Indeed, when looking more detail not only do we find abnormalities within the DNA sequence, but sometimes we see major structural abnormalities within the chromosomes themselves, for example where arms of chromosomes are interchanged, a process called a gene rearrangement or more commonly a gene fusion, because genes are broken up and fused next to each other when they normally wouldn't be.

We have also learnt that the DNA sequence does not all turn into a protein. Within the DNA strand the amount of DNA sequence that encodes a protein is called in exon and it is interspaced with large regions of genomic junk, by large, called introns. Not all exons turn into proteins, and multiple different proteins can be made from the same genetic sequence through a process called alternative splicing. And what we have learned in cancer is that sometimes DNA mutations cause of alternative splicing resulting in an abnormal protein.

Lung cancer is a superb example for how abnormal genetics causes cancer and how that can be identified and drugged. Through multiple large scientific studies of looking at the DNA sequence of lung cancers, we have identified that in a particular type of lung cancer called adenocarcinoma we have found many different types of genetic abnormality, usually mutations, and sometimes gene fusions, all of which function through the same way. They all change the DNA sequence to result in an abnormal protein which normally is not functioning within the cell, but then is abnormally forced to switch on and function without any regulation, out of control of the normal cellular processes, effectively switching "on" that cell and preventing it from dying at the end of its natural life. That cell then divides and forms 2 daughter cells each of which harbours the same genetic mutation as the parents, and those 2 daughters cells become for 4, and those become 8 and so on and so forth. Hence, we can see that the fundamental basis of many different types of adenocarcinomas is in fact a genetic alteration such as a mutation within the DNA of 1 particular cell. These are otherwise known as a driver genetic alteration which drives the cell to growing and dividing and replicating, with the same genetic abnormality being found in all of the daughter cells identified within the cancer.

Sometimes we find other abnormalities within the chromosomes when parts of the chromosomes with multiple copies of the same region, a process called amplification. This also causes cancer in some cancer types.

The second major event that occurred, was mapping of the human receptor protein tyrosine kinome. Protein tyrosine kinases were at that time a newly identified major type of protein which is usually found on the cell surface. These function as receptors and sense outside factors and transduce these signals to the inside of the cell to instruct the nucleus what to do. Our understanding of the genetic sequence of what encodes these proteins was also critical as we discovered exactly what these kinases do and how critical they are in regulating the cell and causing it to grow and divide when stimulated.

The third major event that occurred was that our medicinal chemists identified a new class of drug. These were called kinase inhibitors and whilst the first kinase inhibitor was developed for hypertension, these turned out to be particularly important in cancer where these were called tyrosine kinase inhibitors. This is a entirely new class of drug: a pyrido[2,3-d] pyrimidine derivative that was able to highly effectively inhibit a kinase protein.

So, putting this all together, we discovered that genetic alterations occurring within the DNA of the cell resulted in abnormal receptor protein kinases sitting usually at the surface of the cell which regulated the growth of the cell, and these genetic alterations resulted in kinases being activated all the time when they would normally be under very tight control. These activated kinases resulted in a signal for growth and cell division to be propagated to the cell to force it to grow when it otherwise would not have. This whole process could be effectively terminated with simple straightforward tablets called tyrosine kinase inhibitors which effectively switch off the activity of the abnormally activated kinase.

The final event that has transformed our understanding of genomics in the cancer field is the development of genetic technologies and their costs. Here, mapped against Moore's law where the cost of computing has decreased over the years, the costs of genomics have markedly reduced over time, allowing complex technologies to be routinely implemented into clinical care.

Indeed, when trying to understand the genetic sequence, the gene in the cancer of a human being can be analysed through many ways. We could for example do a very basic test when we perform something called PCR to look for mutations or deletions within a gene, or we could consider more complex technology

call whole exome sequencing when we are trying to sequence all of the coding regions of the DNA: sequencing hundreds or thousands of genes, or we could go even further to look at the whole genome itself. Each of these different processes have different benefits and different limitations and can be used in different scenarios. We may find that we actually want to know more about the RNA than to DNA as this may help us to better identify gene rearrangements and this can be pursued through RNA sequencing through a variety of technologies.

I want now to spend some time talking about non-small cell lung cancer and specifically adenocarcinoma as a paradigm for precision oncology. We all know that Sir Richard Doll presented his seminal paper back in 1954 where he took a cohort of doctors born between 1900-30 and followed them up and demonstrated that the cigarette smokers were dying on average 10 years earlier than never smokers and postulated a direct relationship between tobacco and death from cancer, specifically lung cancer.

And indeed, when we think about lung cancer, we typically think of the character on the left of the screen who always has a cigarette in his mouth. However up to 20% of all lung cancers occur in people that have never smoked. Indeed, famous faces have developed this condition including the late Roy Castle, and the late James Brokenshire. Indeed, many advocates would say that anyone can get lung cancer, not just smokers: If you have lungs, you can get lung cancer it just does not take tobacco to cause this diagnosis.

Now when we look at lung cancer in never smokers it remains a major cause of death, given that 20% of a big problem itself is still a big problem, accounting for more deaths than cervix cancer, pancreas cancer and indeed prostate cancer.

Now one of the fundamental problems we have with lung cancer is that it is a malignancy of an internal organ. And hence picking it up at an early stage is problematic because we do not know that there is a cancer growing inside as the lungs can take on quite a bit of growth before any abnormalities become clinically evident. As a consequence, lung cancer has usually spread to other parts of the body at the time of diagnosis, and indeed this means that most patients have stage IV cancer at the time of diagnosis, which in itself is incurable.

Now one of the great examples of the implementation of genomics into routine care has been the gefitinib story.

Here, in 2002 AstraZeneca were testing a new kinase inhibitor and remember this is a targeted therapy tablet, called AZD1839. This drug switched off the kinase of the EGFR protein, known to be important in lung cancer signalling. And in a seminal publication of a phase 1 trial published by Professor Ranson from Manchester he demonstrated quite remarkable benefit in a handful of patients with widespread metastatic lung cancer with just this simple tablet. However, many patients derived no benefit whatsoever, and it remained quite unclear why some patients benefited with remarkable rapid tumour shrinkage and others did not. This early benefit of AZD1839 which was subsequently called gefitinib, was so marked in some patients that it resulted in approval by the FDA. Indeed, in the US and in parts of Europe this tablet was in widespread use, but it remained entirely unclear exactly who would benefit. It certainly seemed to provide a benefit particularly in groups of people who were never smokers or indeed had Southeast Asian ethnicity.

Subsequently AstraZeneca performed a clinical trial where they randomly allocated several hundred patients to receive either gefitinib or not. Unforutnatly and shockingly to many, the trial demonstrated that on average it did not prolong survival. And on this basis, the FDA withdrew gefitinib from the market. It continued to be used in other parts of the world where clinicians still reported a small proportion mainly of never smoker's and patients with from East Asia were most likely to benefit.

This all changed in 2004 where 2 groups from the US reported seminal findings that told us the biological basis of benefit from gefitinib. Here, they reported that when they took the lung tissue of patients with lung adenocarcinoma and sequenced the EGFR gene in patients that were being treated with gefitinib. Lo and behold they identified that patients that harboured a genetic mutation in the EGFR gene that switched on the EGFR kinase protein, all these patients responded to gefitinib, whereas those that by and large did not harbour a mutation in the EGFR did not respond, opening up the first concept of precision oncology: giving an EGFR kinase inhibitor only to patients with activating mutations in the EGFR gene as these patients benefitted and others without the mutation, did not. On the basis of this and other clinical trial data, the FDA and other regulatory authorities gave approval for gefitinib for EGFR mutant NSCLC: the first genotype-specific approval for a common cancer.

We know a lot about these mutations about these mutations now. We know that they are not heritable by enlarge, that they occur de novo within the lungs, as a founder driver mutation, the first mutation that

occurs in an airways cell to cause cancer, and that they are predominantly seen in never smokers, are sometimes seen in former and at a low level in current smokers. Data also demonstrates that there is an increased incidence of these mutations in patients in Asia. Therefore, we cannot tell just by simply looking at the patient if the tumour harbours an EGFR mutation, we actually have to take the tissue, sequence the DNA of the EGFR gene and see whether the mutation is present.

This is one of the first cases of EGFR mutation we identified in our clinic in routine clinical care, a young patient who was a never smoker who was hospitalised with a large tumour in her left chest causing significant breathlessness and oxygen dependency. Her tumour was biopsied, and shown to have an EGFR mutation, and we were able to access the EGFR kinase inhibitor gefitinib and she started it immediately. Within 3 days her cough had disappeared. Within 1 week she was able to walk up the stairs. When I later saw her after 2 weeks, she had come off oxygen completely. When we scanned her after 8 weeks her tumour had mostly melted away. This is a remarkable benefit, and the benefit that we would have never seen with chemotherapy.

Hence moving forward from 2004 where we were classifying lung cancer according to what it looked like under the microscope with a traditional view of tumours being mostly adenocarcinoma or squamous cell carcinoma, moving forward now we now give non-small cell lung cancer a molecular diagnosis, wanting to know what the genetic abnormality status is at multiple different genes in routine clinical care. Indeed, in order to optimally treat patients these days, we cannot do so and cannot select the right drug for them unless we know the genetic sequence of the tumour to allow us to better understand whether there is an opportunity to use a highly effective kinase inhibitor.

Indeed, as the science developed, we identified further genetic alterations in lung cancer, the next one being identified was a gene rearrangement in the ALK gene otherwise known as an ALK fusion. At that time Pfizer was developing a drug called PF-02341066, which later became known as crizotinib, a highly potent ALK kinase inhibitor. These scans were one of the first patients we identified to have an ALK fusion with widespread bilateral disseminated lung cancer, you can see on the slide here, who was treated with crizotinib, and again had a very similar dramatic response as a previous patient with marked resolution of symptoms and cancer within days of starting treatment.

Bring us forward to 2024, we can now take a group of patients with non-small cell lung cancer at time of diagnosis, take their tissue, and analyse it, with gene sequencing to identify which of a group of genetic alterations is present, which then determines whether we treat the patient upfront with immunotherapy with or without chemotherapy, or whether we find a activating genetic alteration which allows us to use a highly effective kinase inhibitor resulting in dramatic improvements.

But does this really matter? Does this really transform the outcomes of our patients? Back in 2014 our US colleagues reported on a study they had been performing for several years, the Lung Cancer Mutation Consortium LCMC study. Here, a group of centres in the US took their patients with newly diagnosed stage IV non-small cell lung cancer, analysed the tumours at a number of different genes, and if their tumour had a genetic alteration they were allocated to a tablet kinase inhibitor and they looked at the outcome in those that receive the tablet, those that had a genetic alteration but did not receive kinase inhibitor, and those that were allocated standard treatment at that time called chemotherapy. What they demonstrated was a clear benefit for identifying a driver mutational genetic alteration and giving that patient targeted therapy, increasing the median survival by 1 1/2 years.

Now in 2024, in routine clinical practice, patients with non-small cell lung cancer particularly adenocarcinomas should have their tumour sequenced at at least 8 different genes, to look for particular genetic alterations. If these are identified, the tumour should be matched to an appropriate kinase inhibitor, many of which are now approved for use, having proven their safety and efficacy often over chemotherapy in randomised clinical trials.

Indeed, when we look at what is available here in the NHS, we can clearly see that the vast majority of approved kinase inhibitor is against genetic altered targets in lung cancer are funded within the NHS.

However, we have a problem. Because these treatments are not curing our patients. Ultimately after a period of time be it 1 year, 2 years, 3 years and sometimes even longer, the cancer recurs. And what we have identified is that applying the selection pressure of drug therapy changes cancer, which evolves to become resistant to the drugs that it is being exposed to.

And since then, we have learnt quite a lot about resistance mechanisms to drugs, mostly through the great help and support our patients who have aided us through research efforts. Drugs may stop being effective

through pharmacokinetic effects. Patients may refuse to take the drugs usually because of difficult side-effects. There may be drug-drug interactions resulting in a reduced bioavailability of the drug, or the drug may simply not penetrate well enough to certain parts of the body, typically the brain, resulting in growth of the tumour in such sanctuary sites.

Studies of biopsies of patients with tumour growth despite initial benefit and subsequent growth, have demonstrated a number of new different genetic changes that have occurred within the tumour to cause the drug resistance. The commonest thing that we see is that the genetic alteration initially seen is still there, but a second genetic alteration has occurred, to prevent the drug from binding to the kinase inhibitor. But we also see other new genetic alterations occurring called bypass tracks where cells learn to bypass the signalling cascade which is being inhibited and switch on either pathway and sometimes cells themselves transform to more aggressive subtypes looking completely different under the microscope.

We can see that genetic alterations occur in patients with cancer, particularly lung cancer, and that identifying these are particularly important. So how do we deliver tumour genetic testing at scale in routine clinical care in the clinic with high quality and confidence?

Well, biomarker testing and reporting is a complex process. Somebody once told me that it's not that difficult, it's not rocket science, and I would retort that actually it is really quite a complex process. We have to remember that a patient has been newly diagnosed with cancer in the lungs, and is often really quite unwell, with respiratory symptoms and usually rather significant medical complications. We have to obtain the pathological sample; it has to be analysed in the genetic lab the genetic lab has to report on the findings that they identify and that report has to reach the treating oncologist.

However, there are many steps in this process where problems can occur. Who orders the molecular tests? Is it the respiratory physician or the pathologist as the oncologist has never met the patient at that point. How long does it take for the sample to get from where the biopsy is sitting, which is usually in a different place to where the molecular laboratory is, and where is the actual sample sitting, because the pathology laboratory may be a remote location from where the samples actually got taken from the CT scanning suite or the bronchoscopy suite. About the lab, they generally do a great job in analysing the specimen. But has the best test been performed for the question that is being asked, often this is bit of a gray zone in the laboratory just performs a standard test for all lung cancers. And finally, who receives the report? The person that receives the report is the actioning oncologist who is often not the person that orders the test in the first place. The person ordering the test is usually the pathologist and then when the oncologist receives the report, it is usually in often complicated genomic language. So how does the oncologist interpret the report when they have not been trained to do so as the whole area of genomics has been implemented and been mushrooming within recent years.

We also have to recognise that there are many different tests that can be performed on cancer specimens. Indeed, all molecular tests have their limitations, and one would argue that the gold standard test is actually whether the patient benefits from the drug when you actually give it. We use single-gene tests which are fast but waste DNA and can give false negatives as they are not that sensitive and are usually looking to see if a predefined set of mutations is present or not. We have now generally moved over to panel-based sequencing of DNA and RNA which allows us to examine many 10s to 100s of genes at the same time which is a highly efficient way of analysing tumours. But this technology can be slow, and interpreting the results can be really quite problematic. We can use simple straightforward pathology techniques called immunohistochemistry which are fast and efficient, but false negatives and false positives can be quite common depending on the test, and the sample may need molecular verification, anyway. And for certain questions we can use a technology called FISH which for gene rearrangements had been the gold standard but again can be slow and is prone to false negatives and positives and does not give us genomic breakpoint data that may be important.

The other thing that we have identified is that it's not only what is in the foreground that we are looking for IE the driver genetic alteration, but that the background genomic architecture matters. Hence whilst we are looking for certain genetic alterations such as mutations or fusions, we have to be aware of what is lurking in the background, specifically which other genetic alterations are there, that may impact on how the drug works, and how the patient fares.

So far, I have spent a lot of time talking about genetic alterations occurring within the tissue of the cancer itself but what about heritable mutations, mutations that patients are born with, throughout all of their cells in their body. Can these mutations cause cancer, and then their presence to help us with drug decision making?

The answer to both of these questions is yes, it is important, and it does help us. A classic example is that of BRCA 1 and 2 mutation detection which cause sensitivity to PARP inhibitors. Heritable BRCA1 and BRCA2 mutations are very rare in general, and predispose to the development of breast, gynae, and prostate cancers. Seminal work performed in our institution, identified that tumours with these mutations in them resulted in sensitivity to a type of drug called PARP inhibitor, as the cells were no longer able to repair their own DNA. This led to the first phase 1 trials of PARP inhibitors in patients with heritable with BRCA mutations and cancer led by my colleagues at our institution and subsequently globally, identifying that the PARP inhibitor olaparib resulted in quite dramatic benefit in patients born with these mutations that had subsequently developed cancer attributable to these mutations. Indeed, olaparib is now routinely used in breast, ovarian, prostate and pancreatic cancers, in patients with these BRCA genetic alterations, a true example of translation of science into routine effective care, from an academic laboratory to globally changing the face of cancer.

Well, what about lung cancer? Do we see similar issue in lung cancer? Here the story is more complex. BRCA mutations tend not to cause lung cancer. And by and large, the mutations that we find in lung cancer occur within the lung cancer itself, and patients generally are not born with a heritable susceptibility to lung cancer. At least that is what we thought. In our institution we have recently reported on the data where we have mainstreamed testing patients for mutations that they were born with alongside mutations within their cancer in specific groups. Indeed, when we took patients with EGFR mutant lung cancer and looked at their own genes that they were born with we identified that just under 1 in 10 of these patients actually had a heritable susceptibility to cancer. This clearly has huge implications for those patients' families.

And now we are testing patients' tumours in extensive detail, one question asked is "can we identify these germline or heritable mutations through simply testing the cancer itself?" Well, the answer is yes, these can be identified, although not directly and we see signals of this but cancer gene test reports do not often make it obvious that the genetic abnormality identified may be in the tumour as well as being in each of the cells of the individual, and hence potentially a heritable mutation.

But much of what I have been talking about has been on the basis of taking a tissue sample of the cancer and genetically analysing that tissue. A process by which we have to put either a needle into the chest to get a sample of the tumour, or a camera in the airways otherwise known as a bronchoscope to obtain a sample of the tissue directly. Procedures that none of us I think would want to volunteer to have if we did not need to.

Over recent years we have learnt much from analyzing the blood. We have actually identified that from a simple blood draw we can look at the plasma, which is the part of blood without any cells, and at a very, very, very, low level, we can sometimes find fragments of DNA which have originated from tumour cells in the patient. Indeed, we now have technologies available, which can simply, from a blood draw, analyse these small fragments of DNA, to tell us about the tumour genetics, called circulating tumour DNA or ctDNA.

So, one of the questions I often get asked is if you find a genetic alteration from the tissue does it actually relate to that same genetic alteration you can find swimming in the blood and at a very, very, low level. And the answer is that most of the time it does: there is concordance between the mutations of genetic alterations in tissue and the blood. But often we find patients who have a genetic alteration in the tissue, but we cannot detect it in the blood and that can be for a variety of reasons such as the tumour is not shedding adequate amounts to leach out into the blood. The alternative is that we can find genetic alterations in the blood, but not identified them in the tissue. This sometimes occurs when the tissue sampled does not reflect the whole of the genetics of the tumour because of where the needle went in may have a slightly different genetic profile to another area, or perhaps the blood genetic abnormality is not coming from the tumour but leaching out from the white cells of the blood, a process called clonal haematopoiesis.

Indeed, understanding and analysing ctDNA is complex, because when we look at the genetic material from plasma, we have to understand whether the genetic alteration we are looking at is derived from the tumour, which is what we would like to see, from the white cells leaching out into the blood, a process called clonal haematopoiesis, or indeed whether the mutation directly represents a mutation present in every cell of the body, particularly the white cells called a heritable or germline mutation. Therefore, analysing blood genomics through circulating tumour DNA is a complex task.

Nevertheless, ctDNA testing may revolutionize clinical care. Here is a typical clinical pathway for what happens to patients revered from their GP to hospital when lung cancer is suspected. They attend the

hospital respiratory clinic and have usually had a CT scan just before the clinic. The respiratory physician looks at the CT scan and suspects lung cancer. Patients then undergo additional investigations such as PET scans or brain scans if necessary and some sort of biopsy to get a piece of tissue to make the diagnosis, either an image guided biopsy or a bronchoscopy, usually. Thereafter the pathologist will review the tissue biopsy and confirm the diagnosis of NSCLC and the case will be discussed in the multidisciplinary team meeting. Molecular evaluation of the sample will be requested, and the patient will then begin drug treatment according to the molecular diagnosis. However, the time frame from tissue sampling until staring drug therapy can be 3-4 weeks if not longer: a very unsatisfactory state of affairs. Could this whole wait time be truncated by the use of ctDNA? What would happen if we tested patients for ctDNA with a simple blood draw alongside their tissue biopsy undergoing their usual analysis?

We evaluated this process at our Institution in just under 250 patients and published our results. What we found was that there was extremely good concordance for important genetic alterations (tier 1 genotypes) between tissue and ctDNA genotyping. However, ctDNA analysis identified 25% more genetic alterations that tissue missed and similarly tissue 25% more genetic alterations that ctDNA missed: thus, using both in parallel is complimentary and increased the number of patients with important genetic alterations by nearly half (46%).

But the implications of bringing in this assay were even more important. ctDNA results were available in a median of 8 days from time of sampling versus 22 days for tissue biopsies and as a consequence, results were available to clinicians much more rapidly. Indeed, time to gentotyping results improved by 63% when using ctDNA over tissue testing. This has major implications. Because genetic information is so important for drug decision making, the time to starting treatment improved from a median of 35 days to 16 days: an improvement of 54%.

But these data have other implications, what happens if the scan suggests lung cancer in a very frail patient, but the biopsy is complex or risky or if a patient has undergone a biopsy, but the tissue material is insufficient for molecular testing? Again, ctDNA can be very useful in planning if a biopsy is really worth the while in patients with major risk factors and prevent re-biopsy in patients with insufficient tissue for molecular testing.

But what happens if the timing of the blood draw is not later in the pathway, once the tissue biopsy has confirmed lung cancer, but much earlier in the pathway, right at the beginning, at the time the patient goes to the hospital and has a CT scan where lung cancer is suspected. Well, we performed a pilot study of this process when COVID hit, as all lung biopsies were initially paused and massively delayed. Here, we tested 49 patients, and in one fifth (22%) we were able to start a targeted therapy either with or without the tissue or molecular results.

Indeed, here is a case of a lady with a mass in the chest in whom a tissue biopsy was delayed due to COVID. Here, ctDNA confirmed a druggable MET14 alteration and she was able to commence tepotinib, a MET inhibitor, at the same time as the biopsy was being planned with good and durable shrinkage and of the tumour thereafter.

We have now liaised with NHS England and with our guidance they are implementing ctDNA testing into the respiratory clinic at the time of first appointment, in a pilot study to evaluate what the clinical benefits are and its cost effectiveness. Phase 1 of 700 patients in England has completed and we are nearly completing phase 2 implementation of a further 1800 patients. I do hope that this programme will result in ctDNA implemented routinely in the clinic and if so, England will be the World's first country to implement this ctDNA at scale. If successful, I am sure this concept will be taken forward in other tumour types.

So, what does the future hold? You won't be surprised to hear that there is a huge amount of activity in genomic technologies and their clinical implementation ongoing.

When it comes to tissue analysis, our understanding of the complexities of the human and cancer genome are very much in their infancy. The more we analyze of the genome the more we find. Implementing exome and whole genome sequencing in the clinic is now possible, although there are still huge limitations on the bioinformatics aspects. For example, we know that cancer genetic alterations can be clustered into at least 21 different unique signatures, which can help us identify how that cancer arose; for example a tumour may have a genomic signature typical of tobacco exposure or of UV light exposure. But how this information will be useful in the clinic remains unknown.

Similarly, there is an ongoing explosion in research for implementation of ctDNA. For example, if a patient has undergone curative surgery, its fairly straightforward to do a ctDNA test to see if there is any residual

tumor-derived DNA identified, as this may predict those that need additional consolidation treatment sich as chemotherapy or not: IE the technical utility has been proven. However, whether we should or not remains unproven: what are the pros and cons of this technology (such as false positives and false negatives) and what is the cost effectiveness? Similarly, we can test a group of healthy individuals with ctDNA to see if this is present implying that they may have cancer on board. Again, the technical utility has bene proven, but whether we should or not, ie the clinical utility, remains unproven, and the subject of ongoing research with initial data suggesting that ctDNA is effective in identifying some cancers in otherwise healthy individuals but less sensitive when limited to early stage curable cancers, and whether this improves mortality or not, remains unknown.

And with that, I'd like to thank you for your attention.