Cancer genetics or cancer genomics in the era of genomic medicine?

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Introduction

This paper will outline the field of cancer genetics and will identify how knowledge of this area can facilitate better care for cancer patients and their families. Using breast/ovarian cancer as a model, the issues surrounding inherited predisposition to cancer will be explored in terms of models of tumour development, clinical presentation, genetic testing and management interventions. Our limited understanding of the mechanisms whereby molecules, pathways and networks contribute to the evolution of the malignant phenotype will be set forth.

Genetics is the study of single genes and their effects. Genomics, on the other hand, is a relatively new term describing the study of the function and interaction of all the genes in the genome [1]. Cancer was described almost 30 years ago as representing the phenotypic expression of accumulated genetic damage [2]. The malignant phenotype can rarely, if ever, occur as a result of a single genetic defect. As such we should probably now discard the term cancer genetics and talk of cancer genomics among the repertoire of diseases encompassed by genomic medicine [1, 3, 4].

Aspects of genomic medicine relevant to cancer as a multifactorial disorder include inherited mutations that confer an increased predisposition, somatic mutations in the process of carcinogenesis and genetic variations that may be pathological or protective in the context of disease expression or management. An understanding of pharmacogenomics is increasingly relevant to the management of certain malignancies; for example, acute lymphoblastic leukaemia where genetic predisposition to toxicity from agents such as methotrexate and mercaptopurine can be predicted and appropriate dose adjustments made.

Genetic testing has been described as “the analysis of human DNA, RNA, chromosomes, proteins and certain metabolites in order to detect heritable disease-related genotypes, mutations, phenotypes or karyotypes for clinical purposes” [3]. Some tests from this broad range have been applied for many years and include the detection of chromosomal breakage syndromes and the measurement of stimulated calcitonin levels in people at risk of familial medullary thyroid cancer. With the location and cloning of genes conferring inherited predisposition to common cancers [5, 6] much of the interest of the oncology community is now focused on issues surrounding genetic diagnosis and screening for mutations in cancer susceptibility genes [7]. A further component is the application of predictive testing to unaffected, at-risk individuals [6]. This new activity in cancer care requires on-going careful assessment of the risks and benefits to people entering the process. Although evidence is accumulating that certain interventions can help to reduce the risk of dying from cancer for those who carry deleterious, inherited mutations, it is likely that, in general, tests that assess risk are likely to be developed faster than our capacity to offer risk reduction measures. We must, therefore, keep ourselves, our patients and our partners in medical care delivery informed of developments in this area towards developing rational policies in clinical practice.

Models of tumour development

Tumour development may be associated with mutations or the inappropriate expression of specific normal cellular genes involved in growth control [9–14]. These genes are referred to as oncogenes in this context, and dominant and recessive oncogenes have been described. Activation of dominant oncogenes leads to the malignant transformation of cells that contain them. Examples of this are the oncogene activations that occur with chromosomal translocations in diseases such as chronic myeloid leukaemia [t(9:22)] and Burkitt’s lymphoma [t(8:14)] [13]. In such circumstances, the mutations that lead to oncogene activation are dominant at the cellular level and only one copy of the gene requires to be mutated for the malignant phenotype to develop. Assays for dominantly acting oncogenes have been developed whereby transfection into recipient cells can be undertaken and the number of transformed colonies measured [15–17].

In other circumstances where neoplasms arise through an inherited predisposition, tumour development is associated with two recessive mutations that inactivate oncogenes. These genes are referred to as recessive oncogenes, anti-oncogenes or tumour suppressor genes [10–12]. Cancers can also arise sporadically through this mechanism. When cancer risk is inherited the first of the two recessive mutations comes via the germ cell line and is present in all cells. The recessive mutation, however, is not expressed until a second mutation within a somatic cell causes loss of the normal functioning allele. Where similar tumours arise through inherited predisposition and through sporadic mechanisms, the difference is that with inherited predisposition the first mutation occurs in the germ cell whereas with a sporadic tumour both mutations occur in the somatic cell. It can be readily seen, therefore, that the risk of malignancy in an individual who has not inherited the germline mutation is much smaller given that both mutations must occur by chance in the same somatic cell.

The model that describes two (or more) mutations in the development of tumours is known as the ‘two hit’ or Knudson’s hypothesis [18]. The normal function of tumour suppressor genes appears to be the regulation of growth and differentiation within cells. This model serves to explain the apparent paradox whereby...
Inherited cancer syndromes are due to recessive mutations but are dominantly inherited within families. An individual who has inherited the first recessive mutation will have a huge number of cells at risk within the target tissue(s) and the loss of a remaining wild-type allele within a predisposed cell is an event highly likely to occur. Such a cell with two hits will ultimately give rise to a tumour and a high percentage of those with a germline mutation will develop malignancy even though they inherited just one mutated copy of the recessive gene. A major feature related to the ‘second hit’ that occurs in the somatic cell is the frequent large scale deletion of chromosomal material, which causes the loss of the remaining functioning allele [10–12]. By comparing DNA sequence polymorphisms in leukocytes and tumour tissue in the individual patient, the loss of the normal allele can by detected. This has greatly facilitated the investigation of genetic abnormalities associated with tumour development and the detection of loss of heterozygosity (LOH) can represent the unmasking of a mutated gene on the other allele, in other words the germline mutation.

### Clinical cancer genetics

With this background knowledge of tumour development a number of inherited cancer syndromes can be recognised [7]. Some are rare and some are relatively common, the latter representing a percentage of common malignancies such as bowel, breast and ovarian cancer. As described above, the inheritance is mainly autosomal dominant and mainly associated with inactivation of a tumour suppressor gene (Table 1). This inheritance pattern may also be associated with oncogene activation (Table 2) and, with loss of genomic integrity, there are a number of autosomal recessive diseases associated with the hereditary cancer syndromes (Table 3).

### Table 1. Autosomal dominant inheritance: loss of tumour suppressor gene

<table>
<thead>
<tr>
<th>Cancer syndrome</th>
<th>Involved gene(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hereditary retinoblastoma</td>
<td>RB1</td>
</tr>
<tr>
<td>von Hippel–Lindau</td>
<td>VHL</td>
</tr>
<tr>
<td>Nevus basal cell carcinoma</td>
<td>PTCH</td>
</tr>
<tr>
<td>Cowden syndrome</td>
<td>PTEN</td>
</tr>
<tr>
<td>Peutz–Jeghers syndrome</td>
<td>STK11/LKB1</td>
</tr>
<tr>
<td>Hereditary melanoma</td>
<td>CDKN 2A</td>
</tr>
<tr>
<td>Neurofibromatosis type 1</td>
<td>NF1</td>
</tr>
<tr>
<td>Neurofibromatosis type 2</td>
<td>NF2</td>
</tr>
<tr>
<td>Familial adenomatous polyposis</td>
<td>APC</td>
</tr>
<tr>
<td>Hereditary diffuse gastric cancer</td>
<td>CDH1</td>
</tr>
<tr>
<td>Multiple endocrine neoplasia type 1</td>
<td>MEN1</td>
</tr>
<tr>
<td>Familial cutaneous and uterine leiomyomatosis</td>
<td>FH</td>
</tr>
<tr>
<td>Prostate cancer</td>
<td>?RNase L</td>
</tr>
<tr>
<td>Hereditary breast/ovarian cancer</td>
<td>BRCA1/BRCA2*</td>
</tr>
<tr>
<td>Li–Fraumeni syndrome</td>
<td>TP53*</td>
</tr>
<tr>
<td>Hereditary non-polyposis colorectal cancer</td>
<td>MLH1/MSH2/MSH6*</td>
</tr>
</tbody>
</table>

*Genes involved in DNA repair.

### Table 2. Autosomal dominant inheritance: oncogene activation

<table>
<thead>
<tr>
<th>Cancer syndrome</th>
<th>Involved gene</th>
</tr>
</thead>
<tbody>
<tr>
<td>Multiple endocrine neoplasia types 2A, 2B</td>
<td>RET</td>
</tr>
<tr>
<td>Familial medullary thyroid cancer</td>
<td>RET</td>
</tr>
<tr>
<td>Hereditary papillary renal cell carcinoma</td>
<td>MET</td>
</tr>
<tr>
<td>Hereditary gastrointestinal stromal tumours</td>
<td>KIT</td>
</tr>
<tr>
<td>Familial cutaneous malignant melanoma</td>
<td>CDK4</td>
</tr>
</tbody>
</table>

### Table 3. Autosomal recessive inheritance: loss of chromosomal integrity

<table>
<thead>
<tr>
<th>Syndrome and cancers</th>
<th>Involved genes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ataxia-telangiectasia, breast, lymphoma</td>
<td>ATM</td>
</tr>
<tr>
<td>Bloom syndrome, various solid tumours</td>
<td>BLM</td>
</tr>
<tr>
<td>Xeroderma pigmentosum, skin cancer</td>
<td>XPA-G</td>
</tr>
<tr>
<td>Fanconi anaemia, leukaemia</td>
<td>FANCA,C,D,E</td>
</tr>
</tbody>
</table>

Whereas the rarer syndromes are fascinating at a clinical and laboratory level [14], in day to day oncology practice it is the common cancers, such as breast/ovarian and bowel malignancies, which mainly pose management problems. Numerically, hereditary breast and ovarian cancers associated with BRCA1 and BRCA2 mutations are the most important and will, therefore, be the subject of further discussion.

### Breast/ovarian cancer

In most European countries breast cancer is the most common cancer among women, with a lifetime risk of ~8%, and ovarian cancer is the most common cancer of gynaecological organs, with a lifetime risk approaching 1%. Approximately 5–10% of breast and ovarian cancers are caused by inheritance of a mutation in a cancer predisposition gene, but the inheritance of such a mutation, as we have seen, greatly increases the risk of developing cancer [19]. Although the proportion of cases arising because of inherited predisposition is small, the absolute numbers are large because breast cancer in particular is a common disease.

The clinical features that should lead to suspicion of inherited predisposition to cancer in this setting are early age at onset, bilateral breast cancer, multiple cases of breast cancer and ovarian cancer in the family, male breast cancer, breast and ovarian cancer in the same individual and Ashkenazi Jewish origin [19–23]. A key initial step is to gather accurate data on family members, both affected and unaffected, in at least three generations to verify cases of cancer from medical records and death certificates, and to compile a pedigree with these details included, which can serve as a basis for further decision-making and education. Our goal in cancer genetics, as in all areas of cancer medicine, is to prevent deaths from cancer. Genetic testing for hereditary risk of breast and ovarian cancer has been available since 1996 and since then there have been numerous reports that extend our knowledge,
shape our medical management and help to guide risk-reduction strategies for women who have inherited deleterious mutations in the BRCA1 and BRCA2 genes [24–26]. Currently, three basic approaches have been adopted: increased cancer surveillance, the use of chemoprevention and prophylactic surgery. These options need to be considered not only for those unaffected women who carry BRCA1 or BRCA2 mutations, but also for women in this situation who have already developed cancer and who remain at high risk of developing a second malignancy. Early detection and risk reduction options commonly recommended are outlined in Table 4.

### Secondary prevention

Breast cancer surveillance is a modality that is increasingly accepted for early detection of breast cancer in women at high risk, and the hypothetical risk from low-dose radiation associated with screening mammography is outweighed by the benefit of early disease detection. Definitive results from studies using magnetic resonance imaging as a screening method are awaited [27]. Further reassurance regarding lack of increased radiation sensitivity among BRCA mutation carriers is offered by experience in treating such women with limited breast surgery followed by irradiation [28]. No evidence of increased disease recurrence or toxicity has been noted among women with high-risk BRCA-associated breast cancer and those with sporadic disease in this setting.

While screening for ovarian cancer with transvaginal ultrasound (TVU), often using colour-flow Doppler imaging to enhance sensitivity, is widely used along with CA 125 measurement, this remains an unproven screening method. Ideally, TVU should be performed in the early follicular phase of the menstrual cycle to avoid false-positive findings, but even then there is a tendency for this methodology to be oversensitive and non-specific. It is important that this is made clear to women who seek and opt for this particular form of screening.

### Prophylactic surgery

In recent years a number of studies have proven the efficacy of prophylactic mastectomy for women who are at high-risk, including BRCA mutation carriers [29, 30]. While early risk reduction is >90% with bilateral mastectomy, this is a difficult and major decision for women to take. The majority of women appear content with their decision to have such surgery, but the provision of psychological support complemented by proper surgical care, with access to breast and nipple reconstruction, is key to a satisfactory outcome for women who seek this option. Subcutaneous mastectomy is not an acceptable procedure and total mastectomy is the recommended operation.

Given the difficulties with screening for ovarian cancer, prophylactic oophorectomy is a consideration strongly recommended for women who harbour BRCA mutations [31, 32]. Women should consider this option once they have fulfilled their child-bearing ambitions, usually from their late thirties onwards. Fortunately, the peak of ovarian cancer occurs at a later age than breast cancer. Prophylactic oophorectomy reduces the risk of ovarian cancer by >95%, though a small risk remains related to small residual nests of Müllerian tissue within the peritoneum. There is, therefore, a 2–4% residual risk of ‘ovarian’ cancer for women who opt for this prophylactic surgery and the potential for development of this serous surface malignancy should be carefully explained preoperatively.

An added benefit to prophylactic oophorectomy is the reduced risk of breast cancer among BRCA mutation carriers. This has recently been shown to be ~50%, and surprisingly, this risk reduction also applied to women who received hormone replacement therapy (HRT) following ovarian removal. There is no agreed protocol for the use of HRT in women at high risk of breast cancer. In general, doctors are very cautious about the prescription of systemic hormone replacement, with many preferring to use non-hormonal agents such as clonidine or selective serotonin re-uptake inhibitors, along with topical oestrogen preparations for vaginal atrophy and related symptoms [33]. Others take a more liberal approach and use conventional oestrogen replacement up to 50 years of age and then switch to a selective oestrogen receptor modifier (SERM). Lifestyle studies currently underway in this area will hopefully offer guidelines for the future.

### Chemoprevention

The use of SERMs as chemopreventive agents for women with BRCA mutations remains somewhat controversial. While tamoxifen
was shown in the National Surgical Adjuvant Breast and Bowel Project (NSABP) study to reduce the risk of breast cancer by almost 50% in high-risk women [34], including those with a strong family history, no study has specifically reported on BRCA mutation carriers [34–36]. However, women with mutations diagnosed, with breast cancer and taking tamoxifen as adjuvant therapy for 2–4 years had a reduction in risk of developing contralateral breast cancer of 75% [37]. These data encourage us to believe that SERMS will be effective in reducing the risk of breast cancer among unaffected BRCA mutation carriers and offer the rationale for the frequent current use of tamoxifen in this setting. Food and Drug Administration (FDA) approval has been granted in the USA for using tamoxifen 20 mg daily for 5 years for this purpose. Given the small risk of endometrial cancer among tamoxifen users [38], the wisdom of combining hysterectomy with oophorectomy in women who opt for prophylactic ovarian surgery has been widely discussed. A balance between the further risk reduction offered by hysterectomy and the operative risks of more extensive gynaecological surgery needs to be struck and the issue decided on an individual basis depending on the woman’s wishes, the availability of surgical expertise and other relevant factors.

Raloxifene is a newer SERM and data from the Multiple Outcomes of Raloxifene Evaluation (MORE) trial describe a 72% reduction in risk of invasive breast cancer among post-menopausal women using the drug [39, 40]. This is in addition to other beneficial effects such as prevention of osteopenia and maintaining a healthy blood lipid profile with lowering of the risks associated with atheroma. Future trials with this agent will define whether it has a role as a chemopreventive agent among BRCA mutation carriers.

It has been shown that the use of oral contraceptives reduces the risk of ovarian cancer in the general population, and this benefit appears to extend to BRCA mutation carriers [24]. However, the risk of breast cancer appears to rise with the use of oral contraceptives and a previous study that led to the use of these agents being advocated towards prevention of ovarian cancer [41] must now be put in the context of further information reported more recently regarding hormone replacement [42]. Again, the use of an oral contraceptive is a decision for the individual woman, but any BRCA mutation carrier who elects to take such hormonal agents must be carefully monitored for breast cancer risk and cannot be reassured that her risk reduction of ovarian cancer will be more than 60%. Trials are underway of other agents such as vitamin E, aspirin and other non-steroidal anti-inflammatory drugs and oral contraceptives with a high progesterin content for ovarian cancer prevention. At this point, however, no definitive recommendations can be made and the cautious approach outlined in the recent report from the US Preventive Services Task Force appears appropriate [43]. The recommendation on chemoprevention of breast cancer states: “the task force recommends that clinicians discuss chemoprevention with women who are at high risk for breast cancer and at low risk for adverse effects from the drugs.”

The genetic testing process

Testing for cancer predisposition is a clinical activity that is complex and time-consuming. It is also supported by laboratory investigations whose interpretation can range from being simple to being difficult [3, 4, 44–49]. There is no currently available genetic test for cancer susceptibility which can be appropriately applied to broad population screening. An important initial step, therefore, is to identify those who may truly benefit from genetic testing [50]. This usually means genetic screening of an affected individual who has a personal or family history suggesting an inherited cancer syndrome and, if a deleterious mutation in a specific gene is detected, offering the opportunity for predictive testing to unaffected family members to see whether they carry the mutation or not. Appropriate genetic counselling must be provided for those participating in the process both before and after genetic testing [51]. Education and counselling before genetic testing should outline clearly the benefits, risks and shortcomings of the process for individuals and families. It is important to listen and assess perceptions of cancer risk, learn of prior experiences with cancer within the family and provide an ample opportunity for questions to be asked and truly informed decisions to be made. All forms of coercion must be avoided and testing should be voluntary with the option to decline testing always available.

Provided there is a reasonable likelihood that: (i) a person carries a mutation in a cancer susceptibility gene; (ii) a genetic test to detect such a mutation is available and the test can be readily interpreted; (iii) the result is relevant to medical management; and (iv) an informed individual wishes to be tested, it is then appropriate to proceed with genetic testing. Once a test is undertaken, further critical components of clinical care are the disclosure of results in a post-test counselling session, further counselling and support as required, and for both mutation carriers and also high-risk individuals in whom a mutation has not been found, the provision of clinical care through a customised plan for each individual throughout their lifetime.

For BRCA mutation carriers and others at high risk of developing breast cancer, the available options have been discussed earlier. Both individuals and families experience various psychological reactions in the context of genetic testing and issues frequently arise that offer challenges to health carers involved in this area [52–59]. Many have lingering hurt and unresolved grief relating to cancer diagnoses and deaths within their families and these frequently surface during discussions. Guilt is a frequent reaction, with some showing guilt for transmitting a mutation to their children, and others showing survivor guilt when it is shown that they do not have the deleterious mutation. Various emotional reactions are encountered ranging from anxiety and fear to depression and loss of self-esteem. These feelings may be extreme and may lead to communication difficulties, problems with daily living and, importantly, may compromise the capacity of individuals to co-operate with their carers in developing and participating in care programmes. The family dynamic may be upset and various conflicts may arise, often adding to simmering differences present for years.

Therefore, at a time when individuals need the warmth and support of the family to nurture them through a period of difficulty,
this may not be forthcoming because of family strife. It should be remembered that cancer predisposition is transmitted through paternal and maternal lineages. All the issues described pertain to men as well as to women [60–62] and, as in life in general, differences in emotional reactions between the sexes are encountered in clinical practice in this area. Women tend to be supportive of each other, whereas men who are usually unaffected by disease appear more isolated and seem to benefit from close involvement of their partners. Avoidance of stigmatisation is important for both families and ethnic groups within which a particular mutation is transmitted.

As well as considering the psychological issues for individuals and families related to genetic testing, there are broader social, ethical and legal factors to be considered too [44]. Clearly there will be certain differences among communities and states worldwide and a comprehensive review is not possible here. Given that some practice in this area is now in mainstream medicine and some remains within the sphere of laboratory and clinical research, it is vital in addressing the ethical, legal and social dimensions of it is vital in addressing the ethical, legal and social dimensions of setting and inevitably conflict will arise between the concepts of some remains within the sphere of laboratory and clinical research, some practice in this area is now in mainstream medicine and will be certain differences among communities and states world-

ethical and legal factors to be considered too [44]. Clearly there could be modified and focused more keenly with such additional information. To facilitate this and, in particular, to develop effective pharmacological agents, a better understanding of disease mechanisms at the molecular level is required. The recent development of targeted therapies, such as imatinib mesylate for chronic myelogenous leukaemia, other chronic myeloproliferative diseases and gastrointestinal stromal tumours, has signalled how successful this approach can be [66–68]. As well as the BCR-ABL and c-KIT target, other validated targets are HER2/NEU (Herceptin) and PML-RARα and PLZF-RARα (retinoic acid) in breast cancer and promyelocytic leukaemia, respectively.

Molecular mechanisms

In looking at the BRCA genes and their protein products as potential therapeutic targets, it is important to have an understanding of their function. Both BRCA1 and BRCA2 have been shown to be involved in the recognition and repair of DNA damage and to operate in collaboration with multiple other genes in transcriptional regulation of gene expression, transcription-coupled repair of oxidative DNA damage, chromatin remodelling and in control of cell cycle checkpoints [69]. Genes such as RAD51, ATM, CHK2 and the mismatch repair genes MSH2 and MSH6 are among the other genes offering potential targets in this complex activity [69, 70].

While much has been learned, it will take much further unraveling of the molecular mechanisms related to BRCA activity before targeted therapies to compensate for the deficiencies of these proteins will become available. Current impressions are that ~60% of hereditary predisposition to breast cancer is associated with BRCA genes and at least 35% with genes yet to be discovered. Almost 5% is related to other known genes: TP53 (Li–Fraumeni syndrome), LKB1 (Peutz–Jeghers syndrome), PTEN (Cowden’s syndrome) and the mismatch repair genes of hereditary non-polyposis colon cancer (HNPPC). Unravelling the molecular mechanisms involved in these syndromes is also in progress [71–73]. Most of the genes involved in hereditary predisposition to breast and ovarian cancer are not, with current knowledge, involved in the majority of sporadic cancers of these organs. Understanding the molecular mechanisms underlying these sporadic cancers therefore offers an independent and even greater challenge.

Cancer genomics as an important component of genomic medicine has arrived. While medical oncologists have not hitherto been
specifically trained in medical genetics, they must now identify patient care in this area as within their remit. The American Society of Clinical Oncology has set an example in this, with the development of a curriculum in Cancer Genetics and Cancer Pre-disposition testing, as well as holding frequent review meetings on clinical cancer genetics [74]. The European Society for Medical Oncology must also embrace this activity and identify it as a key component of cancer care in Europe and throughout the world.

References

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