A Clinical Systematic Review of BRCA1 and BRCA2 Genetic Testing for Breast and Ovarian Cancers

This report and the French version entitled *Étude méthodique et clinique du dépistage génétique de BRCA1 et de BRCA2 dans la détermination de la prédisposition au cancer du sein et au cancer ovarien* are available on CCOHTA’s web site.

Agence d’évaluation des technologies et des modes d’intervention en santé (AÉTMIS) and the Canadian Coordinating Office for Health Technology Assessment (CCOHTA) collaborated to systematically examine the available evidence regarding the analytical and clinical validity of available molecular technologies, and review inherent issues associated with testing. The results pertaining to molecular methods, analytical validity, psychosocial impact, ethical implications, and clinical management are presented in this report. Results related to prevalence, penetrance, risk assessment, clinical validity, and genetic counselling will be presented separately in forthcoming AÉTMIS monographs.

Production of this report is made possible by financial contributions from Health Canada and the governments of Alberta, British Columbia, Manitoba, New Brunswick, Newfoundland and Labrador, Northwest Territories, Nova Scotia, Nunavut, Ontario, Prince Edward Island, Québec, Saskatchewan, and Yukon. The Canadian Coordinating Office for Health Technology Assessment takes sole responsibility for the final form and content of this report. The views expressed herein do not necessarily represent the views of Health Canada or any provincial or territorial government.

Reproduction of this document for non-commercial purposes is permitted provided appropriate credit is given to CCOHTA.

CCOHTA is funded by Canadian federal, provincial and territorial governments.

Legal Deposit – 2006
National Library of Canada
ISSN: 1203-9012 (print)
ISSN: 1481-4501 (online)

PUBLICATIONS MAIL AGREEMENT NO. 40026386
RETURN UNDELIVERABLE CANADIAN ADDRESSES TO
CANADIAN COORDINATING OFFICE FOR HEALTH TECHNOLOGY ASSESSMENT
600-865 CARLING AVENUE
OTTAWA ON K1S 5S8
A Clinical Systematic Review of BRCA1 and BRCA2 Genetic Testing for Breast and Ovarian Cancers

March 2006


CCOHTA takes sole responsibility for the final form and content.
BRCA1 and BRCA2 Predictive Genetic Testing for Breast and Ovarian Cancers: A Systematic Review of Clinical Evidence

**Technology**
Tests to detect mutations in Breast Cancer susceptibility genes BRCA1 and BRCA2

**Condition**
Some individuals are more likely to have BRCA1 and BRCA2 gene mutations. These mutations have been linked to hereditary breast and ovarian cancers, which account for 5% to 10% of the roughly 24,000 new cases diagnosed annually. Individuals diagnosed with hereditary breast cancer have one or both mutations 84% of the time. The prevalence of BRCA1/2 mutations is between one in 500 and one in 1,000.

**Issue**
Genetic testing for these mutations is available in Canada, and can be accessed as a clinical laboratory service or through a research study. There is a need to better understand the benefits and harms that are associated with testing, the available tests and how they compare with each other, the social factors that influence testing, and the psychological and ethical issues that are associated with testing.

**Methods and Results**
Literature was identified through a defined search strategy and selection criteria. The analytical performance of tests was evaluated from 27 unique studies. Sixty-eight reports with quality measures of psychosocial and ethical issues were identified and synthesized. Eighty-four reports that described the clinical outcomes in prophylactic or therapeutic studies were identified and synthesized to assess the benefit and harm of testing.

**Implications for Decision Making**
- **Other factors need consideration when choosing BRCA1/2 testing.** There is no clear evidence to suggest testing will lead to decisions that result in long-term health benefits.
- **Psychological and social implications require consideration.** Knowledge about the association of cancer and genetics is limited in the general population. Test results influence individual risk perception, emotional states, and social issues. Counselling reduces the perceived risk and the associated anxiety, and increases the uptake of testing.
- **There is no compelling evidence that one test performs better than another.** Until better information becomes available, other factors such as test availability, ease of implementation, regulatory considerations, and price should be considered in deciding the method used for testing.
- **Decisions regarding BRCA1/2 testing need to be revisited.** Scientific data are accumulating rapidly. If the expansion of testing and the creation of best practices are pursued, this report should be updated. Decision makers who adopt this technology should consider the value of gathering information that can contribute to future analyses.

1 Introduction

Breast and ovarian cancers are among the leading causes of cancer-related deaths in Canadian women; 5,200 women die from breast cancer, and 1,500 women die from ovarian cancer each year.\(^1\) Between 5\% and 10\% of all breast and ovarian cancers are hereditary,\(^2\) and approximately 20\% of those can be attributed to mutations in the \textit{Breast Cancer} susceptibility genes \textit{BRCA1} and \textit{BRCA2 (BRCA1/2)}.

If one copy of either \textit{BRCA1/2} is altered, a subsequent mutation to the other copy may result in uncontrolled cell growth or cancer. A variety of evidence suggests that these genes are involved in deoxyribonucleic acid (DNA) repair and genome stability.\(^3\) Studies suggest that women who develop breast cancer before age 50 have a one in four chance of carrying a \textit{BRCA1/2} mutation if they have any relative (first-, second-, or third-degree) who develops the disease before age 50.\(^4\) Mutation carriers have a 50\% chance of transmitting an affected gene to their offspring.

Hereditary breast cancer is clinically distinct from sporadic cancer: it occurs at an earlier age, it more often affects both breasts, and it is associated with other cancers. Not all women who carry a \textit{BRCA1/2} mutation will develop breast or ovarian cancer.\(^5\) The penetrance varies between families and between studies. In \textit{BRCA1} mutation carriers, the lifetime risk of developing breast or ovarian cancer is as high as 85\% and 42\% to 63\% respectively.\(^5\) In \textit{BRCA2} carriers, the lifetime risk of developing breast or ovarian cancer is as high as 86\% and 27\% respectively. More recently, the cancer risk in carriers has been estimated from less selected families and population-based studies. By age 70, the average cumulative risk of breast cancer for \textit{BRCA1} carriers is 65\%, while that for ovarian cancer is 39\%. The corresponding cumulative risks for breast and ovarian cancers among \textit{BRCA2} carriers are 45\% and 11\% respectively.\(^6\)

Mutations that are more common in a well defined population are sometimes traceable to a common ancestor. Founder mutations are common in individuals of Jewish, Icelandic, UK, Spanish or Latin American, and French Canadian ancestry. There is evidence that a dozen other ethnic groups have a higher prevalence of specific \textit{BRCA} mutations (up to eight times that of the general population).\(^7\)

Factors that increase the likelihood of inherited breast or ovarian cancer include multiple cases of breast or ovarian cancer in a family, diagnosis of breast cancer before age 35, presence of male breast cancer in the family, or a combination of Jewish ancestry and breast or ovarian cancer.

Mathematical models, a multifactorial likelihood-ratio model, and computer programs have been developed to help determine cancer risk, but the interpretation of test results remains complex. The clinical significance of some variants is uncertain, and there are advantages and disadvantages to each of the molecular methods that are used to detect mutations. No one method can detect 100\% of gene mutations.

Genetic testing is performed on an individual’s DNA or mRNA (messenger deoxyribonucleic acid) after it is extracted from peripheral blood leukocytes. Most of the commonly used genetic techniques require that portions of a gene be replicated or amplified in vitro using the polymerase chain reaction (PCR).
**DNA amplification**

The most frequently used detection techniques rely on PCR amplification of the starting genomic DNA or mRNA. PCR is based on a specialized polymerase enzyme that synthesizes a complementary DNA strand in a mixture of DNA bases and two DNA primer fragments. The mixture is repeatedly heated and cooled, which exponentially multiplies the target DNA sequence.

**Direct sequence analysis (DSA)**

DNA sequencing pinpoints the location of a mutation and may indicate its effect on the encoded protein. This is useful when the full sequence is available through a public database, the type and frequency of mutations are well known, and a frequently updated catalogue is available.

**Multi-step analysis**

This involves pre-screening a gene for mutations using other kinds of analysis, such as single strand conformation polymorphism analysis, before using DSA.

**Protein analysis**

Because it is thought that most mutations result in protein truncations, gene proteins are synthesized, and then compared to normal proteins using the protein truncation test.

Most technologies are insensitive to large deletions or splice mutations that remove entire exons. This is true even of DSA, although it is considered to be the gold standard. According to the United States Task Force on Genetic Testing, a genetic test must be useful to those tested, and have analytical and clinical validity. It must be able to detect specific mutations in an individual’s gene (analytical validity) that will likely result in the person developing breast or ovarian cancer (clinical validity) (Appendix 1 of the full Technology Report).

In Canada, clinical geneticists in each province have developed clinical criteria, based on the family or personal history of cancer, which are used to establish an individual’s eligibility for testing. Alberta, British Columbia, Manitoba, Ontario, Quebec, and Nova Scotia have regional genetic laboratories, and may do tests for those in other jurisdictions.

Proprietary forms of BRCA1/2 testing are available. Myriad Genetics Inc. of Salt Lake City, Utah, holds a series of US and Canadian patents awarded from October 2000 to April 2001. Their fees range from US$450 (C$600) to US$2,600 (C$3,850), depending on the test.

2 **Objective**

The objective of the systematic review is to evaluate the analytical and clinical validity of BRCA1 and BRCA2 genetic testing; assess the contribution of molecular testing to genetic counselling and clinical management; and discuss the ethical and psychosocial issues inherent in testing.
3 Clinical Review Methods

A comprehensive search strategy was designed and tested a priori to identify published, grey, and unpublished literature in each subject area. Searches for all subject areas covered in this report included the years from 1994 until January 2003. An updated search was performed for all subjects in July 2004. Electronic databases searched included PubMed®, Cochrane Library and a Dialog® OneSearch® on MEDLINE®, CANCERLIT®, EMBASE®, Biosis Previews®, PASCAL, and PsycINFO®. Grey literature was identified by searching relevant web sites, clinical trial registries, clinical practice guidelines, and specialized databases. The commercial developer of the BRCA1/2 tests and primary researchers were also contacted to find unpublished studies, which are included in Appendix 3 of the full Technology Report.

Selection criteria

To be considered for review, studies on analytical validity and molecular methods had to include ≥20 individuals at risk for inherited breast or ovarian cancers with testing done in a research or clinical setting, using any molecular method to detect a BRCA1/2 mutation, and reporting a measure of analytical validity (sensitivity or specificity comparing the test result with genotype, with sequence analysis, or with >1 test; or any new technique for BRCA analysis).

For studies on genetic counselling, psychosocial impact, and ethical issues, the studies had to include ≥20 individuals at risk for inherited breast or ovarian cancers, with testing done in a research or clinical setting using any molecular method to detect a BRCA1/2 mutation and reporting an outcome qualitative in nature, showing the contribution of testing to counselling, and the psychosocial or ethical implications.

For clinical management, studies could be of any design, and the population had to be individuals at risk of developing inherited breast or ovarian cancer with certain factors that are indicative of a BRCA1/2 mutation (e.g., multiple cases of breast or ovarian cancer). Page 21 of the full Technology Report contains a full list of factors. The intervention was a molecular method to detect a BRCA1/2 mutation, and the outcome could be any clinical outcome from subsequent prophylaxis or therapy.

Two reviewers independently screened all citations using the preceding criteria, ordered the full articles if necessary, and made the final selection of relevant studies. Differences were resolved by consensus. Two reviewers then independently extracted data using a data extraction form that was designed a priori.

Quality assessment

Reviewers evaluated the robustness of study design, conduct, analysis, and interpretation, using the study summary and quality assessment forms. Studies on analytical validity were measured against the Standards for Reporting Diagnosis Accuracy statement, known as the “STARD.”

13
Data analysis
For the subjects areas covered in this report, only analytical validity was amenable to having measures of effect calculated. Analytical sensitivity and specificity values were calculated when data were sufficient. Subgroup analysis was performed by population, technique, or mutation type when feasible.

4 Results

Analytical validity and molecular methods
From a total of 881 citations, 28 reports describing 27 unique trials met the selection criteria for study. Many were disqualified, because index and reference techniques were not applied to all samples. Most selected trials were conducted at single hospitals, on individuals from a variety of ethnic groups and with unknown mutation status. A lack of information and a high degree of heterogeneity precluded any quantitative synthesis of the data. The most common reference test used was DSA, but no two studies used the same index test and unit of analysis. It was not possible to determine the most analytically valid molecular technique for detecting BRCA1/2 mutations because of the heterogeneity between studies, which precluded any quantitative analysis of the evidence.

There was a high degree of variability in the mutations tested, tests examined, and reference test used. No two studies used the same index test and the same unit of analysis, precluding any direct comparison of the data. Methodological limitations were identified in selected studies, regarding blinding, intra- and inter-observer reliability, and sources of bias.

Clinical management
From a total of 488 citations, 84 relevant reports met the selection criteria. On review, it became clear that there are no randomized controlled trials of genetic testing and treatment programs (i.e., where a program of testing and treatment has been compared to a program with no testing or treatment). Nor are there comprehensive uncontrolled studies of tested populations to determine the rate of false-positives and false-negatives. Studies only focus on patient cohorts who have been tested and treated.

Management of unaffected BRCA1/2 carriers
Women who test positive have three choices: prophylactic surgery (mastectomy or oophorectomy), drugs, or intensive surveillance. Recent studies have shown that surgery is associated with a reduced risk of breast and ovarian cancers in the short term (i.e., <5 years). The most convincing evidence for mastectomy was a prospective study of 139 women with pathogenic BRCA1 and BRCA2 mutations.14 None of the 76 women who underwent surgery developed breast cancer after a mean period of 2.9 years, whereas eight of the 63 women who chose not to have surgery developed the disease. For oophorectomy, a prospective study of 170 women found that the risk of cancer was reduced from 6.9% to 3.1% after a mean period of two years.
The acceptance of prophylactic surgery varies among countries where it is offered. One Dutch study reported that 51% of unaffected women with an identified mutation opted for bilateral mastectomy, and 64% for oophorectomy. This is higher than rates reported in US test centres. Chemopreventive therapy, particularly the antiestrogenic drug tamoxifen, is another choice for women who are at high risk of breast cancer. There is conflicting evidence on its effectiveness in women with BRCA1/2 mutations, because 80% of BRCA1-related breast cancers are estrogen-receptor negative. In women with BRCA2 mutations, which is estrogen receptor-positive, the difference in the rate of cancer in one randomized double-blind study of 13,338 women was not statistically significant between those who received tamoxifen and those who did not.

Surveillance programs, including mammography, magnetic resonance imaging (MRI), ultrasound, and breast self examination (BSE), are all used to detect breast cancer, but no studies show that they are of particular benefit to BRCA1/2 carriers. The first three may all lead to unnecessary biopsies, because of the possibility of false-positive test results. Mammography also carries the risk of exposure of breast tissue to radiation. Two meta-analyses concluded that BSE does not reduce mortality. While transvaginal ultrasound is the most effective modality for detecting ovarian cancer, it has not been shown to reduce mortality in high risk women.

**Management of affected BRCA1/2 carriers**

Options for managing BRCA1/2 carriers with cancer depend on tumour pathology, differences in survival, radiosensitivity, chemosensitivity, and screening for second primary cancers. BRCA testing at the time of breast cancer diagnosis significantly increases the likelihood of surgery, particularly bilateral mastectomy. Breast tumours that are associated with BRCA1/2 mutations are considered to be more difficult to treat and to have a poorer prognosis than sporadic cancers, yet studies have provided conflicting results. In a study of 49 Dutch BRCA1 carriers, outcomes were about the same; 49% were disease-free at five years compared to 51% for sporadic patients. In two combined studies, the survival rate was worse in 91 women with BRCA1/2 mutations than in those without (62% versus 86% respectively at 10 years, p<0.05).

To better understand the differences in breast tumours, Lakhane et al. studied the immunohistochemical profiles in BRCA1/2 carriers, and concluded that BRCA1 mutations have a distinct morphology and immunohistochemical phenotype, whereas the BRCA2 phenotype is poorly defined. These authors studied the pathological features of ovarian cancers in BRCA1/2 carriers and controls. They found that tumours in BRCA1 carriers were more likely to be invasive serous adenocarcinomas than tumours in age-matched controls (odds ratio 1:84, 95% CI: 1.21; 2.79).

Little evidence of chemoprophylaxis was found among the selected studies. One study showed that BRCA1 carriers who took tamoxifen for up to four years had a significantly increased level of protection. Another study indicated that there was benefit from the antineoplastic drug docetaxel in BRCA1/2 carriers with locally advanced or locally recurrent breast tumours.
Models of clinical management strategies for BRCA1/2 testing and treatment have been created based on certain assumptions about the prevalence of mutations, the proportion of women who accept testing, and the proportion of women who test positive and undergo prophylactic surgery. One study found that a simulated cohort of healthy 30-year-old women who tested positive for BRCA1/2 mutations could prolong their lives (in years) by the following measures: tamoxifen alone (1.8), prophylactic oophorectomy alone (2.6), tamoxifen and prophylactic oophorectomy (4.6), prophylactic mastectomy (3.5), and both prophylactic surgeries (4.9).32

**Health Services Impact**

**Psychosocial Impact**
From a total of 312 studies identified, 59 reports met the selection criteria. These were mainly single-site, cross-sectional studies where the mutation carrier status was unknown at the onset. Populations were mostly American, Canadian, or both.

**Results**
Many of the articles reported results from questionnaires, surveys, interviews, or focus groups not requiring follow-up. In half the articles, study participants were self-referred or volunteers from a program that offered genetic counselling and testing at no cost. As shown in Appendix 7 of the full Technology Report, outcomes were organized into four categories: knowledge and risk perception, interest in and attitudes towards genetic testing, psychological issues, and social issues.

- Participants’ knowledge about the association between breast and ovarian cancers, and genetics was limited. For instance, 56% of participants in the study by Bluman et al. did not know that a father could pass a mutation to his children.33 Self-referred respondents, Caucasian and married respondents with high household incomes, and Ashkenazi Jewish women were most knowledgeable.34,35 In all studies, participants showed a perception of elevated risk, and most overestimated their risk of being a carrier.

- Most study participants (70% to 82%) requested and underwent genetic testing. One study asked those who underwent BRCA1 testing if they would want their children tested, and 17.3% said “yes.”36 People who underwent testing had positive experiences for the most part, with >90% of participants in one study reporting satisfaction, with the exception of the wait for test results.37 In another study, 1% (carriers) regretted being tested, while 8% were unsure.38 In studies that offered genetic counselling, interest in testing was greater after counselling than before.

- The psychological issues included distress, depression, other emotional responses, and coping and support mechanisms. One study found that carriers showed significantly more breast cancer distress at seven to 10 days, and 12 months post-notification than untested women.38 Other studies comparing distress levels between carriers and non-carriers before and after disclosure found little or no difference.39,40 Participants coped by praying; talking with friends, family, or physicians; practising tension-reducing techniques; and changing their eating and exercise habits.

- Some studies asked participants if they shared their mutation status with their children, other family members, and friends. While participants were sometimes anxious about doing so, most disclosed their test results to close relatives, including children, siblings, and parents, but not distant ones.
a) Ethical Issues

Of the 236 citations identified, nine relevant articles met the criteria. These examined issues of informed consent, privacy, confidentiality, and familial implications specific to BRCA1/2 testing. All were observational and most were quantitative in design. The sample size ranged from 30 to 636, and the mean age ranged from 44 to 65, where applicable.

Results

1. Informed consent: To give “informed” consent, participants must understand the medical and non-medical risks and benefits of being tested. The process should encourage participants to consider the implications of testing on other family members, and involve them in counselling whenever possible; communication is critical.

2. Privacy and confidentiality: Most participants indicated a desire to keep test results confidential from employers, insurers, and other family members. The potential for discrimination based on genetic information could deter some from testing. Several countries have passed legislation to prohibit or limit the use of genetic information by third parties to deny health coverage.

3. Familial implications: Privacy may be considered paramount most of the time, but because genetic material is shared by families, health care professionals may feel morally or legally obliged to inform at-risk relatives of test results. Many organizations, such as the World Health Organization, would support them in cases where the harm to relatives is serious and imminent. Given the potential for family discord, it would be sensible for professionals to raise this issue before testing. Testing children is another contentious issue, because hereditary breast and ovarian cancers do not manifest until adulthood but the child may be “stigmatized” if they test positive. The accepted ethical opinion is that children should not undergo testing, even if parents request it.

4. Other implications: These involve patents and the cost-effectiveness of genetic testing. They were not addressed by the selected studies, but warrant exploration. Patenting biotechnology inventions has led to public controversy, because it could threaten equitable and affordable access to testing, and restrict health care research and technology development. Little attention has been paid to the ethical issues relating to the cost-effectiveness of testing. Costs arising from counselling, clinical follow-up, and testing should be considered in any analyses of genetic testing.

5 Conclusion

The integration of BRCA1/2 testing into the health care system has occurred under a variety of conditions and at different rates of uptake. Variations exist in related services such as genetic counselling, and the regional availability of health care professionals and resources to deliver these services.
There is no compelling evidence to suggest that one test performs better than another. Until better information becomes available, other factors such as test availability, ease of implementation, regulatory considerations, and price should be considered in deciding the method for testing. Although DSA is considered to be the “gold standard,” no two tests used the same index test and unit of analysis, thereby precluding comparisons. Clinically relevant mutations may be missed if DSA is used as a primary strategy for detecting $BRCA1/2$ mutations. Consequently, it was impossible to determine the most analytically valid molecular technique.

Prophylactic surgery was associated with a reduced risk of breast and ovarian cancers in short-term (<5 years) cohort studies, whereas surveillance strategies or chemoprophylaxis have not been shown to have a significant effect. Counselling is a critical component of the genetic testing process, informing the patient, and influencing perceived risk, anxiety, and uptake of testing. The public’s knowledge of the association between cancer and genetics is limited. Ethical considerations include informed consent (or refusal), privacy, and confidentiality.

Until better evidence becomes available, each jurisdiction will need to manage $BRCA1/2$ testing in accordance with their regulatory mechanisms, resources, and abilities. Policy and decision makers could conditionally reimburse $BRCA1/2$ testing for selected indications and restrict use to specific centres with identified protocols, or to particular health care providers to gather more information for future analysis.
9 References


