Chapter 1

General introduction
Chapter 1

Chapter outline

As an introduction to this thesis ‘Cancer risk variation in BRCA1/2 mutation families’ several aspects of the Hereditary Breast and Ovarian Cancer (HBOC) syndrome will be discussed briefly. This chapter starts off with a review of the epidemiology and characteristics of breast and ovarian cancer; followed by a description of several aspects of familial and hereditary cancer (e.g. family history and genetic mutations) and of non-genetic risk factors as well as the current clinical practice regarding referral of families to the Family Cancer Clinic, genetic counseling, and risk prevention. This chapter finishes with the aim and outline of this thesis.

Incidence of breast and ovarian cancer

Breast cancer is the most prevalent cancer among women worldwide.\(^1,2\) In the Netherlands approximately 14,000 invasive breast cancer cases are detected each year, of which about 20-25% is detected before the age of 50.\(^3\) The cumulative lifetime risk (CLTR) to develop breast cancer is approximately 12% by age 85 for women in the general population. A positive family history is found in 15-20% of the women with breast cancer, and about 5-10% of all cases is associated with a hereditary mutation.\(^4-7\) The most common high risk mutations associated with the HBOC syndrome are those in the autosomal dominant breast cancer genes 1 and 2 (BRCA1/2), as they account for 1-3% of all the breast cancer cases.\(^8,9\) BRCA1/2 mutation carriers have a CLTR up to 88% to develop breast cancer and also an strongly increased risk to develop ovarian cancer.\(^10-13\) The breast cancer risk of BRCA1/2 mutation carriers starts to increase at an much earlier age as compared to women in the general population. Approximately half of the patients with BRCA1/2 associated breast cancer is diagnosed before their mid- to end-forties. BRCA1/2 mutation carriers also have an increased risk to develop contralateral breast cancer; which risk is approximately 2-3% per year depending on the age of the primary diagnosis.\(^10,14-16\)

Ovarian cancer is less prevalent than breast cancer, but it associated with a worse survival and it is the most lethal gynecologic cancer.\(^3,17\) In the Netherlands every year approximately 1,300 cases are detected, of which 12-16% is detected before the age of 50 years. The CLTR of ovarian cancer for women in the general population is approximately 1.2%.\(^3\) About 10-15% of the ovarian cancer cases are familial, and approximately 5-10% can be attributed to a mutation in the BRCA1/2 genes.\(^18-21\) BRCA1 mutation carriers have a CLTR of ovarian cancer of 30-60%, and BRCA2 mutation carriers of 5-20%.\(^10-13\) Approximately half of the patients with BRCA1/2 associated ovarian cancer is diagnosed before their mid- to end-fifties.
**Tumor characteristics**

Most invasive breast cancers are infiltrating ductal cancers (70-80%). *BRCA1* associated breast cancers most often have a distinct basal-like phenotype, consisting of a higher mitotic index, a higher grade and a triple-negative status (i.e. estrogen receptor (ER), progesterone receptor (PR) and Human Epidermal growth factor Receptor 2 (HER2/neu) negative).\(^{22-26}\) The *BRCA2* associated breast cancers are phenotypically harder to distinguish from sporadic cases.\(^{24, 25, 27, 28}\) They are most often of the luminal type, and are more frequently steroid receptor positive than those associated with a *BRCA1* mutation. *BRCA2* associated breast cancers have a lower proportion of interval cancers, a higher proportion of DCIS, and a significant higher frequency of a favorable tumor size (≤ 2cm) at time of diagnosis compared with *BRCA1* associated breast cancers.\(^{29}\) For *BRCA1* associated breast cancers, the percentage of receptor positive cancers increases with increasing age at breast cancer diagnosis, while in *BRCA2* associated breast cancers the percentage of receptor positive cancers decreases.\(^{30, 31}\)

Ovarian cancer is in about 90% of the cases of epithelial origin, of which many subtypes exist. High-grade serous cancer is the most common (70-80%) and at the same time the most aggressive subtype. *BRCA1/2* associated ovarian cancers are most often of this type.\(^{32, 33}\) No clinicopathological differences are known between *BRCA1* and *BRCA2* associated ovarian carcinoma.\(^{34, 35}\) The site of origin of high-grade serous ovarian cancer is under debate, because ovarian cancer is usually detected in a late stage and precursor lesions are unknown or undetectable. More recently, it has been hypothesized that the fallopian tubes are the site of origin of this ovarian cancer since serous tubal intraepithelial cancer -the first morphological manifestation- has been detected in the tubes.\(^{36-39}\)

**Family history**

About 10-15% of all breast cancer patients has a first-degree relative with breast cancer, and about 15-25% a positive history of breast cancer in first- or second-degree relatives.\(^{4, 40-42}\) Having a first-degree relative with breast cancer increases the risk for breast cancer about 2 times. This risk increase depends on the age of cancer in the relative, and having more affected relatives increases the risk further.\(^{4, 40, 43, 44}\) It is estimated that approximately 30% of the high-risk breast cancer families and 15% of the familial relative risk can be attributed to a pathogenic mutation.\(^7\)

About 15% of all ovarian cancer patients has a first-degree relative with ovarian cancer.\(^{45, 46}\) Having a first-degree relative with ovarian cancer increases the risk for ovarian cancer about 3 times, and having multiple affected relatives increases this risk further.\(^45\) About 25% all familial clustering of ovarian cancers is due to a *BRCA1/2* mutation.\(^{47-49}\) Whether a ovarian cancer patient carries a *BRCA1/2* mutation is not very well predicted by her family history.\(^{41, 46, 50}\)

In *BRCA1/2* mutation families, the cancer risks cannot be contributed completely to the *BRCA1/2* mutation itself.\(^{11, 51}\) Among *BRCA1/2* mutation carriers the breast cancer
risk and ovarian cancer risk increases when the first-degree family history is positive for the same cancer (i.e. breast and ovarian cancer, respectively), however, a family history of the other cancer might decrease the risks.\textsuperscript{51,52}

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**Cancer genetics**

The \textit{BRCA1} and \textit{BRCA2} genes are tumor suppressor genes that contribute to genomic stability through double-stranded DNA repair processes.\textsuperscript{53,54} In the general population the carrier frequency of \textit{BRCA1} and \textit{BRCA2} mutations is estimated to range between 1 in 400 to 1 in 800.\textsuperscript{55,56} Over 5,000 different \textit{BRCA1/2} mutations have been detected to date, and they occur throughout the entire coding region. Apart for these pathogenic mutations, a vast number of mutations has been detected of which the effect is as yet unclassified (variants of unclassified significance, VUS).\textsuperscript{57} The position of the mutation on the \textit{BRCA} gene is statistically significantly correlated to the risk of breast and ovarian cancer, since both ‘breast cancer cluster regions’ and ‘ovarian cancer clusters’ on the \textit{BRCA} genes have been identified.\textsuperscript{58-60} Some specific \textit{BRCA1/2} mutations are more common in populations of a defined geographical or ethnic background (i.e. founder mutations). The \textit{BRCA1/2} carrier frequency can be higher in some populations, such as the Ashkenazi Jewish population.\textsuperscript{61,62} Besides the \textit{BRCA} mutations, other moderate and high risk gene mutations are associated with an increased breast and/or ovarian cancer risk. For breast cancer these are mutations in the genes \textit{PTEN} (Cowden /\textit{PTEN} Hamartoma Tumor Syndrome), \textit{TP53} (Li- Fraumeni syndrome), \textit{NF1} (neurofibromatosis type 1), \textit{MEN1} (Multiple Endocrine Neoplasia type 1), \textit{STK11} (Peutz-Jeghers syndrome), \textit{PALB2} and \textit{CHEK2 1100delC}.\textsuperscript{63-65} For ovarian cancer most of the remaining hereditary ovarian cancer cases are associated with \textit{MSH2}, \textit{MLH1}, \textit{MSH6} and \textit{PMS2} (Lynch syndrome) and \textit{STK11} (Peutz-Jeghers syndrome).\textsuperscript{66} Also common low risk variants, single nucleotide polymorphisms (SNPs), are associated with breast and ovarian cancer. As research on these SNPs is ongoing, the number of SNPs identified is growing and interactions with genetic and non-genetic factors are being identified.\textsuperscript{67,68} Some but not all SNPs validated in the general population are confirmed as risk modifiers in \textit{BRCA1/2} mutations carriers.\textsuperscript{69-75} Therefore, the future use of SNPs in risk prediction models is complicated, and its use might also depend on the population of interest.

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**Risk assessment**

Since the discovery of the \textit{BRCA1} and \textit{BRCA2} genes in the early nineties, multiple studies have been conducted to assess the breast and ovarian cancer risks for mutation carriers, and the probability of carrying a \textit{BRCA1/2} mutation. As a result, multiple risk estimations -both absolute and relative cancer risks- and risk assessment tools are available. Over time, several study designs have been applied to estimate cancer risks, such as case-control studies, kin-cohort studies, retrospective cohort studies and more
recently, prospective cohort studies. The ascertainment of the study population is one of the most important elements when it comes to the estimation of cancer risks. Study populations can roughly be divided in population-based or clinic-based, of which the latter comprise a more selected population based on referral criteria for genetic testing, i.e. one’s personal and family history of cancer. The family history of cancer is a representation of the shared physical environments, lifestyles and genes affecting one’s personal cancer risk. Therefore, all risk assessment tools assessing the cancer risk make use of the family history of cancer, though it depends on the type of tool whether besides breast cancer, also bilateral breast cancer, male breast cancer, ovarian cancer and their ages at diagnosis are considered.\textsuperscript{45, 76-79} Even when a \textit{BRCA1/2} mutation has been detected in a family, apart from the mutation type, the family history still remains relevant for a more tailored risk estimation.\textsuperscript{51, 52, 80} In addition to family history factors, personal characteristics, demographic, lifestyle, reproductive and hormonal factors can be used for a more tailored risk estimation.\textsuperscript{81-87}

### Genetic counseling

Women, who are suspected of carrying a HBOC associated high-risk mutation on the basis of their personal and/or family history of cancer, are referred to a Family Cancer Clinic for counselling and genetic testing.\textsuperscript{88-90} These referral criteria have changed over time and differ between countries. Currently, in the Netherlands a woman can be referred if her personal or family history meets one of the following criteria: \textsuperscript{63, 91}

<table>
<thead>
<tr>
<th>Single cases</th>
</tr>
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<tbody>
<tr>
<td>▪ A history of breast cancer diagnosed &lt;35 years</td>
</tr>
<tr>
<td>▪ A history of epithelial ovarian/fallopian tube cancer any age</td>
</tr>
<tr>
<td>▪ A history of bilateral/multiple primary breast cancer, first cancer &lt;50 years</td>
</tr>
<tr>
<td>▪ A history of both breast and ovarian cancer</td>
</tr>
<tr>
<td>▪ A history of triple negative breast cancer &lt;50 years</td>
</tr>
<tr>
<td>▪ A history of breast cancer diagnosed in a male at any age</td>
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<table>
<thead>
<tr>
<th>Familial cases</th>
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</thead>
<tbody>
<tr>
<td>▪ 2 first-degree relatives with breast cancer, one &lt;50 years</td>
</tr>
<tr>
<td>▪ ≥3 breast cancer cases (one &lt;50 years)</td>
</tr>
<tr>
<td>▪ Breast cancer &lt;50 years and prostate cancer &lt;60 years</td>
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\textit{Nb. For other genetic predispositions than can be associated with breast and/or ovarian cancer, other referral criteria apply.}

The first person in the family who is tested positive for a \textit{BRCA1/2} mutation is called the index case. This index case is most often the person in the family who was diagnosed with cancer at the youngest age, with bilateral breast cancer or with (breast and) ovarian cancer, or being a male breast cancer patient. Genetic testing in family members of the index case is performed following a cascade approach.
In approximately 90% of the referred families, no BRCA1/2 mutation can be detected as an explanation for the strong familiar history of cancer. The current criteria for referral are not very specific as they are associated with a limited positive predictive value.\(^\text{63, 92, 93}\)

After the discovery of the BRCA1 and BRCA2 genes, it was expected that over time a BRCA3 gene would be discovered that could explain the high risk in the families with an unexplained increased risk. However, it is clear by now that there might not be a BRCA3 gene responsible for this risk increase, but that multiple very rare high-risk genes, multiple common moderate-risk genes and multiple common SNPs in combination with environmental factors are driving this risk.\(^\text{94, 95}\)

**Primary and secondary prevention**

Depending on a woman’s cancer risk, different preventive measures are available. For breast cancer screening women are classified as being at population risk or at a slightly increased risk (<20%), at moderate increased risk (20-30%), at high risk (30-40%) or at very high risk (>40%) of breast cancer. Women at population risk or slightly increased risk can opt for breast cancer screening according to the national breast cancer screening program. In the Netherlands this consists of biennial mammographic screening between age 50-75 years. Women with a moderate increased risk get additional screening with annual mammography between age 40-50, and can subsequently enroll in the national screening program, whereas women with a high risk get clinical breast examination between age 35-60 combined with annual mammography, after which they can enter into the national breast cancer screening program. Women at very high risk, i.e. including BRCA1/2 mutation carriers and their untested FDRs, are offered an intensive screening program with an annual clinical breast examination and an annual MRI between age 25-60 years, to which an annual mammogram is added between age 30-60 years. Hereafter they can either continue with annual mammography which will be performed in a hospital or enter into the national screening program till age 75.\(^\text{88, 89}\)

As an alternative to intensive breast cancer screening, BRCA1/2 mutation carriers can opt for preventive surgery. Prophylactic mastectomy reduces the breast cancer risk with approximately 90%, and the residual risk for breast cancer is lower than 5%.\(^\text{96-98}\)

If a woman is already diagnosed with primary breast cancer, mastectomy of the unaffected breast will substantially reduce the risk of contralateral breast cancer.\(^\text{99, 100}\)

Occult breast cancer, i.e. cancer in the absence of mammographic or physical findings of disease in the breast-at the time of mastectomy is rare.\(^\text{101, 102}\)

Up to 2009, annual screening for ovarian cancer with transvaginal ultrasound and serum tumor marker CA-125 testing was offered to woman at high risk of ovarian cancer. However, after several studies this screening was has stopped due to its proven ineffectiveness: ovarian cancer was not detected in an earlier stage, multiple unnecessary surgeries were performed due to false-positive screening results, and mortality was most likely not reduced.\(^\text{103-106}\)

To reduce the risk of ovarian cancer, women are now advised to undergo a timely risk-reducing salpingo-oophorectomy (RRSO)
before the ovarian cancer risk rises. For \textit{BRCA1} mutation carriers RRSO is offered at the age of 35-40 years, and for \textit{BRCA2} mutation carriers at the age of 40-45 years.\textsuperscript{66,90,107} The residual risk of extraovarial ovarian cancer (peritoneal cancer) is about 1% depending on the age at RRSO. Occult invasive ovarian cancer at the time of RRSO is rare.\textsuperscript{39} RRSO was until recently considered to also significantly reduce the breast cancer risk (up to 50%) when performed before the menopause.\textsuperscript{108} However, the evidence contradicting this strong beneficial side-effect is becoming more substantial.\textsuperscript{16,109-111}

\section*{Thesis aims and outline}

\textbf{Aim}
Currently all female \textit{BRCA1/2} mutation carriers receive counseling on the basis of population-averaged CLTRs, while the decision making regarding screening and risk-reducing surgery is drastic. More accurate, age-specific and personalized risk estimation would be beneficial for the medical professionals and their patients.

The aim of this thesis is to improve the accuracy of the breast and ovarian cancer risk estimates in \textit{BRCA1/2} mutation families, by understanding the reasons for risk variation and assessing the effect of suggested risk modifying factors, especially familial factors, more closely. The implications of these CLTRs for breast cancer screening over age 60 in \textit{BRCA1/2} mutation carriers is evaluated.

\textbf{Outline}
Women in \textit{BRCA1} and \textit{BRCA2} mutation families can be classified according to the type of mutation and their carrier status, which is either mutation positive (proven by genetic testing or obligate carrier), proven mutation negative or untested. For proven non-carriers it is under debate whether they are still at increased risk as compared to women in the general population. Chapter 2 discusses the breast and ovarian cancer risk of proven non-carriers in clinic-based \textit{BRCA1/2} mutation families.

For \textit{BRCA1/2} mutation carriers the risk estimates and the reason for the variation in the risk estimates is a topic of debate. For the Northern Netherlands relatively high risks were observed for mutation carriers at older age. Was this a true phenomenon or was it due to bias of differences in methodology? In chapter 3 the breast cancer risk of \textit{BRCA1/2} mutation carriers in the Northern-Netherlands is compared with the rest of the Netherlands using similarly ascertained populations and one method for risk estimation. As methodological differences seemed to impact the CLTRs, in chapter 4 the effect of different methods of risk assessment and bias correction is assessed by applying the different methods to one large clinic-based cohort of \textit{BRCA1/2} mutation carriers.

Many genetic and environmental factors influence the CLTR of breast and ovarian cancer. In population-averaged risk estimates these factors are usually not taken into account, but for the development of more personalized risk estimations the relative risks are of interest. For \textit{BRCA1/2} mutation carriers it is known that the breast cancer risk increases in more recent the birth cohorts, most probably as a consequence of
changing lifestyles and reproductive factors. The existence of such a trend in the ovarian cancer risk in \textit{BRCA1/2} mutation carriers and their background population is discussed in \textbf{chapter 5}. Some risk factors are family-specific or have a high correlation within a family, such as the family history of cancer and type of \textit{BRCA1/2} mutation. The parental origin of a \textit{BRCA} mutation is also such a factor that may affect CLTRs in offspring. In \textbf{chapter 6} it is discussed whether the parental origin of the \textit{BRCA1/2} mutation has an effect on the breast cancer risk of the offspring, when corrected for ascertainment bias.

Breast cancer screening is tailored to a woman’s breast cancer risk classification, as mentioned above. \textit{BRCA1/2} mutation carriers are at very high risk of breast cancer and –when they do not choose for preventive mastectomy- receive intensive screening up to at least age 60. As the breast cancer risk continues to increase after this age, the relevance and necessity of additional screening beyond age 60 is investigated and discussed in \textbf{chapter 7}.

Finally, \textbf{chapter 8} contains a summary and general discussion on the most important results of this thesis.
References


62. Finkelman BS, Rubinstein WS, Friedman S, Friebel TM, Dubitsky S, Schonberger NS,


