Receptor Status in Recurrent Breast Cancer –

A Retrospective Study

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Abbreviations

ER - Oestrogen receptor

PgR - Progesterone receptor

HER2 - Human Epidermal growth factor type 2 receptor

WITS - University of the Witwatersrand

CMAH - Charlotte Maxeke Johannesburg Academic Hospital

HJH - Helen Joseph Hospital

IHC - Immuno-Histochemistry

FISH - Fluorescent in Situ Hybridisation

NHLS - National health laboratory system

SEER -Surveillance, Epidemiology, and End Results Program

Abstract

Background: Breast cancer recurrence is a major clinical event and represents a principal cause of breast cancer related death. A discordance rate between receptor status of primary and matched recurrence tumours has been reported in the literature but the extent of this in our population is unknown. Repeating Immuno-histochemistry (IHC) and fluorescent in-situ hybridization (FISH) studies have financial and workforce implications in a resource-constrained environment. However, the results of these receptor studies have prognostic implications. Therefore it is important to determine the extent of change in receptors in the recurrence.

Aim: To compare the hormone receptor profile between breast cancer primary and matched loco-regional recurrence and to ascertain the extent of receptor discordance.

Methods: All patients who presented to the respective breast care facilities for breast cancer recurrences between 2006 and 2014 were identified using the mammography department records. The specimens for each patient were scrutinized. Oestrogen receptor (ER) and progesterone receptor (PgR) status as well as the Human Epidermal growth factor type 2 receptor (HER2) receptor statuses were noted for each patient and a comparison was made between primary and matching recurrence, with loss and gain being noted.

Results: In the analysis, significant discordance was found for matching hormone receptor status. Discordance in oestrogen receptor status occurred in 14.3% of cases: change occurred both from ER-positive to -negative and vice versa. For progesterone receptor status this occurred in 25.7% of cases. A discordance of 14.8% was noted for HER2 receptor status. These results are not dissimilar to what has been previously reported in the literature. Of note, adverse receptor discordance: positive to negative was noted in a total of 19 receptors (ER 4; PgR 11; HER2 4)

Conclusion: These results confirm the phenomenon of receptor discordance between breast cancer primary and recurrence. The results support the necessity of confirming receptor status on all loco-regional recurrent disease. This reinforces the importance of obtaining a confirmatory biopsy in patients where recurrence is suspected and therefore allowing the appropriate targeted therapy to be selected.

Ethics clearance number M150771

Candidate's declaration

I, Zain Ally, declare that this dissertation is my own work. It is being submitted for the degree of Master in Medicine in the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at this or any other university on this day the March 2017.

Dedication

This dissertation is dedicated to Kimberly Vardy

Chapter 1

Introduction

In South Africa, according to the 2005 cancer registry breast cancer is the most commonly diagnosed cancer among women, with a lifetime risk of 1:33.

Breast cancer recurrence is a serious event and represents a cause of breast cancer related death (1). 20% to 30% of the patients with early breast cancer will eventually relapse (2). In particular, breast cancer patients with loco-regional recurrence have been found to have 5-year disease-free survival rates of 13%–37% and overall survival rates of 21%–50% respectively (3-5).

Women, regardless of their racial or ethnic origin, are at risk of developing breast cancer. Variations in breast carcinoma incidence rates among multicultural populations suggest that etiologic factors differ in their expression and impact on disease outcome (6). The most significant risk factors for breast cancer development are gender (female gender), age and female hormone exposure (7). Gender is the most important factor, with a female to male ratio of 100:1 (7). The incidence of breast cancer increases rapidly after the 4^{th} decade and after menopause the incidence continues to increase but at a much slower rate peaking in the 5^{th} and 6^{th} decades and then slowly levelling off (7). Family history of breast cancer is known to increase the risk of breast cancer. The overall risk depends on the number of relatives, their ages at diagnosis and whether the disease was unilateral or bilateral (7).

A pattern of recurrence between different breast cancer subtypes has been suggested, and it appears that ER-negative breast cancers are associated with a higher risk of recurrence during the initial 5 years after diagnosis, compared to ER-positive breast cancers (8,9).

Apart from hormone receptor status other risk factors associated with loco-regional recurrence include the size of tumour, margin involvement at excision, axillary lymph node status and the nuclear grade and differentiation of the tumour.(10)

The mainstay of treatment for breast cancer is the complete surgical resection with negative margins (11). Apart from surgical resection, adjuvant treatment modalities are used to treat micro-metastatic disease and are aimed at reducing the risk of recurrence, and reducing breast cancer-related morbidity and mortality (12, 13). Adjuvant treatment modalities include radiation, cytotoxic therapy and treatment targeting receptors within cancer cells.

In the case of hormonal treatment, knowledge of the oestrogen(ER) and progesterone (PgR) receptor status, as well as the Human Epidermal Growth factor type 2 receptor (HER2) statuses is fundamental to treatment selection for both patients with early stage disease and those with recurrent disease.

In ER-positive early-stage breast cancer, hormone therapy plays a major role in adjuvant treatment, either alone or in combination with chemotherapy. Hormone treatments function to decrease oestrogen's ability to stimulate existing micro-metastases or dormant cancer cells (14).

In the past, the receptor profile of the primary was a guide to both 1st and to 2nd line therapies. For the recurrence recent studies have demonstrated that discordance may exist between primary and matched recurrence so that therapy chosen for the primary disease may be inappropriate for the recurrence.

The study by Lindstrom et al demonstrated discordance between the receptor status of primary and matched recurrence of 32.4%, 40.7%, and 14.5% for ER, PgR and HER2 respectively (15).

This finding may mean that a primary tumour which was initially sensitive to hormonal treatment may develop a recurrence which lacks hormone receptors and therefore rule out hormonal therapy as a treatment option. Conversely, if a recurrence "switched" from receptor negative to receptor positive this would justify the use of adjuvant anti-oestrogen therapy.

Another important observation regarding receptor change was noted by Dieci et Al (16). These authors demonstrated that patients who experienced loss of receptor sensitivity at recurrence experienced both shorter post recurrence survival and overall survival when compared with concordant receptor profiles.

Tissue confirmation of recurrent breast cancer is recommended by international guidelines (17). Preliminary data indicates that the change of the molecular subtype in the recurrent breast cancer can prompt clinicians to alter the treatment choice in up to 14% of cases (18, 19-21).

The aim of this study is to compare the hormone receptor status between breast cancer primary and matched loco-regional recurrence in South African women treated at two breast centres associated with the University of the Witwatersrand over eight years. The outcomes relating to a change in the receptor profile of the recurrent cancer are noted and will be discussed.

Chapter 2

Methods

Ethical approval to undertake the study was obtained from the University of the Witwatersrand Human Research Ethics Committee, ethics clearance number M150771 (Appendix 1).

All patients attending the mammogram facilities at Charlotte Maxeke Johannesburg Academic Hospital (CMAH) and Helen Joseph Hospital (HJH) are entered into a filing system with a brief history, clinical description and the reason for investigation being noted on a request page. If a suspicious lesion is found on mammogram and ultrasound, a mandatory biopsy is performed and a biopsy/specimen number is attached to the patient request page. All these request pages were checked, beginning in 2006, and all patients with a history or suspicion of a clinical recurrence and subsequent biopsy were selected. Based on the four months spent working at the Helen Joseph Breast ward an estimate of between 40 and 70 patients will be accrued. This is in keeping with the incidence of between 2 and 5% loco-regional recurrence within 5 years. 2874 patients were identified at both centres over the 8 years.

All breast biopsies are done via image guidance, which meant that there should be no side room/examination room breast mass biopsies performed. If a suspicious mass is found at examination, the patient is referred for imaging first and then a biopsy is performed by the mammography department.

This process will identify patients with loco-regional recurrence as the patients may re-present to the breast centres with a breast or an axillary symptom and then referred to the mammography department. Patients with distant recurrence may present to other disciplines for their symptoms. A pleural effusion might manifest as shortness of breath and be managed by the Pulmonology department; similarly a patient with bone metastases might present with back pain and therefore be investigated and managed by the orthopaedic department and initially bypass the breast centres.

The specimen number and hospital number were then cross checked on the National health laboratory system (NHLS) pathology system to ascertain whether recurrence was confirmed and whether hormone receptor status was available for matching primary and recurrence.

Only the receptor status of matching primary and recurrence were recorded on a data sheet (Appendix 2) and numbered chronologically. Patient's personal details were not recorded on the data sheet and in the study.

Those patients in whom hormone receptor status could not be confirmed for both primary and recurrence were not included in the study. 3 patients were recorded as having breast cancer recurrences but no histology of the primary tumour could be found.

Oestrogen receptor(ER) status, progesterone receptor (PgR) status and human epidermal growth factor type 2 (HER2) statuses were noted for each patient and a comparison was made between primary, and recurrence, with loss and gain being noted.

Oestrogen and progesterone receptors were tested for by means of immune-histochemistry (IHC) at primary and recurrence. The intensity of the hormone receptor status was not taken into account. Any positive receptor result, weak or strongly positive was noted as positive, similarly with the negative results, this was done because not all results identified intensity or strength and were simply labelled positive or negative.

All HER2 receptors were tested by means of IHC, with results being labelled as negative (0 or 1+), equivocal (2+) and positive (3+). Those results which were equivocal would be subject to further testing by Fluorescent in Situ Hybridisation (FISH) to ascertain the true status. HER2 results which were equivocal and not submitted for gene copy analysis (FISH) were excluded. Due to the high expense of further testing, the additional testing might not have been undertaken especially during the period when Trastuzumab was not readily available in the State sector.

Statistical analysis

Receptor status was compared for each matched primary and recurrence.

A change in receptor status was noted as discordance and an overall percentage of discordance was calculated. Details of change of receptor status were then documented for each receptor, noting a change from positive to negative, and vice versa.

Chapter 3

Results

Between 2006 and 2014, a total of 70 patients were found to fit the inclusion criteria. Of the 70 patients, 1 patient did not have the HER2 receptor assessed in the primary breast cancer leaving 69 HER2 receptors available for assessment.

The oestrogen and progesterone hormone receptor status was assessed using IHC and was reported as being either weakly or strongly positive or negative. The intensity of each was not taken into account and was recorded only as either positive or negative.

Table 1: Oestrogen receptor results

Oestrogen Receptor Status	Number
Total number assessed	70
ER Positive Primary	45(64.2%)
ER Negative Primary	25(35.7%)
ER Loss (proportion of Positive Primary)	4(8.9%)
ER Gain (proportion of Negative Primary)	6(24.0%)
Total Discordance	10(14.3%)

A total of 70 Oestrogen receptors were assessed and it was found that 45 were positive and 25 were negative in the primary cancer. A total discordance in oestrogen status was observed in 10 cases (14.3%). Among the 45 positive primary receptors, 4 switched to negative at recurrence (8.9%) and 41 maintained positivity. 6 of the 25(24%) negative primary receptors acquired positivity at recurrence.

Table 2: Progesterone receptor results

Progesterone Receptor Status	Number
Total number assessed	70
PgR Positive Primary	34(48.6%)
PgR Negative Primary	36(51.4%)
PgR Loss (proportion of Positive Primary)	11(32.3%)
PgR Gain (proportion of Negative Primary)	7(19.4%)
Total Discordance	18(25.7%)

With regards to progesterone receptor status, 70 patients were included without exclusions and a total of 70 receptors were assessed, 34 positive primary receptors were noted and 36 negative primary receptors were noted. 11(32.3%) receptors "switched" from positive to negative at recurrence and 7(19.4%) switched from negative to positive at recurrence, with a total discordance of 18(25.7%) receptors.

It was found that of the 69 patients with available HER2 receptors, 18 patients had equivocal HER2 results (2+) in either the primary or in the recurrence and only 3 equivocal results were submitted for further testing. Therefore 15 had to be excluded because FISH testing was not performed and the true status could not be ascertained. After exclusion the total number of patients assessed for HER2 discordance was 54.

Table 3: Human epidermal growth factor type 2 receptor results

HER2 Receptor status	Number
Total number assessed	54
Excluded	15
Her2 Positive Primary	12(22.2%)
Her2 Negative Primary	42(77.8%)
Her2 Loss(proportion of Positive Primary)	4(33.3%)
Her2 Gain(proportion of Negative Primary)	4(9.5%)
Total Discordance	8(14.8%)

HER2 positive : 3+ HER2 negative : 1+

It was found that 12 receptors were HER2 positive at primary and 42 receptors were HER2 negative. Of the 3 that were submitted for FISH testing, 2 were concordant and 1 discordant. A change in receptor status was noted in 8 receptors (14.8%) with 4(33.3%) expressing a change from positive to negative and 4(9.5%) from negative to positive.

In summary, the current study showed discordance in 10 ER receptors, 18 PgR receptors and 8 HER2 receptors.

Chapter 4

Discussion

The recurrence of breast cancer is a life threatening event. Unless the recurrent disease is managed appropriately, the risk of breast cancer related mortality will increase. The treatment of breast cancer recurrence, in addition to local modalities such as surgery and radiation, requires systemic therapy. The latter includes cytotoxic drugs and therapies targeted to receptors within cancer cells. Of concern are the findings, widely reported in the literature, that the receptor profile of the primary cancer may have changed in the recurrence. This has serious implications for the selection of systemic treatment for the recurrent disease.

Receptor Discordance

In this study, it was found that of the 70 receptors assessed, there was a discordance of 10 (14.3%) oestrogen receptors (ER). Of the 10 cases which were found to be discordant, 6 expressed cytogenetic gain and 4 lost receptor sensitivity. These results are in keeping with available literature regarding ER discordance. A study by Dieci et al showed a discordance in ER receptors of 13.4% in 119 cases (16), similarly Broom et al showed that the discordance of ER receptors in their study of 100 patients was 17.7 %(22). A study performed by Nishimura et al showed that discordance in ER receptor did exist but to a lesser degree, with 10.3% of 97 patients (23). A meta-analysis of 48 articles performed by Aurelio et al showed a pooled discordance of ER receptor to be 20%. (24).

The progesterone receptor has been the focus of investigation and its importance in regulation and modulation has been highlighted. A recent study published by Mohammed et al has described the role that progesterone may play. The study states that progesterone receptor expression is a biomarker of oestrogen receptor-α (ERα) function which in turn modulates tumour behaviour and is associated with a good clinical outcome. This is due to an increased sensitivity of the tumour to endocrine agents such as Tamoxifen and the aromatase inhibitors (25). Consequently, tumours which display PgR receptor negativity or a negative discordance at recurrence are less susceptible to anti-oestrogen therapy because of the lack of ERα and are therefore at risk of worse outcomes (25).

Of the 70 PgR receptors assessed in this study, a discordance of 18 (25.7%) was noted, with 11 receptors losing sensitivity at recurrence and 7 expressing receptor gain at recurrence. This is not out of keeping with relevant literature. In the same study quoted earlier, Nishimura et al showed 22% discordance in PgR (23). Higher discordance was noted in other studies. Broom et al and Aurelio et al both recorded discordance of 33.3 % in PgR receptors (22, 24). Higher rates of discordance was shown by Dieci et al and Karlsson et al, with rates of 39% and 40.7% respectively (16, 26).

During this study, difficulties were encountered during assessment of the HER2 receptors. 1 of 70 patients did not have a HER2 result for the primary cancer. Of the remaining 69 cases, 18 results were equivocal at either primary or at recurrence. 3 of the 18 were submitted for further testing by in-situ hybridisation, 2 confirming concordant results and 1 discordant result. 15 cases were not submitted for in-situ hybridisation and therefore the results could not be confirmed and were excluded. The reason for this is uncertain but the extra cost of further testing may underlie this omission.

Of the remaining 54 receptors, discordance was noted in 8 (14.8%) with 4 losing receptor sensitivity and 4 gaining sensitivity. Very similar to this result is that of Broom et al, who showed discordance in HER2 receptor status of 14.5% (22). Many other studies have displayed similar rates of discordance. Zidan et al, and Dieci et al showed discordance of HER2 to be 14% and 11.8% respectively (27, 16). Gangcberg et al exclusively used FISH testing results to assess HER2 status and showed a discordance of 7% in their study, similarly the meta-analysis performed by Aurelio et al showed a discordance of 8% for HER2 receptor status (28, 24). Even though 15 patients were excluded, the results of this study are quite comparable to the results found in relevant literature. If the equivocal results were submitted for confirmatory FISH testing, the accuracy of the results would be improved.

Mechanisms of receptor change

Receptor discordance may be the result of a genuine change in the bio-characteristics of the tumour. This is postulated to be due to clonal selection of a less differentiated receptor during the metastatic process (29). The discordance may also result from intra-tumour heterogeneity, or possibly due to a lack of reproducibility and inaccuracy of the testing methods and assays used.

Tumour heterogeneity describes a cancer with different tumour cell populations in the same patient and represents an unavoidable factor for accurate HER2 assessment (30). There are two main types of HER2 tumour heterogeneity, clustered HER2 amplification and mixed HER2 amplification. This disparity makes it difficult to accurately assess HER2 status and has led investigators to conceptualise and develop new novel methods to assess HER2 status with increasing accuracy (30).

By definition, concordance of two results is the product of the accuracy of one testing method multiplied by the accuracy of another (31). Therefore, a 100% accurate test that is correct 100% of the time will have a 100% concordance. Similarly a 90% accurate test that correctly identifies the true receptor status 90% of the time would yield an 81% (0.9 x 0.9) concordance when repeated a second time on the same cases. This means 19% percent of receptor profiles could be discordant by chance alone. This illustrates how vital the accuracy and reproducibility of the testing methods and assays are.

The study by Pusztai et al showed that differences in tissue fixation, antigen retrieval, and staining methods all contribute to variation in results. The subjective scoring of results dependant on the intensity of a stain by different pathologists also led to less than perfect inter-observer reproducibility (31).

Significance of Change

Whether discordance represents a real change in tumour biology or a laboratory error, the results of the tests will have management implications for the patients undergoing testing. A "switch" from negative hormone receptors in the primary to positive receptor status in the recurrence may afford the patient the use of valuable adjuvant hormonal therapy. Conversely a "switch" from a receptor positive primary to a receptor negative recurrence identifies patients who are at an increased risk of unfavourable outcomes because hormonal therapy would not be indicated as adjuvant therapy and it also suggests that, due to the selection of aggressive tumour cell clones, the recurrence is more aggressive than the primary. This is supported by Lower et al which showed that patients with tumours that switched from a positive primary to a negative recurrence experienced significantly shorter median survival {669 days, $p < 0.05$} (32). Similarly Dieci et al during their study showed that the same positive to negative "switch", translated into a worsened overall survival and prognosis compared with the corresponding concordant-positive cases (16).

Discordance may also identify those patients, when the primary cancer was falsely negative, and who then presented with a recurrence found to be receptor positive. This would mean that the patient missed the opportunity of receiving hormonal therapy between the two events. One method of improving testing accuracy is to use the Bayesian misclassification correction method. Sighoko et al applied this method to a data set of 35000 patients from the SEER database and found that 34% of ER "conversion" and 17% of PgR "conversion" was due to technical misclassification (33). Another method of improving the accuracy of results may lie in the standardization of receptor profiling within a central laboratory (32).

The results of this study were consistent with results described in literature (15, 16, 22, 23). The greater part of discordance was due to a receptor loss, chiefly the progesterone receptors. This would therefore identify patients at risk of worse outcomes. Where the treatment is unnecessary, the side effects of that treatment would be avoided.

Conversely, those cases where there is cytogenetic gain of receptor sensitivity will benefit from the addition of targeted treatments.

The aim of personalized medicine is to select therapy based on the basis of the molecular and genomic features of each individual tumour (34, 35).For this reason, re-profiling recurrent breast cancer assumes a major role.

Central laboratories where measurements of hormone receptors and HER2 receptors follow the guidelines established by the American Society of Clinical Oncology and the American Society of Pathology add greater value to check the accuracy of assays done in peripheral laboratories (36). These centres are mostly only available in the well resourced Western centres (36).

Conclusion

These results support the phenomenon of receptor discordance between breast cancer primary and its matched recurrence. It emphasizes the necessity of reassessing receptor status on all recurrent disease to identify discordance and facilitate appropriate systemic treatments.

There seems to be many factors at various levels which may play a role in receptor discordance. By creating a standard format of sampling and testing, patients who undergo true biologic discordance would be identified and this would also limit the amount of discordance associated with inaccurate sampling and testing methods.

Chapter 5

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Appendix 1 : Copy of ethics clearance certificate

R14/49 Dr Zain Ally

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)

CLEARANCE CERTIFICATE NO. M150771

NAME: (Principal Investigator)	Dr Zain Ally
DEPARTMENT:	General Surgery Charlotte Maxeke Johannesburg Academic Hospital and Helen Joseph Hospital
PROJECT TITLE:	Receptor Status In Recurrent Breast Cancer A Retrospective Study
DATE CONSIDERED:	31 July 2015
DECISION:	Approved unconditionally
CONDITIONS:	
SUPERVISOR:	Prof A. Mannell
APPROVED BY:	Professor Angela Woodiwiss, Co-Chairperson, HREC (Medical)
DATE OF APPROVAL:	31/07/2015

This clearance certificate is valid for 5 years from date of approval. Extension may be applied for.

DECLARATION OF INVESTIGATORS

To be completed in duplicate and **ONE COPY** returned to the Secretary in Room 10004, 10th floor, Senate House, University.

I/we fully understand the conditions under which I am/we are authorized to carry out the above-mentioned research and I/we undertake to ensure compliance with these conditions. Should any departure be contemplated, from the research protocol as approved, I/we undertake to resubmit the application to the Committee. **I agree to submit a yearly progress report**

Principal Investigator Signature _____ Date _____

PLEASE QUOTE THE PROTOCOL NUMBER IN ALL ENQUIRIES

Appendix 2 : Data sheet

Patient Number	Primary ER status	Recurrent ER Status	Primary PgR status	Recurrent PgR Status	Primary HER2 Status	Recurrent HER2 Status
01						
02						
03						
04						
05						

Appendix 3 : Research Protocol

Receptor Status in recurrent breast cancer – A retrospective study

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1. Rationale

Breast cancer recurrence is a major clinical manifestation and represents a principal cause of breast cancer related death. The advent of hormone receptor sampling has significantly affected the management of breast cancer. The presence of Oestrogen(ER) and Progesterone (PgR) receptor positivity in breast cancers have been associated with improved prognosis. Receptor screening is performed on all biopsy specimens at initial presentation to determine sensitivity and in the event of recurrence to confirm sensitivity.

This study aims to illustrate the rate of hormone receptor sensitivity discordance in patients with a loco-regional breast cancer recurrence.

2. Introduction

2.1. Background

Breast cancer is the second most common cancer diagnosed worldwide with 1.67 million new cancer cases diagnosed in 2012 (25% of all cancers) (1).

In South Africa, according to the 2005 cancer registry breast cancer is the most commonly diagnosed cancer among women, with a lifetime risk of 1:33.

Breast cancer ranks second as a cause of cancer death in women (after lung cancer), and from 2005 to 2009, rates decreased 3.0% per year in women younger than 50 and 2.0% per year in women 50 and older. The decrease in death rates represents progress in earlier detection, improved treatment, and possibly decreased incidence as a result of declining use of hormonal therapy (3, 4).

The 5-year relative survival rate for female invasive breast cancer patients has improved from 75% in the mid-1970s to 90% today (5).

2.2. Risk factors

Women, regardless of their racial or ethnic origin, are at risk of developing breast cancer. Variations in breast carcinoma incidence rates among multicultural populations suggest that etiologic factors differ in their expression and impact on disease outcome (6).

Key among those factors that affect breast carcinoma development is the roles of genetics and the environment, the reproductive experience and the effects of endogenous and exogenous hormones.

The most significant risk factors for breast cancer development are gender (female gender), age and female hormone exposure (7). Gender is the most important factor, with a female to male ratio of 100:1 (7). The incidence of breast cancer increases rapidly after the 4^{th} decade and after menopause the incidence continues to increase but at a much slower rate peaking in the 5^{th} and 6^{th} decades and then slowly levelling off (7).

Family history of breast cancer is known to increase the risk of breast cancer. The overall risk depends on the number of relatives, their ages at diagnosis and whether the disease was unilateral or

bilateral (7). The highest risk is a young first degree relative with bilateral disease. About 5-10% of breast cancers can be linked to genetic mutations inherited from one's mother or father (8). Women with a BRCA1 mutation have a 55-65% risk of developing breast cancer before age 70 and women with a BRCA2 mutation, have a risk of 45% (9).

Other risk factors implicated in breast cancer development include hormone exposure, whether endogenous or exogenous, age of menarche, age at initiation of regular menstrual cycles, age at first pregnancy and age of menopause all contribute to developing breast cancer. During the Women's Health Initiative, exogenous hormone exposure, in the form of menopausal hormonal therapy was shown to increase the risk of developing breast cancer over a 5 year period by 26% (10, 11)

2.3. Recurrence

A pattern of recurrence between different breast cancer subtypes has been suggested, and it appears that ER-negative breast cancers are associated with a higher risk of recurrence during the initial 5 years after diagnosis, compared to ER-positive breast cancers (12, 13). Thereafter, the risk of recurrence steadily increases in ER-positive breast cancers for the next 10 years, and at 15 years following diagnosis, the risk appears to be equal for both sub-types(14). The triple negative cancers are generally associated with a high risk of recurrence with a particularly high risk of distant metastases, compared to receptor positive tumours (15). Apart from hormone receptor status other risk factors associated with loco-regional recurrence include the size of tumour, margin involvement at excision, axillary lymph node status and the nuclear grade and differentiation of the tumour.(16)

2.4. Treatment

The goals of breast cancer surgery include the complete resection of the primary tumour, with negative margins, to reduce the risk of local recurrences, and pathologic staging of the tumour & axillary lymph nodes to provide necessary prognostic information. Adjuvant treatment for breast cancer involves radiation and systemic therapy (chemotherapeutic, hormonal and biologic agents). Adjuvant treatment is designed to treat micro-metastatic disease and is aimed at reducing the risk of future recurrence, thereby reducing breast cancer-related morbidity and mortality. Knowledge of the oestrogen receptor(ER), progesterone receptor(PgR) and the Human Epidermal Growth factor type 2 receptor(HER)2 status is fundamental for treatment selection among not only patients with early stage disease but also for those with metastatic disease. In ER-positive early-stage breast cancer, hormone therapy plays a major role in adjuvant treatment, either alone or in combination with chemotherapy. Hormone treatments function to decrease oestrogen's ability to stimulate existing micro-metastases or dormant cancer cells (17).

A recent study by Lindstrom et al has demonstrated a discordance rate between receptor status between primary and recurrences of 32.4%, 40.7%, and 14.5% for ER, PgR and HER2 respectively (18).

This change in receptor status translates into a worsened prognosis if the receptor has changed negatively or an improved prognosis if receptor change is positive (19). With such a large discordance rate between primary and recurrence, it would seem prudent for all possible breast cancer recurrence

to be re-staged and evaluated on its own merit and not treated or managed as its matched primary tumour. This may potentially lead to an improved survival and outcome.

The basis of this study is to examine the discordance rate in hormone receptor status between matched primary and recurrent breast cancers within 2 Gauteng based breast units.

2.5. This project in context: Comparison to literature on the topic

Breast cancer management is guided by hormone status

20% to 30% of the patients with early breast cancer will eventually relapse (20). In particular, breast cancer patients with loco-regional recurrence have been found to have 5-year disease-free survival rates of 13%–37% and overall survival rates of 21%–50% respectively (21-23).

Hormone therapy remains the mainstay for hormone receptor–positive breast cancer. The decision for hormone treatment has traditionally relied on assessment of ER and PgR status of primary tumours, with the response generally related directly to ER and PgR content. Patients with ER– and PgR– tumours show virtually no response, whereas patients with breast cancers positive for both hormone receptors demonstrate a significantly higher response (24).

Results and studies have shown that cancer receptors change between primary and recurrence

Broom et al described significant discordance in receptor status between primary and recurrent breast pathology samples. A change in ER status occurred in 17.7% of patients (switching occurred both from ER-positive to -negative and vice versa) and for PgR it occurred in 37.3% of patients (all tumours lost PgR). This meant that a total of 45.1% of patients had some sort of hormone receptor change (25).

Addition of hormone therapy increases survival and if hormone negative, has a worse prognosis

Dieci et al. highlighted the prognostic effect of discordance between matched primary breast cancer and recurrence. Within the discordant group, a loss of a receptor expression rather than gain resulted as the main determinant of poor prognosis (18). In fact, patients who changed their tumour phenotype to triple negative by losing hormone receptor and/or HER2 expression experienced the shortest post recurrence survival and overall survival when compared with concordant cases (18). Moreover, when the impact of single-receptor change was evaluated, ER loss, PgR loss and HER2 loss was associated with a worse post recurrence survival and overall survival (consistently with previous large retrospective reports) (4,26-30) Also ER-positive/PgR-negative metastatic tumours tend to have a more aggressive course and are associated with a reduced overall survival compared to those retaining PR (4, 28). As a result it has been recommended that hormone receptor and HER2 status should be determined for recurrences, any change identified may enlarge treatment possibilities for the patient and improve selection for targeted therapies. This is particularly important for loco-regional recurrence after mastectomy, which carries an unfavourable prognosis with high risk of distant metastases (31).

3. Study Objectives

- To evaluate hormone receptor status in primary and recurrent loco-regional breast cancer patients in 2 Johannesburg specialist breast cancer centres.
- To compare hormone receptor status between matched breast cancer primary and loco-regional recurrence.

4. Methods

Design : cross sectional, descriptive, observational study.

Site of study : Helen Joseph hospital and Charlotte Maxeke Academic hospital

Study population: All patients who presented to the above mentioned breast clinics with histologically proven loco-regional breast cancer recurrences.

Sampling: Sampling will not be needed as the entire population will be included (mentioned above). The database at Helen Joseph begins in 2006, therefore patient results will be accumulated from then till 2014 for both hospitals. Based on the four months spent working at the Helen Joseph Breast ward an estimate of between 40 and 70 patients will be accrued. . The data will be obtained retrospectively from the patient files. The National Health Laboratory Service database will be used to confirm histology results.

Inclusion criteria : All patients at the above mentioned breast units with histologically confirmed loco-regional breast cancer recurrence, that have available hormone receptor sensitivity for both primary and recurrence will be included in this study.

Exclusion criteria: Patients with NHLS data and identification data which cannot be reconciled will be excluded.

Measuring tool: A data collection sheet detailing the hormone receptor status of matching primary and recurrence for every corresponding patient will be used and labelled addendum A.

Data collection: The mammography departments at both hospitals keep records of every patient who has undergone a breast biopsy. All patients detailed to have had a repeat biopsy for a possible recurrence were identified and added to a list. The histology results of all patients on the list were checked and those with available hormone receptor results were earmarked. The hormone receptor status for all primary and recurrent breast cancer as well as the corresponding patient number will be noted and entered onto the data sheet. A comparison can then be made between primary and recurrent receptor status.

5. Data analysis and statistics

The study is purely descriptive; hence pie charts and tables will be used to graphically illustrate the results of the study. Because no comparisons will be made, no tests will be necessary.

6. Ethics

Ethics clearance number M150771

6.1. Data safety

Apart from histology results, no other personal details will be recorded and patients will remain anonymous for the duration of the study.

7. Budget

Funding will not be needed as the study is retrospective in nature and will only require access to the various databases.

8. Anticipated problems

None.

9. Timing

Month of the Year	Jan	Feb	March	April	May-July	Aug-Oct	Nov-March	April	May-June
Literature search									
Reading literature									
Summarising literature									
Preparing Protocol									
Protocol Assessment									
Ethics application									
Collecting data									
Data analysis									
Writing up thesis									
Submit: marking									
Writing up paper									

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Data Sheet

Patient Number	Primary ER status	Recurrent ER Status	Primary PgR status	Recurrent PgR Status	Primary HER2 Status	Recurrent HER2 Status
01	+	–	–	–	+	+
02	–	–	–	–	–	–
03	–	+	–	–		
04	+	+	+	–		
05						
06						
07						