



EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Directorate F - Health, Consumers & Reference Materials (Ispra)
Health in Society

European Commission Initiative on Breast Cancer (ECIBC): European guidelines on breast cancer screening and diagnosis

QUESTION	
Should 70 gene signature test vs. no testing be used for patients who have hormone receptor positive, HER2-negative, lymph node negative or up to 3 lymph nodes positive invasive breast cancer to guide the use of chemotherapy (subgroup: high clinical risk)?	
POPULATION:	Patients with hormone receptor positive, HER2 negative-, lymph node negative or up to 3 lymph nodes positive invasive breast cancer at low clinical risk breast cancer as defined in Cardoso 2016 (modified Adjuvant! Online)
INTERVENTION:	70 gene signature test
COMPARISON:	no testing
MAIN OUTCOMES:	Survival without distant metastasis; Disease free-survival; Overall survival;
SETTING:	European Union
PERSPECTIVE:	Population (National Health System)
BACKGROUND:	<p>Hormone receptor positive, HER2-negative invasive breast cancer represents about 70% of breast cancer diagnosed in western countries (3) . About 60% of these have not spread in lymph nodes at time of diagnosis (3).</p> <p>Approximately 15% of these women diagnosed with a hormone receptor positive, HER2-negative, lymph node negative invasive breast cancer will develop recurrence within 10 years with adjuvant endocrine therapy alone. The risk of recurrence could be reduced by the addition of chemotherapy in 30% of these patients, which translates into an absolute benefit in the rate of freedom from recurrence of up to 5 percentage points (4)(5).</p> <p>Therefore, most of the women would be overtreated if all would receive chemotherapy. It is assumed that this is also true for patients with hormone receptor positive, HER2-negative invasive breast cancer with 1-3 positive lymph nodes (2).</p> <p>Prognostic factors that have been used to predict the risk recurrence in these women include clinicopathologic features such as age, tumour size, grade, percentage of ER- and PR-positive cells as well as Ki67 index (6)(7). Although these factors have been shown to discriminate different prognostic groups, they are limited in their ability to predict who might benefit from chemotherapy and who will not. The meta-analysis of Peto et al. (8) suggested that the relative benefit of chemotherapy is only minimally affected by these conventional clinicopathological parameters. In consequence, adjuvant polychemotherapy was recommended to many patients with ER-/PR-positive, HER2-negative early breast cancer in the past.</p>

	<p>In the last 15 years different multigene tests have been developed to stratify patients with early breast cancer in different risk groups by analysing the activity of various genes. Although these tests analyse different genes and use various techniques (RT-PCR, microarray, etc), they have in common, that they mainly focus on genes involved in cell proliferation. The tests provide an individual risk profile of a breast cancer patient, whereby some of them combine the molecular result with clinicopathological features like tumour size and nodal status to improve the prediction of recurrence risk.</p> <p>The goal to use these tests has changed over time. Initially, studies aimed to identify patients with “favourable” clinical-pathological characteristics who might benefit from the addition of chemotherapy (9)(10). Over time, interest has also grown to identify those patients who gain little or none from chemotherapy and who may safely avoid toxicities (11)(1)(2) , not least because the introduction of mammography screening has increased the proportion of prognostically favourable breast carcinomas in many countries.</p> <p>However, the tests vary not only in the used techniques, the analysed genes, and the number of risk groups (low/high vs. low/intermediate/high), they also differ in the type and scope of the studies analysing their impact. Only two tests have been analysed in prospective trials so far.</p> <p>The interpretation of the published study results and assessment of the clinical utility of these tests is internationally inconsistent, leading to differences in guideline recommendations and refundability.</p> <p>Originally, we aimed to assess the benefit of 4 standardised multigene tests available in Europe (12-gene molecular test, 70 gene signature, 21-gene recurrence score sand PAM50 risk of recurrence score) to support the decision for or against adjuvant chemotherapy compared to endocrine therapy alone based on a multigene test decision strategy in patients who have hormone receptor positive, HER2-negative, lymph node negative or up to 3 lymph nodes positive invasive breast cancer. However, as priority was given to prospective data or randomised controlled trials, only two tests were analysed (21 gene recurrence score, and 70 gene signature).</p>
CONFLICT OF INTEREST:	<p>Management of Conflicts of Interest (Col): Cols for all Guidelines Development Group (GDG) members were assessed and managed by the European Commission Joint Research Centre (JRC) following an established procedure in line with the institutional rules. GDG member participation in the development of the recommendations was restricted, according to Col disclosure. Consequently, for this particular question, the following GDG members were recused from voting: Edoardo Colzani, Susan Knox, Elsa Pérez. Miranda Langendam, as external expert, was also not allowed to vote, according to the ECIBC rules of procedure.</p>

ASSESSMENT

Problem

Is the problem a priority?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
<ul style="list-style-type: none"> ○ No ○ Probably no ○ Probably yes ● Yes ○ Varies ○ Don't know 	<p>70 gene signature</p> <ul style="list-style-type: none"> • CE marked microarray • Designed to assess the risk of distant recurrence within 5 years and whether a woman would benefit from chemotherapy. Intended for use in pre- and postmenopausal women with stage I or stage II breast cancer with tumour size ≤ 5 cm and 0-3 positive LNs • Measures the expression of 70 genes involved in different parts of the metastatic pathway, including growth and proliferation, angiogenesis, local invasion etc. • Requires RNA extracted from formalin-fixed, paraffin-embedded tumour tissue (FFPE) • Test can be performed locally and off-site in a central lab of the Company in the Netherlands. • Uses microarray technology • Results: Discriminates in low and high risk using a predefined cut-off corresponding to a 10% risk of developing distant metastases over the next 10 years without any adjuvant hormone or chemotherapy. <p>Reference: (2)</p>	<p>The GDG prioritised this question for the ECIBC.</p>

Desirable Effects

How substantial are the desirable anticipated effects?

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<div><div><div>○ Trivial</div><div>○ Small</div><div>○ Moderate</div><div>● Large</div><div>○ Varies</div><div>○ Don't know</div></div></div>	<div><div><div>We found evidence from two types of studies, a retrospective subgroup analysis (interaction) from several validation and observational studies (12), and from an RCT comparing clinical outcomes in patients with discordant clinical and genomic risk assessment (2); the results are presented in three separate summaries of findings. Knauer2010</div><table><tr><th>Outcomes</th><th>No of participants (studies) Follow up</th><th>Certainty of the evidence (GRADE)</th><th>Relative effect (95% CI)</th><th colspan="2">Anticipated absolute effects* (95% CI)</th></tr><tr><td rowspan="3">Distant free survival - low genomic risk group</td><td rowspan="3">252 (1 observational study)^{1,a}</td><td rowspan="3"><div><div>⊕○○○</div><div>VERY LOW^{b,c,d,e,f}</div></div></td><td rowspan="3">HR 0.26 (0.03 to 2.02)</td><th>Risk with endocrine therapy</th><th>Risk difference with endocrine therapy plus chemotherapy</th></tr><tr><td colspan="2">Study population</td></tr><tr><td>69 per 1,000</td><td>51 fewer per 1,000 (67 fewer to 65 more)</td></tr></table></div></div>	Outcomes	No of participants (studies) Follow up	Certainty of the evidence (GRADE)	Relative effect (95% CI)	Anticipated absolute effects* (95% CI)		Distant free survival - low genomic risk group	252 (1 observational study) ^{1,a}	<div><div>⊕○○○</div><div>VERY LOW^{b,c,d,e,f}</div></div>	HR 0.26 (0.03 to 2.02)	Risk with endocrine therapy	Risk difference with endocrine therapy plus chemotherapy	Study population		69 per 1,000	51 fewer per 1,000 (67 fewer to 65 more)	<div><div><div>For this high clinical risk group, the GDG agreed the desirable effects were large.</div><div><div>*Definitions of used to classify patients as having high clinical risk (of recurrence or mortality) according to the modified version of Adjuvant! Online tool (see table below).</div><div>Among the patients classified at high clinical risk & low genomic risk: 98% were hormone receptor-positive, 92% were HER2-negative and 52% had negative lymph nodes.</div></div></div><table><tr><th>ER status</th><th>HER2 status</th><th>Grade</th><th>Nodal status</th><th>Tumour size</th></tr><tr><td rowspan="4">HR positive</td><td rowspan="4">HER2 negative</td><td rowspan="2">Well differentiated</td><td>Negative</td><td>3.1-5 cm</td></tr><tr><td>1-3 positive nodes</td><td>2.1-5 cm</td></tr><tr><td rowspan="2">Moderately differentiated</td><td>Negative</td><td>2.1-5 cm</td></tr><tr><td>1-3 positive</td><td>Any size</td></tr></table></div>	ER status	HER2 status	Grade	Nodal status	Tumour size	HR positive	HER2 negative	Well differentiated	Negative	3.1-5 cm	1-3 positive nodes	2.1-5 cm	Moderately differentiated	Negative	2.1-5 cm	1-3 positive	Any size
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Cardoso et al, 2016: high clinical risk - low genomic risk					
Outcomes	No of participants (studies) Follow up	Certainty of the evidence (GRADE)	Relative effect (95% CI)	Anticipated absolute effects* (95% CI)	
				Risk with endocrine therapy	Risk difference with endocrine therapy plus chemotherapy

HER2 positive	Poorly differentiated OR undifferentiated	Negative	2.1-5 cm
		1-3 positive	Any size
	Well differentiated OR moderately differentiated	Negative	2.1-5 cm
		1-3 positive	Any size
HER2 negative	Well differentiated	Negative	2.1-5 cm
		1-3 positive	Any size
	Moderately differentiated OR Poorly differentiated OR undifferentiated	Negative	1.1-5 cm
		1-3 positive	Any size
HR negative	Well differentiated OR moderately differentiated	Negative	1.1-5 cm
		1-3 positive	Any size
	Poorly differentiated OR undifferentiated	Any	Any size

		Poorly differentiated OR undifferentiated	Negative	2.1-5 cm
			1-3 positive	Any size
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			1-3 positive	Any size
	HER2 positive	Well differentiated OR moderately differentiated	Negative	1.1-5 cm
		Poorly differentiated OR undifferentiated	1-3 positive	Any size

Survival without distant metastasis	1228 (1 RCT) ^{1,a,b}	⊕⊕○○ LOW ^{c,d,e,f,g}	HR 0.65 (0.38 to 1.10)	Study population	
				58 per 1,000	20 fewer per 1,000 (36 fewer to 6 more)
Disease free-survival	1228 (1 RCT) ^{1,a,b}	⊕⊕○○ LOW ^{c,d,e,f,g}	HR 0.64 (0.43 to 0.95)	Study population	
				104 per 1,000	36 fewer per 1,000 (58 fewer to 5 fewer)
Overall survival	1228 (1 RCT) ^{1,a,b}	⊕⊕○○ LOW ^{c,d,e,f,g}	HR 0.63 (0.29 to 1.37)	Study population	
				28 per 1,000	10 fewer per 1,000 (20 fewer to 10 more)

1. Cardoso F, van't Veer LJ, Bogaerts J, Slaets L, Viale G, Delaloge S, et al. 70-Gene Signature as an Aid to Treatment Decisions in Early-Stage Breast Cancer.. 2016.

- a. The population was defined as High clinical Risk by the Adjuvant Online!: -Lymph node negative: 52%; HR positive: 98%; HER2 negative: 92%; Size >2cm: 58%
- b. The trial aims to assess non-inferiority results for the test. We report here the per-protocol analysis which is a more conservative approach in this context.
- c. The study design used did not provide the relevant clinical information of interest (i.e. number of chemotherapies avoided, direct comparison between use or not use of the test across genomic risk groups) thus extrapolation of those events from the study's results is required.
- d. There were a low number of events in each genomic risk group. The effect size estimates were imprecise.
- e. Results were reported at 5 years. This follow-up time might be insufficient for the measured outcomes. This treatment effect may become more pronounced over the next five years because more events will occur (i.e. In ER-positive/HER2-negative (luminal-type) breast cancer, evidence shows that recurrences occur after five years in approximately one half of all distant recurrence cases -Sparano JA, Breast Cancer. J Clin Oncol. 2015;33(21):2353-60).
- f. There was imbalance between groups in the proportions of violations to the protocol. Additionally, a group of patients were incorrectly classified by test, and the sample had to be increased to handle the small number of patients recruited.
- g. The MINDACT trial does not have a formal non-inferiority design (would need to have a very large sample size or present a very long follow-up). Instead propose does that if fewer patients are treated with chemotherapy while not adversely affecting survival without distant metastasis, it can be taken to be equivalent.

The PRU members developed a "back of the envelope" model to estimate the downstream consequences of testing patients with 70 gene signatures versus using clinical risk scores.

The general model assumptions were:

1) Results are based on a fixed observation time of 10 years.

3) The observed rate of events at 5 years in the MINDACT trial (Cardoso et al, 2016) will remain constant at 10 years.

4) Rates of events observed in the RCTs were applied to the simulated clinical score arms. It was assumed that basal risk of events for clinical score groups was homogenous between the low and the high clinical risk groups into a given genomic risk group.

An approximately 40% reduction in the women receiving chemotherapy was considered as a desirable effect.

The model assumes that women with low clinical risk are not tested and not treated with chemotherapy in both strategies. Consequently, the first two columns and the fourth one are invariant.

	low clinical low genomic	low clinical high genomic	high clinical low genomic	high clinical high genomic	total		low clinical low genomic	low clinical high genomic	high clinical low genomic	high clinical high genomic	total
Number of women	410	88	232	270	1000		410	88	232	270	1000
Chemotherapies	0.0	0.0	0.0	269.8	269.8		0	0	231.6	269.8	501.4
Distant metastasis recurrence	9.8	5.4	12.0	25.4	52.6		9.8	5.4	7.6	25.4	48.2
Any disease or recurrence	29.5	8.4	22.5	39.7	100.1		29.5	8.4	15.5	39.7	93.1
Deaths	6.6	2.7	6.3	14.3	29.8		6.6	2.7	2.8	14.3	26.3

Undesirable Effects

How substantial are the undesirable anticipated effects?

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- b. The study enrolled patients that were treated more than ten years ago, the chemotherapy regime is very different now which preclude the applicability of the results. The study design used did not provide the relevant clinical information of interest (i.e. number of chemotherapies avoided, direct comparison between use or not of the test) thus extrapolation of those events from the study's results is required.
- c. There were a low number of events in each genomic risk group. The effect size estimates were imprecise which also led to a low power to test interaction.
- d. Follow-up was censored at 5 years. The follow-up time might be short to assess of the outcomes of interest, other studies have shown a high risk of disease recurrence after the 5 years of treatment.
- e. Data included in the patient data pooled analysis were previously used in validation studies. The test was trained to identify the prognosis of these patients which might lead to overfitting in the subsequent analysis.
- f. Results were not provided by strata of negative and positive lymph nodes (up to 3), instead as adjusted estimations. Adjustment might be inappropriate if there is interaction between outcome and number of lymph node positive, which were not assessed in the analysis.

Cardoso et al, 2016: high clinical risk - low genomic risk

Outcomes	No of participants (studies) Follow up	Certainty of the evidence (GRADE)	Relative effect (95% CI)	Anticipated absolute effects* (95% CI)	
				Risk with endocrine therapy	Risk difference with endocrine therapy plus chemotherapy
Survival without distant metastasis	1228 (1 RCT) ^{1,a,b}	⊕⊕○○ LOW ^{c,d,e,f,g}	HR 0.65 (0.38 to 1.10)	Study population	
				58 per 1,000	20 fewer per 1,000 (36 fewer to 6 more)
Disease free-survival	1228 (1 RCT) ^{1,a,b}	⊕⊕○○ LOW ^{c,d,e,f,g}	HR 0.64 (0.43 to 0.95)	Study population	
				104 per 1,000	36 fewer per 1,000 (58 fewer to 5 fewer)
Overall survival	1228 (1 RCT) ^{1,a,b}	⊕⊕○○ LOW ^{c,d,e,f,g}	HR 0.63 (0.29 to 1.37)	Study population	
				28 per 1,000	10 fewer per 1,000 (20 fewer to 10 more)

	<ol style="list-style-type: none"> 1. Cardoso F, van't Veer LJ, Bogaerts J, Slaets L, Viale G, Delaloge S, et al. 70-Gene Signature as an Aid to Treatment Decisions in Early-Stage Breast Cancer. 2016. a. The population was defined as High clinical Risk by the Adjuvant Online!: -Lymph node negative: 52%; HR positive: 98%; HER2 negative: 92%; Size >2cm: 58% b. The trial aims to assess non-inferiority results for the test. We report here the per-protocol analysis which is a more conservative approach in this context. c. The study design used did not provide the relevant clinical information of interest (i.e. number of chemotherapies avoided, direct comparison between use or not use of the test across genomic risk groups) thus extrapolation of those events from the study's results is required. d. There were a low number of events in each genomic risk group. The effect size estimates were imprecise. e. Results were reported at 5 years. This follow-up time might be insufficient for the measured outcomes. This treatment effect may become more pronounced over the next five years because more events will occur (i.e. In ER-positive/HER2-negative (luminal-type) breast cancer, evidence shows that recurrences occur after five years in approximately one half of all distant recurrence cases - Sparano JA, Breast Cancer. J Clin Oncol. 2015;33(21):2353-60). f. There was imbalance between groups in the proportions of violations to the protocol. Additionally, a group of patients were incorrectly classified by test, and the sample had to be increased to handle the small number of patients recruited. g. The MINDACT trial does not have a formal non-inferiority design (would need to have a very large sample size or present a very long follow-up). Instead propose does that if fewer patients are treated with chemotherapy while not adversely affecting survival without distant metastasis, it can be taken to be equivalent. <p>The PRU members developed a "back of the envelope" model to estimate the downstream consequences of testing patients with 70 gene signatures versus using clinical risk scores.</p> <p>The general model assumptions were:</p> <ol style="list-style-type: none"> 1) Results are based on a fixed observation time of 10 years. 3) The observed rate of events at 5 years in the MINDACT trial (Cardoso et al, 2016) will remain constant at 10 years. 4) Rates of events observed in the RCTs were applied to the simulated clinical score arms. It was assumed that basal risk of events for clinical score groups was homogenous between the low and the high clinical risk groups into a given genomic risk group. <p>An approximately 40% reduction in the women receiving chemotherapy was considered as a desirable effect.</p> <p>The model assumes that women with low clinical risk are not tested and not treated with chemotherapy in both strategies. Consequently the first two columns and the fourth one are invariant.</p>	
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	low clinical low genomic	low clinical high genomic	high clinical low genomic	high clinical high genomic	total		low clinical low genomic	low clinical high genomic	high clinical low genomic	high clinical high genomic	total	
Number of women	410	88	232	270	1000		410	88	232	270	1000	
Chemo-therapies	0.0	0.0	0.0	269.8	269.8		0	0	231.6	269.8	501.4	
Distant metastasis recurrence	9.8	5.4	12.0	25.4	52.6		9.8	5.4	7.6	25.4	48.2	
Any disease or recurrence	29.5	8.4	22.5	39.7	100.1		29.5	8.4	15.5	39.7	93.1	
Deaths	6.6	2.7	6.3	14.3	29.8		6.6	2.7	2.8	14.3	26.3	

Certainty of evidence

What is the overall certainty of the evidence of effects?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
<ul style="list-style-type: none"> ○ Very low ● Low ○ Moderate ○ High ○ No included studies 		The GDG agreed by consensus that the certainty of the evidence was low, due to issues around imprecision, and risk of bias (some patients were reclassified according to genomic risk in a post-hoc fashion).

Values

Is there important uncertainty about or variability in how much people value the main outcomes?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
<ul style="list-style-type: none"> ○ Important uncertainty or variability ○ Possibly important uncertainty or variability ● Probably no important uncertainty or variability ○ No important uncertainty or variability 		The GDG agreed by consensus that there is probably no important uncertainty or variability.

○ No known undesirable outcomes		
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Balance of effects

Does the balance between desirable and undesirable effects favor the intervention or the comparison?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
<ul style="list-style-type: none"> ○ Favors the comparison ○ Probably favors the comparison ○ Does not favor either the intervention or the comparison ● Probably favors the intervention ○ Favors the intervention ○ Varies ○ Don't know 		The GDG agreed by consensus that for the high clinical risk group the balance of effects probably favours the intervention.

Resources required

How large are the resource requirements (costs)?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS																										
<div><div><div>○ Large costs</div><div>○ Moderate costs</div><div>○ Negligible costs and savings</div><div>○ Moderate savings</div><div>● Large savings</div><div>○ Varies</div><div>○ Don't know</div></div></div>	<div>The cost of the assay is around EUR 3 153 per patient (HR positive, HER2 negative, lymph node negative) in the included studies (13)(14)</div> <table><tr><th>Study ID</th><th>Country</th><th>Year value</th><th>Cost</th><th>Currency</th><th>US PPP</th></tr><tr><td colspan="6">Lymph-node negative</td></tr><tr><td>1</td><td>Ward</td><td>UK</td><td>2012</td><td>2675</td><td>British Pounds</td><td>3811</td></tr><tr><td>2</td><td>Kondo</td><td>Japan</td><td>2009</td><td>4222</td><td>US Dollar</td><td>4222</td></tr></table>	Study ID	Country	Year value	Cost	Currency	US PPP	Lymph-node negative						1	Ward	UK	2012	2675	British Pounds	3811	2	Kondo	Japan	2009	4222	US Dollar	4222	<div>For the high clinical risk group the GDG considered that there would be large savings with the 70 gene signature test.</div> <div>Cost of testing is large = for 1000 women about EUR 3 million for tests; avoidance of approximately 460 chemotherapies (EUR 10000 per chemotherapy) = EUR 4.6 million for chemotherapy as reported by the back of the envelope calculation - in high clinical risk group.</div> <div>Potential savings of EUR 1.6 million per 1000 women with 70 gene signature are associated with the reduction in the number of women who receive chemotherapy, which is 464/1000 patients according to the 'back of the envelope' model.</div>
Study ID	Country	Year value	Cost	Currency	US PPP																							
Lymph-node negative																												
1	Ward	UK	2012	2675	British Pounds	3811																						
2	Kondo	Japan	2009	4222	US Dollar	4222																						

Certainty of evidence of required resources

What is the certainty of the evidence of resource requirements (costs)?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
<ul style="list-style-type: none"> ● Very low ○ Low ○ Moderate ○ High ○ No included studies 	High certainty of the evidence of 70 gene signature cost. Very low certainty for harms. The studies consistently reported a similar cost of the 70 gene signature. Also, there are no concerns regarding the indirectness nor imprecision of the published costs. Very low certainty regarding the savings due to chemotherapy avoided calculated above.	The GDG agreed by consensus that the certainty of the evidence of required resources was very low.

Cost effectiveness

Does the cost-effectiveness of the intervention favor the intervention or the comparison?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
<ul style="list-style-type: none"> ○ Favors the comparison ○ Probably favors the comparison ○ Does not favor either the intervention or the comparison ○ Probably favors the intervention ○ Favors the intervention ○ Varies ● No included studies 	<p>The cost-effectiveness evaluations available from the literature (15)(16) used models that do not fulfil the assumptions that were used in the 'back of the envelope' model developed ad-hoc for this PICO.</p>	<p>The GDG, therefore, did not include any studies.</p>

Equity

What would be the impact on health equity?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
<ul style="list-style-type: none"> ○ Reduced ● Probably reduced ○ Probably no impact ○ Probably increased ○ Increased ○ Varies ○ Don't know 	<p>No systematic review was carried out.</p>	<p>The GDG agreed by consensus that equity would probably be reduced.</p>

Acceptability

Is the intervention acceptable to key stakeholders?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
<ul style="list-style-type: none"> ○ No ○ Probably no ○ Probably yes ○ Yes ● Varies ○ Don't know 	<p>No systematic review was carried out.</p>	<p>Patients: It would be acceptable</p> <p>Health care providers: varies (due to costs, mainly due to having to send the results to a lab in the Netherlands)</p> <p>Policy makers: Don't know until there is context specific evidence on cost-effectiveness.</p>

Feasibility

Is the intervention feasible to implement?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
<ul style="list-style-type: none"> ○ No ○ Probably no ○ Probably yes ○ Yes ● Varies ○ Don't know 	No systematic review was carried out.	The GDG agreed by consensus that feasibility would vary depending on the availability of resources.

SUMMARY OF JUDGEMENTS

	JUDGEMENT						
PROBLEM	No	Probably no	Probably yes	Yes		Varies	Don't know
DESIRABLE EFFECTS	Trivial	Small	Moderate	Large		Varies	Don't know
UNDESIRABLE EFFECTS	Large	Moderate	Small	Trivial		Varies	Don't know
CERTAINTY OF EVIDENCE	Very low	Low	Moderate	High			No included studies
VALUES	Important uncertainty or variability	Possibly important uncertainty or variability	Probably no important uncertainty or variability	No important uncertainty or variability			No known undesirable outcomes
BALANCE OF EFFECTS	Favors the comparison	Probably favors the comparison	Does not favor either the intervention or the comparison	Probably favors the intervention	Favors the intervention	Varies	Don't know
RESOURCES REQUIRED	Large costs	Moderate costs	Negligible costs and savings	Moderate savings	Large savings	Varies	Don't know
CERTAINTY OF EVIDENCE OF REQUIRED RESOURCES	Very low	Low	Moderate	High			No included studies
COST EFFECTIVENESS	Favors the comparison	Probably favors the comparison	Does not favor either the intervention or the comparison	Probably favors the intervention	Favors the intervention	Varies	No included studies
EQUITY	Reduced	Probably reduced	Probably no impact	Probably increased	Increased	Varies	Don't know

	JUDGEMENT						
ACCEPTABILITY	No	Probably no	Probably yes	Yes		Varies	Don't know
FEASIBILITY	No	Probably no	Probably yes	Yes		Varies	Don't know

TYPE OF RECOMMENDATION

Strong recommendation against the intervention ○	Conditional recommendation against the intervention ○	Conditional recommendation for either the intervention or the comparison ○	Conditional recommendation for the intervention ●	Strong recommendation for the intervention ○
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CONCLUSIONS

Recommendation

For women with hormone receptor positive, HER2-negative, lymph node negative or up to 3 lymph nodes positive invasive breast cancer at high clinical risk* the ECIBC's Guidelines Development Group (GDG), suggests using the 70 gene signature test to guide the use of chemotherapy (conditional recommendation, low certainty of the evidence).

*This recommendation applies to patients with HR-positive, HER2-negative invasive breast cancer which are: G1 and node negative and 3,1-5cm or G1 and 1-3 positive nodes and 2,1-5cm or G2 and node negative and 2.1-5cm or G2 and 1-3 positive nodes and any size or G3 and node negative and 2,1-5cm or G3 and 1-3 positive nodes and any size.

Justification

In the high clinical risk group, the recommendation is conditional in favour of using the 70 gene signature testing to guide the use of chemotherapy as mainly because of the low certainty of the evidence.

Subgroup considerations

The proportion of women with 2 or 3 lymph node positive breast cancer were small, so the results may be less clear in this subgroup.

Implementation considerations

Decreasing cost for the test would support widespread use - price negotiations may be appropriate.

Monitoring and evaluation

None were considered by the GDG.

Research priorities

Longer follow up studies would be needed, as currently there is only a follow-up of five years.

REFERENCES SUMMARY

1. Sparano JA, Gray RJ Makower DF Pritchard KI Albain KS Hayes DF et al. Adjuvant Chemotherapy Guided by a 21-Gene Expression Assay in Breast Cancer.. N Engl J Med.; 2018.
2. Cardoso F, van't Veer LJ, Bogaerts J, Slaets L, Viale G, Delaloge S, et al. 70-Gene Signature as an Aid to Treatment Decisions in Early-Stage Breast Cancer.. 2016.
3. Howlader, N., S. F. Altekruse, C. I. Li, V. W. Chen, C. A. Clarke, L. A. Ries, and K. A. Cronin. "US incidence of breast cancer subtypes defined by joint hormone receptor and HER2 status." J Natl Cancer Inst; 2014.
4. Early Breast Cancer Trialists' Collaborative, Group. Effects of chemotherapy and hormonal therapy for early breast cancer on recurrence and 15-year survival: an overview of the randomised trials. Lancet; 2005.
5. Early Breast Cancer Trialists' Collaborative, Group. Relevance of breast cancer hormone receptors and other factors to the efficacy of adjuvant tamoxifen: patient-level meta-analysis of randomised trials. Lancet; 2011.
6. Coates, A. S., E. P. Winer, A. Goldhirsch, R. D. Gelber, M. Gnant, M. Piccart-Gebhart, B. Thurlimann, H. J. Senn, and Members Panel. Tailoring therapies--improving the management of early breast cancer: St Gallen International Expert Consensus on the Primary Therapy of Early Breast Cancer 2015.. Ann Oncol; 2015.
7. Goldhirsch, A., W. C. Wood, A. S. Coates, R. D. Gelber, B. Thurlimann, H. J. Senn, and members Panel. 2011. Strategies for subtypes--dealing with the diversity of breast cancer: highlights of the St. Gallen International Expert Consensus on the Primary Therapy of Early Breast Cancer 2011. Ann Oncol; 2011.
8. Early Breast Cancer Trialists' Collaborative, Group. Comparisons between different polychemotherapy regimens for early breast cancer: meta-analyses of long-term outcome among 100,000 women in 123 randomised trials. Lancet; 2012.
9. Albain KS, Barlow WE, Shak S, Hortobagyi GN, Livingston RB, Yeh IT, Ravdin P, Bugarini R, Baehner FL, Davidson NE, Sledge GW, Winer EP, Hudis C, Ingle JN, Perez EA, Pritchard KI, Shepherd L, Gralow JR, Yoshizawa C, Allred DC, Osborne CK, Hayes DF, America., Breast, Cancer, Intergroup, of, North. Prognostic and Predictive Value of the 21-Gene Recurrence Score Assay in a Randomized Trial of Chemotherapy for Postmenopausal, Node-Positive, Estrogen Receptor-Positive Breast Cancer. Lancet Oncol; 2010.
10. Paik, . Gene Expression and Benefit of Chemotherapy in Women With Node-Negative, Estrogen Receptor-Positive Breast Cancer. J Clin Oncol; 2006.
11. Sparano, J. A., R. J. Gray, D. F. Makower, K. I. Pritchard, K. S. Albain, D. F. Hayes, et al. Prospective Validation of a 21-Gene Expression Assay in Breast Cancer et al. N Engl J Med; 2015.
12. Knauer M, Mook S, Rutgers EJ, Bender RA, Hauptmann M, van de Vijver MJ, Koornstra RH, Bueno-de-Mesquita JM, Linn SC, van 't Veer LJ.. The predictive value of the 70-gene signature for adjuvant chemotherapy in early breast cancer. Breast Cancer Res Treat; 2010.
13. Ward S, Scope A, Rafia R, Pandor A, Harnan S, Evans P, et al.. Gene expression profiling and expanded immunohistochemistry tests to guide the use of adjuvant chemotherapy in breast cancer management: a systematic review and cost-effectiveness analysis. . Health Technol Assess (Winchester, England); 2013.
14. Kondo M, Hoshi SL, Ishiguro H, Toi M.. Economic evaluation of the 70-gene prognosis-signature (MammaPrint(R)) in hormone receptor-positive, lymph node-negative, human epidermal growth factor receptor type 2-negative early stage breast cancer in Japan. . Breast Cancer Res Treat ; 2012.
15. Blok EJ, . Systematic review of the clinical and economic value of gene expression profiles for invasive early breast cancer available in Europe.. Cancer Treatment Reviews; 2018.
16. Wang H, K resen R, Hervik A, Thoresen SO. Mammography screening in Norway: results from the first screening round in four counties and cost-effectiveness of a modeled nationwide screening. Cancer Causes Control; 2001.