

# **Clinical Significance of Circulating Tumor Cells in Peripheral Blood from Patients with Gastric Cancer**

**Molecular Frontier Surgery  
Surgical Oncology and Digestive Surgery  
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# Recurrence Pattern after Gastrectomy in Patients with Gastric Cancer

Author	Year	No. of Pts.	Hematogenous	Dissemination	Local or LN
Katai	1994	687	25%	44%	23%
Nakajima	1996	1382	32%	40%	23%
Maehara	2000	130	54%	43%	17%
Otsuji	2001	290	26%	57%	17%
Ohno	2003	120	34%	45%	26%
Nashimoto	2004	151	43%	33%	25%

# Purpose

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- **To evaluate Circulating Tumor Cells (CTCs) in patients with gastric cancer using the CellSearch<sup>®</sup> System.**
  - **To clarify the clinical significance of CTCs in resectable and unresectable gastric cancer.**
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# Methods

## CellSearch<sup>®</sup> System



Volume of evaluated blood : 7.5ml

### Cancer cell separation



**EpCAM antibody-coated magnetic beads**

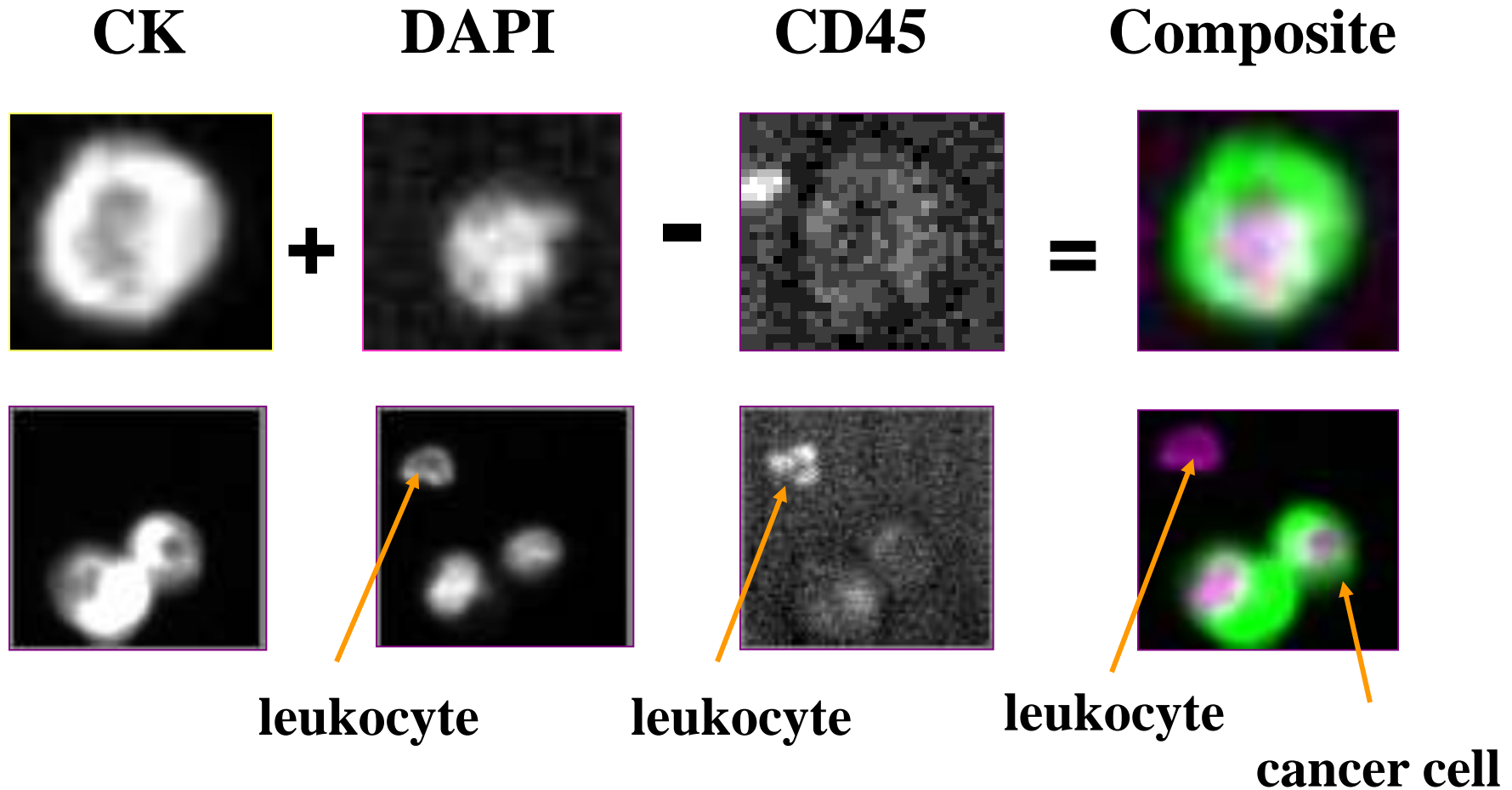
### Immunohistochemical stain

**CK:** Cytokeratin 8, 18, 19-phycoerythrin

**DAPI:** Fluorescent nucleic acid dye 4, 2-diamidino-2-phenylidole dihydrochloride

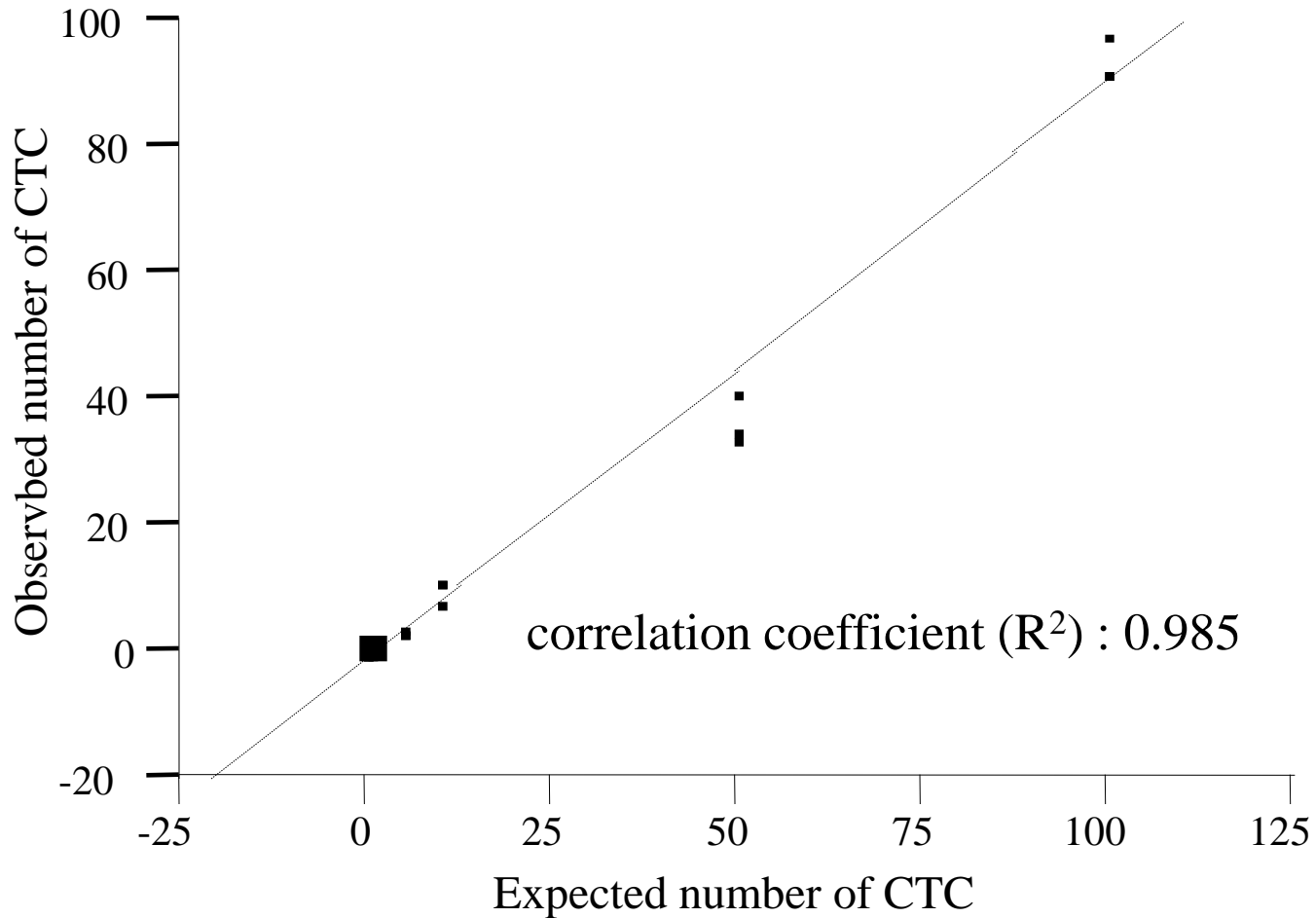
**CD45:** Fluorescently labeled monoclonal antibodies specific for leukocytes

# Definition of CTCs in CellSearch<sup>®</sup> system



**CTCs are defined as nucleated cells lacking CD45 and expressing cytokeratin.**

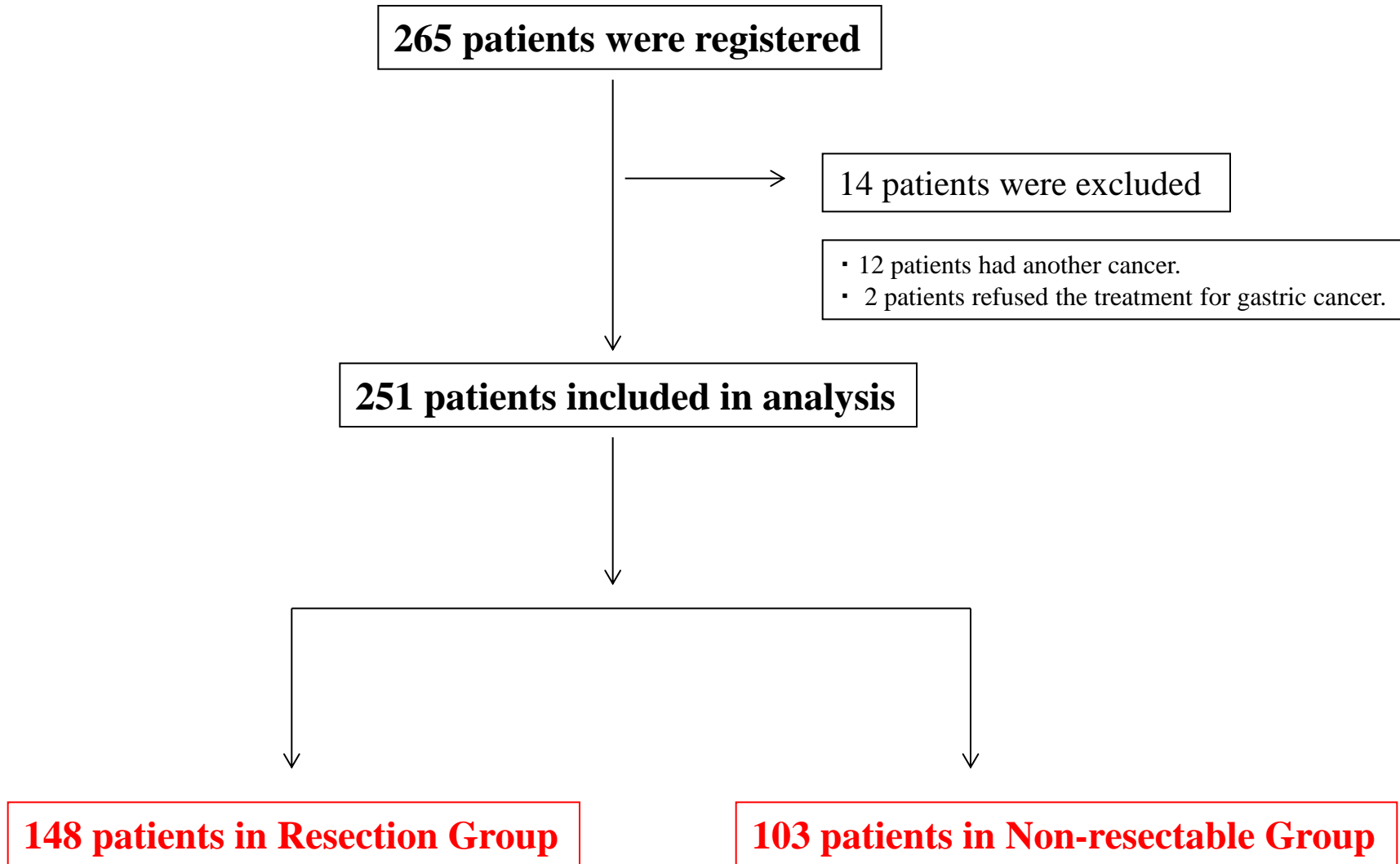
# Regression analysis of gastric cancer cell line using KATO III



Even the single cell spiked into the samples was detected using this system.

# Clinical design in CTC evaluation for gastric cancer

(February 2005 ~ December 2012)



# Patients (n=251)

## ■ Resection Group with gastrectomy (n=148)

**pT1                      63 pts**

**pT2                      18 pts**

**pT3                      37 pts**

**pT4                      30 pts**

**Average of follow-up time after surgery 31.6 month (4 month~5 year)**

## ■ Non-resectable Group (n=103)

**Un-resectable                      72 pts**

**Recurrent                              31 pts**

**Samples of peripheral blood were collected before gastrectomy or chemotherapy.**





# Detection rate of CTCs



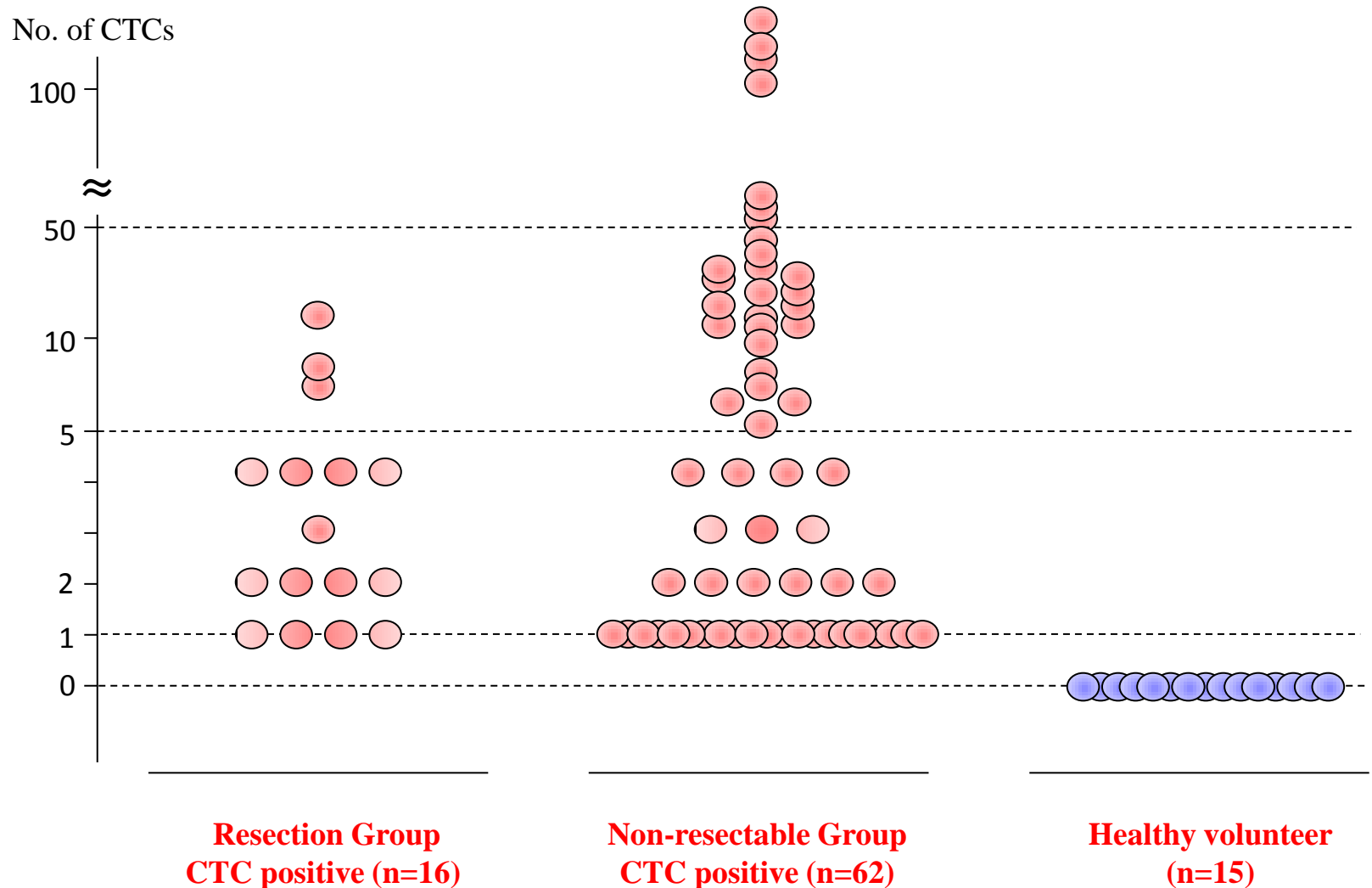
## ■ Resection Group

**16 pts (10.8 %)**

## ■ Non-resectable Group

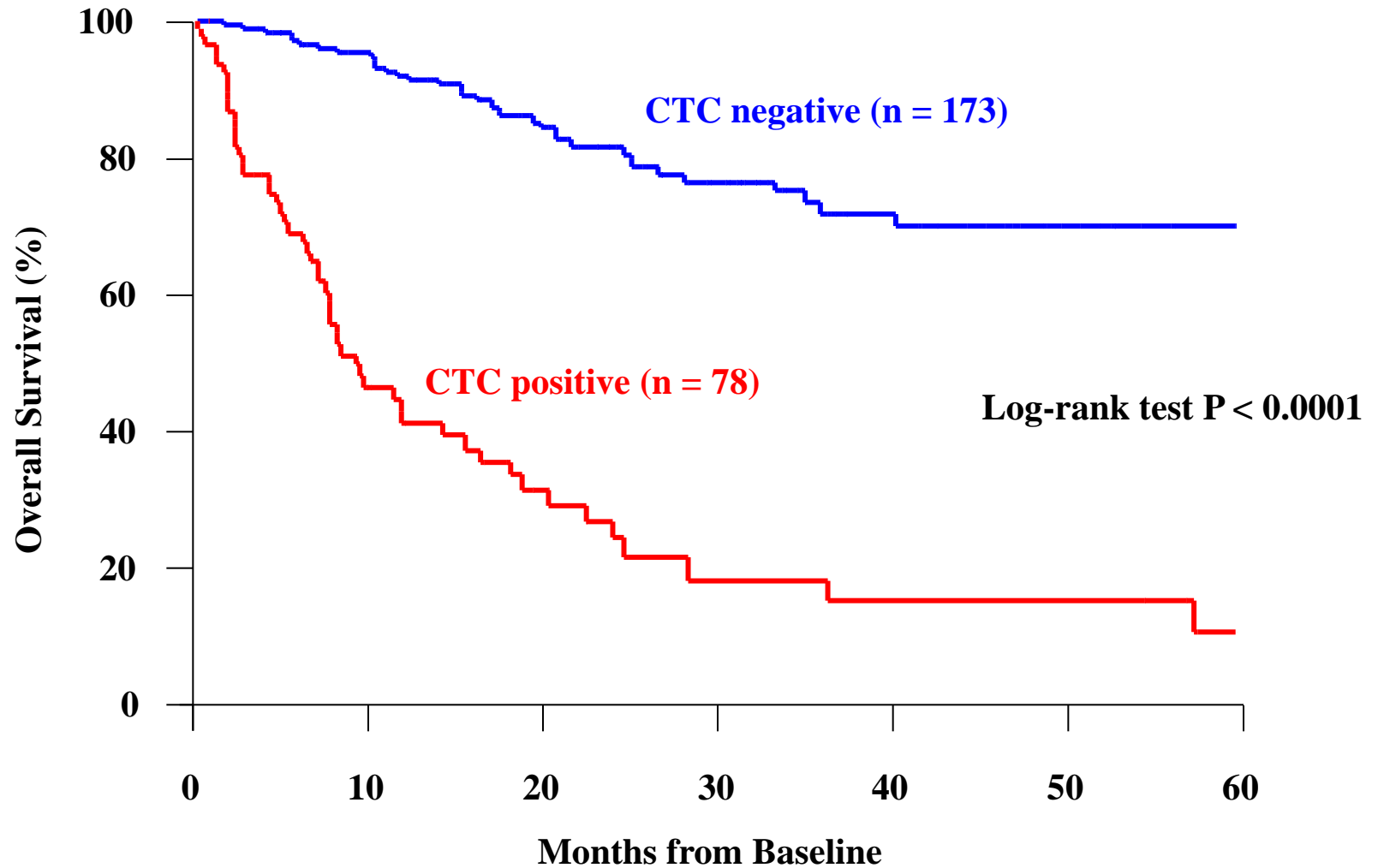
**62 pts (60.2 %)**

# The number of CTC in clinical samples

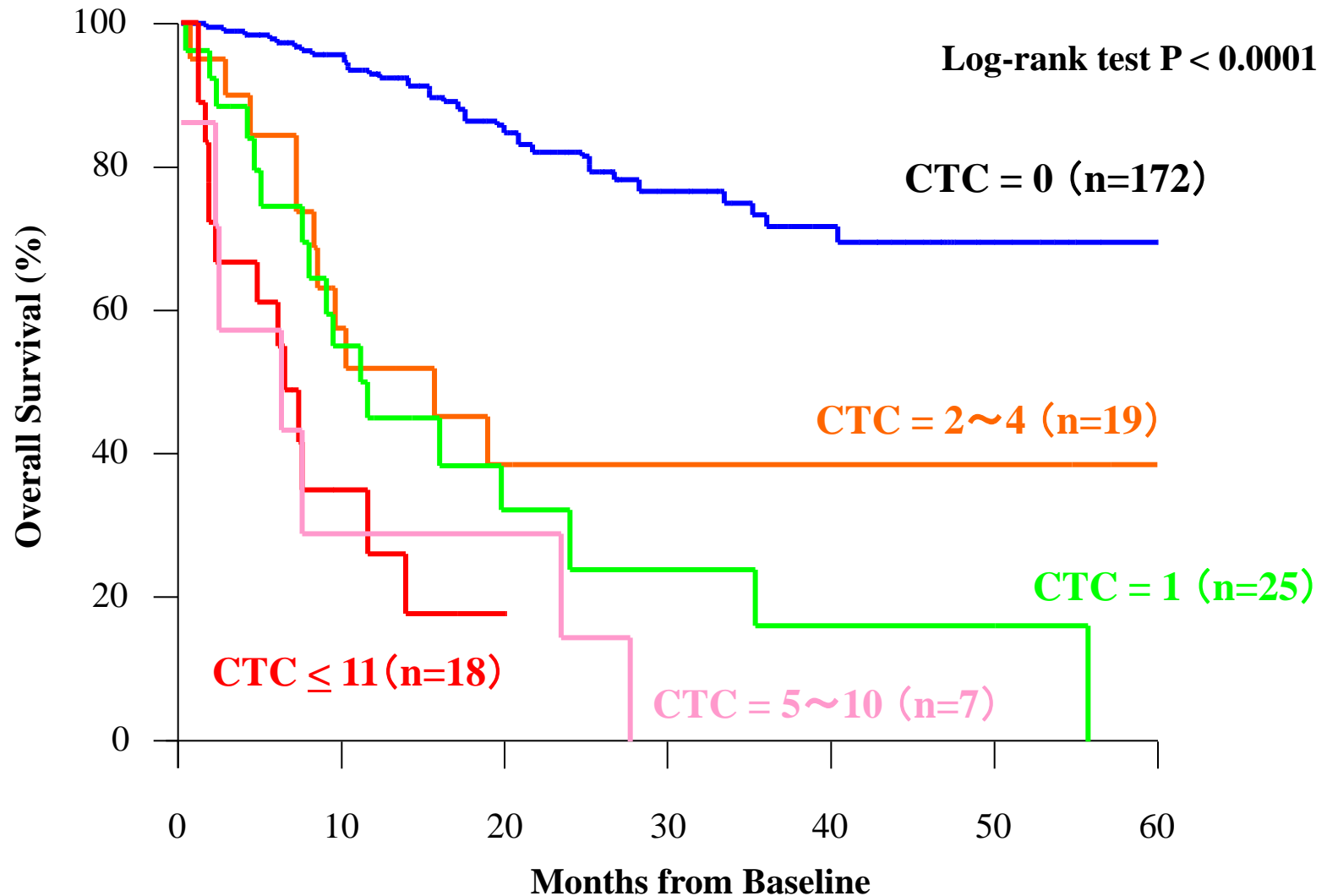


**We defined that one or more CTCs are positive.**

# Overall survival curve according to CTCs in all patients with gastric cancer (n=251)



# Overall survival according to the number of CTCs



# Clinicopathological factor in Resection Group

Variable	CTC (%)		P-value
	Positive (n=16)	Negative (n=132)	
<b>T factor</b>			
1	1 (6.3)	62 (47.0)	<b>0.009</b>
2	2 (12.5)	16 (12.1)	
3	6 (37.5)	31 (23.5)	
4	7 (43.8)	23 (17.4)	
<b>N factor</b>			
0	2 (12.5)	80 (60.6)	<b>&lt;0.0001</b>
1	0 (0)	19 (14.4)	
2	1 (6.3)	17 (12.9)	
3	13 (81.3)	16 (12.1)	
<b>Distant metastasis</b>			
yes	3 (18.8)	5 (3.8)	<b>0.012</b>
no	13 (81.2)	127 (96.2)	
<b>Stage</b>			
I	1 (6.3)	63 (47.7)	<b>0.0002</b>
II	1 (6.3)	25 (18.9)	
III	11 (68.8)	39 (29.5)	
IV	3 (18.8)	5 (3.8)	
<b>Lymphatic invasion</b>			
0	1 (6.3)	71 (53.8)	<b>0.0003</b>
1	15 (93.7)	61 (46.2)	
<b>Vessel invasion</b>			
0	3 (18.8)	73 (55.3)	<b>0.006</b>
1	13 (81.2)	59 (44.7)	

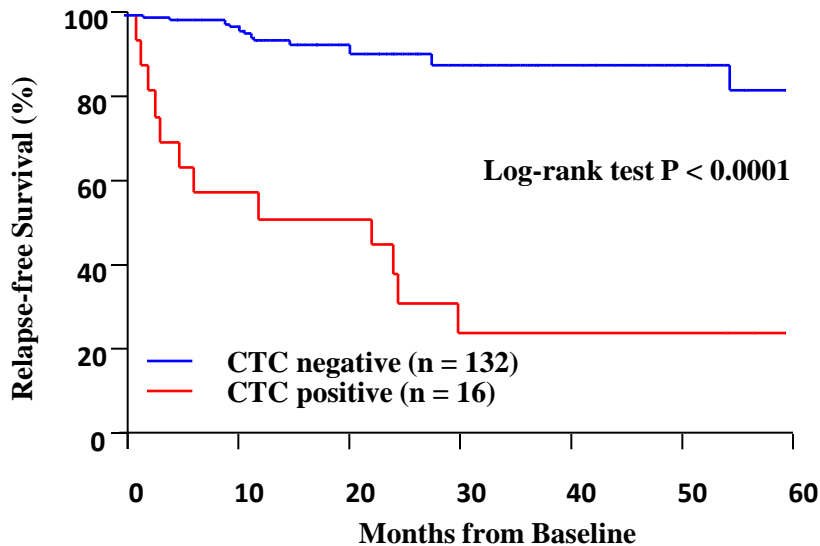
There are no difference in Gender, Age, Pathological differentiation, CEA and CA19-9 in serum.

# Follow-up after gastrectomy in 16 patients with CTC

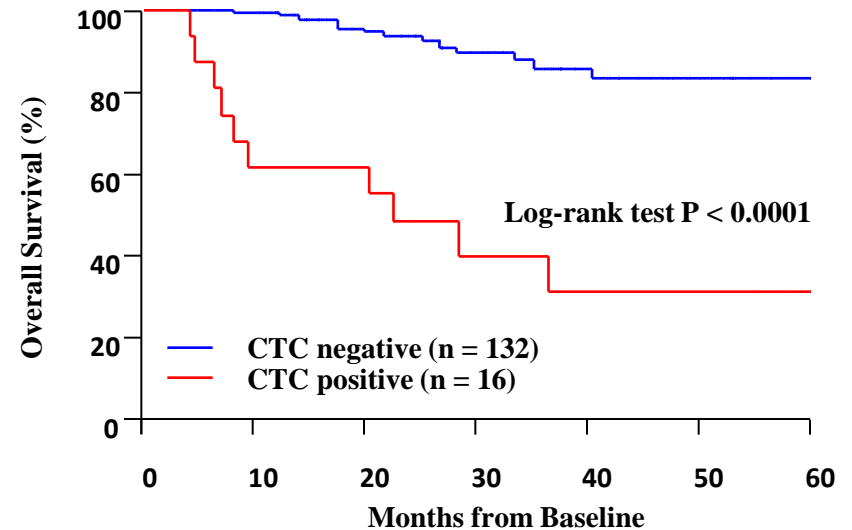
Post gastrectomy	No. of patients
<b>Recurrence pattern</b>	<b>12 (75.0 %)</b>
Peritoneal dissemination	9 (56.3 %)
Liver metastasis	2 (12.5 %)
Bone metastasis	2 (12.5 %)
Adrenal gland metastasis	1 (6.3%)
Lymph node metastasis	1 (6.3 %)
No recurrence	4 (25.0 %)

# Relapse free and Overall survival curve according to CTCs in Resection group

## Relapse free Survival



## Overall Survival

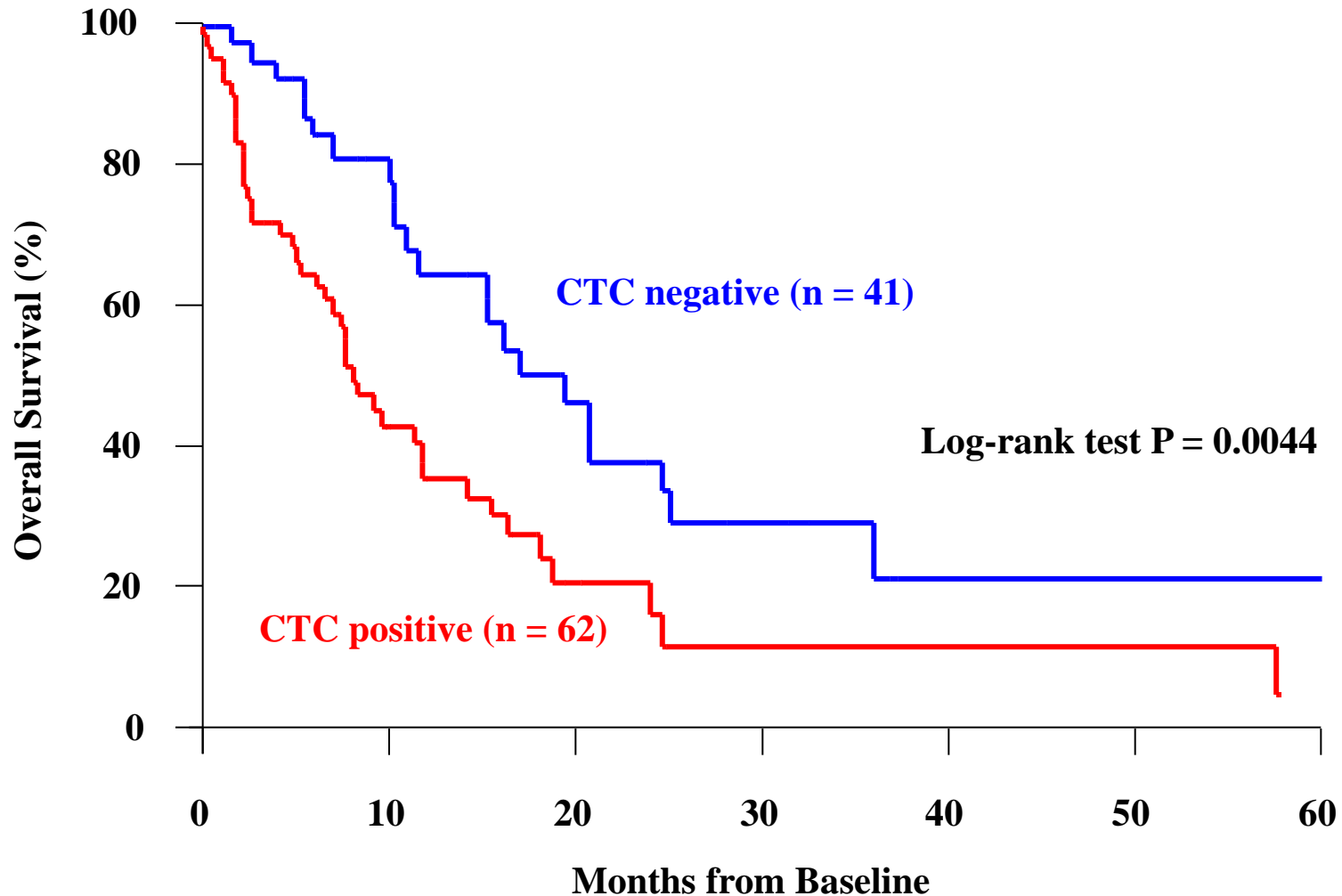


# Independent prognostic predictors of survival in Resection group

Independent factors	Univariate analysis		Multivariate analysis	
	Hazard ratio (95% CI)	P-value	Hazard ratio (95% CI)	P-value
Depth of tumor invasion	2.33 (1.56-3.69)	< 0.0001	1.45 (0.82-2.69)	0.204
Lymph node metastasis	2.60 (1.80-4.04)	< 0.0001	1.53 (0.91-2.69)	0.114
Distant metastasis	3.85 (1.11-10.28)	0.035	0.87 (0.23-2.59)	0.807
Lymphatic invasion	10.41 (3.05-65.08)	< 0.0001	1.73 (0.33-13.90)	0.541
Vessel invasion	7.53 (2.58-31.99)	< 0.0001	1.62 (0.39-9.07)	0.526
<b>CTC</b>	2.77 (1.81-4.18)	< 0.0001	1.73 (1.08-2.77)	<b>0.024</b>



# Survival curve depends on CTCs in Non-resectable Group



# Change in the number of CTC before and after chemotherapy in Non-resectable Group (n=45)

	Partial Response (n=21)	Stable Disease (n=14)	Progression Disease (n=10)	
<b>CTC</b>	<b>Decrease</b>	<b>16 (76.2%)</b>	<b>3 (21.4%)</b>	<b>1 (10%)</b>
	<b>No change</b>	<b>5 (23.8%)</b>	<b>10 (71.4%)</b>	<b>3 (30%)</b>
	<b>Increase</b>	<b>0</b>	<b>1 (7.1%)</b>	<b>6 (60%)</b>

P<0.0001

# Conclusion

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**The evaluation of CTCs in peripheral blood may be a useful tool for predicting tumor progression, prognosis, and the effect of chemotherapy in patients with gastric cancer.**

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