Influence of hospital procedure volume on ovarian cancer survival in Japan, a country with low incidence of ovarian cancer

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The survival of ovarian cancer patients has been reported to be superior at hospitals with a high volume of operations. A population-based study was carried out to assess whether this is true in Japan, where the incidence rate is relatively low as compared with other developed countries. The Osaka Cancer Registry's data were used to investigate associations between hospital procedure volume and survival of ovarian cancer patients. Hospitals were ranked according to the number of operations for ovarian cancer performed per year (high/medium/low/very low). Survival analysis was restricted to the reported 2450 cases who lived in Osaka Prefecture (except for Osaka City) diagnosed in 1975–1995, or those who resided in Osaka City in 1993–1995, since active follow-up data on vital status 5 years after the diagnosis were available. The relative 5-year survival for all ovarian cancer cases was 38.8%, and the survival was higher with greater hospital volume (22.3%/34.2%/46.2%/55.0%). After adjustment for age, histologic type and cancer stage by the Cox regression model, patients receiving care in very-low-volume hospitals were seen to have a 60% higher risk of death than patients receiving care in high-volume hospitals (*P*<0.01). Although some limitations existed in this study, the results indicated that further centralization of operative treatment in high-volume hospitals might improve survival of ovarian cancer patients in Japan. (Cancer Sci 2004; 95: 233–237)

Ovarian cancer is the leading cause of death among gynecologic malignancies in the developed countries, and fewer than half of women diagnosed with ovarian cancer survive 5 years after diagnosis. In Japan, the incidence rate of ovarian cancer is relatively low as compared with other developed countries (i.e., Nordic nations, the United Kingdom, and North America),[1] while the relative 5-year survival for ovarian cancer is less than that in the US (36.4% vs. 43.5%).[2]

The standard treatment of ovarian cancer is cytoreductive surgery followed by combination chemotherapy. Initial surgery is important for accurate staging of the disease and for removing as much of the tumor as possible. Former reports suggest that one of the prognostic factors predicting survival is the diameter of the residual tumor mass after operation,[3, 4] so it has been suggested that the initial operation provides the most important opportunity to improve survival.[5] The question arises, are lower survival rates in Japan, where the incidence rate of ovarian cancer is low, associated with poor technical expertise of physicians and of the teams performing primary surgery?

Relative 5-year survival for malignant ovarian germ cell tumors in Japan is also much lower than that in the US (58.6% vs. 83.7%).[2] Similar differences were observed between Japan and the US in survival for malignant testicular germ cell tumors.[6] Malignant ovarian germ cell tumors are rare compared with testicular germ cell tumors. Most of the advances in the management of the former have been extrapolations from experience with the latter, and developments in surgery and of effective chemotherapy regimens have markedly improved the outcome of treatment of ovarian germ cell tumors.[7, 8] Lower availability of medical oncologists than in the US might have adversely influenced the 5-year survival of both types of germ cell tumor patients in Japan.

In this paper, using the Osaka Cancer Registry's data, we have tried to clarify whether increased hospital procedure volume (i.e., the number of operations performed for ovarian cancer) is associated with increased survival in Japan, and whether centralizing operative treatment for ovarian cancer might improve survival.

Materials and Methods

Data sources. Data on 3523 reported ovarian cancer (ICD Tenth Revision, C56) cases who were newly diagnosed in 1975–1995, and treated in hospitals in Osaka were retrieved from the Osaka Cancer Registry's database; 3030 of these (86.0%) underwent surgery at hospitals. The Osaka Cancer Registry (OCR) has been operating since December 1962, covering Osaka Prefecture, with its population of 8.8 million (2000 census). Cancer incidence data in Osaka have been reported in 'Cancer Incidence in Five Continents' volumes III to VIII.[1] The quality of this data, therefore, can be assumed to have met the standards set by the International Association of Cancer Registries during the last 3 decades. In 1993–1997, the proportion of death certificate only (DCO) cases was 16%, the mortality to incidence (M/I) ratio was 0.62, and the proportion of morphologically verified cases was 73% for ovarian cancer.[1] In the OCR's data processing for cancer statistics, primary facilities for treatment for each cancer were determined and coded according to the following priority: surgery, radiotherapy, transarterial embolization, ethanol injection, chemotherapy, immunotherapy, hormone therapy, laser therapy, and thermotherapy.

Hospitals were ranked according to the number of operations performed on ovarian cancer patients between 1975 and 1995, an approach that has been previously validated.[9–11] Categorization of the hospitals by the volume of operations was carried out by dividing the 3030 operations into 4 quartiles based on the mean annual number of ovarian cancer surgical procedures: 4 high-volume hospitals with a total of 742 operations (average: 8.8 operations per hospital per year), 10 medium-volume hospitals with a total of 832 operations (4.0 operations per hospital per year), 18 low-volume hospitals with a total of 764 operations (2.0 operations per hospital per year), and 175 very-low-volume hospitals with a total of 692 operations (less than one (0.3) operation per hospital per year).

According to the ideas presented by International Agency for Research on Cancer, histologic types of ovarian cancer were classified into five categories: carcinomas, sex cord-stromal tumors, germ cell tumors, other specified cancers, and unspecified cancers. The cancer stage at diagnosis was classified into

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the following three groups: 1) localized: cancer is confined to the original organ: 2) regional: cancer spreads to regional lymph nodes and/or spreads to immediately adjacent tissues; 3) distant: cancer metastasizes to distant organs.

To obtain information on the vital status of registered cases, the OCR has used the following three steps: 1) collation with the annual cancer death file, 2) collation with the annual death certificate file in Osaka, and 3) confirmation of the cases' living status by referring to registers in local municipality offices of inhabitants 5 years after the diagnosis. The final step was, however, not conducted for those residing in Osaka City in 1975–1992. Therefore, survival analysis was restricted to 2450 cases, who lived in Osaka Prefecture (except for Osaka City) in 1975–1995 or resided in Osaka City in 1993–1995, since active follow-up information was available for them.

Statistical analysis. The distributions of patients' characteristics according to hospital volume were assessed with Kruskal-Wallis tests for continuous variables and χ^2 tests for categorical variables. Cumulative observed survival was estimated using the Kaplan-Meier method according to hospital volume. Survival time was computed from the date of first diagnosis to the end-point, defined as death from any cause. Closing date was defined as the date 5 years after the starting point. Fifty-one cases (2.1%) lost to follow-up were treated as censored cases at the latest date when they were confirmed as alive. The log rank tests were used as statistical tests to evaluate the differences between cumulative observed survival curves. The prognostic factors were evaluated by use of the Cox proportional hazards regression model. In this analysis, the dependent variable was vital status 5 years after diagnosis and independent variables were age, histologic type (carcinomas, sex cord-stromal/germ cell tumors, and other specified/unspecified cancers), cancer stage (localized, regional, distant, and unknown) and hospital procedure volume (high, medium, low, and very low). Histologic type was classified into the three categories mentioned above, since our previous study showed similar 5-year survival within each category.[2] The category of other specified/unspecified cancers included cases without microscopic verification. Relative 5-year survival was calculated adjusting for differences in the probability of dying of causes other than cancer among subjects. Relative survival was calculated as the ratio of observed survival to expected survival, the latter being estimated using the survival probability in the general population of Japan of similar subjects with respect to sex, age, and calendar year at diagnosis. The Ederer II method was employed.[12] Differences were considered as statistically significant if P values were less than 0.05 by two-sided test. The SPSS (version 11.0) statistical software package was used for statistical analysis.

Results

In Table 1, the number of hospitals and the patients' characteristics are presented and compared among the four hospital volume groups. In total, 3523 patients with ovarian cancer were treated in 207 hospitals in Osaka. The average age was 52.7 years (standard error (SE) 0.3), though it differed among the groups ($P<0.01$). Patients from the high- and medium-volume hospitals tended to be younger. Surgical treatment also differed somewhat by hospital volume ($P<0.01$). Surgery was performed for 94.6% of the patients treated in high-volume hospitals, but for only 74.2% of the cases treated in very-low-volume hospitals. Complete resection was done for 55.9% of the patients in high-volume hospitals, but for only 24.6% in very-low-volume hospitals. Distribution of histologic types differed by hospital volume ($P<0.01$), and the proportion with no microscopic verification in very-low-volume hospitals (15.9%) was the highest among these groups. The proportion designated as localized was also higher in high-volume hospitals (44.4%) than that in any other volume hospitals.

Table 1. Hospital volume group and characteristics of patients

	Hospital volume group[1]				P value
	Very-low-volume hospital	Low-volume hospital	Medium-volume hospital	High-volume hospital	
No. of hospitals	175	18	10	4	
Total no. of patients	932	880	927	784	
Mean age±SE	56.1±0.5	52.1±0.5	51.0±0.5	51.3±0.5	<0.01
Surgery					<0.01
Done	692 (74.2)[2]	764 (86.8)	832 (89.8)	742 (94.6)	
Complete resection	229 (24.6)	327 (37.2)	483 (52.1)	438 (55.9)	
Incomplete resection	403 (43.2)	383 (43.5)	304 (32.8)	264 (33.7)	
Unknown	60 (6.4)	54 (6.1)	45 (4.9)	40 (5.1)	
Not done	233 (25.0)	114 (13.0)	87 (9.4)	40 (5.1)	
Unknown	7 (0.8)	2 (0.2)	8 (0.9)	2 (0.3)	
Histologic type					<0.01
Carcinoma	639 (68.6)	709 (80.6)	756 (81.6)	705 (89.9)	
Sex cord-stromal tumors	8 (0.9)	14 (1.6)	12 (1.3)	9 (1.1)	
Germ cell tumors	54 (5.8)	48 (5.5)	63 (6.8)	40 (5.1)	
Other specified cancers	6 (0.6)	9 (1.0)	17 (1.8)	6 (0.8)	
Unspecified cancers	225 (24.1)	100 (11.4)	79 (8.5)	24 (3.1)	
Unspecified morphology	77 (8.3)	44 (5.0)	42 (4.5)	15 (1.9)	
No microscopic verification	148 (15.9)	56 (6.4)	37 (4.0)	9 (1.1)	
Cancer stage					<0.01
Localized	142 (15.2)	203 (23.1)	337 (36.4)	348 (44.4)	
Regional	363 (38.9)	368 (41.8)	316 (34.1)	234 (29.8)	
Distant	324 (34.8)	249 (28.3)	187 (20.2)	169 (21.6)	
Unknown	103 (11.1)	60 (6.8)	87 (9.4)	33 (4.2)	

1) High, medium, low, and very low hospital volume averaged 8.8, 4.0, 2.0, and 0.3 operations per hospital per year, respectively.
2) Figures in parentheses are proportions among total number of patients.

Fig. 1 shows 5-year cumulative survival curves of patients with ovarian cancer diagnosed in 1975–1995 by hospital surgical volume. These were significantly different among the hospital volumes (log rank test $P<0.01$). The 5-year survival in high-volume hospitals was estimated as 53.4% (SE 2.0), which was higher than that in medium- (45.1%, SE 2.3), low- (33.3%, SE 1.8), and very-low-volume (21.5%, SE 1.7) hospitals. Similar findings were obtained in cumulative survival curves even if the study subjects were restricted to cases who underwent surgery (log rank test $P<0.01$). The 5-year survival in high-, medium-, low-, and very-low-volume hospitals was estimated as 55.6% (SE 2.1), 49.0% (SE 2.4), 37.3% (SE 1.9), and 28.0% (SE 2.1), respectively.

Table 2 presents relative 5-year survival, crude hazard ratio, and adjusted hazard ratio stratified by various prognostic variables. Figures parenthesized are the results obtained when analysis was restricted to those who underwent surgery. Relative 5-year survival was 38.8% for all subjects diagnosed with ovarian cancer during 1975–1995, and the survival increased with increasing hospital volume. After adjustment for age and other variables using the Cox regression model, we found that the hazard ratio correlated positively with hospital volume (χ^2 test for linear trend: $P<0.01$), and patients receiving care in very-low-volume hospitals had a 60% higher risk of death than patients receiving care in high-volume hospitals. The survival for patients with other specified/unspecified cancers was the lowest among the three histologic types, and the survival for patients with distant stage cancer was also the lowest as compared with other stages of cancer; the adjusted hazard ratios were 1.8 for other specified/unspecified cancers as compared with that for carcinoma, and 9.2 for distant stage as compared with that for localized stage. Similar findings for relative 5-year survival and hazard ratio were obtained even if study subjects were restricted to cases who underwent surgery.

Fig. 2 shows 5-year cumulative survival of germ cell tumor cases by hospital surgical volume. There were only 152 cases with germ cell tumors in Osaka, in 1975–1995; of these 31 cases (20.4%), 43 cases (28.3%), 42 cases (27.6%), and 36 cases (23.7%) were treated in high-, medium-, low-, and very-low-volume hospitals, respectively. Only one case among them did not undergo surgery for germ cell tumor. There were significant differences in the cumulative survival curves among these hospital volumes (log rank test $P<0.05$). The 5-year survival in high-volume hospitals (67.7%, SE 8.4) was higher than that in medium-, low-, and very-low-volume hospitals (the survivals were 59.6%, 65.1%, and 38.9%, respectively). After adjustment for age and cancer stage, hazard rate ratios were 0.8 for medium hospital volume (95% confidence interval (CI) 0.4–1.7), 1.3 for low hospital volume (95% CI 0.6–3.0), and 2.1 for very

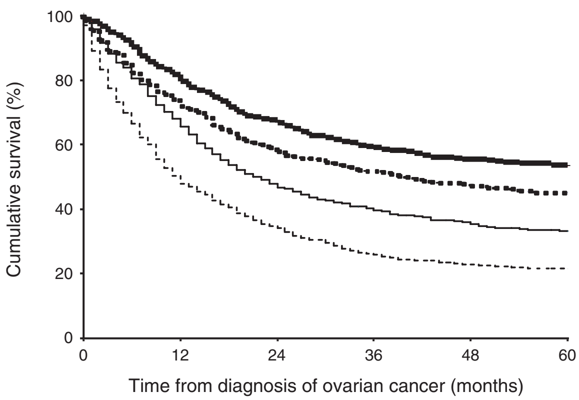

Fig. 1. Kaplan-Meier estimates for cumulative survival of ovarian cancer by hospital case volume in Osaka, Japan. The 5-year survival in high-volume hospitals (thick solid line) was higher than that in medium-volume hospitals (thick dotted line), low-volume hospitals (thin solid line), and very-low-volume hospitals (thin dotted line).

Table 2. Relative 5-year survival and hazard ratio by characteristics of patients with ovarian cancer

	No.	Relative 5-year survival	Standard error	Crude hazard ratio	95% CI	Adjusted hazard ratio	95% CI
Hospital volume							
High	617	55.0	2.1	1.0		1.0	
	(583)[1]	(57.4)	(2.1)	(1.0)		(1.0)	
Medium	481	46.2	2.4	1.2	1.0–1.4	1.1	0.9–1.3
	(439)	(50.2)	(2.5)	(1.1)	(0.9–1.3)	(1.0)	(0.9–1.2)
Low	726	34.2	1.8	1.6	1.4–1.9	1.4	1.2–1.6
	(642)	(38.2)	(2.0)	(1.6)	(1.3–1.8)	(1.4)	(1.2–1.6)
Very low	626	22.3	1.7	2.5	2.1–2.8	1.6	1.4–1.9
	(468)	(28.7)	(2.2)	(2.1)	(1.8–2.5)	(1.6)	(1.3–1.8)
Histologic type							
Carcinoma	1976	40.1	1.1	1.0		1.0	
	(1795)	(43.8)	(1.2)	(1.0)		(1.0)	
Sex cord-stromal/germ cell tumors	180	58.0	3.8	0.6	0.5–0.7	1.1	0.8–1.3
	(177)	(59.0)	(3.8)	(0.6)	(0.5–0.8)	(1.1)	(0.8–1.4)
Other specified/unspecified cancers[2]	294	17.9	2.3	2.2	1.9–2.5	1.8	1.6–2.1
	(160)	(27.8)	(3.7)	(1.6)	(1.3–1.9)	(1.8)	(1.5–2.2)
Cancer stage							
Localized	686	84.1	1.5	1.0		1.0	
	(672)	(84.9)	(1.5)	(1.0)		(1.0)	
Regional	905	25.7	1.5	6.2	5.2–7.4	5.4	4.5–6.4
	(820)	(27.8)	(1.6)	(6.2)	(5.2–7.5)	(5.6)	(4.7–6.7)
Distant	668	10.3	1.2	11.2	9.4–13.4	9.2	7.7–11.0
	(493)	(13.7)	(1.6)	(10.4)	(8.6–12.6)	(8.9)	(7.4–10.8)
Unknown	191	40.5	3.7	4.1	3.2–5.2	3.5	2.7–4.4
	(147)	(50.2)	(4.3)	(3.1)	(2.4–4.1)	(2.9)	(2.2–3.8)

1) Figures in parentheses are results obtained when analysis was restricted to those who underwent surgery for ovarian cancer.
2) Includes 156 cases without microscopic verification.

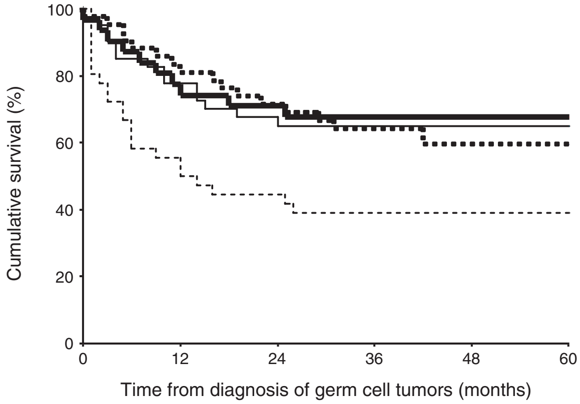

Fig. 2. Kaplan-Meier estimates for cumulative survival of germ cell tumors by hospital case volume in Osaka, Japan. The 5-year survival in high-volume hospitals (thick solid line) was higher than that in medium-volume hospitals (thick dotted line), low-volume hospitals (thin solid line), and very-low-volume hospitals (thin dotted line).

low hospital volume (95% CI 1.0–4.6) as compared with that for high hospital volume, although the differences were not statistically significant.

Discussion

The incidence rate of ovarian cancer has increased during the last few decades in Japan,[13, 14] while the relative 5-year survival has remained approximately constant at a little less than 40%.[2] There are increasing demands for improvement in the treatment of ovarian cancer patients. Our population-based study suggested that increased hospital surgical volume was associated with a better survival for ovarian cancer in Osaka, after adjusting for patient characteristics such as age, histologic type, and cancer stage.

Is the reason for difference in survival by different hospital volume associated with prognostic hospital factors, such as the technical expertise of physicians and of the teams performing primary surgery? The management of ovarian cancer has advanced notably during the 1980s and 1990s and this has been accompanied by increased survival rates. Laurvick *et al.*[5] have reported that the use of specific surgical procedures to treat ovarian cancer (bilateral salpingo-oophorectomy, omentectomy, and lymphadenectomy) has increased dramatically, and that these surgical trends improved relative 5-year survival in the period 1982–1998. Our study indicates that high-volume hospitals had the highest proportion of complete resection of tumors. Our results suggest that the remarkable differences in survival in high/medium/low- and very-low-volume hospital groups might be explained in part by differences in the speed and extent of diffusion of new and effective surgical techniques for treating ovarian cancer in each hospital volume group (e.g., advances in management might have occurred only for high-volume hospitals).

Survival difference by hospital volume was particularly remarkable for ovarian germ cell tumors (67.7%–38.9%), which were expected to show significantly improved survival after the introduction of effective chemotherapeutic regimens, such as the combination of bleomycin, etoposide, and cisplatin. Similar findings were also observed in cases of testicular germ cell tumors in Osaka: 5-year cumulative survivals by hospital volume were 90.2%, 82.0%, 81.3%, and 66.7%, respectively, when categorized into 4 quartiles. These results suggest that there are severe problems with the speed and extent of diffusion of not only effective surgery, but also effective chemotherapy for ovarian cancer in Osaka.

The survival of ovarian cancer patients might be increased by extending the appropriate management and treatment to lower-volume hospitals. However, if we consider the fact that ovarian cancer is still rare in Japan, the patients' survival might be improved more efficiently through centralization of care and treatment in high-volume hospitals. If all ovarian cancer patients had received treatment in high-volume hospitals, the relative 5-year survival would have been expected to improve from 39.0% to 48.1%. (We calculated the latter survival rate by applying the relative 5-year survivals of each cancer stage in high-volume hospitals to the stage distribution of all ovarian cancer in Osaka.) This projected improved survival rate is better than that in the US (43.5%). Under these circumstances, the average number of operations per hospital in high-volume hospitals would increase from just 8.8 to 36.1 per year and the average number of treatment for patients with ovarian cancer per hospital would increase from 9.3 to 41.9 per year.

There are several limitations in this study that should be considered before accepting any of our conclusions. First, survival differences were not analyzed with consideration of co-morbidities: more patients with co-morbidities might have been referred to lower-volume hospitals than to high-volume hospitals. In fact, the proportion of ovarian cancer patients for whom surgery not done was higher in lower-volume hospitals. For these reasons, survival might have been lower in lower-volume hospitals, despite adjustment for cancer stage by using the Cox proportional hazards model. However, similar findings in survival were seen even if study subjects were restricted to cases who underwent surgery. Second, the risk of death in lower-volume hospitals might have been overestimated because of possible so-called stage migration, as well as insufficient adjustment for cancer stage distribution. The classification of the International Federation of Gynecology and Obstetrics (FIGO) is usually used in staging of ovarian cancer which is performed after operation.[15] Thus, if this staging system had been available and used in this study, more reliable results would have been obtained regarding the relation between hospital surgical volume and survival of ovarian cancer. Third, we need to take into consideration differences in completeness of reporting to the cancer registry by various hospitals, because the proportion of DCO cases was 16% for ovarian cancer in the OCR in 1993–1997, which was higher than that in North American or northern/western European registries. Although there is no actual index of this factor, completeness of notification might be expected to be better in high-volume hospitals than in others. Hospitals with poor completeness of notification might show tendencies either of underreporting of patients who are still alive, thus underestimating survival, or of an increased proportion of DCO cases, thus overestimating survival. We need to reanalyze the association between ovarian cancer survival and hospital volume after having achieved satisfactory completeness of notification. Fourth, the present study suggested that increased hospital surgical volume was associated with better survival for ovarian cancer in Osaka by analyzing OCR's database only. It is urgently necessary to ascertain from national population-based cancer registry data whether or not similar problems exist in other locations in Japan.

Despite some limitations inherent in this study, the results suggest that increased hospital surgical volume was associated with a better survival rate for ovarian cancer in Osaka. The authors consider that further centralization of treatments for ovarian cancer patients in high-volume hospitals would be desirable for improving the survival of ovarian cancer patients in Osaka, and that it should be feasible to make this change.

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1. Parkin DM, Whelan SL, Ferlay J, Teppo L, Thomas DB, editors. Cancer incidence in five continents vol. VIII. In: IARC Scientific Publ. no.155. Lyon, France: International Agency for Research on Cancer; 2002.
2. Ioka A, Tsukuma H, Ajiki W, Oshima A. Ovarian cancer incidence and survival by histologic type in Osaka, Japan. *Cancer Sci* 2003; **94**: 292–6.
3. Hacker NF, Berek JS, Lagasse LD, Nieberg RK, Elashoff RM. Primary cytoreductive surgery for epithelial ovarian cancer. *Obstet Gynecol* 1983; **61**: 413–20.
4. Griffiths CT. Surgical resection of tumor bulk in the primary treatment of ovarian carcinoma. *Natl Cancer Inst Monogr* 1975; **42**: 101–4.
5. Laurvick CL, Semmens JB, Leung YC, Holman CD. Ovarian cancer in Western Australia (1982–1998): trends in surgical intervention and relative survival. *Gynecol Oncol* 2003; **88**: 141–8.
6. Oshima A, Kitagawa T, Ajiki W, Tsukuma H, Takenaka S, Iura A. Survival of testicular cancer patients in Osaka, Japan. *Jpn J Clin Oncol* 2001; **31**: 438–43.
7. Williams SD. Ovarian germ cell tumors: an update. *Semin Oncol* 1998; **25**: 407–13.
8. Gershenson DM. Update on malignant ovarian germ cell tumors. *Cancer* 1993; **71**: 1581–90.
9. Begg CB, Cramer LD, Hoskins WJ, Brennan MF. Impact of hospital volume on operative mortality for major cancer surgery. *JAMA* 1998; **280**: 1747–51.
10. Hillner BE. Is cancer care best at high-volume providers? *Curr Oncol Rep* 2001; **3**: 404–9.
11. Kumpulainen S, Grenman S, Kyyronen P, Pukkala E, Sankila R. Evidence of benefit from centralized treatment of ovarian cancer: a nationwide population-based survival analysis in Finland. *Int J Cancer* 2002; **102**: 541–4.
12. Estev J, Benhamou E, Raymond L. Statistical methods in cancer research vol. IV: Descriptive epidemiology. In: IARC Scientific Publ. no. 128. Lyon, France: International Agency for Research on Cancer; 1994. p. 231–45.
13. The Research Group for Population-based Cancer Registration in Japan. Cancer incidence and incidence rates in Japan in 1997: estimates based on data from 12 population-based cancer registries. *Jpn J Clin Oncol* 2002; **32**: 318–22.
14. Tamakoshi K, Kondo T, Yatsuya H, Hori Y, Kikkawa F, Toyoshima H. Trends in the mortality (1950–1997) and incidence (1975–1993) of malignant ovarian neoplasm among Japanese women: analyses by age, time, and birth cohort. *Gynecol Oncol* 2001; **83**: 64–71.
15. Richardson GS, Scully RE, Nikrui N, Nelson JH Jr. Common epithelial cancer of the ovary. *N Engl J Med* 1985; **312**: 415–24.