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ORIGINAL ARTICLE

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Efficacy of adjuvant Immunochemotherapy with polysaccharide K for patients with curatively resected colorectal cancer: a meta-analysis of centrally randomized controlled clinical trials

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Abstract The benefits of immunochemotherapy employing the biological response modifier polysaccharide K (PSK) for patients with curatively resected colorectal cancer was reassessed by means of a meta-analysis of data with center randomization from 1,094 patients enrolled in three clinical trials. In all three trials, patients were followed up for at least 5 years after surgery and enrollment of the last patient and outcomes for standard chemotherapy were compared with those for chemotherapy plus PSK. The endpoints were overall survival and disease-free survival; and intent-to-treat analysis was performed without patient exclusion. Data were analyzed using the weighted average of the individual log hazard ratios. The overall survival risk ratio for all eligible patients was 0.71 (95% confidence interval (CI) : 0.55–0.90; $P = 0.006$), and the disease-free survival risk ratio was 0.72 (95% CI: 0.58–0.90; $P = 0.003$). The results of this meta-analysis suggest that adjuvant immunochemotherapy with PSK can improve both survival and disease-free survival of patients with curatively resected colorectal cancer.

Keywords Adjuvant immunochemotherapy · Colorectal cancer · Meta-analysis · Polysaccharide K

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Introduction

Colorectal cancer is the third most common cancer in the world (9.4% of all new cancers) with two-thirds of cases occurring in developed countries [1]. In 2000, an estimated 94,500 new cases were diagnosed worldwide with 492,000 deaths (fourth highest after lung, stomach, and liver cancer). In Japan alone, the number of deaths was 80,000 in 2000; and is expected to climb to 110,000 by the year 2020.

At present, surgery and adjuvant chemotherapy with 5-FU/ Leucovorin/ Oxaliplatin have become the standard treatment for curatively resected colon cancer in Western countries [2]. In Japan, on the other hand, the efficacy of oral fluorinated pyrimidines in an adjuvant setting was demonstrated in a meta-analysis of randomized trials [3, 4]. Furthermore, the addition of immunotherapy to adjuvant chemotherapy can be expected to exert a synergistic effect [5]. One of the immunotherapy agents is polysaccharide K (PSK; Kureha Chemical Industry Co., Ltd., Tokyo, Japan), extracted from mycelia of the Coriolus versicolor strain CM-101, a member of the basidiomycetes [6]. It consists of proteins and polysaccharides, and its major glycoside portion is β-1, 4-gulcan with an average molecular weight of approximately 100 kDa. PSK is a potent inducer of gene expression for several interleukins, tumor necrosis factor, and monocyte activating factors (MCP-1). The antitumor activity of PSK is thought to be based on its regulation of the host immune response, and this nonspecific immunopotentiator is able to inhibit the immunosuppression of cancer patients. In Japan, it is

widely prescribed for oral ingestion [7]. The beneficial effects of PSK have been attributed to the activation of leucocyte chemotactic locomotion and phagocytic activity [8]. Adjuvant chemotherapy with PSK and oral 5-FU, subsequent to intravenous mitomycin C (MMC), significantly increased the disease-free survival rate as compared with 5-FU alone in patients with curatively resected colorectal cancer [9]. It has been confirmed that PSK is more effective in the treatment of gastric cancer when intermittently administered followed by alternating chemotherapy [10]. However, the immunochemotherapeutic efficacy of PSK plus chemotherapeutic agents for colorectal cancer remains unclear.

The aim of the study presented here was to evaluate the combined data from published and unpublished randomized clinical trials for a comparison of the effect of adjuvant immunochemotherapy using PSK with that of chemotherapy alone for patients who had undergone curative resection of colorectal cancer.

Materials and methods

Literature search

In December 2004, a search of the MEDLINE database was performed and inquiries were made from the pharmaceutical industry regarding clinical trials published since 1980, with an abstract in either English or Japanese, of polysaccharide-K (PSK), or Krestin used for colorectal cancer patients. The collected reports were summarized independently by two of the authors using a standard form. The two resultant summaries for each report were compared to ensure there were no discrepancies. The following questions were used to decide the eligibility of the trial: (1) Was the aim of the study the evaluation of the effect of adjuvant chemotherapy with or without PSK administration? (2) Was it a randomized trial with central assignment to avoid bias? (3) Was adjuvant therapy administered after curative tumor resection? (4) Was there a control arm that received the same adjuvant chemotherapy as the therapeutic arm?

The adjuvant chemotherapy regimens, the number of patients in each arm, the number of deaths in each arm, and the follow-up period were recorded for each trial selected on the basis of these four questions.

Table 1 The list of potentially relevant papers in this meta-analysis

Authors	Excluded reasons
Saji et al. [33]	*Without a randomized control arm*
Saeki et al. [42]	*Dealt with non-resectable cases*
Nakazato et al. [53]	Included (retrieved for detail evaluation)
Matsunaga et al. [14]	*Pre-clinical study*
Yasutomi et al. [27]	*Review article*
Nishizawa et al. [43]	*Dealt with non-resectable cases*
Takashima et al. [64]	Included (retrieved for detail evaluation)
Mitomi et al. [55]	Included (retrieved for detail evaluation)
Kimura et al. [31]	*Randomization with other modalities*
Ishigaki et al. [44]	*Dealt with non-resectable cases*
Torisu et al. [8]	Included (retrieved for detail evaluation)
Masayoshi et al. [45]	*Dealt with non-resectable cases*
Mitomi et al. [56]	Included (retrieved for detail evaluation)
Ebina et al. [12]	*Pre-clinical study*
Nio et al. [36]	*Examined immunologic parameters as the endpoint*
Hayashibe et al. [37]	*Examined immunologic parameters as the endpoint*
Toshino et al. [46]	*Dealt with non-resectable cases*
Mitomi et al. [9]	Included (retrieved for detail evaluation)
Kanoh et al. [15]	*Pre-clinical study*
Suo et al. [16]	*Pre-clinical study*
Kaneoka et al. [47]	*Dealt with non-resectable cases*
Noguchi et al. [17]	*Pre-clinical study*
Ebina et al. [18]	*Pre-clinical study*
Kobayashi et al. [6]	*Review article*
Sugimachi et al. [38]	*Examined immunologic parameters as the endpoint*
Sugiyama et al. [19]	*Pre-clinical study*
Harada et al. [13]	*Pre-clinical study*
Kamei et al. [20]	*Pre-clinical study*
Ikeda et al. [48]	*Dealt with non-resectable cases*
Ogihara et al. [21]	*Pre-clinical study*
Matsunaga et al. [22]	*Pre-clinical study*
Sakamoto et al. [28]	*Review article*
Matsunaga et al. [23]	*Pre-clinical study*
Kidd et al. [29]	*Review article*
Mukai et al. [49]	*Dealt with non-resectable cases*
Kudo et al. [50]	Included (retrieved for detail evaluation)
Okuzawa et al. [44]	*Pre-clinical study*
Fisher et al. [30]	*Review article*
Munemoto et al. [34]	*Without a randomized control arm*
Shibata et al. [39]	*Examined immunologic parameters as the endpoint*
Ohwada et al. [51]	Included (retrieved for detail evaluation)
Wada et al. [25]	*Pre-clinical study*
Koda et al. [32]	*Randomization with other modalities*
Ito et al. [52]	Included (retrieved for detail evaluation)
Ohwada et al. [41]	Included (retrieved for detail evaluation)
Yoshikawa et al. [26]	*Pre-clinical study*
Munemoto et al. [25]	*Without a randomized control arm*
Alliot et al. [40]	*Letter commenting*

Statistical methods

We used the weighted average of the individual log hazard ratios to analyze pooled estimates of the common relative risk and its confidence interval [11]. The chi-square test was used to test heterogeneity among the individual hazard ratios.

For each individual trial, the log hazard ratio and its variance were calculated from the number of observed events, the number of randomized patients, and the *P*-value for the log-rank test obtained with the indirect log hazard ratio and variance estimation method [11]. A relative risk of 1.0 indicated no effect of immunochemotherapy on crude mortality and recurrence of disease, whereas a ratio of less than 1.0 meant a beneficial effect of immunochemotherapy, and a ratio above 1.0 meant a harmful effect. SAS for Windows, release 8.02 (SAS Institute Inc., Cary, NC, USA) was used for all analyses.

Table 2 Randomized clinical trials using PSK in patients with colorectal cancer

Authors	Randomization	Patient accrual	Treatment/control arms	No. of patients		Conclusion
				With PSK	Without PSK	
Nakazato et al. [53]	Envelope	1977.07–1980.06	esquinon ± PSK	75	36	NS, $P = 0.018$ in subset (Dukes'C & long(> 6weeks) administration)
Takashima et al. [54]	Envelope	1980.09–1983.12	MMC → tegafur supp. ± PSK	53	71	NS $P < 0.05$ in ly (+) subset
Mitomi et al. [55]	*(see article no.56)*					
Torisu et al. [8]	Double blind	not described	PSK only vs. placebo	56	55	$P < 0.05$
Mitomi et al. [9]	*(see article no.56)*					
Mitomi et al. [56]	Envelope	1985.03–1987.02	MMC → oral 5-FU ± PSK	221	227	$P = 0.0325$(OS), 0.0302(DFS)
Kudo et al. [50]	Not described	1995.09–1997.11	oral fluoropyrimidine ± PSK	48	10	$P = 0.0467$ in DFS, (OS; not described)
Ohwada et al. [51]	*(see article no.41)*					
Ito et al. [52]	Center	1991.02–1993.03	5-FUciv → oral 5-FU ± PSK	220	221	NS (cancer death; significant)
Ohwada et al. [41]	Center	1994.10–1997.03	MMC → oral UFT ± PSK	137	68	DFS $P = 0.016$ (OS; $P = 0.056$)

Results

Review and evaluation of retrieved reports

The 48 papers identified as potentially relevant are summarized in Table 1, together with the reasons for exclusion. Screening of the 48 selected abstracts for relevant trials yielded 10 reports that qualified for retrieval. Of the remaining 38 abstracts, 12[12, 13, 14–26] were of preclinical studies, five [6, 27–30] of review articles, two [31] and [32] of studies using randomization with other modalities (adjuvant chemotherapy [31] or preoperative radiation [32]) than PSK administration, three [33–35] of adjuvant immunochemotherapy studies without a randomized control arm, four [36–39] of studies that examined immunologic parameters as the endpoint. In addition, one [40] was a letter commenting on an article [41], and eight articles [42–49] dealing with non-resectable cases.

Relevant trials

The references of the ten qualifying articles were also reviewed (Table 2). The quality of the study design and the reporting of the results were evaluated, including protocol therapies of both—PSK and control arm—the method of randomization, the definition of ineligible patients, and the number of exclusions and subjects lost to follow-up. Six reports were published in English [8, 9 and 50–52], and four in Japanese [53–55 and 56]. A comparison of the number of the cases and the period of study, indicated that three articles [55, 9, 56] seemed to deal with the same study, while two [51, 41] appeared to be descriptions of the same cases. Three reports [8, 53, 54] were considered to be inadequate for this meta-analysis, because of comparison with a surgery alone group [53], with suppository usage of tegafur [54], or because it was an analysis of simple immunotherapy without combination with chemotherapy [8]. One study [50] did not seem to be randomized. Eventually, three studies [9, 51, 52] covering 1,094 patients were analyzed. In these studies, the effect of immunochemotherapy including PSK and oral fluorinated pyrimidines was compared with that of chemotherapy alone.

In each of these three trials, the chemotherapy and immunochemotherapy arms were balanced with regard to the number of patients 1:1 [9, 51], 1:2 [52]. Although there were slight differences in the control chemotherapy regimen used for the three trials, chemotherapy usually consisted of induction with mitomycin C plus long-term administration of oral fluorinated pyrimidines. In all trials, the effect of immunochemotherapy including PSK was compared with that of chemotherapy alone.

Results of meta-analysis

Survival duration for all three trials was calculated starting from the date of the colorectal cancer operation.

It can therefore be assumed that the combined survival odds ratio of the three trials is well synchronized and appropriate. For our meta-analysis, only the results for at least 5-year survival were used. For one trial [41], the maximum follow-up time was 7 years.

The 5-year overall survival curves are shown in Fig. 1. Survival rates were calculated based on the published results of the three trials [9, 52, 41]. After 5 years, 121 of the 578 patients allocated to the immunochemotherapy group had died (21.0%; survival rate: 79.0%) and 143 of the 516 patients allocated to chemotherapy alone had died (27.8%; survival rate: 72.2%).

The survival odds ratios and 95% confidence intervals (CIs) calculated for the qualifying trials are shown in Fig. 2. The odds ratio for the immunochemotherapy group was not significantly different from that for the chemotherapy group in two of the individual trials. The test for heterogeneity of the data yielded $P = 0.50$, thus lending strong support to the hypothesis of homogeneity. Combining the data for the trials yielded an estimated common odds ratio of 0.71 with a 95% confidence interval (CI) of 0.55–0.90, and a significant effect of PSK ($P = 0.006$).

The 5-year disease-free survival curves are shown in Fig. 3. After 5 years, recurrence was noted in 161 of the 578 patients allocated to the immunochemotherapy group (27.8%; survival rate: 72.2%) and 176 of the 516 patients allocated to chemotherapy alone (34.1%; survival rate: 65.9%).

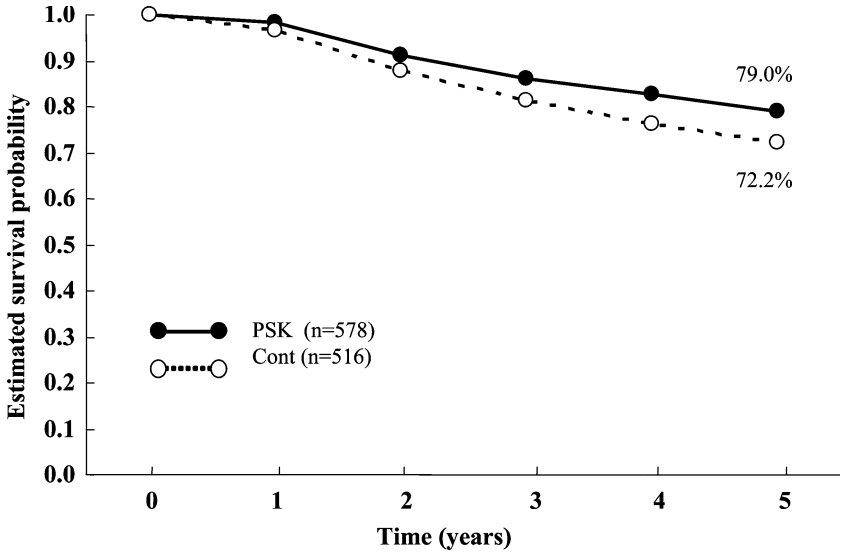

Fig. 1 Five-year overall survival curves for all eligible patients in the immunochemotherapy group (*solid line*) and chemotherapy group (*dotted line*)

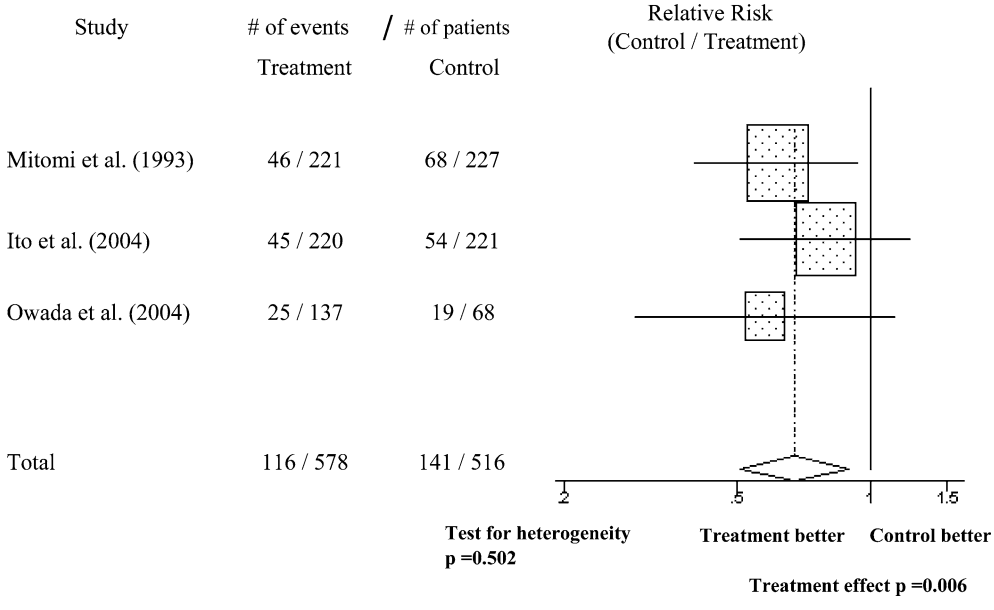

Fig. 2 Survival odds ratios and their 95% CIs for each trial and overall (test for treatment effect $P = 0.006$)

The disease-free survival odds ratios and 95% CIs calculated for the qualifying trials are shown in Fig. 4. The odds ratio for the immunochemotherapy group was not significantly different from that for the chemotherapy group in one of the individual trials. The test for heterogeneity of the data yielded $P = 0.22$, i.e., similar support for the hypothesis of homogeneity as in the case of survival. Combining the data for the trials yielded an estimated common odds ratio of 0.72, with a 95% CI of 0.58–0.90, and a significant effect of PSK ($P = 0.003$).

Discussion

Immunotherapy is added to adjuvant chemotherapy in an attempt to overcome the immunosuppression caused by chemotherapeutic agents as well as to exert a synergistic effect. The combination of fluorinated pyrimidines with the immunotherapeutic agent OK-432 has proved to be effective as adjuvant chemotherapy for both gastric cancer and lung cancer [57, 58]. In the case of colorectal cancer, the combination of 5-FU with levamisole (a biologic modifier) was also confirmed to be significantly more effective for Dukes' C patients after curative resection [59].

PSK is a nonspecific immunopotentiator and exerts its immunomodulatory action by inducing the production of interleukin-2 and interferon-γ, thereby stimulating lymphokine activated killer cells (LAK) and enhancing natural killer cells [12, 13, 60–65] . PSK also has a favorable effect on the activation of leucocyte chemotactic locomotion and phagocytic activity [8].

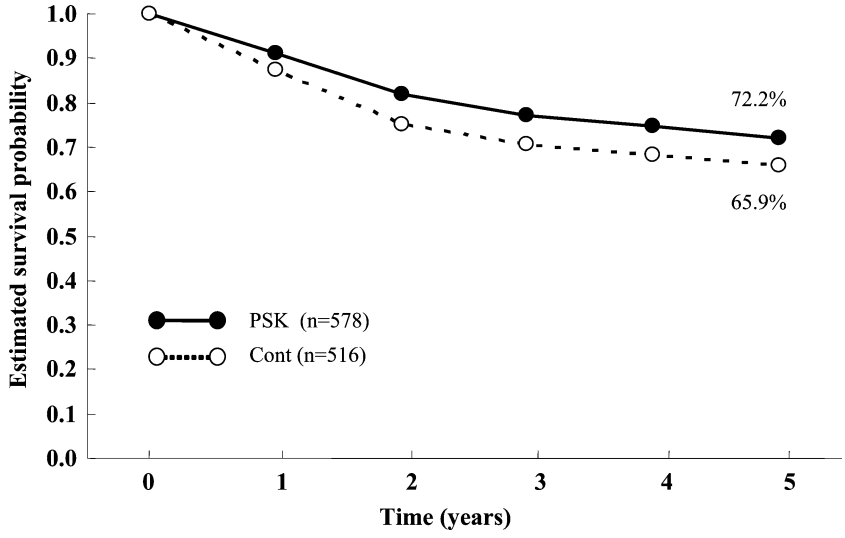

Fig. 3 Five-year disease-free survival curves for all eligible patients in the immunochemotherapy group (*solid line*) and chemotherapy alone group (*dotted line*)

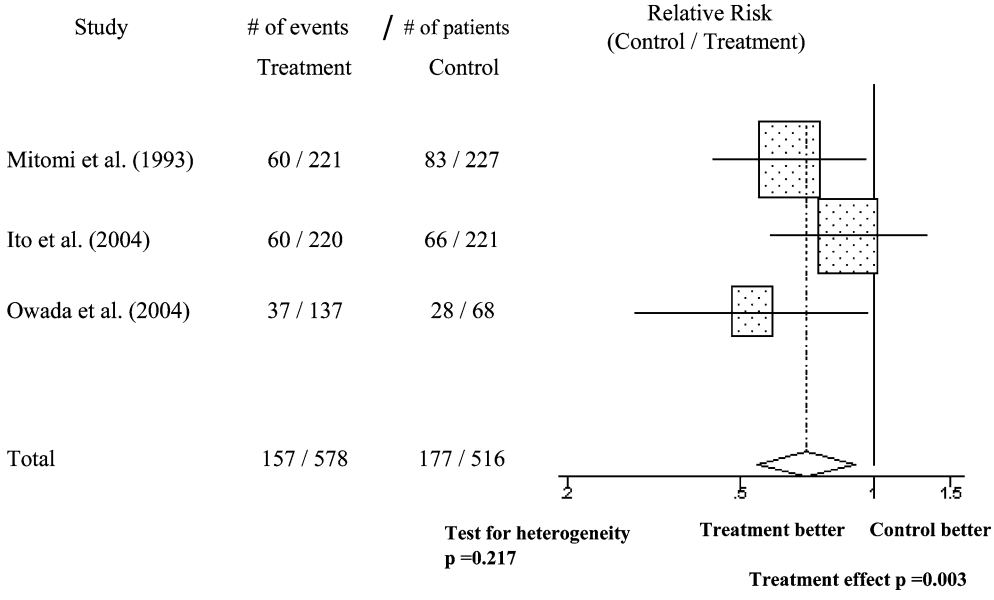

Fig. 4 Disease-free survival odds ratios and their 95% CIs for each trial and overall (test for treatment effect $P = 0.003$)

For the meta-analysis reported here, we focused on the effect of adding PSK therapy to adjuvant chemotherapy after curative resection of colorectal cancer. In Japan, oral fluorinated pyrimidines have been developed as standard chemotherapy for colorectal cancer. Recently reported results have shown a favorable effect of oral fluorinated pyrimidine therapies, such as oral FU, which uses tegafur, tegafur-uracil (UFT), carmofur and 5'DFUR, over surgery alone, and a meta-analysis demonstrated a significant benefit of the treatment with oral fluorinated pyrimidines compared to surgery alone for resectable tumors of the colon and rectum [3, 4]. However, the same type of meta-analysis regarding the immunochemotherapeutic efficacy of PSK plus chemotherapeutic agents for colorectal cancer has not been published. Therefore, we assessed a number of randomized clinical trials designed to examine the effect of adding PSK to oral fluorinated pyrimidines.

Although a considerable number of trials have been performed to compare the effect of immunochemotherapy combined with PSK with that of chemotherapy alone, not all of these studies have been published. In an effort to broaden our search, we therefore also screened presentations made at conferences and made inquiries of doctors performing clinical trials as well as of pharmaceutical industry representatives. As a result, we succeeded in detecting a number of trials meeting our selection criteria. The final three trials were selected by checking the study procedures, especially in terms of central randomization and completeness of the data. Among the individual clinical trials performed so far, only one [9] has shown a survival advantage, while none of other trials demonstrated a significant effect of PSK.

However, combining data from the three relevant studies revealed an odds ratio of 0.71 for survival and of 0.72 for disease free survival, so that our meta-analysis indicated a significant improvement in survival as a result of immunochemotherapy with PSK ($P = 0.006$ for survival and 0.003 for disease-free survival). This improvement may be not only statistically but also clinically significant. A reduction of the death rate by 29% and of recurrence by 28% could be substantial and may justify the inconvenience and financial burden of long-term administration of PSK.

It is difficult to explain why the combination therapy with PSK and oral fluorinated pyrimidines is more effective for curatively resected colorectal cancer. Two largescale clinical trials showed the effectiveness of levamisole plus fluorouracil as adjuvant immunochemotherapy [5, 66]. While oral fluorinated pyrimidine has already been approved as the standard adjuvant chemotherapy in Japan, an even higher efficacy as a result of the addition of immunopotentiator PSK could open new possibilities for a better therapeutic modality for colorectal cancer. Since PSK, OK-432 and levamisole all have similar immunomodulatory effects, but are not necessarily effective as monotherapy against malignancies, the effect of immunochemotherapy with PSK after curative resection of colorectal cancer may be the result of restoration of immunity in patients who show immunosuppression because of surgery and subsequent chemotherapy. Our previous findings from experimental and human studies also support this concept [67, 68].

In conclusion, the results of the meta-analysis presented here suggest that addition of PSK to the standard oral fluorinated pyrimidine based chemotherapy used in Japan offers a significant advantage over chemotherapy alone, in terms of both overall and disease-free survival for patients with curatively resected colorectal cancer. A meta-analysis of individual patient data is now in progress which is expected to provide more detailed and useful information regarding the effects of immunochemotherapy with PSK.

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