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FROM TRANSLATIONAL RESEARCH TO A LARGE RANDOMIZED CLINICAL TRIAL: A LONG AND STREANUOUS WAY FROM BENCH TO BEDSIDE

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ABSTRACT

Several complicated steps are should be necessary to bring bioscientific products successfully to market. Products generated on the bench first have to be called screened for safety in a Phase I clinical trial. This step is called translational research, indicating that the material must be proven safe not only for experimental animals, but also for human subjects. Once the Phase I study is completed, Phase II determines the safety of the bioscientific products on humans, establishing both its maximum tolerated dose and recommended dose. In a Phase II study, the material should be tested for its efficacy against certain diseases. Once it is found to be effective, a comparison between the current actual standard therapy and the new therapy using the newly developed material will be implemented in a large or against scale randomized trial, after which a final decision must be made for its approval as a new drug. The authors have illustrated the steps of: a Phase I translational research by showing the data of a monoclonal antibody A33; a Phase II trial with several combination chemotherapies of chemotherapeutic agents; a Phase III clinical trial through a comparison of antihypertension agents to show the evolutionary process from translational research to a large randomized trial based on their own experience.

Key Words: Translational research, Clinical trials, Large-scale randomized clinical trial.

INTRODUCTION

There is no doubt that bioscience will be the leading industry in developed countries over the next five to ten years. At the moment, however, only six countries (i.e., USA, Japan, UK, France, Germany, and Switzerland) have the ability to produce promising bioscientific materials. Unlike automobiles or refrigerators, the characteristic feature of bioscientific products is that researchers must first assess their direct and indirect effects on human beings. In this regard, any type of bioscientific product should be carefully examined for its acute and chronic toxicity as well as its novel efficacy in humans.

Since the ultimate target of a new scientific material is human beings themselves, it is essential that carefully prepared authentic clinical trials should be conducted to examine its benefits as well as any possible risks to human health. Regardless of whether answer a new substance is destined for diagnostic use or for actual treatment, it is necessary that however promising material appeared to be in the laboratory, it must be verified by those steps of clinical trials. Those new products should be investigated for safety in Phase I trial, efficacy in Phase II, and should be

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compared with pre-existing standard treatments in Phase III trials.

In the present manuscript, such basic and clinical research as an the authors themselves directed and were involved in, are described and presented as an example of successful large scale randomized trial of antihypertensive drug conducted at the Kyoto University EBM Collaborative Research Center, where he also directed the practical implementation of an additional large randomized clinical trial.

How to Introduce Bioscientific Products from Bench to Bedside

When thinking about the possible clinical applications of a material invented in the laboratory for clinicaluse, the first step should be the confirmation of its safety. Working as a research fellow in the Department of Tumor Immunology in the Memorial Sloan-Kettering Institute from 1981 to 1985, the authors discovered three tumor-related antigen systems as well as the monoclonal antibodies that specifically recognize them, after screening over 8000 hybridomas that produce various kinds of antigens. Two antigens against the Lewis blood group antigen system, i.e. Lewis^{a} and Lewis^{b}, were highly useful in distinguishing the "secretor" and "non-secretor" of the Lewis antigen in humans.[1)] In terms of diagnosis, these tumor-related antigens were confirmed to be strongly expressed in colon cancer and gastric cancer.[2,3,)] Despite these new findings, both of these antigens proved to be not applicable to the immunological treatment of cancers.

The third antigenic system the author discovered was totally different from the above blood group-related antigens. This antigen A33, was not soluble in blood. Was a heat stable glycoprotein, and structurally, resembled one of the immunoglobulin superfamily. Immunohistochemical analysis revealed that this antigen was expressed on the surface of the small and large intestine, but was not present in the intracellular substances of the normal cells. However, A33 was abundantly expressed in the cytoplasm and perinuclear region of the colorectal cancer cells,[4)] and monoclonal antibody anti-A33 was considered to be one of the leading candidates for the immunotherapy of colorectal cancer.

After mass production and purification of the antibody, it was tested for contamination of any type of human and mouse viruses such as C-particles. The purified antibody was tested for its toxicity in mice, rats, rabbits, and horses. One to ten doses of the LD_{10} from the most susceptible animal was administered to the colorectal cancer patients in the Phase I clinical trial as a novel investigational drug (IND) be tasted in the United States.

The Phase I study and radioimmuno-localization test on the labeled antibody to the tumor site specifically by a single injection,[5)] demonstrated the absolute safety of IND A33. Following Phase I/II trials implemented by multiple injections of raw antibodies, however, human-anti-mouse -antibody was detected in the patient's blood. To solve that problem, humanized A33 antibody (hu A33) was established through several stages of genetic engineering.

At present, larger Phase I/II studies have been conducted in the United States and Australia using a more refined huA33.[6)] In Japan, a phase I radioimmuno–localization clinical trial against locally advanced cancer using huA33 was conducted in collaboration with Gunma University, and accumulation of huA33 was confirmed in the gastric tumor site of half of patients, especially those with in diffuse type tumors[7)]. We are now preparing confirmatory Phase I/II clinical trials of huA33 against gastric cancer and for gastric cancer-associated ascites fluid.

Investigator-oriented Phase I clinical trials are more popular in the field of combination chemotherapy for cancers. Trials have been implemented to investigate the efficacy of combination chemotherapy using TS-1 and Paclitaxel,[8)] Paclitaxel and 5-FU,[9)] Cisplatin and Paclitaxel,[10)] and a triple combination of 5FU, Paclitaxel, and Cisplatin.[11)] Those clinical studies are of substantial importance in evaluating the safety of such combination therapies. Those Phase I clinical trials should prove useful in determining both the maximum tolerated dose and recommended dose

(RD) for future Phase II studies to evaluate the efficacy of the treatment.

Assessment of Efficacy in Phase II Clinical Trial

Once the safety of a new drug, new treatment modality, or a new combination of therapies has been corroborated by the Phase I trials, and RD for each treatment has been determined, the next step is a Phase II study to evaluate their efficacy. We have performed Phase II studies on Capecitabine in registration trials for colorectal and gastric cancer,[12,13] and have obtained a favorable response to it. With regard to the investigator-oriented trial, a combination chemotherapy of 5'DFUR and Irinotecan was tested for advanced colorectal cancer, and was demonstrated to be safe, showing a response rate as high as 55%, which is comparable to other more toxic regimens.[14] The rate of Paclitaxel-5FU Phase II trial, and a Cisplatin-Paclitaxel trial have already been submitted to journals are expected to be published in the near future. A TS-1 and Paclitaxel trial is now ongoing, and results are anxiously awaited.

Evaluating Benefits of New Treatment in Phase III Study in Comparison with Best Current Treatment

Even though a high response rate might be obtained in a Phase II clinical study, basing a decision on a single arm trial poses several problems. For instance, a porta-caval shunt that had been routinely used to ameliorate esophagogastric bleeding from liver cirrhosis in the United States showed significant benefits in 10 out of 15 Phase II clinical trials. However, once a comparison was done between the porta-caval shunt and other supportive care measures in 6 Phase III trials, all of those trials demonstrated any beneficial impact of the shunt operation on patient survival. After the publication of these results, the porta-caval shunt was abandoned as a treatment for esophageal varices caused by liver cirrhosis. This example clearly demonstrates that the response rate obtained in a Phase II study is not an ultimate proof of efficacy compared to other existing treatment modalities. Any Phase III trial should be implemented by comparing the newly developed treatment with the pre-existing best standard treatment of care for a disease.

The comparison of two or more treatments in a Phase III study is regulated by a very strict rule. At present, such a comparison was stipulated to be performed by randomization, and sample-size analysis and other analytical methods shad to be specified before the trial to avoid intentional subset analysis or intermediate analysis during treatment and follow-up. An assessment of the results of a randomized Phase III trial was originally mandated to be performed by an intention-to-treat analysis. The author had managed a Phase III trial comparing chemotherapy and immuno- chemotherapy and has succeeded resulting demonstrating that the addition of immuno- therapy could bring about a significantly superior prognostic impact over chemotherapy alone.[15] The expression "evidence-based medicine (EBM)" has currently become popular even among those who do not clearly recognize what it implies. In western countries, the results from large randomized Phase III trials, and/or the meta-analysis of randomized trials are now acknowledged to be EBM. In recognition of that consensus, the scale of Phase III randomized trials has grown over, and larger, and the trend has been extended to evaluate the treatment of various kinds of the diseases.

Large Randomized Clinical Trial in Japan

To detect modest but humanly worthwhile differences between treatments or diagnostic methods, a single large randomized trial is now considered to be the best modality. Where it is impossible to carry outs such a trial, a meta-analysis of several small size trials could compromise the flaw in certain rare morbidities, a single large randomized trial, like in pancreatic cancer or in pediatric leukemia.

At present, though Japan is recognizing the value of large randomized trials, single institutional trials, or university-directed trials remain the mainstream in Japan. However, such old-fashioned trials will eventually be abandoned as being inadequate to produce any useful clinical information in terms of the benefits or disadvantages. Therefore, the author can see no future in the concept of single institution or university-driven clinical trials.

Despite those major obstacles, some large randomized trials have been planned and started in Japan. In the field of lung cancer, Japanese investigators have confirmed the significant efficacy of oral fluorinated pyrimidine – UFT for curatively resected early lung cancer.[16)]

In the EBM Collaborative Research Center (EBM center), where the author has been working since 2001, a large randomized Phase III trial was counducted. In this trial, called the Candesartan Antihypertensive Survival Evaluation in Japan (CASE-J), high-risk hypertensive patients were allocated to either an angiotensin II antagonist receptor (Candesartan) or a calcium blocker (Amlodipine). This CASE-J trial is a prospective, multicenter, randomized, open-labeled, active-controlled, two-arm parallel group comparison with a response-dependent dose titration and blinded assessment of endpoints. The subjects are high-risk hypertensive patients treated with either of the two above-mentioned drugs.[17)]

The essentials required to implement and accomplish such large scale randomized trials are: well designed systems and protocols; abundant funding; enthusiastic recruiting of participating physicians; and training of clinical research coordinator (CRC) staffs that support the doctors who registered their patients for the trial. The author was appointed managing director of the EBM center, and the practical head of the steering committee of this CASE-J trial. He was also involved in and responsible for the management and allocation research funding that amount to 3,000,000,000 yen, the recruitment of physicians as well as from throughout Japan, the development of an electronic data capturing system of methods to accelerate the accrual, proper follow-up, and the training and education of the CRCs and data managers in the EBM center. A description of each activity follows.

Funding

To utilize the funding in a decent way is one of the most important elements in managing a large-scale trial. Funding for the CASE-J trial was provided by Takeda Chemical Industries, etd., and the office staff of Kyoto University has been inspecting it's legislate and proper use. Each participating physician contracted with Kyoto University to receive 40.000 yen for registration expenses and each semi-annual report that accounted to be more than seven times. Therefore, participating doctors were supposed to receive a total of at least 280,000 yen per registered patient. This remuneration was made sufficiently generous to attract physicians to enroll their patients in the trial. In addition, the development of a new electronic data capturing system cost over 200,000,000 yen, including the additional purchase of the computers that accounted for another 300,000,000 yen in total. Salaries for over 10 CRCs and data managers also called for a great deal of money. Moreover, to remind patients of their periodical examination, and to help doctors who were reluctant to write reports, we sent our CRCs to the doctors' offices. Their travel expenses and additional incentive for those CRCs who visited all over Japan, also cost a great deal.

Promotion of registration for clinical trial

To successfully accomplish and lead a clinical trial, the preliminary hurdle is the recruiting of eligible patients. The eligibility criteria for the CASE-J study were: 1) age between 20 and 85 years; 2) systolic blood pressure of >140 mmHg or >160 mmHg depending on the patient's age or diastolic blood pressure of >90 mmHg; and 3) presence of at least one of the high-risk

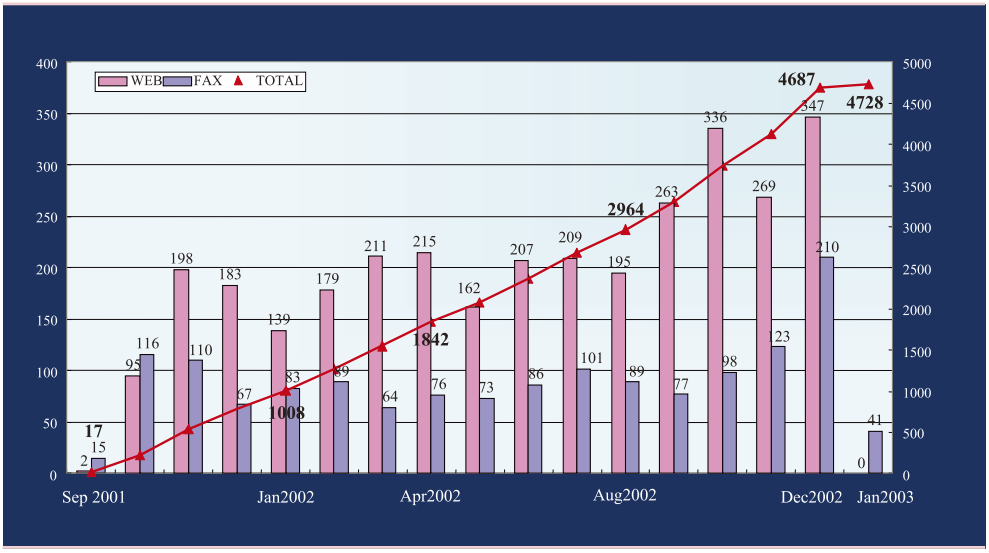

Fig. 1 Cumulative number of patients from the randomization

factors.[17] The first step is to hold orientation meetings in many regions of Japan. These meetings, allow participating doctors to become aware of the importance of the CASE-J trial, as well to acfuaine them with the remuneration for registering each patient. The second step is the leasing of special computers for electronic registration. Nearly 600 computers were distributed to the physicians who agreed to participate in the study. Some computers were retrieved from doctors who failed to register any patients, by the end of the accrual period. Doctors who had registered patients were allowed to keep the computers until the end of the study in order to record follow-up data or to receive information from the EBM center. Since by the end of the study the depreciation of those computers makes them practically worthless the EBM center dose not make the retrievd all of the computers compulsory. The third step is communication with the drug companies who are handling the medicine allocated to the control arm. The EBM center representatives, Dr.Fukui, and Sakamoto, have visited Pfizer and Sumitomo Pharmaceutical Industries to explain the purpose of the clinical trial and to reassure them of joint ownership of any kind of information, especially with regard to toxicity. The last step was restrained efforts for the accrual of participating doctors. As study manager, I traveled around Japan, visiting many individual hospitals and doctors to persuade them to join the CASE-J trial.

Through the above we eventually succeeded in accumulating 4728 cases in 16 months (Fig. 1). That number of patients and the speed of accrual are comparable with those in similar trials performed in the United States or Europe.

Accumulation of follow-up report and event evaluation

Unlike cancer clinical trials where the endpoints are comparatively obvious, verification and confirmation of important events marking for endpoints in such trials as ours are not easy to confirm; e.g., events such as angina or a transient ischemic attack of the brain might simply disappear after a certain period of time, With no way of reassuring a recurrence. Therefore, it is crucial that participating physicians periodically examine registered patients. To encourage their compliance, we have established an automated reminder system. Physicians receive a first

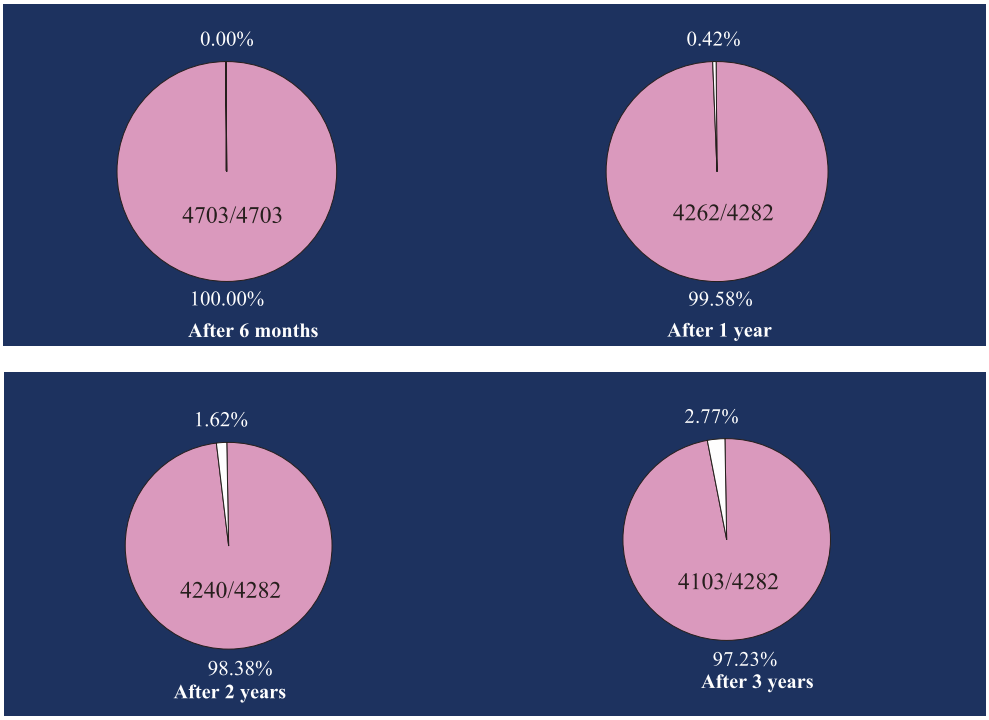

Fig. 2 Scheduled Follow-up rate in CASE-J

reminder 2 months and another month before the next visit of a patient, to reconfirm whether the necessary examinations will have been planned by the time the patient visits the clinic or hospital. Reminders are still sent to the doctors at the time of patient visit, and if data was not loaded on to the electronic data capturing system at the EBM center, they are sent follow-up reminders one 2 and 4 weeks, respectively, after the expected time of the patient visit.

If the EBM center did not receive a sufficient periodical report, the CRC, who is in charge of the patients (one CRC has charge of 600 to 700 patients), first sends an e-mail to the doctor, and then a FAX to remind him/her about the semi-annual reports. If a doctor is the report, the CRC in charge makes an appointment to visit the doctor's hospital or clinic and fill out the data instead of the doctors. Some doctors do not respond to phone calls queries by CRCs, or EBM Center representations, including the author. Thus, one must cold the doctor, make an appointment, then visit him or her with the CRC to complete the case report form.

As for the event verification, a similar process was implemented to collect the necessary data. To avoid the preoccupations and biases of the event evaluation, the EBM Center sends all data to an Event Evaluation Committee (Chairperson; Dr. Fukiyama). Four members of the Committee discuss the situation by e-mail, and a final recommendation is sent to the EBM Center. Likewise, an independent data monitoring committee operates in a similar way so that the EBM Center can be kept in a blind-data-handling condition.

To address the various problems inherent in a randomized trial of the kind, the number of CRC was increased from 0 at the time of the start of the study to 10 at the end of the follow-up. Three additional data managers and clerks were also hired to handle of the payment and business

administration details of the trial. Thanks to these tremendous efforts and funding collections, not only the accrual but also the follow-up rates might well have surpassed the world standard[19] (Fig. 2). The results of this trial confirmed that of the new drug was in no way superior to the current therapeutic agents.

DISCUSSION AND CONCLUSIONS

As can be understood from this report the human application of bioscientific products requires long and strenuous procedures. However, although such procedures seem to be incredibly cumbersome, we should not omit or abridge any of the stipulated steps. In the universities of Japan, people started to realize the importance of the clinical application of the new products or diagnostic methodologies rather than writing high-impact factor papers to acquire grants and funding so small as to contribute very little to contemporary clinical science. The top priority for licensed physicians is to conduct or participate in a well-designed clinical trial, from translational research to Phase I, II, and III trials, examining safety, efficacy, and comparing them with those of pre-existing methods.

From that point of view, the Japanese system of grant applications and/or promotion criteria in the universities have become woefully outdated. In order to compete with other developing countries that put enormous effort into the development and application of bioscientific results, the current Japanese system of concluding clinical science needs to be totally reconsidered.

In this regard, the author is now interested in the medical administration system and medical policy in Japan. If we could succeed in exchanging the old concept of a medical policy favored by the medical "establishment" and bureaucrats for a new strategy of bioscientific reform in Japan, we can and will become competitive with the new bioscientific development systems of the Western world.

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