

Histological evaluation of nerve sparing technique in robotic assisted radical prostatectomy

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ABSTRACT

Introduction: The objective of this study is to compare intrafascial nerve-sparing (NS), interfascial NS and non-NS prostatectomy specimens to assess the feasibility of NS technique in Robot-assisted radical prostatectomies (RARP).

Materials and Methods: The records of the first 43 consecutive patients (86 prostatic sides (lobe) who underwent NS RARP (6 intrafascial NS, 46 interfascial NS, 34 non-NS) were reviewed and histopathological examinations were performed. The presence and distribution of periprostatic neurovascular structures were histologically evaluated using mid-gland section of each prostate lobe in the prostatectomy specimen and it was immunostained with the S-100 antibody for quantitative analysis of nerves.

Results: The average number of nerve fibers per prostatic half was 37.2 ± 20.6 . The number of resected peri-prostatic nerves counted was 13.7 ± 13.5 , 30.5 ± 15.0 and 50.4 ± 20.4 in intrafascial NS, interfascial NS and non-NS specimens, respectively. The difference in the number of nerve bundle counts in the three groups was statistically significant ($P < 0.05$). Patients with urinary continence at 6 months after surgery had significantly less number of nerve fibers resected with the prostate than the incontinence group ($P = 0.013$) and the number of nerve fibers resected in the potent group were lower than in the impotent group but did not reach statistical significance ($P = 0.057$).

Conclusions: Our study showed that NS RARP could be performed according to surgeons' intention (intrafascial, interfascial or non-NS) and urinary continence significantly correlated to the number of nerve fibers resected with the prostate.

Key words: Histological evaluation, nerve-sparing, robot-assisted radical prostatectomy

INTRODUCTION

World-wide dissemination of the da Vinci surgical system has been associated with an increase in the number of robot-assisted radical prostatectomies (RARP) performed in Asia. The Japanese government included this surgery under medical insurance coverage as of April, 2012 and, since then, RARP has dramatically increased in Japan. Currently, 52 da Vinci

surgical systems are installed in Japan and this number is increasing such that Japan has become one of the countries, which have the largest number of robotic surgeries among Asian countries.^[1]

One of the main reasons for the spread of robotic surgery is its precision, especially while dealing with the prostatic apex and urethra, bladder-urethra anastomosis and nerve-sparing (NS) technique.^[2] In particular, the NS technique may be important both for preservation of erectile function and urinary continence post-operatively.^[3] In Japan, there are few reports on the outcomes of robotic surgery.^[4]

Several NS techniques are described which differ in the plane of dissection between the peri-prostatic fascia and prostatic capsule and are referred to as intrafascial NS, interfascial NS and non-NS (extra-fascial) techniques.^[5,6] In this study, we evaluated our consecutive initial RARP cases to determine if our technique of NS could be in fact performed according to the surgeons' intentions and the relationship between NS and postoperative continence and sexual function, using histological nerve identification with

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immunohistochemical (IHC) staining in the prostatectomy specimens.

MATERIALS AND METHODS

Patients

The initial 43 patients who underwent RARP with NS techniques by the same surgical team at our institution between October 2010 and July 2011 were enrolled in this study. All cases were consecutive without any exclusion in order to reflect the real clinical situation and findings. The records of NS RARP in these patients were reviewed retrospectively. The data were analyzed focusing on whether the surgeons' intention to perform intrafascial NS, interfascial NS and non-NS surgery correlated with the degree of nerve preservation in the histopathological examination of the prostatectomy specimens, postoperative continence and sexual function.

Surgeries

RARPs were performed using the method of Menon *et al.*:^[7] (1) The first camera port (2.5 cm in diameter) was inserted via an open laparotomy method just above the umbilicus and the 3 da Vinci ports (0.8 cm in diameter) and 2 laparoscopic ports (1.2 cm in diameter) were inserted under laparoscopic observation; (2) prostatectomy was performed via the antegrade and lateral approach; (3) bladder-urethral anastomosis was performed using a continuous suture; (4) bilateral pelvic lymphadenectomies were performed; (5) a single intrapelvic drain was inserted; (6) peritoneum, fascia of rectus muscle, subcutaneous tissue and skin were closed. In the NS technique, surgeons aimed to spare and detach the neurovascular bundle (NVB) from prostate as described by Ko *et al.*^[8] and the layer to spare was decided according to the relationship between the prostatic fascia, peri-prostatic fascia and prostate capsule. However, Walz *et al.* stated that the prostatic fascia consists of several layers (not a single sheet) and the extent to spare the fascia depends on the surgeon, particularly considering that its thickness has individual variation.^[9] We therefore considered that the layer to spare is between the prostatic capsule and the prostatic fascia for intrafascial NS and between the prostatic fascia and the lateral pelvic fascia or peri-prostatic fascia for interfascial NS.^[6] The indication for NS was based on clinical T stage (cT1c or T2), from digital rectal examination and magnetic resonance imaging (MRI) findings, the patient's sexual function on the International Index of Erectile Function (IIEF-5) as well as patient preferences. The indication for NS was decided separately for each prostate lobe, resulting in 86 prostate lobes to examine in this study.

IHC analyses

The presence and distribution of peri-prostatic neurovascular structures were histologically evaluated in the prostatectomy specimen. A mid-gland section of each prostate was immunostained with the S-100 antibody for quantitative analysis of nerves. Nerves and ganglions

exclusively in peri-prostatic locations were counted. We noted staining especially around the prostatic capsule to identify the distribution of nerves in NS cases (or sides) and non-NS cases (or sides), comparing them especially for the difference between intrafascial and interfascial NS. The histopathology of prostatectomy specimens was assessed on hematoxylin and eosin stained slides. IHC staining of tissue specimens was performed by a Dako Autostainer Plus System (Dako Corporation, Carpinteria, CA) using the rabbit polyclonal antibody against S-100 (1:1000, Leica Biosystem Newcastle Ltd, Newcastle, UK) or by the BenchMark XT IHC/ISH Staining Module (Ventana Medical Systems, Inc., Tucson, AZ) using the rabbit polyclonal antibody against S-100 (1:2000, Thermo ELECTRON Corporation, Waltham, MA). Tissues were deparaffinized, rehydrated and subjected to 15-min double endogenous enzyme block, 3-min triple wash, 10-min non-specific protein absorption and overnight primary antibody reaction followed by a 3-min triple wash and 10-min 2nd antibody (biotin signaling anti mouse/rabbit immunoglobulin, Dako Corporation) incubation at room temperature. Antigenic signals were detected by adding diaminobenzidine as a chromogen of horseradish peroxidase and slides were counterstained by hematoxylin. All the reagents were obtained from Dako Corporation unless otherwise indicated. For quantification, 100 cells at 3 randomly selected areas in which the stained lesions tended to look more than other parts were assessed at $\times 100$ magnification and the positive S-100 stained cells were recorded with the positive intensity of S-100 staining distributed from (-) to (++). Two independent observers scored the quantification of positively stained nerves and their averages were evaluated. They were blinded to the information of NS as assessed by the surgical team.

Evaluation

We counted the number of S-100 positively stained nerve fibers in prostatectomy specimens and compared these data with the surgeon's intention to perform intrafascial NS, interfascial NS and non-NS surgery. Middle section of prostate specimen was evaluated for NS in this study. The urinary continence and sexual function (potency) were also evaluated at 6 months after the operation. The definition of urinary continence was pad free and that of potency was decided by whether vaginal penetration was possible.

Statistical analyses

Statistical analyses were performed by Student's *t* tests using JSTAT (Java Virtual Machine Statistics Monitoring Tool) with $P < 0.05$ considered to indicate statistical significance.

RESULTS

Patient backgrounds

Patient prostate specific antigen (PSA) level was 8.05 ± 3.70 ng/ml. The stage distribution was pT2a in 10 cases,

pT2b in two cases, pT2c in 22 cases, pT3a in six cases and pT3b in one case (2 were non-gradable). However, six out of these seven cases were diagnosed as clinical T2c and one of them had a suspicion of T3 disease but the patient strongly desired NS. Fortunately, this patient did not have any PSA failure post-operatively in 2 years follow-up. The Gleason score was 3 + 3, 3 + 4, 4 + 3 and 4 + 4 in 11, 13, 6 and 11 cases, respectively [Table 1]. Nerve sparing was performed in 43 patients; 6 intrafascial NS, 46 interfascial NS and 34 non-NS [Table 2].

Histopathological findings in NS

Figure 1 shows the IHC for each NS method (intrafascial NS, interfascial NS and non-NS). Quantitative S-100 IHC data showed that the average number of nerve fibers counted per prostatic half was 37.2 ± 20.6 in the 86 prostatic lobes. The detailed distribution of resected nerve fibers using NS revealed 13.7 ± 13.5 , 30.5 ± 15.0 and \pm counts with intrafascial NS, interfascial NS and non-NS, respectively. The differences in the number of nerve bundle counts in the three groups was statistically significant ($P < 0.05$) [Table 2 and Figure 1].

Functional outcomes and NS

Patients who were continent at 6 months after surgery had significantly less number of nerve fibers resected with the prostate than the incontinent group ($P = 0.013$) [Table 3]. The number of nerve fibers resected in the potent group tended lower than the impotent group but the difference was not significant ($P = 0.057$).

DISCUSSION

Robotic surgery has spread rapidly, in part because of the facilitation of delicate surgical techniques^[10] and many surgeons feel that this technique may make it easier to offer nerve sparing than open or laparoscopic surgeries.^[11,12] In addition, robotic surgery offers better surgical vision due to the wide range of the endoscopic camera with three-dimensional surgical vision, allowing surgeons to recognize the different layers.^[13] Robotic surgery is expected to enable better cancer control and better post-operative quality-of-life (QOL) than other modalities partly due to the higher costs of robotic surgeries than open or laparoscopic surgeries.^[14] Two of the most important aspects of QOL are urinary continence and erectile function^[15] and these are expected to be improved using delicate techniques such as NS procedures.^[16]

After conventional NVB sparing surgery was first developed by Walsh and Talcott *et al.*,^[17,18] several new concepts have emerged such as the Veil technique and the recognition of the thickness of prostatic fascia for NS.^[19,20] The former was reported by Kaul *et al.*^[21] who found that nerve fibers exist not only in the postero-lateral parts but also in the lateral and ventral parts around the prostate^[22] and surgeons need to be conscious of this fact when selecting surgical layers for NS.^[23] We define as intrafascial NS when no prostate fascia tissue remains on the prostatic capsule and as interfascial NS when parts of prostatic fascia are left on the prostatic capsule.^[24] There are very few reports on the correlation between intrafascial NS, interfascial NS and non-NS and

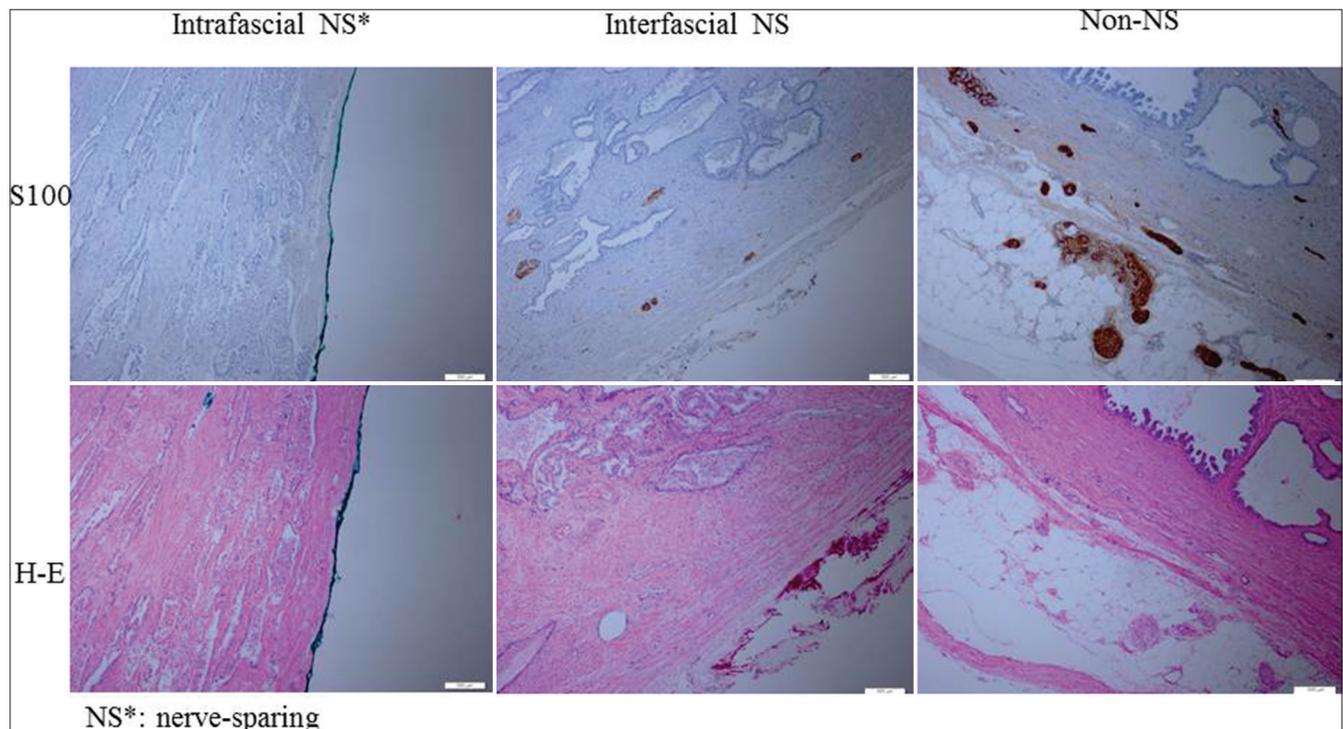


Figure 1: Immunohistochemical (S-100) analyses are shown. More positively-stained neurons are seen with non-nerve sparing than with intrafascial or interfascial NS. These differences are statistically significant [Table 2]

Table 1: Patients' backgrounds

N	43
Age (median (range))	65 (51-79)
BMI (median (range))	23.5 (17.6-32.9)
PSA (ng/ml)	8.05±3.70
Clinical stage	
cT1c	3
cT2a	23
cT2b	0
cT2c	16
cT3a	1
Pathological stage	
pT2a	10
pT2b	2
pT2c	22
pT3a	6
pT3b	1
Non-gradable	2
Gleason score	
3+3	11
3+4	13
4+3	6
4+4	11
4+5	2
Surgical time (console) (min)	277±46
Blood loss (ml)	284±388

BMI=Body mass index; PSA=Prostate specific antigen

Table 2: NS (nerve sparing) in 86 prostatic half (43 patients)

	The number of resected nerve fiber	P value		
		1 vs. 2	2 vs. 3	1 vs. 3
1 Intrafascial NS (n=6)	13.7±13.5	P=0.005		
2 Interfascial NS (n=46)	30.5±15.0	P<0.0001		
3 Non-NS (n=34)	50.4±20.4	P=0.001		

NS=Nerve sparing

Table 3: Comparison of the number of resected nerve fibers 6 months after operation

	Number of nerve fiber		P value
	Continence +	Continence -	
	63.0±33.0	89.5±34.5	0.013
Potency +		Potency -	
	60.3±31.1	81.3±36.4	0.057

the distribution of nerves in the resected prostates based on histopathological data, especially in robotic surgery.^[22] In open or laparoscopic surgery, Shigemura *et al.* and Greco *et al.* showed that NS procedures left more nerve tissues

than non-NS surgery^[25,26] and similar studies were also reported in robotic surgeries.^[27]

In our country, the number of RARPs is dramatically increasing and it is necessary to examine NS outcomes. Our study demonstrates that NS can be performed even by surgeons with relatively little experience of RARP without comprising surgical margin status even though we included high Gleason score cases in about 30% cases (data not shown). Long-term outcomes in RARP NS and the number of cases needed to reach proficiency in NS as verified by histopathological examinations such as those in this study are needed.

We also evaluated functional outcome based on the number of nerve fibers resected. The number of nerve fibers in continent group was significantly lower than the incontinent group. As for potency, that in potent group was lower than in potent group but there was also a not significant difference. These results mean that our technique of NS was appropriate not only histologically but also functionally. Previous studies have reported that high degree of NS technique significantly correlated with early return of urinary continence^[28] and improving potency outcomes,^[29] suggesting our study and results were supported by those of high volume center with larger experience.

We suggest performing independent NS surgeries on each prostate lobe in T2 or T1c prostate cancer based on the location and aggressiveness of cancer. This is partly possible due to the improved diagnostic power of MRI and the fact that the number of NS RARP procedures in Japan is still comparatively low due to its recent adoption.^[28] We have several limitations. First, this is a retrospective study with a small number case from our initial experience and this may explain the comparatively fewer nerve fibers than other studies.^[30] This may suggest success of NS even for comparatively less experienced surgeons and that our spared layers might be closer to the prostatic capsule. Second, we lack the histopathological data of resected tissues of prostatic fascia and its distribution between intrafascial, interfascial and non-NS. These will be studied in our future work.

CONCLUSION

Our data showed that NS RARP could be performed as per the surgeons' intention and this was verified quantitatively by the number of nerve fibers resected with the prostate. This also significantly correlated with urinary continence.

REFERENCES

1. Fujisawa M. Introduction and surgical procedure of robot-assisted radical prostatectomy. *Jpn J Endourol* 2012;25:202.
2. Mottrie A, Gallina A, De Wil P, Thüer D, Novara G, Ficarra V. Balancing

- continence function and oncological outcomes during robot-assisted radical prostatectomy (RARP). *BJU Int* 2011;108:999-1006.
3. Stolzenburg JU, Nicholas M, Minh D, Dietel A, Hicks J. Does a nerve-sparing technique for potency affect continence after open radical retropubic prostatectomy? *BJU Int* 2009;103:1292.
 4. Vora AA, Dajani D, Lynch JH, Kowalczyk KJ. Anatomic and technical considerations for optimizing recovery of urinary function during robotic-assisted radical prostatectomy. *Curr Opin Urol* 2013;23:78-87.
 5. Stolzenburg JU, Kallidonis P, Do M, Dietel A, Häfner T, Rabenalt R, *et al.* A comparison of outcomes for interfascial and intrafascial nerve-sparing radical prostatectomy. *Urology* 2010;76:743-8.
 6. Potdevin L, Ercolani M, Jeong J, Kim IY. Functional and oncologic outcomes comparing interfascial and intrafascial nerve sparing in robot-assisted laparoscopic radical prostatectomies. *J Endourol* 2009;23:1479-84.
 7. Menon M, Tewari A, Peabody J, VIP Team. Vattikuti Institute prostatectomy: Technique. *J Urol* 2003;169:2289-92.
 8. Ko WJ, Hruby GW, Turk AT, Landman J, Badani KK. Pathological confirmation of nerve-sparing types performed during robot-assisted radical prostatectomy (RARP). *BJU Int* 2013;111:451-8.
 9. Walz J, Burnett AL, Costello AJ, Eastham JA, Graefen M, Guillonneau B, *et al.* A critical analysis of the current knowledge of surgical anatomy related to optimization of cancer control and preservation of continence and erection in candidates for radical prostatectomy. *Eur Urol* 2010;57:179-92.
 10. Iselin CE. Treatment of localized prostate cancer: The role of robotic radical prostatectomy. *Rev Med Suisse* 2008;4:2626-8.
 11. Chauhan S, Coelho RF, Rocco B, Palmer KJ, Orvieto MA, Patel VR. Techniques of nerve-sparing and potency outcomes following robot-assisted laparoscopic prostatectomy. *Int Braz J Urol* 2010;36:259-72.
 12. Asimakopoulos AD, Pereira Fraga CT, Annino F, Pasqualetti P, Calado AA, Mugnier C. Randomized comparison between laparoscopic and robot-assisted nerve-sparing radical prostatectomy. *J Sex Med* 2011;8:1503-12.
 13. Yoshioka K, Hatano T, Nakagami Y, Ozu C, Horiguchi Y, Sakamoto N, *et al.* First 24 Japanese cases of robotic-assisted laparoscopic radical prostatectomy using the daVinci Surgical System. *Hinyokika Kyo* 2008;54:333-8.
 14. van der Poel HG, de Blok W, Joshi N, van Muilekom E. Preservation of lateral prostatic fascia is associated with urine continence after robotic-assisted prostatectomy. *Eur Urol* 2009;55:892-900.
 15. Duffey B, Varda B, Konety B. Quality of evidence to compare outcomes of open and robot-assisted laparoscopic prostatectomy. *Curr Urol Rep* 2011;12:229-36.
 16. Sooriakumaran P, Calaway A, Sagalovich D, Roy S, Srivastava A, Joneja J, *et al.* The impact of multiple biopsies on outcomes of nerve-sparing robotic-assisted radical prostatectomy. *Int J Impot Res* 2012;24:161-4.
 17. Walsh PC. Anatomic radical retropubic prostatectomy. In: Walsh PC, Retik AB, Vaushan ED, Wein AJ, editors. *Campbell's Textbook of Urology*. 7th ed. Philadelphia: WB Saunders; 1997. p. 2565-88.
 18. Talcott JA, Rieker P, Probert KJ, Clark JA, Wishnow KI, Loughlin KR, *et al.* Patient-reported impotence and incontinence after nerve-sparing radical prostatectomy. *J Natl Cancer Inst* 1997;89:1117-23.
 19. Menon M, Kaul S, Bhandari A, Shrivastava A, Tewari A, Hemal A. Potency following robotic radical prostatectomy: A questionnaire based analysis of outcomes after conventional nerve sparing and prostatic fascia sparing techniques. *J Urol* 2005;174:2291-6, 2296.
 20. Takenaka A, Hara R, Soga H, Murakami G, Fujisawa M. A novel technique for approaching the endopelvic fascia in retropubic radical prostatectomy, based on an anatomical study of fixed and fresh cadavers. *BJU Int* 2005;95:766-71.
 21. Kaul S, Bhandari A, Hemal A, Saveria A, Shrivastava A, Menon M. Robotic radical prostatectomy with preservation of the prostatic fascia: A feasibility study. *Urology* 2005;66:1261-5.
 22. Tewari A, Peabody JO, Fischer M, Sarle R, Vallancien G, Delmas V, *et al.* An operative and anatomic study to help in nerve sparing during laparoscopic and robotic radical prostatectomy. *Eur Urol* 2003;43:444-54.
 23. Lavery HJ, Prall DN, Abaza R. Active patient decision making regarding nerve sparing during radical prostatectomy: A novel approach. *J Urol* 2011;186:487-92.
 24. Hinata N, Sejima T, Takenaka A. Progress in pelvic anatomy from the viewpoint of radical prostatectomy. *Int J Urol* 2013;20:260-70.
 25. Shigemura K, Yamanaka N, Yamashita M. Veil nerve-sparing technique and postoperative urinary continence in open antegrade radical prostatectomy. *Urol Int* 2012;89:283-9.
 26. Greco F, Hoda MR, Wagner S, Reichelt O, Inferrera A, Magno C, *et al.* Bilateral vs unilateral laparoscopic intrafascial nerve-sparing radical prostatectomy: Evaluation of surgical and functional outcomes in 457 patients. *BJU Int* 2011;108:583-7.
 27. Xylinas E, Ploussard G, Salomon L, Paul A, Gillion N, Laet KD, *et al.* Intrafascial nerve-sparing radical prostatectomy with a laparoscopic robot-assisted extraperitoneal approach: Early oncological and functional results. *J Endourol* 2010;24:577-82.
 28. Srivastava A, Chopra S, Pham A, Sooriakumaran P, Durand M, Chughtai B, *et al.* Effect of a risk-stratified grade of nerve-sparing technique on early return of continence after robot-assisted laparoscopic radical prostatectomy. *Eur Urol* 2013;63:438-44.
 29. Tewari AK, Srivastava A, Huang MW, Robinson BD, Shevchuk MM, Durand M, *et al.* Anatomical grades of nerve sparing: A risk-stratified approach to neural-hammock sparing during robot-assisted radical prostatectomy (RARP). *BJU Int* 2011;108:984-92.
 30. Saveria AT, Kaul S, Badani K, Stark AT, Shah NL, Menon M. Robotic radical prostatectomy with the "Veil of Aphrodite" technique: Histologic evidence of enhanced nerve sparing. *Eur Urol* 2006;49:1065-73.

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