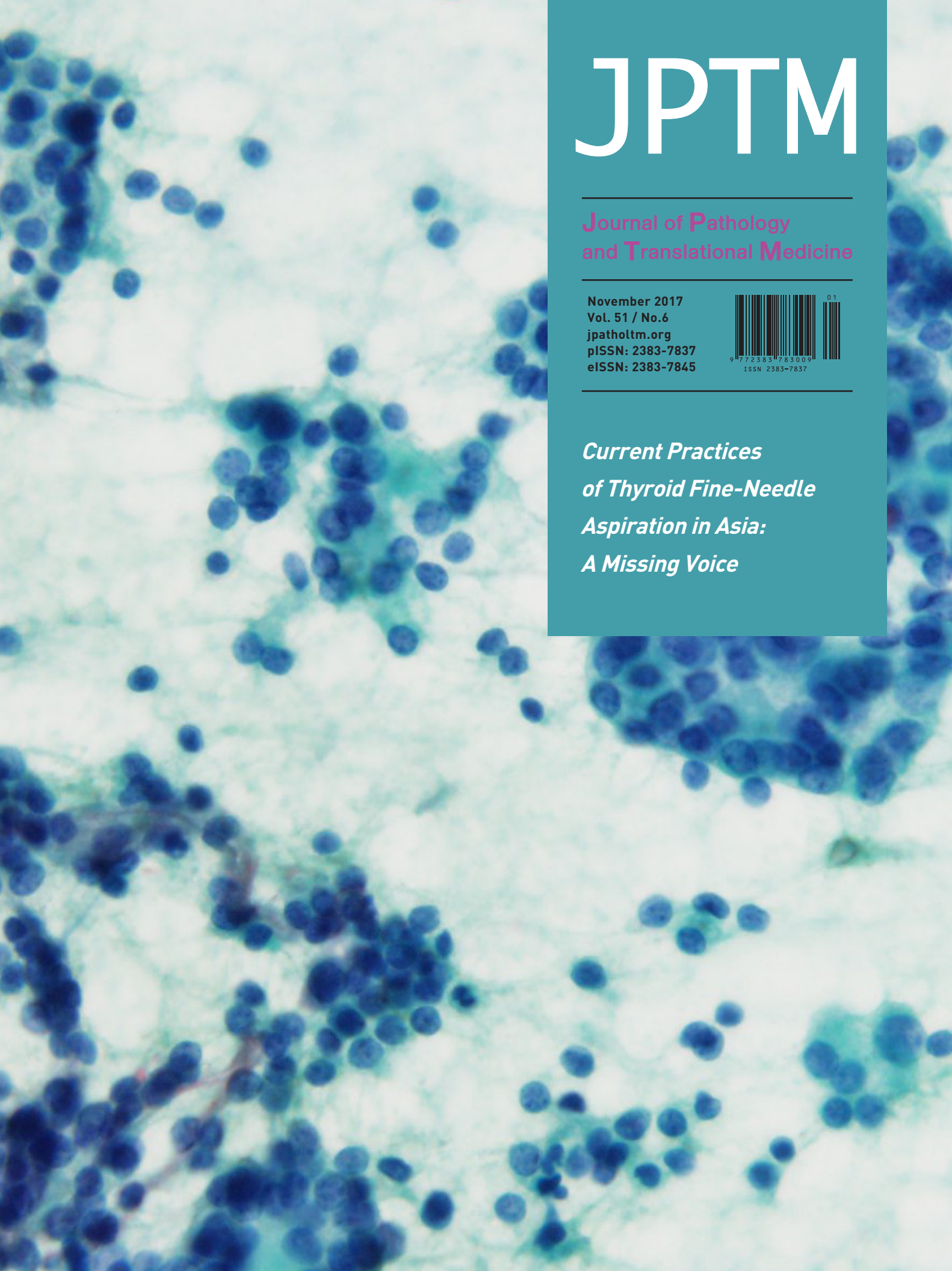


JPTM

Journal of Pathology and Translational Medicine

November 2017
Vol. 51 / No.6
jpatholtm.org
pISSN: 2383-7837
eISSN: 2383-7845

Current Practices of Thyroid Fine-Needle Aspiration in Asia: A Missing Voice

Journal of Pathology and Translational Medicine

Volume 51 • Number 6 • November 2017 (bimonthly)
Published since 1967
Printed on 11 November 2017 Published on 15 November 2017

J Pathol Transl Med
pISSN: 2383-7837
eISSN: 2383-7845

Aims & Scope

The Journal of Pathology and Translational Medicine is an open venue for the rapid publication of major achievements in various fields of pathology, cytopathology, and biomedical and translational research. The Journal aims to share new insights into the molecular and cellular mechanisms of human diseases and to report major advances in both experimental and clinical medicine, with a particular emphasis on translational research. The investigations of human cells and tissues using high-dimensional biology techniques such as genomics and proteomics will be given a high priority. Articles on stem cell biology are also welcome. The categories of manuscript include original articles, review and perspective articles, case studies, brief case reports, and letters to the editor.

Subscription Information

To subscribe to this journal, please contact the Korean Society of Pathologists/the Korean Society for Cytopathology. Full text PDF files are also available at the official website (http://jpatholtm.org). *Journal of Pathology and Translational Medicine* is indexed by Emerging Sources Citation Index (ESCI), PubMed, PubMed Central, Scopus, KoreaMed, KoMCI, WPRIM, Directory of Open Access Journals (DOAJ), and CrossRef. Circulation number per issue is 700.

Contact the Korean Society of Pathologists/the Korean Society for Cytopathology

Publishers: Han Kyeom Kim, MD, Hye Kyoung Yoon, MD
Editors-in-Chief: Soon Won Hong, MD, Chong Jai Kim, MD
Published by the Korean Society of Pathologists/the Korean Society for Cytopathology

Editorial Office
Room 1209 Gwanghwamun Officia, 92 Saemunan-ro, Jongno-gu, Seoul 03186, Korea
Tel: +82-2-795-3094 Fax: +82-2-790-6635 E-mail: office@jpatholtm.org

#1508 Renaissancetower, 14 Mallijae-ro, Mapo-gu, Seoul 04195, Korea
Tel: +82-2-593-6943 Fax: +82-2-593-6944 E-mail: office@jpatholtm.org

Printed by iMiS Company Co., Ltd. (JMC)
Jungang Bldg. 18-8 Wonhyo-ro 89-gil, Yongsan-gu, Seoul 04314, Korea
Tel: +82-2-717-5511 Fax: +82-2-717-5515 E-mail: ml@smileml.com

Manuscript Editing by InfoLumi Co.
210-202, 421 Pangyo-ro, Bundang-gu, Seongnam 13522, Korea
Tel: +82-70-8839-8800 E-mail: infolumi.chang@gmail.com

Front cover image: Cytological findings of indeterminate A3. The specimen was aspirated from a minimally invasive follicular carcinoma (Fig. 2). p551.

This journal was supported by the Korean Federation of Science and Technology Societies Grant funded by the Korean Government.

Journal of Pathology and Translational Medicine

J Pathol Transl Med
pISSN: 2383-7837
eISSN: 2383-7845

Volume 51, Number 6, November 2017

CONTENTS

EDITORIAL

517 Current Practices of Thyroid Fine-Needle Aspiration in Asia: A Missing Voice
Andrey Bychkov, Kennichi Kakudo, SoonWon Hong

REVIEWS

521 Thyroid Fine-Needle Aspiration Cytology Practice in Korea
Yoon Jin Cha, Ju Yeon Pyo, SoonWon Hong, Jae Yeon Seok, Kyung-Ju Kim, Jee-Young Han, Jeong Mo Bae, Hyeong Ju Kwon, Yeejeong Kim, Kyueng-Whan Min, Soonae Oak, Sunhee Chang

528 History and Practice of Thyroid Fine-Needle Aspiration in China, Based on Retrospective Study of the Practice in Shandong University Qilu Hospital
Zhiyan Liu, Dongge Liu, Bowen Ma, Xiaofang Zhang, Peng Su, Li Chen, Qingdong Zeng

533 Thyroid Cytology in India: Contemporary Review and Meta-analysis
Shipra Agarwal, Deepali Jain

548 Thyroid Cytology: The Japanese System and Experience at Yamashita Thyroid Hospital
Shinya Satoh, Hiroyuki Yamashita, Kennichi Kakudo

555 Thyroid Fine-Needle Aspiration Practice in the Philippines
Agustina D. Abelardo

560 Thyroid Fine-Needle Aspiration in Taiwan: The History and Current Practice
Jen-Fan Hang, Chih-Yi Hsu, Chiung-Ru Lai

565 Current Status of Thyroid Fine-Needle Aspiration Practice in Thailand
Somboon Keelawat, Samreung Rangdaeng, Supinda Koonmee, Tikamporn Jitpasutham, Andrey Bychkov

571 The Use of Fine-Needle Aspiration (FNA) Cytology in Patients with Thyroid Nodules in Asia: A Brief Overview of Studies from the Working Group of Asian Thyroid FNA Cytology
Chan Kwon Jung, SoonWon Hong, Andrey Bychkov, Kennichi Kakudo

CONTENTS CONTINUED

ORIGINAL ARTICLE

579 Current Cytology Practices in Korea: A Nationwide Survey by the Korean Society for Cytopathology
Eun Ji Oh, Chan Kwon Jung, Dong-Hoon Kim, Han Kyeom Kim, Wan Seop Kim, So-Young Jin, Hye Kyoung Yoon,
The Fellowship Council and Committee of Quality Improvement of the Korean Society for Cytopathology

CASE REPORT

588 Aggressive Supratentorial Ependymoma, RELA Fusion-Positive with Extracranial Metastasis: A Case Report
Seong-Ik Kim, Yoojin Lee, Seung Ki Kim, Hyoung Jin Kang, Sung-Hye Park

Instructions for Authors for *Journal of Pathology and Translational Medicine* are available at http://jpatholtm.org/authors/authors.php

Journal of Pathology and Translational Medicine 2017; 51: 517-520
https://doi.org/10.4132/jptm.2017.09.27

■ EDITORIAL ■

Current Practices of Thyroid Fine-Needle Aspiration in Asia: A Missing Voice

Andrey Bychkov · Kennichi Kakudo[1] · SoonWon Hong[2]

Department of Pathology, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand;
[1]Department of Pathology, Nara Hospital, Kindai University Faculty of Medicine, Nara, Japan;
[2]Department of Pathology, Yonsei University College of Medicine, Seoul, Korea

Journal of Pathology and Translational Medicine (JPTM) is pleased to announce a special issue devoted to the current practices of thyroid fine-needle aspiration (FNA) cytology in Asian countries as a joint effort of members of the Working Group of Asian Thyroid FNA Cytology. Currently, this growing network of Asian thyroid pathologists includes representatives from China, India, Japan, the Philippines, South Korea, Taiwan, Thailand, Turkey, and Vietnam.

Asia, as the largest and most populous continent, comprises several geographic regions with notable ethnic, cultural and religious diversity. The different levels and pace of economic growth within these regions determines the development of local health systems. Most Asian countries are well integrated into the modern international medical community. Contemporary practices in various fields of medicine were established under a strong Western influence. On the other hand, Asian philosophy, traditional Chinese medicine, Ayurveda, and other conventional medical practices with deep historical roots are often integrated into advanced medical approaches.[1,2]

Consistent with its huge population, the Asian continent is the largest contributor to the worldwide prevalence of thyroid cancer. According to the GLOBOCAN estimates, 48% of all new thyroid cancer cases are diagnosed in Asia.[3] In addition, the absolute number of patients with thyroid cancer increases each year, and this growth has recently been labeled a thyroid cancer epidemic, due to unexpectedly high increases compared to estimates.[4] South Korea is the best example of this thyroid cancer "epidemic," which has been proven to be largely caused by opportunistic screening.[5] The same epidemic in many Western countries with a growing incidence of thyroid cancer was later attributed to opportunistic screening also.[6] As a result of these epidemiological shifts, thyroid cancer and thyroid nodules have attracted considerable attention from the medical community worldwide.

Leading medical centers in well-developed Asian countries have promoted innovative approaches and disseminated high-quality evidence, which has profoundly influenced international practice.[7,8] Numerous publications from Japan and Korea have laid the ground for national guidelines on the management of thyroid cancer and thyroid nodules, which are finely tuned to the local settings.[9,10] At the same time, guidelines from less advanced Asian countries largely rely on international recommendations partially adjusted to lower economic standards.[11-13]

Thyroid FNA is a mainstay of preoperative diagnosis of thyroid nodules, which drives further decision making.[14] This simple procedure is available globally and has been accepted as the first-line intervention in the workup of thyroid nodules, which can significantly reduce unnecessary surgery. Unlike in gynecologic cytopathology, evaluation of thyroid FNA is usually performed by surgical pathologists who tended to apply their own classification schemes used in histopathologic diagnosis of thyroid nodules. As a result, cytologic diagnoses applied to thyroid FNAs were not uniform and clear, but rather variable and vague. Furthermore, a substantial number of samples are found to be uncertain, indeterminate, equivocal, or suspicious, which can create confusion for the clinicians with regard to treatment planning.

In the effort to standardize thyroid cytologic terminology and

Corresponding Author
Andrey Bychkov, MD, PhD
Department of Pathology, Faculty of Medicine, Chulalongkorn University, Rama IV Rd., Pathumwan, Bangkok 10330, Thailand
Tel: +66-2-256-4235, Fax: +66-2-652-4208, E-mail: andrey.b@chula.ac.th

Received: September 14, 2017 **Accepted:** September 25, 2017

pISSN 2383-7837
eISSN 2383-7845

to improve communication between pathologists and clinicians, a new reporting system encompassing six diagnostic categories was proposed around 10 years ago.[15] Since that time, the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) has received universal acclaim and has been endorsed by numerous national and international societies in the fields of endocrinology, thyroidology, and cytopathology.[16] Beyond TBSRTC, several countries, including the UK, Italy, and Japan, have established their own systems for reporting thyroid cytopathology.[17-19] Nevertheless, the terminology used in non-Bethesda reporting systems is easily adjustable to Bethesda diagnostic categories, which is important for comparison.

Each of the modern systems for reporting thyroid cytopathology provides important statistical outputs, which serve as the quality control criteria. These criteria include (1) distribution of thyroid FNA samples by diagnostic category; (2) resection rate (RR), measured as a ratio of surgically excised nodules to all sampled thyroid nodules within a certain category; and (3) risk of malignancy (ROM) or the percentage of malignant nodules among all FNAs. ROM is important because it indicates the necessity of surgical treatment. The original TBSRTC estimated ROM ranges for diagnostic categories based on preceding literature.[15] These estimates were further modified in meta-analyses to provide the actual ROM, summarized from the numerous single- or multi-center studies.[20-23]

Most meta-analyses on thyroid FNA and TBSRTC have not included Asian publications.[20-22] Only one meta-analysis included a fair number of original studies from Turkey, Korea, and Arabic countries.[23] In fact, experience with thyroid FNA in Asia has been extensively reported. Recently, the results of a nationwide study covering more than 42,000 FNAs were presented by the Korean Society of Endocrine Pathologists.[24] Japanese institutions have also shared their experience with the Japanese system of reporting thyroid FNA.[18] There is a growing number of publications from India and China. Reports on thyroid cytology from Southeast Asia are less abundant and often non-systematic. Notwithstanding, we should note that even low-resource countries, for example, Bangladesh and Nepal, have been able to publish their experience with thyroid FNA.[25,26] Once again, despite the efforts of Asian cytopathologists to share their data with the international community, their voice has not been recognized. Hopefully, output data on the use of TBSRTC from major Asian countries summarized in this special issue will contribute to future meta-analyses of the Bethesda system.

An important lesson learned after comparison of Asian and Western series is that the Asian experience varies in several aspects. Thyroid FNA studies disclosed low RR and high ROM for indeterminate nodules in Asian practice.[27] This could be explained by

Fig. 1. Members of the Working Group of Asian Thyroid FNA Cytology and hosts during the inaugural meeting of the 12th Asia and Oceania Thyroid Association (AOTA) congress in Busan, Korea (March 16, 2017). FNA, fine-needle aspiration. *Left-to-right*: Z. Liu (China), K. Kakudo (Japan), C.R. Lai (Taiwan), S. Satoh (Japan), S. Keelawat (Thailand), S. Canberk (Turkey), A. Bychkov (Thailand), S.W. Hong (Korea), D.E. Song (Korea), C.K. Jung (Korea), H.J. Kwon (Korea), M. Hirokawa (Japan), H.K. Chang (Korea).

 https://doi.org/10.4132/jptm.2017.09.27

the more conservative management approach for indolent thyroid tumors compared to Western practice.[28] As a result, borderline thyroid tumors, such as noninvasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP) and well-differentiated tumor of uncertain malignant potential, are histologically rare in Asian countries.[29,30] These differences are not acknowledged worldwide, which continues to create confusion among experts due to this lack of communication. It should be reiterated that the Asian continent is a major contributor to the global prevalence of thyroid cancer, and that local experience cannot be ignored.

International communication is a key factor in disseminating knowledge and staying up-to-date. There are several international forums held annually for pathologists in Asia, but until recently there were no active well-established networks for those practicing within the thyroid niche. The Working Group of Asian Thyroid FNA Cytology was established recently to promote communication among Asian pathologists and cytopathologists, to share experience in Asian practice, and to conduct multi-institutional studies. An inaugural meeting took place at the 12th Asia and Oceania Thyroid Association (AOTA) Congress in Busan, Korea on March 16, 2017 (Fig. 1). Despite its recent formation, several achievements have resulted from this joint effort. Senior group members released a book *Thyroid FNA Cytology: Differential Diagnoses and Pitfalls*, which was published in 2016 as the first English language textbook on thyroid FNA cytology from Asia.[31] Several authors contributed to a special NIFTP issue of the *Journal of Basic and Clinical Medicine*.[32-35] More original studies and reviews have been published[28,29,36] or are currently in process.

Presented herein is a collection of articles on the current practices of thyroid FNA cytology in Asian countries that highlights important aspects of this diagnostic technique, including details on operators and readers, sampling and preparation, and reporting systems and audit programs. Also included are original data collected from the authors of previous publications and statistics from literature review. The authors wish to thank JPTM for hosting this special issue. We hope that our contemporary reviews will serve as a useful reference for a wide variety of specialists involved in the management of patients with thyroid nodules and thyroid cancer.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

REFERENCES

1. Dong J. The relationship between traditional Chinese medicine and modern medicine. Evid Based Complement Alternat Med 2013; 2013: 153148.
2. Shrivastava SR, Shrivastava PS, Ramasamy J. Mainstreaming of Ayurveda, Yoga, Naturopathy, Unani, Siddha, and Homeopathy with the health care delivery system in India. J Tradit Complement Med 2015; 5: 116-8.
3. Ferlay J, Soerjomataram I, Ervik M, *et al*. GLOBOCAN 2012 v1.0, Cancer incidence and mortality worldwide: IARC CancerBase No. 11 [Internet]. Lyon: International Agency for Research on Cancer, 2013 [cited 2016 Oct 18]. Available from: http://globocan.iarc.fr.
4. Roman BR, Morris LG, Davies L. The thyroid cancer epidemic, 2017 perspective. Curr Opin Endocrinol Diabetes Obes 2017; 24: 332-6.
5. Ahn HS, Kim HJ, Welch HG. Korea's thyroid-cancer "epidemic": screening and overdiagnosis. N Engl J Med 2014; 371: 1765-7.
6. Vaccarella S, Franceschi S, Bray F, Wild CP, Plummer M, Dal Maso L. Worldwide thyroid-cancer epidemic? The increasing impact of overdiagnosis. N Engl J Med 2016; 375: 614-7.
7. Ito Y, Uruno T, Nakano K, *et al*. An observation trial without surgical treatment in patients with papillary microcarcinoma of the thyroid. Thyroid 2003; 13: 381-7.
8. Leboulleux S, Tuttle RM, Pacini F, Schlumberger M. Papillary thyroid microcarcinoma: time to shift from surgery to active surveillance? Lancet Diabetes Endocrinol 2016; 4: 933-42.
9. Japan Thyroid Association. Guidelines for clinical practice for the management of thyroid nodules in Japan 2013. Tokyo: Nankodo Co., Ltd., 2013.
10. Yi KH. The revised 2016 Korean Thyroid Association guidelines for thyroid nodules and cancers: differences from the 2015 American Thyroid Association guidelines. Endocrinol Metab (Seoul) 2016; 31: 373-8.
11. Chindavijak S, Panchan V, Chaiwerawattana A, Imsamran W. Guidelines for the diagnosis and treatment of thyroid cancer. Bangkok: Kosit Press Co., Ltd., 2015.
12. Sison CM, Obaldo J, Matsuo J, Uy GL, Jaring C. University of the Philippines - Philippine General Hospital revised clinical practice guidelines for the management of well-differentiated thyroid carcinoma of follicular cell origin. J Asean Fed Endocr Soc 2012; 27: 49-61.
13. Unnikrishnan AG, Kalra S, Baruah M, *et al*. Endocrine Society of India management guidelines for patients with thyroid nodules: a position statement. Indian J Endocrinol Metab 2011; 15: 2-8.
14. Haugen BR, Alexander EK, Bible KC, *et al*. 2015 American Thyroid Association management guidelines for adult patients with thyroid

nodules and differentiated thyroid cancer: the American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer. Thyroid 2016; 26: 1-133.
15. Cibas ES, Ali SZ; NCI Thyroid FNA State of the Science Conference. The Bethesda System For Reporting Thyroid Cytopathology. Am J Clin Pathol 2009; 132: 658-65.
16. Pusztaszeri M, Rossi ED, Auger M, *et al*. The Bethesda System for Reporting Thyroid Cytopathology: proposed modifications and updates for the second edition from an international panel. Acta Cytol 2016; 60: 399-405.
17. Cross P, Chandra A, Giles T, *et al*. Guidance on the reporting of thyroid cytology specimens. 2nd ed. London: Royal College of Pathologists, 2016.
18. Kakudo K, Kameyama K, Miyauchi A, Nakamura H. Introducing the reporting system for thyroid fine-needle aspiration cytology according to the new guidelines of the Japan Thyroid Association. Endocr J 2014; 61: 539-52.
19. Nardi F, Basolo F, Crescenzi A, *et al*. Italian consensus for the classification and reporting of thyroid cytology. J Endocrinol Invest 2014; 37: 593-9.
20. Bongiovanni M, Spitale A, Faquin WC, Mazzucchelli L, Baloch ZW. The Bethesda System for Reporting Thyroid Cytopathology: a meta-analysis. Acta Cytol 2012; 56: 333-9.
21. Krauss EA, Mahon M, Fede JM, Zhang L. Application of the Bethesda classification for thyroid fine-needle aspiration: institutional experience and meta-analysis. Arch Pathol Lab Med 2016; 140: 1121-31.
22. Sheffield BS, Masoudi H, Walker B, Wiseman SM. Preoperative diagnosis of thyroid nodules using the Bethesda System for Reporting Thyroid Cytopathology: a comprehensive review and meta-analysis. Expert Rev Endocrinol Metab 2014; 9: 97-110.
23. Straccia P, Rossi ED, Bizzarro T, *et al*. A meta-analytic review of the Bethesda System for Reporting Thyroid Cytopathology: has the rate of malignancy in indeterminate lesions been underestimated? Cancer Cytopathol 2015; 123: 713-22.
24. Kim M, Park HJ, Min HS, *et al*. The use of the Bethesda System for Reporting Thyroid Cytopathology in Korea: a nationwide multicenter survey by the Korean Society of Endocrine Pathologists. J Pathol Transl Med 2017; 51: 410-7.
25. Choudhary PK, Nepal N, Mainali N, Meenakshi B. Implementation of Bethesda system in thyroid aspirate: a cyto-histopathological correlation study. J Pathol Nepal 2016; 6: 902-5.
26. Hassan MQ, Hasanat MA, Fariduddin M, *et al*. Fine needle aspiration cytological diagnosis of thyroid nodule with its clinical correlation. Bangabandhu Sheikh Mujib Med Univ J 2013; 6: 108-15.
27. Kakudo K. How to handle borderline/precursor thyroid tumors in management of patients with thyroid nodules. Gland Surg 2017 Aug 2 [Epub]. https://doi.org/10.21037/gs.2017.08.02.
28. Kakudo K, Higuchi M, Hirokawa M, Satoh S, Jung CK, Bychkov A. Thyroid FNA cytology in Asian practice–Active surveillance for indeterminate thyroid nodules reduces overtreatment of thyroid carcinoma. Cytopathology 2017 [Epub]. https://doi.org/10.1111/cyt.12491.
29. Bychkov A, Hirokawa M, Jung CK, *et al*. Low rate of noninvasive follicular thyroid neoplasm with papillary-like nuclear features in Asian practice. Thyroid 2017; 27: 983-4.
30. Liu Z, Zhou G, Nakamura M, *et al*. Encapsulated follicular thyroid tumor with equivocal nuclear changes, so-called well-differentiated tumor of uncertain malignant potential: a morphological, immunohistochemical, and molecular appraisal. Cancer Sci 2011; 102: 288-94.
31. Kakudo K, Liu Z, Hirokawa M. Thyroid FNA cytology: differential diagnoses and pitfalls. Nara: Kakudo Medical Education, 2016.
32. Canberk S, Baloch ZW, Ince U, Schmitt F. Diagnosis of non-invasive follicular tumor with papillary-like nuclear features (NIFTP): a practice changer for thyroid fine-needle aspiration interpretation. J Basic Clin Med 2017; 6: 38-43.
33. Jung CK, Kim C. Effect of lowering the diagnostic threshold for encapsulated follicular variant of papillary thyroid carcinoma on the prevalence of non-invasive follicular thyroid neoplasm with papillary-like nuclear features: a single-institution experience in Korea. J Basic Clin Med 2017; 6: 26-8.
34. Kakudo K. Unsettled issues in non-invasive encapsulated/well-circumscribed follicular thyroid neoplasm with papillary-like nuclear features (NIFTP) and precursor thyroid tumors. J Basic Clin Med 2017; 6: 3-7.
35. Kakudo K, Liu Z, Satoh S, Higuchi M, Hirokawa M. Non-invasive follicular thyroid neoploasm with papillary-like nuclear features (NIFTP): diagnosis and differential diagnoses. J Basic Clin Med 2017; 6: 14-21.
36. Bychkov A, Keelawat S, Agarwal S, *et al*. Impact of noninvasive follicular thyroid neoplasm with papillary-like nuclear features on the Bethesda System for Reporting Thyroid Cytopathology: a multi-institutional study in five Asian countries. Pathology. Forthcoming.

Journal of Pathology and Translational Medicine 2017; 51: 521-527
https://doi.org/10.4132/jptm.2017.09.26

■ REVIEW ■

Thyroid Fine-Needle Aspiration Cytology Practice in Korea

Yoon Jin Cha · Ju Yeon Pyo
SoonWon Hong · Jae Yeon Seok[1]
Kyung-Ju Kim[2] · Jee-Young Han[3]
Jeong Mo Bae[4] · Hyeong Ju Kwon[5]
Yeejeong Kim[6] · Kyueng-Whan Min[7]
Soonae Oak[8] · Sunhee Chang[9]

Department of Pathology, Gangnam Severance Hospital, Yonsei University College of Medicine, Seoul; [1]Department of Pathology, Gachon University Gil Medical Center, Incheon; [2]Department of Pathology, Yeungnam University College of Medicine, Daegu; [3]Department of Pathology, Inha University Hospital, Incheon; [4]Department of Pathology, Seoul National University Hospital, Seoul; [5]Department of Pathology, Wonju Severance Christian Hospital, Yonsei University Wonju College of Medicine, Wonju; [6]National Health Insurance Service Ilsan Hospital, Goyang; [7]Department of Pathology, Hanyang University Guri Hospital, Hanyang University College of Medicine, Guri; [8]Department of Pathology, Ilsin Christian Hospital, Busan; [9]Department of Pathology, Inje University Ilsan Paik Hospital, Goyang, Korea

Received: August 12, 2017
Revised: September 16, 2017
Accepted: September 25, 2017

Corresponding Author
SoonWon Hong, MD, PhD
Department of Pathology, Gangnam Severance Hospital, Yonsei University College of Medicine, 211 Eonju-ro, Gangnam-gu, Seoul 06273, Korea
Tel: +82-2-2019-3540
Fax: +82-2-3463-2103
E-mail: soonwonh@yuhs.ac

We reviewed the current status of thyroid fine-needle aspiration cytology (FNAC) in Korea. Thyroid aspiration biopsy was first introduced in Korea in 1977. Currently, radiologists aspirate the thyroid nodule under the guidance of ultrasonography, and cytologic interpretation is only legally approved when a cytopathologist makes the diagnosis. In 2008, eight thyroid-related societies came together to form the Korean Thyroid Association. The Korean Society for Cytopathology and the endocrine pathology study group of the Korean Society for Pathologists have been updating the cytologic diagnostic guidelines. The Bethesda System for Reporting Thyroid Cytopathology was first introduced in 2009, and has been used by up to 94% of institutions by 2016. The average diagnosis rates are as follows for each category: I (12.4%), II (57.9%), III (10.4%), IV (2.9%), V (3.7%), and VI (12.7%). The malignancy rates in surgical cases are as follows for each category: I (28.7%), II (27.8%), III (50.6%), IV (52.3%), V (90.7%), and VI (100.0%). Liquid-based cytology has been used since 2010, and it was utilized by 68% of institutions in 2016. The categorization of thyroid lesions into "atypia of undetermined significance" or "follicular lesion of undetermined significance" is necessary to draw consensus in our society. Immunocytochemistry for galectin-3 and BRAF is used. Additionally, a molecular test for BRAF in thyroid FNACs is actively used. Core biopsies were performed in only 44% of institutions. Even the institutions that perform core biopsies only perform them for less than 3% of all FNACs. However, only 5% of institutions performed core biopsies up to three times more than FNAC.

Key Words: Bethesda; Fine needle aspiration cytology; Thyroid neoplasms; Korea

In this review, we surveyed the current status of thyroid fine-needle aspiration cytology (FNAC) in Korea and briefly described the history of FNAC in Korea. The multiple topics covered in this review include thyroid cytology sample collectors and interpreters, cytotechnician training programs, preparation methods for thyroid cytology samples, staining of thyroid cytology samples, thyroid cytology reporting systems and data distribution, the use of "atypia of undetermined significance" and "follicular lesion of undetermined significance" (AUS/FLUS) as diagnostic categories, the thyroid cytology audit program, correlation between cytology and histology, external quality assurance for thyroid FNAC, and the status of ancillary testing including core biopsy.

The 2016 survey project was designed to create a short communication that was compiled by a limited number of members of the Korean Society for Cytopathology (KSC). The 2012 survey[1] is more comprehensive than this 2016 survey, but we compared 2012 and 2016 data in this paper. We asked the KSC to administer the survey to the 210 institutions under its quality control.

pISSN 2383-7837
eISSN 2383-7845

Thirty-eight institutions responded to this survey within 3 weeks. The survey items included thyroid cytology sample collectors and interpreters, preparation methods for thyroid cytology samples, thyroid cytology reporting systems and data distribution, the use of AUS/FLUS categorization, correlation between cytology and histology, and the status of core biopsy. Twelve among the 38 responders answered distribution data for each class of the Bethesda reporting system. Eight institutions also sent correlation data between cytology and histology. Graphs and statistical analysis of Student's t test for categorical diagnosis rates were constructed using Excel Software. Categorical variables were expressed as percentage, frequency, and range. Differences were considered statistically significant at $p < .05$.

BRIEF HISTORY OF THYROID FINE-NEEDLE ASPIRATION CYTOLOGY

In Europe, thyroid FNAC was introduced in the 1950s,[2] but in the United States, it began to be more actively used in the 1970s.[3] Thyroid aspiration biopsy was first introduced in Korea by a physician in 1977.[4] As for the introduction of FNAC to pathologists, it was started by a pathologist who was experienced in aspiration cytology from cytopathology training in Europe in the 1980s.[5] In the early stages of use, aspirations were initially performed by a radiologist who had undergone European training, and the aspirates were sent to pathologists for interpretation. At that time, clinicians directly interpreted the aspiration cytology slide; however, with the development of quality control and insurance coverage, the frequency of clinician's interpretation has decreased. Initially, aspiration cytology was performed only on palpable lesions of superficial organs, but with the development of imaging system, it has become possible to accurately locate and aspirate nonpalpable lesions with very high diagnostic accuracy. Therefore, FNAC has become prevalent at the majority of medical institutions.[6] In 2006, pathologists created management guidelines for patients with thyroid nodules and thyroid cancer as a mainstay of the Korean Endocrine Society.[7] As a result, the Endocrine Pathology Study Group was created in 2007. In 2008, eight thyroid-related societies made up the Korean Thyroid Association. The main goal of the Endocrine Pathology Study Group and the KSC is to update the cytologic diagnostic guideline.[8] According to recent data, 196,000 cases of thyroid FNAC are performed each year in Korea, accounting for 60% of all FNACs performed.[9] Due to a perceived over-diagnosis of thyroid cancer, the performance of FNAC as a whole decreased by 19.6% in 2015 compared to 2014, based on the data of our survey.

THYROID CYTOLOGY SAMPLE COLLECTORS AND INTERPRETERS

In the early stages of FNAC, endocrinologists, surgeons, radiologists, and pathologists aspirated palpable thyroid nodules and interpreted them using a Giemsa stain. After ultrasonography was introduced, radiologists began to aspirate thyroid nodules more than any other specialists. Now, national insurance covers the aspiration fee only when radiologists aspirate the thyroid nodule by guided ultrasonography. While radiologists perform the aspiration, legal standards dictate that a diagnosis can only be made by a cytopathologist. Therefore, cytologic interpretation has been carried out at more than 200 cytology laboratories throughout the country, all of which are subject to quality control by the KSC.

CYTOTECHNICIAN TRAINING PROGRAM

In 1981, the Korean National Medical Center, under the auspices of the World Health Organization (WHO), launched a nationwide education program for cytotechnicians to work as cytology screeners. This is considered to be the first systematic cytologic education program in Korea. Under the direction of the WHO, this course was planned to increase early detection of cervical cancer, the most frequent cancer at the time, and it was a first step toward a national cancer eradication project (National Cancer Control Program) planned by the Health and Social Affairs Department (present Health and Welfare Department) in 1978. The need for systematic cytologic screening and education was widely supported by the government. To establish the Cytology School of the National Medical Center's Pathology Department, a Swedish cytotechnician, Barbro Nilsson, came to Korea in 1981 as a WHO adviser who worked as the primary cancer screening personnel (cytotechnician) educator. The WHO supported this adviser by providing educational equipment such as microscopes and lanterns; other healthcare professionals, like Swedish doctors Nils, who was the then president of the International Academy of Cytology (IAC), and Stormby, also donated slide teaching materials and textbooks for gynecological cytology. At that time, cytotechnician education was carried out at the National Medical Center with the main purpose of improving the quality of cytopathology examination conducted at university hospitals and general hospital nationwide. Under the direction of anatomic pathologists, we selected 10 cytotechnicians

 https://doi.org/10.4132/jptm.2017.09.26

who were in charge of cytology screening, nine of whom attended their first training course in November 1981, and in the second course of the following year, we again selected other 10 cytotechnicians and educated.[6]

Thyroid FNAC is included in the cytotechnician education program; however, participation in cytology screening depends on the institutional policy.

PREPARATION OF THYROID CYTOLOGY SAMPLES

At the time of introduction, FNACs were analyzed with air-dried Giemsa stain; since 2000, they have been read using an alcohol-fixed conventional smear with Papanicolaou staining and cell block preparation. Currently, a few institutions still use the air-dried Giemsa stain.

Liquid-based cytology (LBC), which decreases the rate of cell paucity, was introduced for FNAC preparation despite some inherent disadvantages such as unfamiliar cellular morphology. Since 2010, LBC had been widely adopted and was used by 44% of institutions in 2012 and by 68% in 2016. In the 2012 survey, LBC alone was not sufficient for diagnosis, so 22 of 33 institutions (67%) used LBC combined with conventional methods. In 2016, the number of institutions adding a supplemental method dropped to 12 of 23 (52%). This reduction can be attributed to increased experience in LBC interpretation.[1] For FNAC specimens, ThinPrep (45.0%) was most commonly used, followed by SurePath (33.6%), EASY Prep (12.9%), and Huro Path (4.5%).[9]

STAINING OF THYROID CYTOLOGY SAMPLES

Staining was initially performed using air-dried Giemsa stain, but the Papanicolaou stain has become predominant. However, some institutions still use hematoxylin and eosin or Giemsa stains.

THYROID CYTOLOGICAL REPORTING SYSTEMS AND DATA DISTRIBUTION

Until the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) appeared in 2008,[10] thyroid FNAC was diagnosed in a variety of ways; at some institutions, it was diagnosed using descriptive terms, in the same format as a pathology diagnosis.

The diagnosis of thyroid FNAC was made based mainly on the Papanicolaou Society Guidelines;[11] as a result, the Korean Endocrine Society published management guidelines for patients with thyroid nodules and thyroid cancer in 2006.[7]

After that, the Endocrine Pathology Study Group was created in 2007. In 2008, eight thyroid-related societies made up the Korean Thyroid Association, and TBSRTC was introduced to Korea in 2009. The Endocrine Pathology Study Group and the KSC have been updating the cytologic diagnostic guideline

Table 1. Cytologic diagnosis rates according to TBSRTC

Institution	Category					
	I	II	III	IV	V	VI
1	8.6	61.7	6.1	5.5	3.4	14.7
2	16.2	51.9	18.9	0.6	3.3	9.1
3	8.0	61.0	2.3	0.6	4.5	23.6
4	15.4	51.5	16.5	0.5	4.8	11.4
5	8.1	60.0	11.8	1.1	2.9	16.2
6	11.2	56.9	7.6	1.0	4.3	19.0
7	20.1	34.1	21.9	0.7	5.7	17.4
8	20.3	44.5	18.1	2.0	3.0	12.2
9	32.6	61.5	3.2	0.2	1.9	0.6
10	2.9	86.5	2.7	0.0	0.6	7.3
11	0.0	74.9	9.7	0.0	3.4	12.0
12	5.5	50.0	6.6	22.3	6.8	8.9
Average (%)	12.4	57.9	10.4	2.9	3.7	12.7
Range (%)	0–32.6	34.1–86.5	2.3–21.9	0–22.3	0.6–6.8	0.6–23.6
Average (%)[12]	12.9	59.3	9.6	10.1	2.7	5.4
Range (%)[12]	1.8–23.6	39.0–73.8	3.0–27.2	1.2–25.3	1.4–6.3	2.0–16.2
p-value[a]	.790	.723	.992	.053	.385	.008

TBSRTC, the Bethesda System for Reporting Thyroid Cytopathology.
[a]Statistical analysis of student's t test for categorical diagnosis rate between this survey data and reference data were constructed from Excel Software. Differences for which $p < .05$ were considered significant.

based on TBSRTC.[8] According to the survey conducted in 2012, 60 of the 74 responding institutions (80%) used TBSRTC,[1] while the 2016 survey showed that 94% (34/36) are using TBSRTC.

Twelve institutions reported distribution data according to class in the Bethesda reporting system. Eight institutions also sent correlation data between cytology and histology.

The categorical diagnosis rate of each institution by TBSRTC is as follows: I (12.4%), II (57.9%), III (10.4%), IV (2.9%), V (3.7%), and VI (12.7%), as shown in Table 1 and Fig. 1.

The average value is not very unique compare to worldwide reports,[12] but some categories show significant differences between this present survey data and Bongiovanni's meta-analysis data.[12] Categories 1, 2, 3, and 5 showed rates similar to Bongiovanni's data. However, category 6 was significantly higher in this Korean survey than in Bongiovanni's data (12.7% vs 5.4%, p = .008). Additionally, category 4 was lower in this Korean data than in Bongiovanni's data, but the difference was not statistically significant (2.9% vs 10.1%, p = .053).

This survey is limited in that the proportion of referral hospitals is higher than the distribution of nationwide laboratories in Korea.

USE OF ATYPIA OF UNDETERMINED SIGNIFICANCE AND FOLLICULAR LESION OF UNDETERMINED SIGNIFICANCE

In this survey, we looked at how to appropriately use the AUS/FLUS terminology of TBSRTC. In 66% of institutions, the two terms are used to indicate the same lesion, although most use only the term AUS. Only two institutions use the term AUS/FLUS.

In the remaining 34% of institutions, the two terms are used differently. AUS is used in cases with nuclear atypia, and FLUS is used in cases where there is architectural atypia. This is not consistent with the original intent of TBSRTC. In the case of AUS/FLUS, subcategorized studies are often reported by multiple institutions.[13] Confusion exists surrounding this term, and it is necessary to establish consensus on the proper use of this term.

THYROID CYTOLOGY AUDIT PROGRAM: CORRELATION BETWEEN CYTOLOGY AND HISTOLOGY

The quality control system is managed by each institution, and reports on the quality control of thyroid FNACs have been published since 1996.[14,15]

Accuracy was assessed based on malignancy rates using cytohistological correlations, but some reports assessed the accuracy of each specific diagnostic entity. There are about 12 papers addressing this accuracy, of which about four demonstrated accuracy using TBSRTC.[1,16-18]

The malignancy rates for overall cytologic diagnoses are as follows for each category: I (1.8%), II (0.7%), III (6.3%), IV (19.1%), V (51.9%), and VI (63.5%) (Fig. 2A). The malignancy rates for surgical cases are as follows for each category: I (28.7%), II (27.8%), III (50.6%), IV (52.3%), V (90.7%), and VI (100.0%) (Fig. 2B).

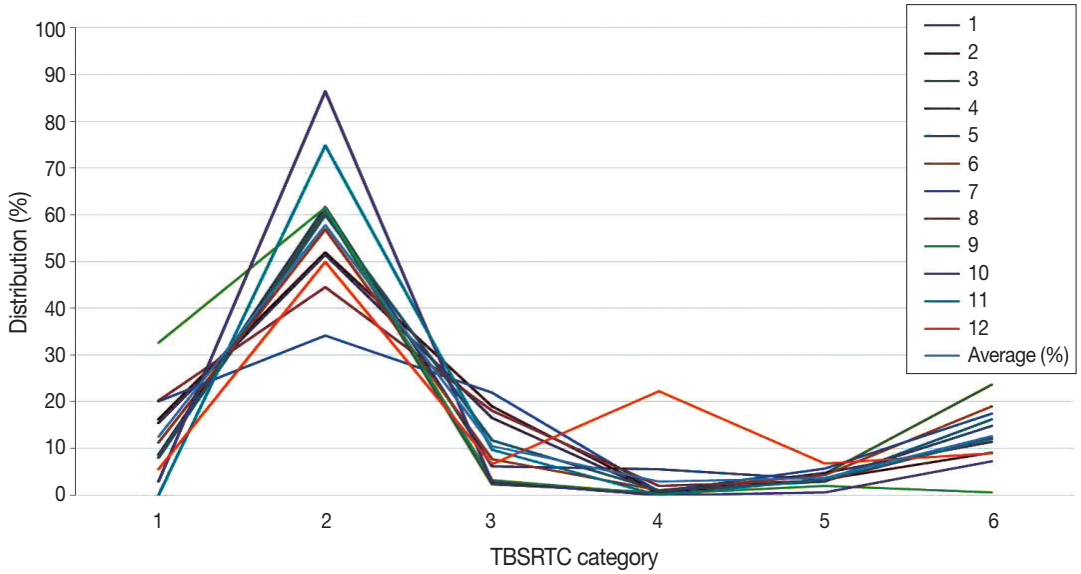


Fig. 1. The diagnosis rate of each institution by the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) category.

https://doi.org/10.4132/jptm.2017.09.26

EXTERNAL QUALITY ASSURANCE FOR THYROID FINE-NEEDLE ASPIRATION CYTOLOGY

Since 1995, the KSC has been in charge of quality control of fine needle aspiration smears, and the quality control findings have been reported since 1999.[6] Quality evaluation by the Korean Society of Pathologists was added in 2007, and each institute is making efforts to improve the quality of thyroid FNAC, as well as overall cytopathology.[19]

THE STATUS OF ANCILLARY TESTING INCLUDING CORE BIOPSY

Immunocytochemical staining could theoretically be used to make cytologic diagnoses, but immunocytochemical targets useful for thyroid cytology have not been identified. There have been many reports of galectin-3 being applied to cytologic diagnosis since 2008, but the results have been controversial.[20-22]

β-Catenin, CXCL12, and other rare immunocytochemical stains are also used for cytology. There are current reports of immunocytochemistry for BRAF, and some institutions use it for actual analysis.[23-26] Immunostaining for cytologic examination is also covered by insurance.

Since 2010, a molecular pathology approach, including the *BRAF*V600E mutation, has been utilized, and various molecular pathological studies are underway. However, only the BRAF test is actively used at present and approved by insurance.[27-30]

In recent years, core biopsies have been performed mainly in Korea and Italy. However, core biopsies were performed in only 17 of the 36 institutions (44%) in this survey. Even institutions that perform core biopsy used the procedure in less than 3% of FNACs.

However, at two of these institutions, core biopsy was performed up to three times more than FNAC. There are 45 articles on core biopsies written by Korean authors, most of which are reported by radiologists who prefer core biopsy.

These radiologists asked pathologists to establish a standardized classification for core biopsy compatible with the TBSRTC. Pathologists agreeing with this idea have published a paper that standardizes core biopsy readings.[31]

In Korea, pathologists are interpreting both pathology and cytopathology. Therefore, for pathologists, histologic interpretation of a core biopsy per se is easier than the cytological interpretation of FNAC if difficulties or side effects accompanying the core biopsy procedure are not considered.

However, considering that we often cannot distinguish adenomatous hyperplasia from follicular neoplasm, even if we examine all surgical specimens, it is controversial to say that it is more accurate to make this distinction with core biopsy than with FNAC.

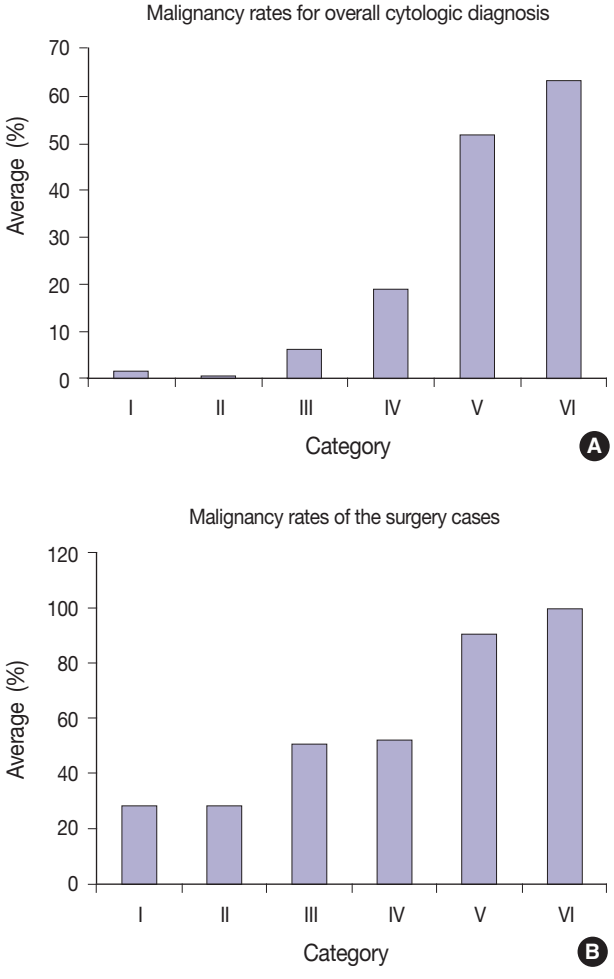


Fig. 2. (A) The malignancy rates for overall cytologic diagnoses are as follows for each category: I (1.8%), II (0.7%), III (6.3%), IV (19.1%), V (51.9%), and VI (63.5%). (B) The malignancy rates for surgical cases are as follows for each category: I (28.7%), II (27.8%), III (50.6%), IV (52.3%), V (90.7%), and VI (100.0%).

CONCLUSION

Currently, radiologists aspirate thyroid nodules under the guidance of ultrasonography, and legal standards dictate that a diagnosis can only be made by a cytopathologist.

The TBSRTC was used by up to 94% of institutions in 2016. The average diagnosis rates are as follows for each category: I (12.4%), II (57.9%), III (10.4%), IV (2.9%), V (3.7%), and VI (12.7%). The malignancy rates in surgical cases are as follows for

each category: I (28.7%), II (27.8%), III (50.6%), IV (52.3%), V (90.7%), and VI (100.0%).

Since 2010, LBC has been used and was implemented by 68% of institutions in 2016.

It is necessary to draw consensus on the use of the terms AUS and FLUS.

Immunocytochemistry is used in galectin-3 and BRAF assays. For molecular tests in thyroid FNACs, BRAF is actively used. Core biopsies are performed only rarely in a few institutions.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

Acknowledgments

We would like to thank the participants of members of the Korean Society of Cytopathology: Dr. Young Lyun Oh, Samsung Medical Center, Sungkyunkwan University School of Medicine, Seoul; Dr. Ju Hie Lee, Kyung Hee University Medical Center, Seoul; Dr. Ae Ri An, Chonbuk National University Hospital, Jeonju; Dr. Hee Sung Kim, Chung-Ang University College of Medicine, Seoul; Dr. Jin Hee Sohn, Kangbuk Samsung Hospital, Sungkyunkwan University School of Medicine, Seoul; Dr. Yosep Chong, College of Medicine, The Catholic University of Korea, Seoul; Dr. Dong Eun Song, Asan Medical Center, University of Ulsan College of Medicine, Seoul; Dr. Hyun Ju Lee, Soonchunhyang University Cheonan Hospital, Soonchunhyang University College of Medicine, Cheonan; Dr. Tae Sook Hwang, Konkuk University School of Medicine, Seoul; Dr. Mi Kyung Shin, Hallym University Kangnam Sacred Heart Hospital, Seoul; Dr. Kang Min Han, Dongguk University Ilsan Hospital, Ilsan; Yeong-Seon Hong, Good Samsun Hospital, Busan; Dr. Myoung Jin Ju, Presbyterian Medical Center, Jeonju; Dr. Hye-Sun Kim, Cheil General Hospital & Women's Healthcare Center, Dankook University College of Medicine, Seoul; Dr. Yoon Hee Jin, Seongnam Central Hospital, Seongnam; Dr. Dongyoul Choi, GC Labs, Youngin, Dr. Byung Doo Lee, Seegene Medical Foundation, Seoul; Dr. Yoo Duk Choi, Chonnam National University Medical School, Gwangju; Dr. Song-Yi Choi, Chungnam National University School of Medicine, Daejeon; Dr. Min Gyoung Pak, Dong-A University College of Medicine, Busan; Dr. Ji-Young Choe, Hallym University Sacred Heart Hospital, Anyang; Dr. Hyojin Kim, Seoul National University Bundang Hospital, Seongnam; Dr. Songmi Noh, CHA Gangnam Medical Center, CHA University, Seoul; and Dr. Hyunju Yoo, Daerim Saint Mary's Hospital Thyroid Center, Seoul, Korea.

REFERENCES

1. Kim M, Park HJ, Min HS, *et al*. The use of the Bethesda System for Reporting Thyroid Cytopathology in Korea: a nationwide multicenter survey by the Korean Society of Endocrine Pathologists. J Pathol Transl Med 2017; 51: 410-7.
2. Lipton RF, Abel MS. Aspiration biopsy and the thyroid in evaluation of thyroid dysfunction. Am J Med Sci 1944; 208: 736-42.
3. Crile G Jr, Hawk WA Jr. Aspiration biopsy of thyroid nodules. Surg Gynecol Obstet 1973; 136: 241-5.
4. Lee MH. Thyroid biopsy. Korean J Med 1977; 20: 731-6.
5. Park HS. Cytohistopathologic comparative study of aspiration biopsy cytology from various sites. Korean J Cytopathol 1991; 2: 8-19.
6. Lee K. The history of the Korean Society for Cytopathology. Seoul: The Korean Society for Cytopathology, 2006; 15-9.
7. Kim WB, Kim TY, Kwon HS, *et al*. Management guidelines for patients with thyroid nodules and thyroid cancer. J Korean Endocr Soc 2007; 22: 157-87.
8. Yi KH, Lee EK, Kang HC, *et al*. 2016 Revised Korean Thyroid Association management guidelines for patients with thyroid nodules and thyroid cancer. Int J Thyroidol 2016; 9: 59-126.
9. Oh EJ, Jung CK, Kim DH, *et al*. Current cytology practices in Korea: a nationwide survey by the Korean Society for Cytopathology. J Pathol Transl Med 2017 Sep 27 [Epub]. https://doi.org/10.4132/jptm.2017.08.11.
10. Cibas ES, Ali SZ; NCI Thyroid FNA State of the Science Conference. The Bethesda System For Reporting Thyroid Cytopathology. Am J Clin Pathol 2009; 132: 658-65.
11. Guidelines of the Papanicolaou Society of Cytopathology for the examination of fine-needle aspiration specimens from thyroid nodules. The Papanicolaou Society of Cytopathology Task Force on Standards of Practice. Diagn Cytopathol 1996; 15: 84-9.
12. Bongiovanni M, Spitale A, Faquin WC, Mazzucchelli L, Baloch ZW. The Bethesda System for Reporting Thyroid Cytopathology: a meta-analysis. Acta Cytol 2012; 56: 333-9.
13. Jung YY, Jung S, Lee HW, Oh YL. Significance of subcategory atypia of undetermined significance/follicular lesion of undetermined significance showing both cytologic and architectural atypia in thyroid aspiration cytology. Acta Cytol 2015; 59: 370-6.
14. Kwon KH, Jin SY, Lee DW. A study of usefulness of fine needle aspiration cytology of the thyroid lesions. Korean J Cytopathol 1996; 7: 111-21.
15. Park KM, Ko IH. Diagnostic accuracy of fine needle aspiration cytology in thyroid lesions: analysis of histologically confirmed 153 cases.

Korean J Cytopathol 1996; 7: 122-33.
16. Park JH, Yoon SO, Son EJ, Kim HM, Nahm JH, Hong S. Incidence and malignancy rates of diagnoses in the bethesda system for reporting thyroid aspiration cytology: an institutional experience. Korean J Pathol 2014; 48: 133-9.
17. Lee YB, Cho YY, Jang JY, *et al*. Current status and diagnostic values of the Bethesda system for reporting thyroid cytopathology in a papillary thyroid carcinoma-prevalent area. Head Neck 2017; 39: 269-74.
18. Lee K, Jung CK, Lee KY, Bae JS, Lim DJ, Jung SL. Application of Bethesda system for reporting thyroid aspiration cytology. Korean J Pathol 2010; 44: 521-7.
19. The Korean Society of Pathologist. The History of the Korean Society of Pathologists 2006~2015. Seoul: The Korean Society of Pathologist, 2016; 20-1.
20. Jung CK, Lee A, Jung ES, Choi YJ, Jung SL, Lee KY. Split sample comparison of a liquid-based method and conventional smears in thyroid fine needle aspiration. Acta Cytol 2008; 52: 313-9.
21. Kim ES, Lim DJ, Lee K, *et al*. Absence of galectin-3 immunostaining in fine-needle aspiration cytology specimens from papillary thyroid carcinoma is associated with favorable pathological indices. Thyroid 2012; 22: 1244-50.
22. Gweon HM, Kim JA, Youk JH, *et al*. Can galectin-3 be a useful marker for conventional papillary thyroid microcarcinoma? Diagn Cytopathol 2016; 44: 103-7.
23. Koo JS, Jung W, Hong SW. Cytologic characteristics and beta-catenin immunocytochemistry on smear slide of cribriform-morular variant of papillary thyroid carcinoma. Acta Cytol 2011; 55: 13-8.
24. Jung YY, Park IA, Kim MA, Min HS, Won JK, Ryu HS. Application of chemokine CXC motif ligand 12 as a novel diagnostic marker in preoperative fine-needle aspiration biopsy for papillary thyroid carcinoma. Acta Cytol 2013; 57: 447-54.
25. Lee SR, Yim H, Han JH, *et al*. VE1 antibody is not highly specific for the *BRAF* V600E mutation in thyroid cytology categories with the exception of malignant cases. Am J Clin Pathol 2015; 143: 437-44.
26. Kim YH, Yim H, Lee YH, *et al*. Evaluation of the VE1 antibody in thyroid cytology using ex vivo papillary thyroid carcinoma specimens. J Pathol Transl Med 2016; 50: 58-66.
27. Kwak JY, Kim EK, Kim JK, *et al*. Dual priming oligonucleotide-based multiplex PCR analysis for detection of *BRAF*V600E mutation in FNAB samples of thyroid nodules in *BRAF*V600E mutation-prevalent area. Head Neck 2010; 32: 490-8.
28. So YK, Son YI, Park JY, Baek CH, Jeong HS, Chung MK. Preoperative *BRAF* mutation has different predictive values for lymph node metastasis according to tumor size. Otolaryngol Head Neck Surg 2011; 145: 422-7.
29. Lee SE, Hwang TS, Choi YL, *et al*. Molecular profiling of papillary thyroid carcinoma in Korea with a high prevalence of *BRAF*V600E mutation. Thyroid 2017; 27: 802-10.
30. Kim Y, Choi KR, Chae MJ, *et al*. Stability of DNA, RNA, cytomorphology, and immunoantigenicity in residual ThinPrep specimens. APMIS 2013; 121: 1064-72.
31. Jung CK, Min HS, Park HJ, *et al*. Pathology reporting of thyroid core needle biopsy: a proposal of the Korean Endocrine Pathology Thyroid Core Needle Biopsy Study Group. J Pathol Transl Med 2015; 49: 288-99.

Journal of Pathology and Translational Medicine 2017; 51: 528-532
https://doi.org/10.4132/jptm.2017.09.12

■ REVIEW ■

History and Practice of Thyroid Fine-Needle Aspiration in China, Based on Retrospective Study of the Practice in Shandong University Qilu Hospital

Zhiyan Liu[1,2] · Dongge Liu[3,4]
Bowen Ma[5] · Xiaofang Zhang[1,2]
Peng Su[1] · Li Chen[6] · Qingdong Zeng[7]

[1]Department of Pathology, School of Basic Medical Science, Shandong University, Jinan; [2]Department of Pathology, Shandong University Qilu Hospital, Jinan; [3]Department of Pathology, Beijing Hospital, Beijing; [4]Chinese Cytology Association, Beijing; [5]Department of Cytopathology, Cancer Hospital of Xinjiang Medical University, Wulumuqi; [6]Department of Endocrinology, Shandong University Qilu Hospital, Jinan; [7]Department of General Surgery, Shandong University Qilu Hospital, Jinan, China

Received: July 26, 2017
Revised: September 8, 2017
Accepted: September 12, 2017

Corresponding Author
Zhiyan Liu, MD, PhD
Department of Pathology, School of Basic Medical Science, Shandong University, Jinan, Shandong, China
Tel: +86-18560081167
Fax: +86-53182679225
E-mail: zhiyanliu@sdu.edu.cn

Cytology in China developed from nothing and underwent a long journey from gynecologic cytology to that of all organs, laying a solid foundation for new developments in the 21st century. Thyroid fine-needle aspiration (FNA) was primarily developed in an endocrinology department and then in the clinical laboratory department or pathology department in the 1970–80s. Wrights staining is popular in endocrine and clinical laboratory departments, while hematoxylin and eosin staining is common in pathology. Liquid based cytology is not common in thyroid FNA cytology, while *BRAF*^{V600E} mutation analysis has been the most popular molecular test. The history and practice of thyroid FNA practice in China were reviewed based on retrospective study of the practice in Qilu Hospital of Shandong University.

Key Words: Thyroid fine needle aspiration; Practice; China; Qilu Hospital

THE BRIEF HISTORY OF CHINESE CYTOPATHOLOGY

In the 1950s, Dr. Dawang Yang returned to China to start cervical cytology after completing her academic studies in the United States.[1,2] She began the practice of Papanicolaou cervical smear classification and a cervical cancer screening program in Peking Union Hospital.[3] Vaginal Cytopathology was published by Dr. Dawang Yang in 1952, which was the first Chinese cytology book and marked the start of modern cytopathology in China.

Esophageal balloon cytology was developed for screening of esophageal cancer, and a series of English publications from China made it well known around the world since the 1960s.[4-6] Fine-needle aspiration (FNA) was applied first on the body surface and then in deep organs in the 1970–80s. Bone tumor cytodiagnosis was developed by hematologist and cytologist Dr. Xiaojing Peng,[7] who published the first Chinese FNA book, Atlas of Clinical Cytology, in 1972. The Chinese Academy of Cytology was founded and the first National Clinical Cytology Conference was held in 1985, which was a milestone of cytology in China.[8]

New ancillary techniques such as immunocytochemistry, flow cytometry and DNA alteration analysis were applied in addition to cytopathology starting at the end of 1980s.[1,9] Liquid-based cytology was initially applied to the cervical smear in the 1990s and greatly improved slide quality and accuracy of diagnosis. The Bethesda System (TBS) for reporting cervical cytology replaced

pISSN 2383-7837
eISSN 2383-7845

Papanicolaou classification, and computer-aided analysis started to play a role.

Cytology has started to play a more prominent role in diagnosis, and quality control of cytology has further improved during the recent 10 years. The Cytology Operational Manual and Quality Control Standards were proposed by the Cytology Section of the Chinese Pathology Association in 2007. A Cytology Quality Control Expert Team was formed in 2010 to supervise clinical diagnosis and academic training, and international exchanges became more popular.

THYROID FINE-NEEDLE ASPIRATION IN CHINA

Thyroid FNA was primarily developed in endocrinology departments and became popular around China in the 1970–80s. Wright's stain was originally the most popular staining method and was founded by hematologist and cytologist Dr. Xiaojing Peng. In 1987, a national conference posited that FNA cytology should be a branch of pathology, and the cytologist should have a background of surgical pathology. Thyroid FNA began to increase in popularity in some pathology departments, and hematoxylin and eosin staining was applied because it increased the ease of comparing histological samples. Ultrasound-guided thyroid FNA (UG-FNA) became popular gradually, and molecular examination was applied as an additional diagnostic method for papillary thyroid carcinoma (PTC).

MOLECULAR TESTING OF THYROID FINE-NEEDLE ASPIRATION IN CHINA

Next-generation sequencing was proposed recently to improve the diagnosis of thyroid FNA specimens with indeterminate cytology; however, it is expensive and not well accepted in clinics in China.[10-12] *BRAF*^{V600E} mutation analysis was recommended by American Thyroid Association (ATA) guidelines as an auxiliary diagnostic method for thyroid FNA cytology.[13,14] The amplification refractory mutation system is efficient and inexpensive and is the most popular method for detection of *BRAF*^{V600E} mutation in China. Zhang *et al*.[14] recently studied the Thyroid Imaging Reporting and Data System (TIRADS), Bethesda System for Reporting Thyroid Cytopathology (BSRTC), and the *BRAF*^{V600E} mutation analysis as molecular tools in diagnosing thyroid carcinoma. The TIRADS was applied for selecting patients for FNA biopsy and *BRAF*^{V600E} mutation analysis. They found that *BRAF*^{V600E} mutation detection had the best sensitivity, specificity, and accuracy among the three methods. Both TIRADS and *BRAF*^{V600E} detection showed increased sensitivity and accuracy when combined with BSRTC. Of all methods, a combination of BSRTC and *BRAF*^{V600E} mutation detection demonstrated the best diagnostic efficiency.[14]

PRACTICE IN QILU HOSPITAL, SHANDONG PROVINCE

Thyroid FNA is not yet well accepted in China, and most general hospitals use frozen sectioning as a diagnostic method instead of thyroid FNA. Some hospitals began to use UG-FNA around the 1990s, along with diagnostic frozen sectioning. UG-FNA is more popular in local hospital than in general hospital. Qilu Hospital is one of the epitomes of thyroid FNA in China, and began performing non-ultrasound guided thyroid FNA cytology in the endocrinology department (including both FNA and cytopathology) in 1991. UG-FNA for thyroid nodules began in 2014, the pathologist began to sign the thyroid FNA report instead of the endocrinologist.

Another point to note is that most Chinese patients choose to undergo diagnostic surgery in the presence of unfavorable clinical and ultrasonographic features, no matter what the size is. There were 2,612 thyroid surgeries, all with a rapid intraoperative pathological diagnosis using diagnostic frozen section, from 2015 to 2016 in our department.[15] However, only 791 thyroid FNA patients (30.3%) underwent thyroid surgery. This indicates that there is still ample opportunity for advancement of thyroid FNA in China. Thyroid FNA should be performed to avoid unnecessary surgery for benign thyroid lesions.

Materials and methods

A retrospective study was conducted of all patients with UG-FNA between January 2014 and April 2017 in Qilu Hospital, Shandong University. All patients had available thyroid ultrasound records. Approval was obtained from Qilu Hospital ethics committee, and the patients provided written informed consent. Criteria for FNA were those of Dr. Zhu *et al*.[16] Hematoxylin and eosin–stained slides of all tumors were reviewed by three pathologists (Z.L., X.Z., and P.S.). Diagnosis was made according to BSRTC, as shown in Table 1; the only exception was that the cystic-only group was classified into the benign group rather than as nondiagnostic.[17]

Criteria for surgical treatment

Thyroid surgery was recommended to all patients with suspicion of malignancy or malignancy reports. Diagnostic surgery and

frozen sectioning were suggested for patients with high-risk clinical or ultrasonographic features. Patients with at least one atypia of undetermined significance or follicular lesion of undetermined significance (AUS/FLUS) report were recommended for repeat FNA or diagnostic partial thyroid lobectomy. Once malignant histological evidence was demonstrated by frozen section, lobectomy was performed for papillary thyroid microcarcinoma (PTMC), and total thyroidectomy was usually performed for tumors larger than 1 cm with lymph node metastasis. Patients refusing surgical treatment or with high surgical risk were recommended for clinical and ultrasonographic follow-up. Patients with benign FNA cytology diagnosis underwent surgery only when there were clinical symptoms. Patients with nondiagnostic reports were recommended for repeat FNA or clinical and ultrasonographic follow-up.

Results and discussion

As shown in Table 1, FNA performed on 2838 thyroid nodules showed 3.6% nondiagnostic specimens, 44.7% benign nodules, 7.1% indeterminate (6.9% AUS/FLUS and 0.2% follicular neoplasm or suspicious for a follicular neoplasm [FN/SFN]), 14.1% suspicious for malignancy, and 30.6% positive for malignancy. The correlation between FNA cytology and histological diagnosis is shown in Table 2.

Table 1. Results of FNA cytology according to the Bethesda System for reporting thyroid FNA

Bethesda category	Nodule
Nondiagnostic	101 (3.6)
Benign	1,268[a] (44.7)
AUS/FLUS	195 (6.9)
FN/SFN	5 (0.2)
Suspicious for malignancy	401 (14.1)
Malignancy	868 (30.6)
Total	2,838 (100)

Values are presented as number (%).
FNA, fine-needle aspiration; AUS/FLUS, atypia of undetermined significance or follicular lesion of undetermined significance; FN/SFN, follicular neoplasm or suspicious for a follicular neoplasm.
[a]151 cyst fluid only.

The most common FNA diagnosis rendered in our practice was benign, which was nearly the same as the Bethesda expected incidence. Ninety cases of benign nodules were followed with clinical management because of high-risk ultrasound results, 31 cases were proved to be PTMC less than 5 mm in diameter, and one case was mucosa-associated B-cell lymphoma. This positive rate of FNA for PTMC less than 5 mm is worse than other practices in China, probably due to the unskillful operation of the UG-FNA for thyroid nodules less than 5 mm in diameter.[13] Nodules less than 1 cm in size should be followed up closely according to the ATA guideline,[13,18] which is not well accepted in China. Two cases of FN/SFN were proved to be follicular carcinoma (FTC), and one was proved to be follicular tumor of uncertain malignant potential, which indicated the difficulty to cytologically differentiate atypical follicular adenoma from minimally invasive FTC.[19]

Fifteen of 21 cases of AUS/FLUS were proved to be histologically malignant. These results were different from another study in China, as shown in Table 2. The risks of malignancy (ROMs) of AUS/FLUS and suspicious for malignancy were nearly the same, likely due to the different diagnostic criteria for the cytopathologist; when there are less than 30 atypical cells, we make the diagnosis of "AUS/FLUS" instead of "suspicious of malignancy." This is an attempt by the pathologist to suggest a repeat FNA instead of diagnostic surgery for such lesions. In the suspicious of malignancy group, there were three cases of hyalinizing trabecular tumor (HTT). As shown in Fig. 1A and B, there were obvious pseudoinclusions in the cell smear of HTT. Although the nuclear clearing is not conspicuous, the cells were of variable size with nuclear atypia and nuclear grooves, which led to the over-diagnosis of PTC. However, filament-like hyalinizing material was found around the tumor cells, suggesting the possibility of HTT, which is classified as borderline tumor in the new World Health Organization classification of endocrine tumors.[20]

Patients in the "nondiagnostic" group will not undergo immediate operation, and repeat FNA is recommended after 3-month follow-up in our practice. Only six cases were followed by operation,

Table 2. Correlation of FNA cytology with histological diagnosis for the 791 cases

Bethesda category	Nondiagnostic	Benign	AUS/FLUS	FN/SFN	Suspicious for malignancy	Malignancy	Total
Cases for surgery	6	90	21	3	200	471	791
Malignancy on surgery	0	31	15	2	168	457	673
ROM (%)	0	34.4	71.4	66.7	69	97.0	85.1
ROM from Zhang *et al.*[14] (%)[a]	27.9	7.9	45.5	75.5	98.5	100	65.5
ROM from Haugen[13] (%)[b]	20	2.5	14	25	70	99	Not available

FNA, fine-needle aspiration; AUS/FLUS, atypia of undetermined significance or follicular lesion of undetermined significance; FN/SFN, follicular neoplasm or suspicious for a follicular neoplasm.
[a]Risk of malignancy (ROM) in another two thyroid FNA practices in China; [b]ROM in practice in United States.

https://doi.org/10.4132/jptm.2017.09.12

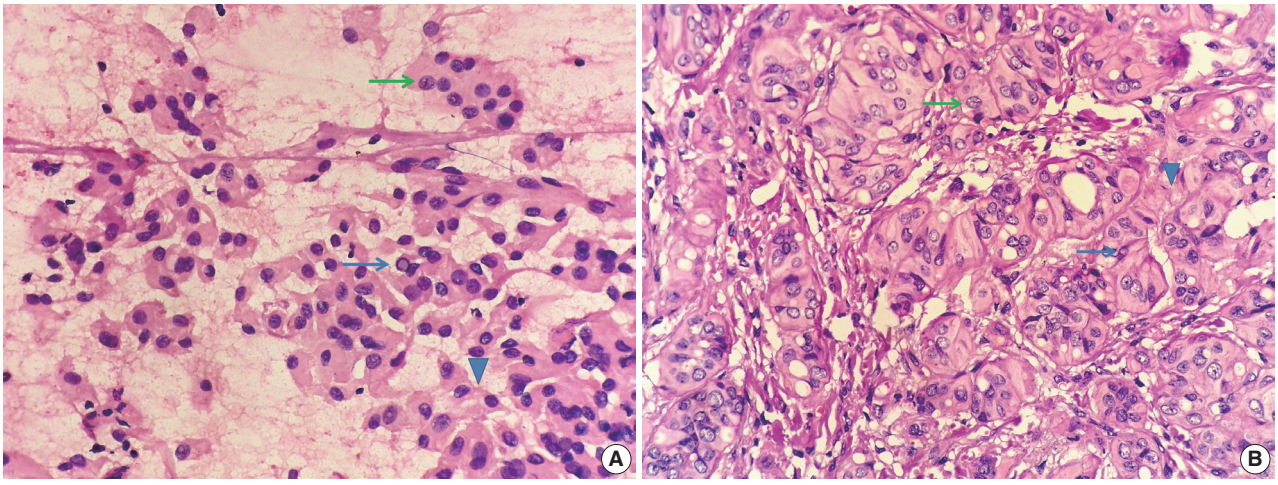


Fig. 1. (A) Cytology of hyalinizing trabecular tumor. The thin green arrow shows nuclear groove, and the thin blue arrow shows pseudoinclusion. The arrowhead shows filament-like hyalinizing material between the tumor cells. (B) Histology of hyalinizing trabecular tumor. The thin green arrow shows nuclear groove, and the thin blue arrow shows pseudoinclusion. The arrowhead shows hyalinizing material around the cell nests.

and none of them was malignant. The ROM of this category was different from that of another practice in China and Dr. Ali's practice in the United Sates.[14,21] Routine *BRAF*^{V600E} mutation analysis was applied to all FNA samples in Dr. Zhang's practice, which could improve the accuracy rate of FNA.[14] This is likely one reason that the operation rate was low in our practice for patients in the nondiagnostic category. Another reason could be a significant number of patients went to another hospital instead of our hospital because of bed tension or economic issue. Meanwhile, Dr. Zhang's result suggests that routine *BRAF*^{V600E} mutation analysis is necessary to improve the efficiency, sensitivity, and specificity of thyroid FNA cytology.

Above all, thyroid FNA is an important part of preoperative diagnosis, but it is still in an early stage in China compared with histopathology. Ultrasound, radiology, physical presentation, and molecular features of thyroid nodules are critical to achieve an accurate cytologic diagnosis. The last decade has witnessed rapid development of thyroid FNA in China. The next decade will offer exciting opportunities, and international exchange and cooperation are necessary for cytologists to develop thyroid FNA to a higher level.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

REFERENCES

1. Shu YJ, Kan X. Cytopathology in China. Zhonghua Bing Li Xue Za Zhi 2005; 34: 625-7.
2. Liu DG. Cytology development in recent 10 years and the future outlook in China. J Am Soc Cytopathol 2016; 5: 10-3.
3. Liu DG. Importance of cytopathology. Zhonghua Bing Li Xue Za Zhi 2009; 38: 799-805.
4. Shen O, Liu SF, Dawsey SM, *et al*. Cytologic screening for esophageal cancer: results from 12,877 subjects from a high-risk population in China. Int J Cancer 1993; 54: 185-8.
5. Shen Q, Qiu S, Zhao H. Report on cytopathological diagnosis for esophageal carcinoma screening. Chin J Pathol 1963; 7: 19-24.
6. Li FP, Shiang EL. Screening for oesophageal cancer in 62 000 Chinese. Lancet 1979; 2: 804.
7. Peng XJ, Yan XC. Bone tumor cytodiagnosis with fine-needle aspiration: a preliminary report. Chin Med J (Engl) 1983; 96: 611-8.
8. Zhang Y. Review and prospect of fifty years of Chinese clinical cytology. Chin J Diagn Pathol 1999; 6: 252-5.
9. Fan YB, Wu X, Fu ZM, Wu GP. Amplification of the human telomerase gene in liquid-based preparations is associated with cervical dysplasia and carcinoma. Int J Gynecol Pathol 2010; 29: 157-64.
10. Nikiforov YE, Carty SE, Chiosea SI, *et al*. Impact of the multi-gene ThyroSeq next-generation sequencing assay on cancer diagnosis in thyroid nodules with atypia of undetermined significance/follicular lesion of undetermined significance cytology. Thyroid 2015; 25: 1217-23.
11. Nikiforov YE, Carty SE, Chiosea SI, *et al*. Highly accurate diagnosis of cancer in thyroid nodules with follicular neoplasm/suspicious

for a follicular neoplasm cytology by ThyroSeq v2 next-generation sequencing assay. Cancer 2014; 120: 3627-34.
12. Le Mercier M, D'Haene N, De Nève N, *et al*. Next-generation sequencing improves the diagnosis of thyroid FNA specimens with indeterminate cytology. Histopathology 2015; 66: 215-24.
13. Haugen BR. 2015 American Thyroid Association management guidelines for adult patients with thyroid nodules and differentiated thyroid cancer: what is new and what has changed? Cancer 2017; 123: 372-81.
14. Zhang YZ, Xu T, Cui D, *et al*. Value of TIRADS, BSRTC and FNA-*BRAF* V600E mutation analysis in differentiating high-risk thyroid nodules. Sci Rep 2015; 5: 16927.
15. Liu Z, Song Y, Han B, Zhang X, Su P, Cui X. Non-invasive follicular thyroid neoplasm with papillary-like nuclear features and the practice in Qilu Hospital of Shandong University, China. J Basic Clin Med 2017; 6: 22-5.
16. Zhu Y, Dai J, Lin X, Wu H, Wang T. Fine needle aspiration of thyroid nodules: experience in a chinese population. J Basic Clin Med 2015; 4: 65-9.
17. Cibas ES, Ali SZ; NCI Thyroid FNA State of the Science Conference. The Bethesda System For Reporting Thyroid Cytopathology. Am J Clin Pathol 2009; 132: 658-65.
18. Haugen BR, Alexander EK, Bible KC, *et al*. 2015 American Thyroid Association management guidelines for adult patients with thyroid nodules and differentiated thyroid cancer: the American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer. Thyroid 2016; 26: 1-133.
19. Kini SR. Thyroid cytopathology: an atlas and text. Philadelphia, PA: Lippincott Williams & Wilkins, 2008.
20. Lloyd R, Osamura R, Kloppel G, Rosai J. WHO classification of tumours: pathology and genetics of tumours of endocrine organs. 4th ed. Lyon: IARC Press, 2017.
21. Ali SZ, Cibas E. The Bethesda System for Reporting Thyroid Cytopathology: definitions, criteria and explanatory notes. New York: Springer, 2010.

Journal of Pathology and Translational Medicine 2017; 51: 533-547
https://doi.org/10.4132/jptm.2017.08.04

■ REVIEW ■

Thyroid Cytology in India: Contemporary Review and Meta-analysis

Shipra Agarwal · Deepali Jain

Department of Pathology, All India Institute of Medical Sciences, New Delhi, India

Received: July 22, 2017
Accepted: August 4, 2017

Corresponding Author
Deepali Jain, MD, DNB, FIAC
Department of Pathology, All India Institute of Medical Sciences, New Delhi 110029, India
Tel: +91-9868895112
Fax: +91-11-26588641
E-mail: deepalijain76@gmail.com

Fine-needle aspiration cytology (FNAC) is a screening test for triaging thyroid nodules, aiding in subsequent clinical management. However, the advantages have been overshadowed by the multiplicity of reporting systems and a wide range of nomenclature used. The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) was formulated in 2007, to give the world a uniform thyroid cytology reporting system, facilitating easy interpretation by the clinicians. Here, we review the status of thyroid FNAC in India in terms of various reporting systems used including a meta-analysis of the previously published data. An extensive literature search was performed using internet search engines. The reports with detailed classification system used in thyroid cytology were included. The meta-analysis of published data was compared with the implied risk of malignancy by TBSRTC. More than 50 studies were retrieved and evaluated. TBSRTC is currently the most widely used reporting system with different studies showing good efficacy and interobserver concordance. Ancillary techniques have, as of now, limited applicability and acceptability in thyroid cytology in India. Twenty-eight published articles met the criteria for inclusion in the meta-analysis. When compared with TBSRTC recommendations, the meta-analysis showed a higher risk of malignancy for categories I and III. Thyroid FNAC is practiced all over India. TBSRTC has found widespread acceptance, with most institutions using this system for routine thyroid cytology reporting. However, reasons for a high malignancy risk for categories I and III need to be looked into. Various possible contributing factors are discussed in the review.

Key Words: Cytology; Fine-needle aspiration cytology; Thyroid FNA; The Bethesda System for Reporting Thyroid Cytopathology; Review; Meta-analysis; India

Thyroid cancer is the most common endocrine malignancy, constituting 0.1%–0.2% of all cancers in India with an age-adjusted incidence of 1 per 100,000 in males and 1.8 per 100,000 in females.[1] As per the latest three-year report of 27 population based cancer registries from 2012 to 2014 issued by the National Cancer Registry Program, the incidence was particularly high among females of Papumpare District in Arunachal Pradesh (age adjusted rate 20.7 per 100,000 population), followed by Thiruvananthapuram (13.3) and Kollam Districts (12.0) in Kerala.[2]

Thyroid cancers most commonly present as a solitary thyroid nodule. The consensus guidelines from the Endocrine Society of India published a summary of current medical evidence for thyroid nodule management and optimized the guidelines for the clinical practice setting in India.[3] It includes a strong recommendation (Level A) for evaluation of all thyroid nodules >1 cm, including both palpable and radiologically distinct non-palpable nodules.[3] Prevalence of palpable nodules in India is about 12.2%.[4] India being an endemic area for goiter due to iodine deficiency, it is important to differentiate benign thyroid nodules from malignant ones.

Numerous studies have demonstrated that fine-needle aspiration cytology (FNAC) is a valid procedure for evaluation of thyroid nodules in adults and pediatric population. The role of FNAC is increasing in recent years in management and risk assessment of thyroid nodules.

Cytological evaluation of thyroid swellings is a rapid, easy and inexpensive diagnostic procedure which is widely used as a screening tool. It helps in triaging the patients into candidates for surgical or conservative management. However, the technique has its own shortcomings mainly due to interobserver and intraobserver variability, especially in indeterminate cases. In addition, there is also a lack of uniformity in the reporting systems used, which vary not only from country to country but also from laboratory to laboratory and even among individuals working at the same laboratory. This hampers accurate interpretation by the clinician, thus affecting patient management. To address this common issue, the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) was introduced based upon the proceedings of "The NCI Thyroid Fine Needle Aspiration State of the Science Conference" held in Bethesda, Maryland, in

pISSN 2383-7837
eISSN 2383-7845

2007.[5] TBSRTC encompasses six thyroid cytology categories, with each category having an implied cancer risk and the best modality of management.[5]

Here we review the available Indian literature on the status of thyroid aspiration cytology in India along with a meta-analysis of the reviewed data. We performed an extensive literature search in PubMed and Google Scholar databases using the following keywords: "India," "thyroid," "cytology," "cytopathology," "audit," "cytology-histology correlation," "the Bethesda system," "TBSRTC," and "FNAC." Those reports in which a recognizable classification system was used to categorize thyroid cytology smears were included. Case reports and case series were excluded. Cross-references of the selected articles were also checked to look out for additional studies. For meta-analysis, publications with available histopathological correlation were evaluated. The publications which had not used TBSRTC but had used a four or higher-tier system (including "unsatisfactory" category) were reclassified to fit into one of the TBSRTC categories. Hence, "indeterminate cases" were categorized as "atypia of undetermined significance/follicular lesion of undetermined significance" (AUS/FLUS, category III), while "follicular neoplasm," "follicular patterned lesion" and "Hurthle cell lesion/neoplasm" were classified as "follicular neoplasm/suspicious for a follicular neoplasm" (category IV). Wherever possible, the risk of malignancy (ROM) and the risk of neoplasm (RON) were calculated.[6] Reports with just two categories (benign and neoplastic) besides unsatisfactory and those using histopathological terminologies for imparting cytological diagnoses were not included in the meta-analysis. Papers just providing statistical measures of performance (sensitivity, specificity, negative and positive predictive values) of cytology in comparison with histopathology but not providing details of the histopathological diagnoses were also excluded from the meta-analysis. It should be noted that approximately half of the analyzed articles were published in non-PubMed indexed journals, which may raise an issue regarding quality of the publications. As per our evaluation, studies employed for this review contained sufficient amount of raw or processed data, and hence were eligible for inclusion.

BRIEF HISTORY OF THYROID FINE-NEEDLE ASPIRATION IN INDIA

FNAC was first used for cytological diagnosis in 1930s;[7] however, the method has been widely used after 1952.[8] In India, FNAC has been introduced in early 1970s.[9] First publication on FNAC appeared in 1975 by Gupta *et al*.,[10] which was published in the Indian Journal of Cancer. Needle biopsy of the thyroid had been attempted for the first time in 1965 in India,[11] whereas the first paper on FNAC of the thyroid dates back to 1987 by Rege *et al*.[12] Needle aspiration was initially started without any guidance. Later, with the advent of interventional radiology, the lesional localization was improved. Ultrasound-guided fine-needle aspiration (FNA) is a widely-acclaimed technique in investigating thyroid nodules/lesions. Approximately 50 reports so far have been published based on the Bethesda system and otherwise fulfilling our abovementioned criteria (Table 1).[13-63]

Most studies reclassified cases as per TBSRTC, and compared their distribution data and the ROM in each of the categories (Table 2).[13-63] Few compared diagnostic accuracy and inter-observer variation of previously used classification systems with TBSRTC. Old classification systems have been used in few of the studies which evaluated sensitivity and specificity of thyroid cytology in accurate diagnoses.

OPERATOR OF THYROID FINE-NEEDLE ASPIRATION

Review of available literature in India and our personal experience suggest that most blind, palpation guided FNAs of thyroid are done by pathologists, whereas clinicians or radiologists perform the FNAC under image guidance and leave the interpretation to pathologists. Although an occasional publication does provide evidence of at least some cases being aspirated by surgical medical officers, by-and-large, palpation-guided thyroid FNA is mostly performed by cytopathologists and ultrasound-guided aspiration by radiologists.[60]

In India, palpation-guided FNA appears to be the most commonly used technique, probably being more cost-effective (Table 1). Ultrasound-guided FNA is usually reserved for small or deep-seated poorly palpable nodules. It is also preferable to use ultrasound guidance to aspirate predominantly cystic lesions and for repeat aspiration of a previously non-diagnostic/unsatisfactory aspirate. Only a few centers are using the ultrasound-guided technique for all patients irrespective of the type of the thyroid nodule (Table 1).[34,58]

Interpretation can be done immediately after procedure at the site of FNA or later in the laboratory after staining of aspiration smears. Without rapid on-site evaluation (ROSE), a significant subset of thyroid FNAs are diagnosed inadequate/unsatisfactory for interpretation, which potentially leads to repeat aspirations and additional procedures. The basic purpose of ROSE is to increase the adequacy rate, diagnostic yield, and accuracy of the procedure. Systemic reviews and meta-analysis showed significant reduction

 https://doi.org/10.4132/jptm.2017.08.04

Table 1. Review of the published studies on thyroid FNA in India

No.	Study	Place	Needle size and technique	Wet fixation	Staining technique used		Reporting system followed
					Wet fixation	Air-dried smears	
1	Mandreker *et al.* (1995)[13]	Goa	n/s	n/s	n/s		Unsatisfactory, benign, SFM, malignant
2	Sirpal (1996)[14]	Delhi	21G	n/s	Pap, H&E	Leishman-Giemsa	Malignant, non-neoplastic, FN, Hurthle cell tumors, thyroglossal cyst, extrathyroidal, inconclusive when unsatisfactory
3	Handa *et al.* (2008)[15]	Chandigarh	23G ± aspiration, manual[a]	n/s	Pap, H&E	MGG	n/s
4	Guhamallick *et al.* (2008)[16]	Kolkata	23–24G + aspiration	n/s	Pap, H&E	Leishman-Giemsa	Unsatisfactory, non-neoplastic, indeterminate, malignant
5	Gupta *et al.* (2010)[17]	Jammu and Kashmir	n/s	Ether-95% alcohol solution	Pap	n/s	Benign, FN, SFM, malignant
6	Bagga and Mahajan (2010)[18]	Haryana	23–25G, non-aspiration[a]	95% Ethanol	H&E	MGG	Unsatisfactory, benign, SFM, malignant
7	Sengupta *et al.* (2011)[19]	Bihar	22–23G + aspiration[b]	n/s	n/s	MGG	Colloid goiter, granulomatous thyroiditis, FA, FC, anaplastic carcinoma
8	Renuka *et al.* (2012)[20]	Andhra Pradesh	22G ± aspiration, USG in some	95% Methanol	Pap, H&E	MGG	TBSRTC
9	Sharma and Mathur (2012)[21]	Rajasthan	23G + aspiration	Ether-95% alcohol solution	Pap, H&E	Giemsa	Unsatisfactory, non-neoplastic, FN, SFM, malignant (RCP)
10	Patel *et al.* (2013)[22]	Gujarat	23–24G ± aspiration[a]	95% Ethanol	Pap, H&E	MGG	Non-neoplastic, neoplastic, others
11	Mondal *et al.* (2013)[23]	West Bengal	n/s, USG in some[a]	n/s	Pap	Leishman-Giemsa	TBSTRC
12	Kukar *et al.* (2013)[24]	Punjab	n/s[a]	95% isopropanol	Pap, H&E	MGG	Non-neoplastic, neoplastic
13	Bhasin *et al.* (2013)[25]	Punjab	n/s	n/s	n/s; MGG as per figures		TBSRTC
14	Borgohain *et al.* (2014)[26]	Assam	n/s	n/s	n/s	MGG	Non-neoplastic, neoplastic
15	Mangshetty *et al.* (2014)[27]	Karnataka	22–24G + aspiration	Alcohol	Pap	MGG	Unsatisfactory, benign, malignant
16	Panchal *et al.* (2014)[28]	Maharashtra	22/23G + aspiration	95% Ethanol	Pap	n/s	Unsatisfactory, benign, SFM, malignant
17	Pathak *et al.* (2014)[29]	Delhi	n/s	n/s	Pap	MGG	TBSRTC
18	Sukumaran *et al.* (2014)[30]	Kerala	n/s	95% alcohol	Pap	n/s	TBSRTC
19	Arul and Masilamani (2015)[31]	Tamil Nadu	n/s	n/s	n/s, H&E as per figures		TBSRTC
20	Arul *et al.* (2015)[32]	Tamil Nadu	n/s, USG if small lesion[a]	n/s	H&E	MGG	TBSRTC
21	Sekhar *et al.* (2015)[33]	Karnataka	23–24G + aspiration	95% Ethanol	Pap, H&E	MGG	TBSRTC
22	Mehra and Verma (2015)[34]	Delhi	n/s, USG in all	n/s	Pap	MGG	TBSRTC
23	Agrawal *et al.* (2015)[35]	Uttar Pradesh	23G + aspiration	95% Ethanol	Pap, H&E	MGG	TBSRTC
24	Sharma (2015)[36]	Tamil Nadu	n/s, manual	n/s	n/s		Unsatisfactory, benign, follicular pattern lesions, suspicious (includes atypical), malignant
25	Thakkar *et al.* (2015)[37]	Gujarat	22/24G + aspiration[a]	n/s	H&E	MGG	TBSRTC
26	Garg *et al.* (2015)[38]	Gujarat	23–24G + aspiration[a]	n/s	n/s		TBSRTC
27	Kathirvel (2015)[39]	Tamil Nadu	25–27G	100% Isopropanol	H&E		TBSRTC

(Continued to the next page)

Table 1. Continued

No.	Study	Place	Needle size and technique	Wet fixation	Staining technique used		Reporting system followed
					Wet fixation	Air-dried smears	
28	Alagarsamy *et al.* (2015)[40]	Tamil Nadu	23G ± aspiration	100% Isopropanol	H&E	n/s	Colloid goiter, adenoma, carcinoma, others
29	Mamatha *et al.* (2015)[42]	Telangana	n/s	n/s	n/s	n/s	Unsatisfactory, colloid cyst/goiter, follicular lesions/ neoplasm, indeterminate, SFM, malignant as well as TBSRTC
30	Gupta *et al.* (2015)[43]	Uttar Pradesh	n/s	n/s	Pap	Diff-Quick	Histopathological equivalents as well as TBSRTC
31	Hathila *et al.* (2016)[41]	Gujarat	23G, non-aspiration	95% Ethanol	Pap, H&E	MGG	Benign, malignant
32	Shankar *et al.* (2016)[44]	Tamil Nadu	n/s + aspiration	n/s	Pap	n/s	TBSRTC
33	Prathima *et al.* (2016)[45]	Karnataka	n/s, USG in some	Alcohol	Pap, H&E	Giemsa	TBSRTC
34	Mehrotra *et al.* (2016)[46]	Karnataka	n/s ± aspiration, USG in some	95% Ethanol	H&E	MGG	TBSRTC
35	Tagore *et al.* (2016)[47]	Karnataka	22G + aspiration	Alcohol	Pap	MGG	TBSRTC
36	Kalita and Das (2016)[48]	Assam	23G ± aspiration	n/s	n/s	MGG	TBSRTC
37	Bhartiya *et al.* (2016)[49]	Bihar	23–24G + aspiration, USG in some[a]	Wet fixed	Pap	Leishman-Giemsa	TBSRTC
38	Kulkarni *et al.* (2016)[50]	Madhya Pradesh	n/s	n/s	Pap	n/s	TBSRTC
39	Lohiya *et al.* (2016)[51]	Rajasthan	23/24G	n/s	n/s	MGG	TBSRTC
40	Kasliwal *et al.* (2016)[52]		24–26G + aspiration	95% Ethanol	H&E	MGG	TBSRTC
41	Khatib *et al.* (2016)[53]	Maharashtra	n/s, USG if unsatisfactory	n/s	Pap	Giemsa	TBSRTC
42	Pantola *et al.* (2016)[54]	Tamil Nadu	23G	95% Alcohol	Pap, H&E	MGG	TBSRTC
43	Babu *et al.* (2016)[55]	Tamil Nadu	23G	Ether-95% alcohol solution	Pap	n/s	Unsatisfactory, benign, malignant
44	Solanki *et al.* (2016)[56]	Rajasthan	n/s	n/s	H&E	MGG	TBSRTC
45	Aramani and Gururajaprasad (2017)[57]	Karnataka	24–25G + aspiration	95% Ethanol	Pap, H&E	MGG	Benign, malignant
46	Sunder and Khan (2017)[58,b]	Telangana	23/25/26G + aspiration	95% Ethyl alcohol or isopropanol	Pap, H&E	MGG	Benign, malignant
47	Garg *et al.* (2017)[59]	Maharashtra	n/s ± aspiration, USG in all	n/s	Pap	MGG	TBSRTC
48	Kannan *et al.* (2017)[60]	Karnataka	n/s[a,b]	n/s	n/s	n/s	TBSRTC
49	Mahajan *et al.* (2017)[61]	Chandigarh	n/s	n/s	n/s	n/s	TBSRTC
50	Chandra *et al.* (2017)[62]	Uttarakhand	26–28G	Alcohol	Pap, H&E	MGG	TBSRTC
51	Laishram *et al.* (2017)[63]	Manipur	n/s	n/s	n/s	MGG	TBSRTC

FNA, fine needle aspiration; n/s, not specified; SFM, suspicious for malignancy; Pap, Papanicolaou stain; H&E, hematoxylin and eosin; FN, follicular neoplasm; MGG, May-Grünwald-Giemsa; FA, follicular adenoma; FC, follicular carcinoma; USG, ultrasound-guided aspirate; TBSRTC, The Bethesda System for Reporting Thyroid Cytopathology; RCP, Royal College of Pathologists guidelines.
[a]FNA performed by cytopathologist; [b]FNA performed by clinician.

in inadequacy rate of thyroid FNAs with ROSE.[64,65] Acquisition of ROSE in routine practice depends upon the infrastructure of the institute which includes availability of manpower, location of the procedure room, case volume, and resources. In our institute, ROSE is offered to all thyroid FNAs under guidance, and it is performed by cytopathologists. Studies on comparison analysis of adequacy assessment of thyroid FNA with and without ROSE were not found in Indian literature. We believe ROSE is practiced only in a few academic institutions in India.

CYTOTECHNICIAN TRAINING PROGRAM AND QUALITY CONTROL IN INDIA

The Indian Academy of Cytologists (http://www.cytoindia.com)

Table 2. Descriptive data with the risk of malignancy

No.	Study	Thyroid FNA (operated nodules)	Distribution of the Bethesda categories and corresponding risk of malignancy (%)					
			I	II	III	IV	V	VI
1	Mandreker *et al.* (1995)[13]	1,992 (238)	12.7	78.2 (5.5)	-	-	7.6 (26.3)	1.5 (91.7)
2	Sirpal (1996)[14]	1,114[a] (128)	0.6[b] (0)	97.1 (0)	-	1 (11.1)	-	1.3 (100)
3	Handa *et al.* (2008)[15]	434 (66)	5.1	87.8 (1.9)	-	3.2 (0)	-	3.9 (100)
4	Guhamallick *et al.* (2008)[16]	288 (75)	13.5	68.4 (3.1)	-	9.4 (30)	-	8.7 (95.6)
5	Gupta *et al.* (2010)[17]	75 (75)	-	60 (6.7)	-	24 (16.7)	4 (0)	12 (100)
6	Bagga and Mahajan (2010)[18]	252 (32)	1.6	90.5	-	-	6.7	1.2
7	Renuka *et al.* (2012)[20]	564	17	70.5	1.9	4.2	2.6	3.5
8	Sharma and Mathur (2012)[21]	94 (76)	2.1	53.2 (0)	-	35.1 (10)	2.1 (100)	7.4 (100)
9	Mondal *et al.* (2013)[23]	1,020 (323)	1.2 (0)	87.5 (4.5)	1 (20)	4.2 (30.6)	1.4 (75)	4.7 (97.8)
10	Bhasin *et al.* (2013)[25]	80	1.2	61.2	10	20	3.8	3.8
11	Panchal *et al.* (2014)[28]	300 (36)	-	98.7	-	-	0.3	1
12	Pathak *et al.* (2014)[29]	454	25.7	59	6	4	1.8	3.5
13	Sukumaran *et al.* (2014)[30]	248 (248)	6 (6.7)	12.5 (12.9)	4.4 (54.6)	13.3 (87.9)	4 (100)	59.7 (100)
14	Arul and Masilamani (2015)[31]	483 (209)	5 (8.3)	44.5 (1.1)	2.9 (0)	21.5 (11.5)	15.3 (96.9)	10.8 (100)
15	Arul *et al.* (2015)[32]	603 (392)	2.7 (0)	65.2 (1.2)	10 (24.4)	10.6 (28.9)	5.3 (70.8)	6.3 (100)
16	Sekhar *et al.* (2015)[33]	150 (64)	2.6 (0)	76.6 (0)	0.7	12.7 (5.9)	2.7 (100)	4.7 (66.7)
17	Mehra and Verma (2015)[34]	225 (40)	7.2 (0)	80 (13)	4.9 (100)[c]	2.2 (25)	3.5 (50)	2.2 (100)
18	Agrawal *et al.* (2015)[35]	281 (134)	2.5 (0)	87.9 (1.8)	3.9	2.5 (18.2)	1.8 (80)	1.4 (100)
19	Sharma (2015)[36]	724 (724)	-	87.7 (1.3)	-	-	2.9 (52.4)	9.4 (97.1)
20	Thakkar *et al.* (2015)[37]	134 (24)	4.5 (0)	85.8 (0)	0.7	7.5 (33.3)	0.7	0.7
21	Garg *et al.* (2015)[38]	100 (60)	6 (20)	78 (0)	4 (25)	5 (20)	3 (66.7)	4 (100)
22	Kathirvel (2015)[39]	59	15.3	16.9	15.3	16.9	15.3	20.3
23	Mamatha *et al.* (2015)[42]	240 (214)	10.8	59.2 (0)	4.2 (50)	15 (6.7)	4.2 (60)	6.6 (100)
24	Gupta *et al.* (2015)[43]	300	11	78	2	3	1	5
25	Shankar *et al.* (2016)[44]	402 (92)	10.7 (0)	81.6 (1.6)	1.2 (0)	1.7 (28.6)	2 (71.4)	2.7 (80)
26	Prathima *et al.* (2016)[45]	178 (60)	11.7 (33.3)	77.5 (7.1)	1.1 (50)	3.9 (25)	2.2 (66.7)	3.3 (100)
27	Mehrotra *et al.* (2016)[46]	175 (34)	4.6	68.6 (0)	5.7	17.1 (0)	1.1[c] (0)	2.9[c] (100)
28	Tagore *et al.* (2016)[47]	100	3	81	0	9	3	4
29	Kalita and Das (2016)[48]	664	12	72.3	1.8	4.8	1.5	7.5
30	Bhartiya *et al.* (2016)[49]	238 (105)	5.9	84 (2)	1.3	2.9 (0)	2.5	3.4 (100)
31	Kulkarni *et al.* (2016)[50]	151 (16)	11.2 (0)	76.8 (0)	0	9.3 (25)	0.7	2 (100)
32	Lohiya *et al.* (2016)[51]	250	4	88	2	1.6	0.8	3.6
33	Kasliwal *et al.* (2016)[52]	411 (97)	0.5	94.2 (2.6)	0	3.5 (22.2)	0	1.7 (100)
34	Khatib *et al.* (2016)[53]	287 (287)	0.7 (0)	87.8 (3.3)	3.5 (20)	4.2 (25)	1.7 (80)	2.1 (100)
35	Pantola *et al.* (2016)[54]	218 (44)	5.5 (0)	69.3 (0)	10.5 (8.3)	8.2 (10)	2.3 (100)	4.1 (100)
36	Solanki *et al.* (2016)[56]	1,287 (62)	22 (18.2)	73.9 (2.6)	0.7 (0)	1.5 (50)	0.4 (50)	1.3 (100)
37	Kannan *et al.* (2017)[60]	404 (243)	7.7 (28.6)	40.8 (13)	24.3 (41.7)	10.6 (46.9)	6.9 (96.3)	9.7 (100)
38	Mahajan *et al.* (2017)[61]	4,532 (335)	3.5 (50)	79.6 (7.8)	2.5 (50)	3.9 (23.6)	0.5 (75)	9.8 (85.4)
39	Chandra *et al.* (2017)[62]	971[d]	5.5	74.9	6.4 (51.4)	2.6	3.2	7
40	Laishram *et al.* (2017)[63]	576 (11)	5.2	89.9	0	2.2 (40)	0.3	2.2 (100)

FNA, fine-needle aspiration.
[a]Excluding nine extrathyroidal; [b]After adjustment of "inconclusive" to the Bethesda terminology; [c]Only one case with available histology; [d]Included 35 cases of category III with surgical follow-up.

conducts examination for cytotechnicians and cytotechnologists. There are few centers which run cytotechnician and cytotechnologist training programs for certification. Cytotechnologists work as cytoscreeners; however, in India only limited institutions have cytoscreeners. Their work allocation depends upon the institutional work requirement and administration policies.

The External Quality Assurance Programme of the Indian Academy of Cytologists is aimed to maintain and monitor the quality of reporting on all cytopathology specimens, in which over 100 cytopathology laboratories from all over the India participate for FNA, exfoliative specimens and cervical smears. In terms of thyroid FNA, only straightforward diagnoses (such as lymphocytic thyroiditis or carcinomas) are assessed and so TBSRTC is not strictly followed, unlike cervical smears where it is mandatory to diagnose lesions according to the Bethesda classification (personal communication with Prof. Radhika Srinivasan, Postgraduate Institute of Medical Education and Research, Chandigarh, India).

PREPARATION AND STAINING OF THYROID CYTOLOGY SAMPLES

The needles used in thyroid FNA vary in size from 21G to 28G, with or without aspiration for fine needle aspiration cytology and fine needle capillary sampling, respectively (Table 1). The most commonly used needle was 23G followed by 24G in published studies. Since the thyroid is a highly vascular organ, and given the risk of hemorrhagic complications, it is advisable to use a small-bore needle (25–27 gauge). The unstained smear may then be visually evaluated for tissue fragments and/or colloid, and if required, a larger bore needle may be used for subsequent aspirations.[5,66] Larger diameter needles are also preferable for draining thick colloid.[5]

Most institutes use direct smears in which the material is smeared onto the glass slide and either kept air-dried for Romanowsky stains (May-Grünwald-Giemsa, Leishman, Giemsa) stain or wet-fixed with common fixatives (95% ethyl alcohol, 95% methanol, 95% isopropyl alcohol, or a solution of ether and 95% alcohol) for Papanicolaou and/or hematoxylin and eosin stain (H&E). Papanicolaou and H&E stains help in characterization of nuclear features whereas Romanowsky stains better define cytoplasmic characteristics. In case of cystic nodules, aspirated fluid is centrifuged and smears are prepared from the sediment. Most cytopathologists in India use a combination of Romanowsky and Papanicolaou stains. However, H&E is preferred in a few institutions due to its cost effectiveness and better familiarity of the stain from surgical pathology.

Liquid-based cytology (LBC) is another adjunctive technique in thyroid FNA which is associated with better preservation of cellular details. It removes obscuring hemorrhage, cellular debris and inflammatory cells to a large extent from the background. Keyhani *et al*.[67] have compared conventional, cell block and LBC preparations in a cohort of 100 patients with thyroid nodules. While a significant percentage (87%) of cases yielded informative results using LBC method, only 69% of the samples processed for cell blocks were informative. Both techniques had almost equal sensitivity (95% for LBC vs 96% for cell block), but the specificity of LBC (31%) was reported to be higher than that of the cell block (24%). It has been suggested that LBC may be used as a supplementary technique to conventional smears to improve the diagnostic yield of thyroid aspiration cytology. In a more recent study by Prasad *et al*.,[68] LBC slides from 41 cases of thyroid swellings (23 nodular colloid goiter, 14 thyroiditis, and 4 carcinoma) were assessed and compared with conventional smears. Importantly, the authors cautioned against the regular use of LBC in thyroid cytology. While the amount of background colloid was reported to be significantly diminished, they found nuclear features (grooves and pseudoinclusions) of papillary thyroid carcinoma less forthcoming on LBC. Another study evaluated 18 cases of thyroid swellings (10 colloid goiter, four thyroiditis, and four carcinoma cases) by LBC and compared the results with conventional smears.[69] While the technique was not found to be of much use in benign thyroid diseases, it was beneficial in diagnosis of neoplastic lesions. However, the small number of cases evaluated precludes any definite interpretation. To conclude, data reported from Indian institutions suggest that LBC may be used as an adjunct method but cannot replace conventional smears in thyroid cytology.

THYROID CYTOLOGY REPORTING SYSTEMS

Reporting of thyroid aspirate smears has evolved tremendously over the past decade. Studies from pre-Bethesda era showed usage of a range of formats for reporting. These include descriptive reporting, use of histopathology equivalents, and variably tiered classification systems, ranging from just two categories (non-neoplastic and neoplastic) to four or five categories (Table 1). In the past and even today in some centers across the country, a range of formats are being used. Histopathology correlates when used are easily interpretable by the clinicians, however, they may not be perfectly applicable to all thyroid aspirates, especially the gray zone lesions: for example, follicular neoplasms, hyperplastic

 https://doi.org/10.4132/jptm.2017.08.04

thyroid nodules versus follicular adenoma, papillary hyperplasia versus papillary thyroid carcinoma, or reactive change versus papillary thyroid carcinoma. These cytodiagnostic categories do not provide management guidelines to clinicians. The 2-tier system suffers from similar shortcomings, and can lead to over- as well as under-treatment. A 3- or 4-tiered system has a drawback of inadvertent clubbing of benign and malignant cases. Benign lesions such as hyperplastic nodules and Hashimoto's thyroiditis with nuclear atypia are combined together with follicular/Hurthle cell neoplasms, non-invasive follicular thyroid neoplasm with papillary like nuclear features (NIFTP) and carcinomas with poor preservation or less cellularity.

After introduction of the 6-tiered TBSRTC,[5] several cytopathologists tested its efficacy and reported the ROM in different categories. Pathak *et al*.[29] reclassified over 400 thyroid aspirates as per TBSRTC and found strong agreement level among the three observers (Fleiss' kappa score, 50.7) and a significant reduction in the number of inconclusive diagnoses ($p < .001$) while using TBSRTC. In another study, TBSRTC was also found to be superior in terms of sensitivity (100% vs 77%) and specificity (82.5% vs 69%) in comparison with the conventional system.[42] Upon comparing three thyroid cytology reporting systems, which included conventional (unsatisfactory, benign or negative for malignancy, follicular lesions, indeterminate, positive for malignancy), the British Thyroid Association/the Royal College of Pathologists (BTA/RCP) (Thy 1–Thy 5 categories) and TBSRTC, TBSRTC and BTA/RCP were found to be better in terms of approachability, classification of thyroid lesions, treatment and follow-up than the conventional reporting system.[70]

NON-DIAGNOSTIC CRITERIA FOR CYTOLOGICAL DIAGNOSIS

In cytology, every FNA from any organ system must be evaluated for adequacy in the proper context of clinical and radiological findings. To decrease the false-negative rate, TBSRTC has laid down criteria for adequacy. For accurate interpretation, TBSRTC recommended any thyroid FNA specimen to be considered satisfactory for evaluation when at least six groups of well-preserved, well-stained, and well-visualized follicular cells are seen on the aspirate and each group is composed of at least 10 follicular epithelial cells, preferably on a single slide.[5] There are certain exceptions to this rule, such as solid nodules with cytologic atypia, solid nodules with inflammation and colloid nodules. Cyst fluid with less than six groups is considered non-diagnostic/unsatisfactory unless clinical and radiological features are suggestive of a benign cyst.[5]

Pre-Bethesda era studies considered an FNA as unsatisfactory/non-diagnostic when there was less cellularity (no objective quantification) and when excessive blood or poor technical quality obscured smears such as overtly thick smears and air drying of alcohol-fixed smears. Now, most cytopathologists use objective adequacy criteria laid down by TBSRTC, except for a few who use the Royal College guidelines (Table 1).[36]

Since inception of TBSRTC in 2008, most laboratories in India have adopted it. However, a few studies have used a different set of criteria.[47] While TBSRTC requires 5–6 groups of well-preserved follicular epithelial cells with 10 or more cells per group, Tagore *et al*.[47] claimed that in case of large clusters of follicular epithelial cells, 10 clusters were needed with each having more than 20 cells. In case of presence of tissue fragments, the minimum number of fragments required was 8.[47] The Royal College of Pathologists (RCP) guidelines were used in one of the studies, which were similar to TBSRTC in terms of cellularity and approach to cystic lesions, i.e., minimum of six groups of follicular cells across all the submitted slides, each with at least 10 well-visualized epithelial cells.[36] Samples containing mostly macrophages but lacking enough cells and/or abundant colloid were also considered "unsatisfactory," similar to cases with cellular details obscured by blood/clotting or crushing artifact/poor fixation/poorly spread smears. However, there were others who have just mentioned "blood only/lack of cellularity/poor quality smears/presence of obscuring factors" as the reason for calling a sample unsatisfactory.[15,22,24] Still, some studies have not specified the criteria used for adequacy.[27,28,57]

ANCILLARY TECHNIQUES

Of the various ancillary techniques which can be utilized in cytology as diagnostic aid, cell block, immunocytochemistry (ICC) and flow cytometry are probably the most commonly used. Ancillary studies in thyroid cytology are more useful in rare borderline cases of medullary carcinoma,[71] anaplastic carcinoma,[72] metastases and lymphomas,[73] than in the more common papillary thyroid carcinoma. ICC could be used for diagnosing tuberculous thyroiditis, in which Ziehl-Neelsen staining had failed to reveal acid-fast bacilli.[74] ICC is a cost-effective and easy technique which can be performed on alcohol or acetone-fixed unstained as well as destained cytology smears and better, if available, on cell blocks.[75-79] Nevertheless, its role in differentiating benign from malignant thyroid nodules of follicular cell origin is limited with contradictory results in different studies.[75]

Cell block is a complementary method of assessing cytology material, which gained importance because of the advantages it has over conventional cytology smears. Cell block is similar to a mini-biopsy, since it imparts better-preserved tissue architecture and provides several sections, which can be utilized to perform a battery of ancillary tests including special stains, immunohistochemistry, ultrastructural studies, and molecular tests.[80] Although few studies have shown utility of cell blocks as an adjunct to conventional cytology in diagnosis of thyroid tumors,[81] their use in everyday clinical practice is limited by their low cellularity, enhanced cost and turnover time.[80] As per the available literature, the technique is being done in routine only in rare centers across India.[44]

Molecular testing for *BRAF* mutation and other molecular alterations, as per American Thyroid Association 2015 guidelines, may be used to supplement malignancy risk assessment, especially in indeterminate cytology.[82] However, there is not much published data on the utility of thyroid FNA molecular testing in India. Despite a thorough search, we could find only one abstract, whereby the authors had retrospectively evaluated 40 thyroid aspirate samples for *BRAF* mutation by Sanger sequencing, with effective amplification achieved in over half of them.[83] The mutation was detected in 12% of papillary thyroid carcinomas, which was significantly lower than the expected rate.[83,84] Rare reports of fluorescence *in-situ* hybridization on cytology smears of thyroid are also available.[73]

While FNAC is the most common primary diagnostic modality for diagnosing follicular-derived thyroid tumors, cases with clinical, cytological or radiological features suggestive of non-follicular cell derived thyroid malignancy are subjected to tru-cut or open biopsy, as it gives the additional advantage of architectural preservation and performing immunohistochemistry. It is of use especially for hematolymphoid neoplasms.[85-88]

META-ANALYSIS OF THE RISK OF MALIGNANCY IN THE BETHESDA DIAGNOSTIC CATEGORIES

Owing to the variable number of cases included in various studies, in order to get an accurate overall assessment of the ROM in different categories, a metaanalysis was performed. Of the 52 articles selected for the review, 28 met the inclusion criteria for meta-analysis (Table 3). It was done using STATA ver. 12.0 (Stata Corp, College Station, TX). Random effects model was used to calculate the pooled estimate. A $p < .05$ was considered statistically significant.

TBSRTC has ascribed a particular ROM to every Bethesda category (Table 3). Meta-analysis revealed a higher ROM for the category III as compared to TBSRTC estimates, 34% versus 5%–15% (Figs. 1–6). Recent meta-analyses by Straccia *et al*.[89] and Krauss *et al*.[6] also found a high ROM (27% and 24%, respectively) for category III. Interestingly, we have also found a ROM of 34.3% (unpublished data) at our institute. Category III includes cases which are neither undoubtedly benign nor can be categorized into higher categories of IV and higher. It is so heterogeneous that sub-classification of AUS/FLUS has been recommended by some authors, based on the presence of architectural and/or nuclear atypia, improving cancer risk estimation.[90-93] Although TBSRTC defines AUS/FLUS category as last resource category and should only be ≤7% of total thyroid FNAs, it was found out to be a heterogeneous category which ranged from < 1% to > 5% in different studies (Table 2). We have an approximately 11% rate of AUS/FLUS category in our institute (unpublished data).

Cases with poor cellularity and/or technical quality having some atypia are also huddled into this category. Our institute is a teaching hospital, where aspiration is performed by residents. Hence, lack of adequate experience of the aspirator results in hemodilution and poor smear preparation and may contribute to a higher ROM of category III. Re-aspirating such cases along with radiological correlation, to some extent, decreases the proportion of cases in this category as well as the ROM. Category I also had a higher ROM and may be explained by the same reason.

Impact of the recent reclassification of non-invasive encapsulated follicular variant of papillary carcinoma of thyroid into NIFTP is not evident in this review as most studies included are from pre-NIFTP era. Studies post-NIFTP introduction have not specified it in their histopathological diagnoses. It is likely that the impact, particularly a decrease of ROM for the indeterminate diagnostic categories,[94] is dependent on the incidence of NIFTP, which is relatively low in our settings (unpublished data), consistent with Asian data.[95]

In addition, a wide 95% confidence interval (CI) was noted for categories III (23%–45%), IV (15%–36%), and V (55%–84%). The wide range may be attributed to interobserver variation and differences in experience levels of the pathologists. As the extreme entities (benign and malignant) are the easiest to categorize in a well-prepared aspirate, the CI values in the two categories were low. Krauss *et al*.[6] in their recent meta-analysis also reported a wide CI for categories III, IV, and V. The authors ascribed it to subjective differences in the interpretation of the Bethesda criteria for diagnosis of these categories, and recommended introduction of a performance measure such as ratio of AUS/FLUS to total

Table 3. Summary of meta-analysis

Category	ROM					RON			
	Studies included for ROM	Pooled ROM (95% CI, %)	I^2 (%)	p-value	ROM as per TBSRTC (%)[5]	Studies included for RON	Pooled RON (95% CI, %)	I^2 (%)	p-value
I	18	15 (6–24)	11.8	.34	1–4	15	34 (17–52)	65.5	.01
II	28	3 (2–4)	54	.00	0–3	23	8 (6–10)	80.3	.00
III	15	34 (23–45)	57.9	.01	5–15	11	62 (44–81)	77.4	.00
IV	27	26 (15–36)	87.2	.00	15–30	23	81 (73–89)	77.2	.00
V	21	69 (55–84)	87.7	.00	60–75	17	76 (62–90)	64.1	.00
VI	28	94 (89–98)	56.3	.03	97–99	24	95 (90–99)	58.1	.05

ROM, risk of malignancy; CI, confidence interval; TBSRTC, the Bethesda System for Reporting Thyroid Cytopathology; RON, risk of neoplasia.

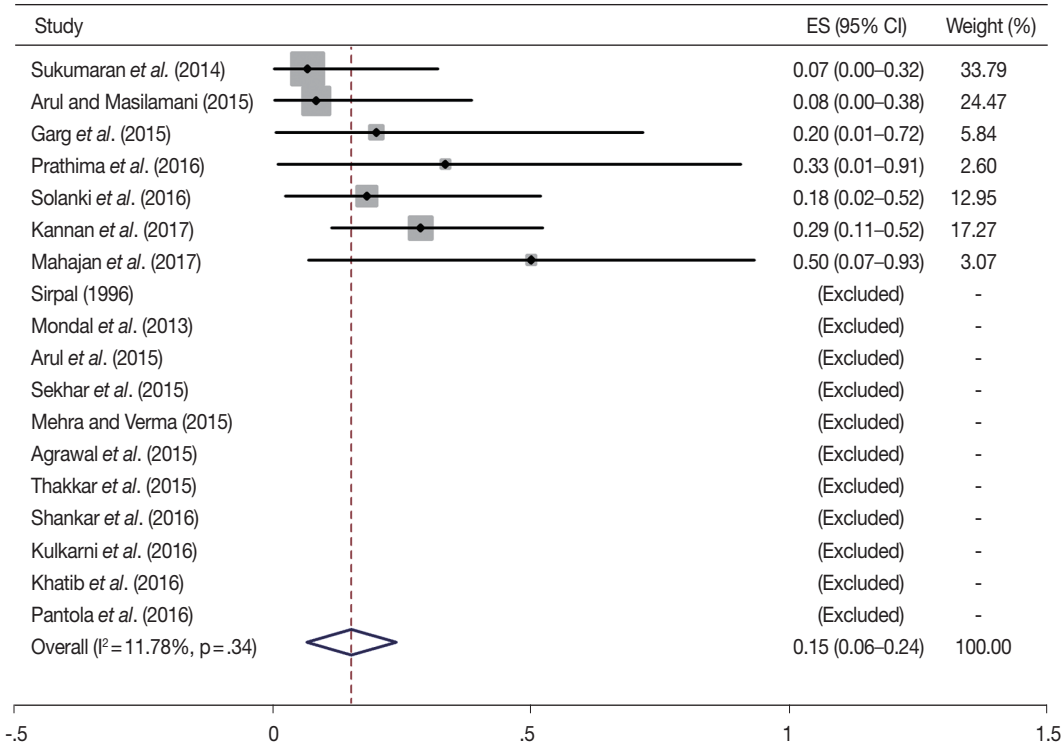


Fig. 1. Forest plot of meta-analysis on the risk of malignancy for Bethesda category I (non-diagnostic).[14,23,30-35,37,38,44,45,50,53,54,56,60,61] ES, effect size; CI, confidence interval.

thyroid FNAs for each laboratory to follow. As expected, RON which includes benign tumors (the most common being follicular adenoma) was higher than the ROM (Table 3).

CONCLUSION

Thyroid FNAC is practiced all over India in academic and private institutes as well as private hospitals and laboratories. In India, most thyroid aspiration samples are collected by pathologists, using manual palpation. Most centers prepare both alcohol-fixed and air-dried smears stained with Papanicolaou/H&E and May-Grünwald-Giemsa, respectively. TBSRTC is currently the most widely used reporting system with different studies showing good efficacy and interobserver concordance. Ancillary studies including core biopsy and molecular testing, as of now, have limited applicability and acceptability in thyroid cytology in India. Category III is the most heterogeneous category with a wide range of ROM and RON. Case to case discussion among the clinicians and pathologists supplemented by radiological correlation may help improve the management of these patients.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

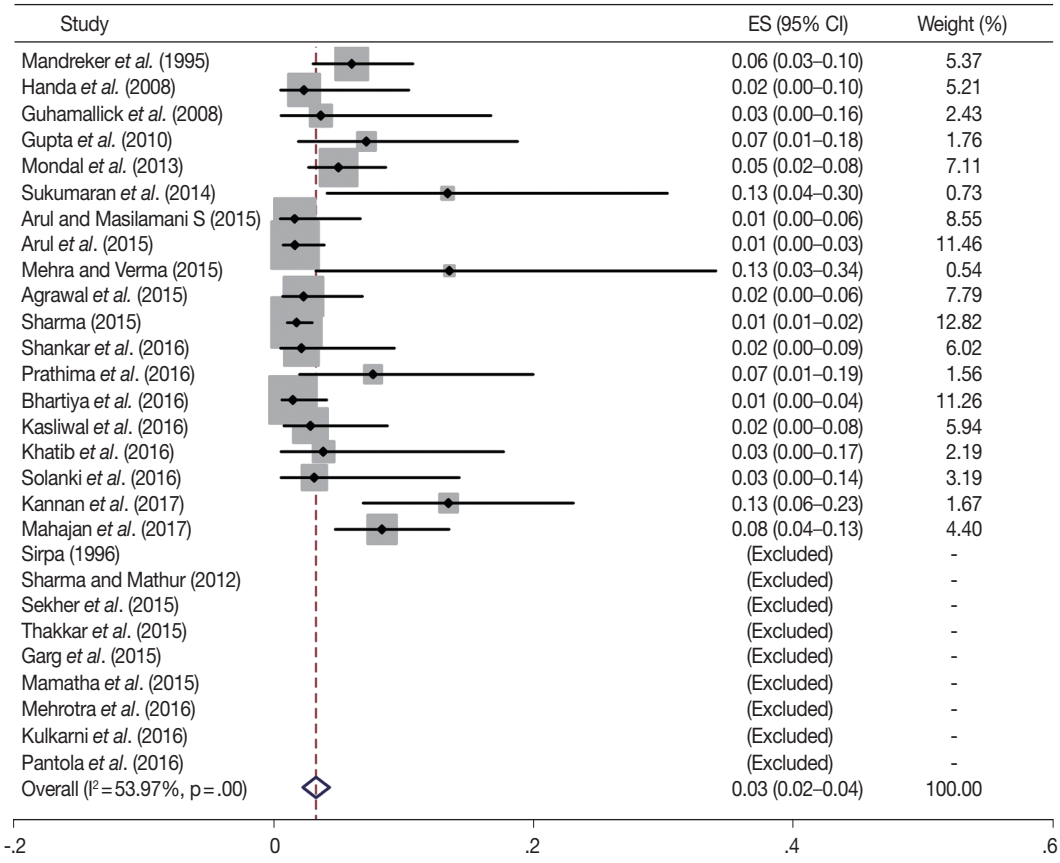


Fig. 2. Forest plot of meta-analysis on the risk of malignancy for Bethesda category II (benign).[13-17,21,23,30-38,42,44-46,49,50,52-54,56,60,61] ES, effect size; CI, confidence interval.

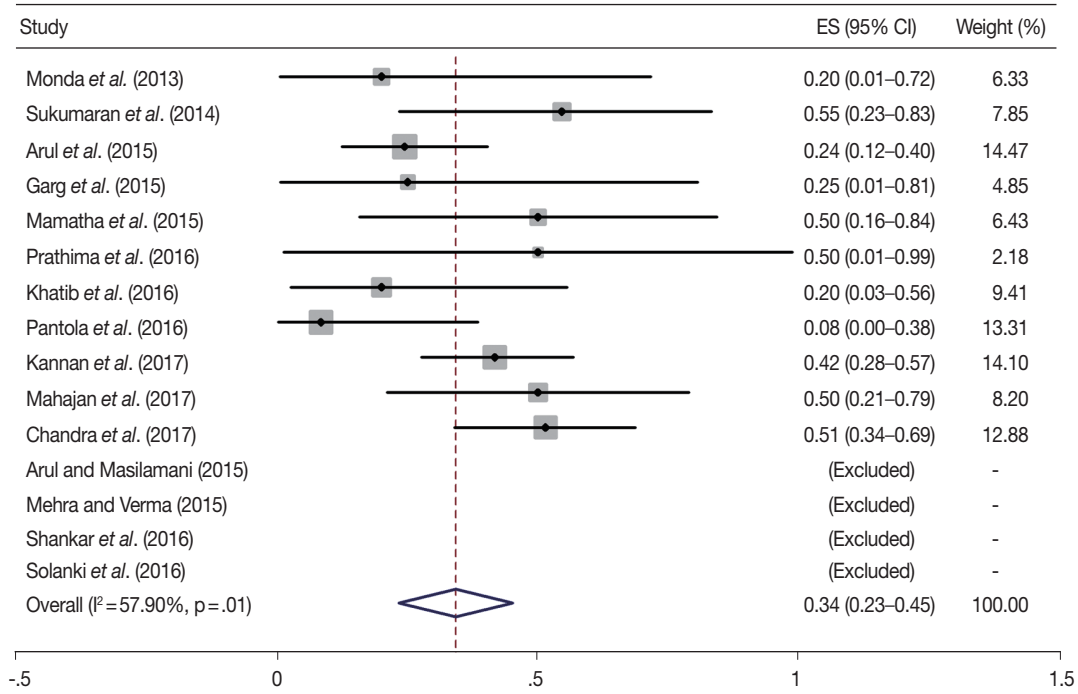


Fig. 3. Forest plot of meta-analysis on the risk of malignancy for Bethesda category III (atypia of undetermined significance/follicular lesion of undetermined significance).[23,30-32,34,38,42,44,45,53,54,56,60-62] ES, effect size; CI, confidence interval.

https://doi.org/10.4132/jptm.2017.08.04

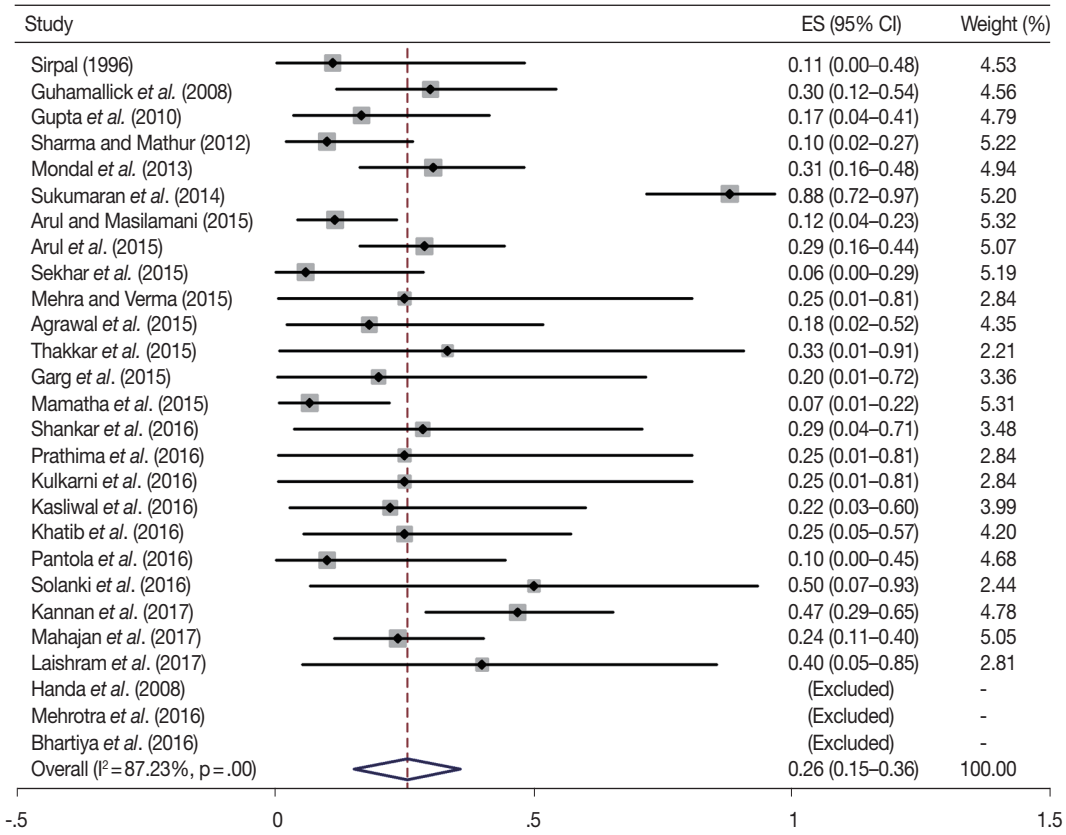


Fig. 4. Forest plot of meta-analysis on the risk of malignancy for Bethesda category IV (follicular neoplasm/suspicious for a follicular neoplasm).[14-17,21,23,30-35,37,38,42,44,45,46,49,50,52-54,56,60,61,63] ES, effect size ; CI, confidence interval.

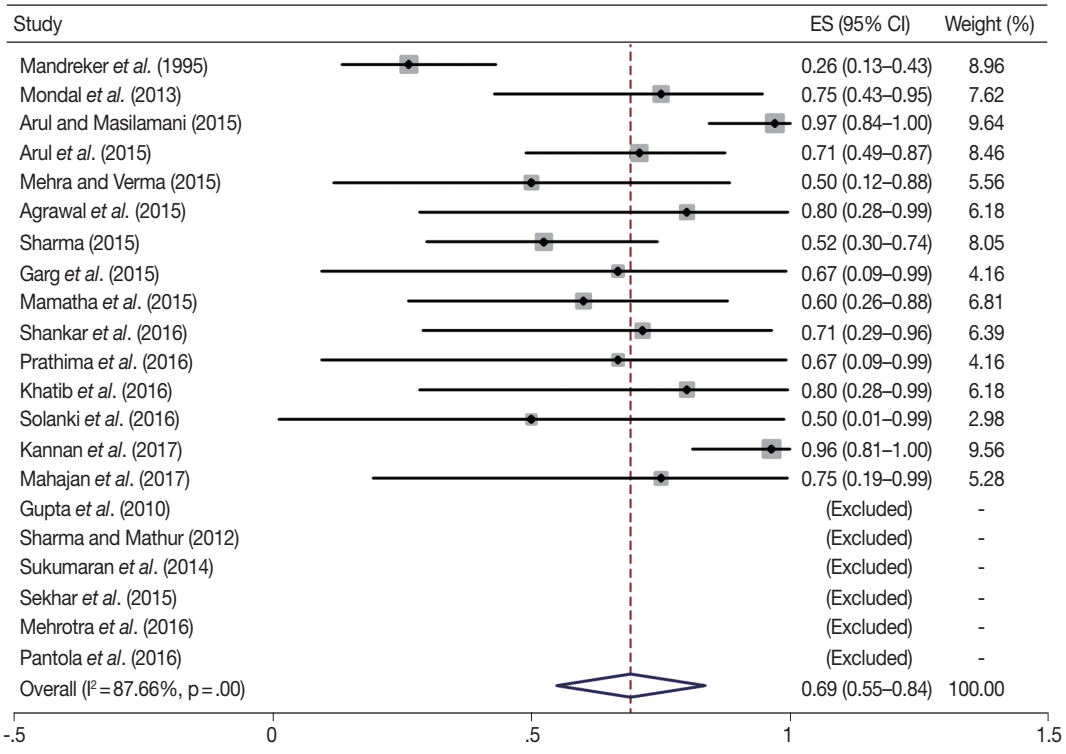


Fig. 5. Forest plot of meta-analysis on the risk of malignancy for Bethesda category V (suspicious for malignancy).[13,17,21,23,30-36,38,42,44,45,46,53,54,56,60,61] ES, effect size; CI, confidence interval.

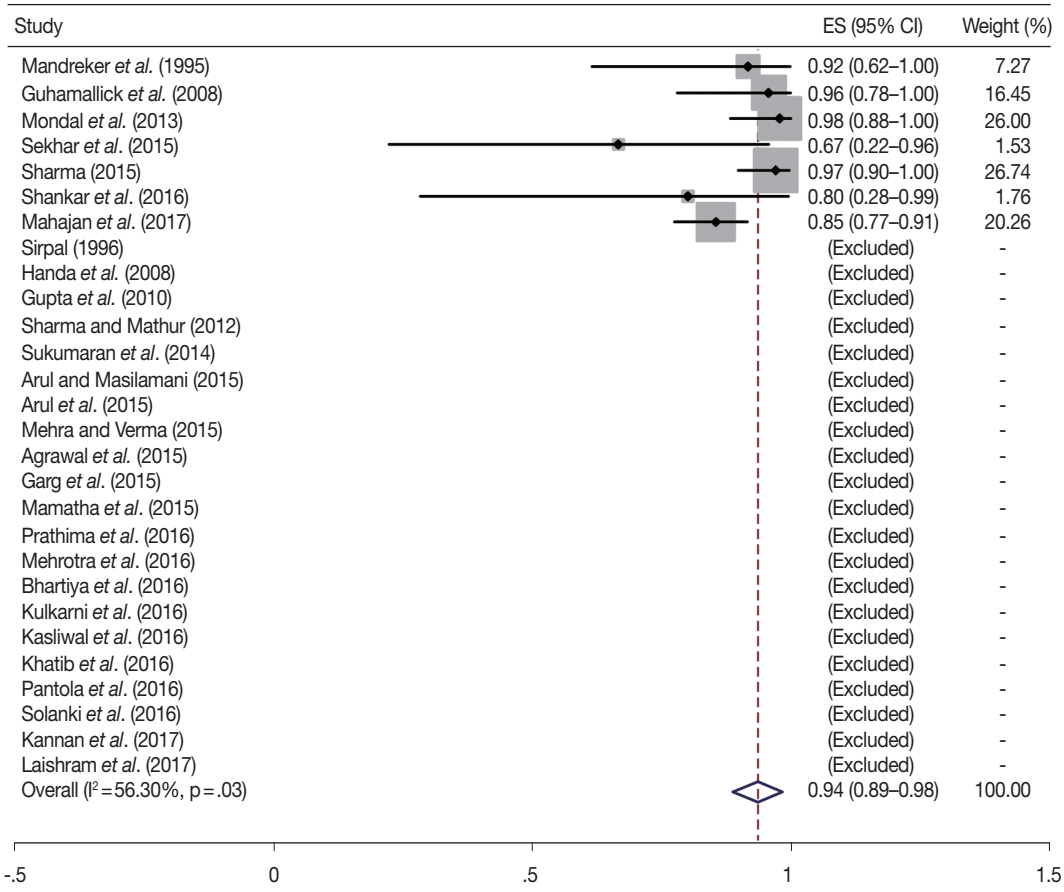


Fig. 6. Forest plot of meta-analysis on the risk of malignancy for Bethesda category VI (malignant).[13-17,21,23,30-36,38,42,44,45,46,49,50,52-54,56,60,61,63]

Acknowledgments

We sincerely thank Dr. Andrey Bychkov, MD, PhD, Department of Pathology, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand, for his expert guidance and help in manuscript editing.

REFERENCES

1. Gangadharan P, Nair MK, Pradeep VM. Thyroid cancer in Kerala. In: Shah AH, Samuel AM, Rao RS, eds. Thyroid cancer: an Indian perspective. Mumbai: Quest Publications, 1999; 17-32.
2. National Centre for Disease Informatics and Research; National Cancer Registry Programme Indian Council of Medical Research. Three-year report of population based cancer registries 2012-2014. Bengaluru: NCDIR-NCRP, 2016.
3. Unnikrishnan AG, Kalra S, Baruah M, *et al*. Endocrine Society of India management guidelines for patients with thyroid nodules: a position statement. Indian J Endocrinol Metab 2011; 15: 2-8.
4. Usha Menon V, Sundaram KR, Unnikrishnan AG, Jayakumar RV, Nair V, Kumar H. High prevalence of undetected thyroid disorders in an iodine sufficient adult south Indian population. J Indian Med Assoc 2009; 107: 72-7.
5. Baloch ZW, LiVolsi VA, Asa SL, *et al*. Diagnostic terminology and morphologic criteria for cytologic diagnosis of thyroid lesions: a synopsis of the National Cancer Institute Thyroid Fine-Needle Aspiration State of the Science Conference. Diagn Cytopathol 2008; 36: 425-37.
6. Krauss EA, Mahon M, Fede JM, Zhang L. Application of the Bethesda classification for thyroid fine-needle aspiration: institutional experience and meta-analysis. Arch Pathol Lab Med 2016; 140: 1121-31.
7. Martin HE, Ellis EB. Biopsy by needle puncture and aspiration. Ann Surg 1930; 92: 169-81.
8. Soderstrom N. Puncture of goiters for aspiration biopsy. Acta Med Scand 1952; 144: 237-44.
9. Das DK. Fine-needle aspiration cytology: its origin, development, and present status with special reference to a developing country, India. Diagn Cytopathol 2003; 28: 345-51.
10. Gupta SK, Dutta TK, Aikat M, Gupta BD, Talwar BL, Aikat BK. Evaluation of fine needle aspiration biopsy technique in the diagnosis

of tumours. Indian J Cancer 1975; 12: 257-67.
11. Singh P, Khanna SD, Manchanda RL. Needle biopsy of thyroid. Arch Surg 1965; 91: 646-51.
12. Rege JD, Nath AR, Bijlani JC, Trivedi DR, Deshpande DV. Fine needle aspiration cytology in solitary cold nodules of thyroid. J Assoc Physicians India 1987; 35: 819-21.
13. Mandreker SR, Nadkarni NS, Pinto RG, Menezes S. Role of fine needle aspiration cytology as the initial modality in the investigation of thyroid lesions. Acta Cytol 1995; 39: 898-904.
14. Sirpal YM. Efficacy of fine needle aspiration cytology in the management of thyroid diseases. Indian J Pathol Microbiol 1996; 39: 173-8.
15. Handa U, Garg S, Mohan H, Nagarkar N. Role of fine needle aspiration cytology in diagnosis and management of thyroid lesions: a study on 434 patients. J Cytol 2008; 25: 13-7.
16. Guhamallick M, Sengupta S, Bhattacharya NK, *et al*. Cytodiagnosis of thyroid lesions: usefulness and pitfalls: a study of 288 cases. J Cytol 2008; 25: 6-9.
17. Gupta M, Gupta S, Gupta VB. Correlation of fine needle aspiration cytology with histopathology in the diagnosis of solitary thyroid nodule. J Thyroid Res 2010; 2010: 379051.
18. Bagga PK, Mahajan NC. Fine needle aspiration cytology of thyroid swellings: how useful and accurate is it? Indian J Cancer 2010; 47: 437-42.
19. Sengupta A, Pal R, Kar S, Zaman FA, Sengupta S, Pal S. Fine needle aspiration cytology as the primary diagnostic tool in thyroid enlargement. J Nat Sci Biol Med 2011; 2: 113-8.
20. Renuka IV, Saila Bala G, Aparna C, Kumari R, Sumalatha K. The Bethesda System for Reporting Thyroid Cytopathology: interpretation and guidelines in surgical treatment. Indian J Otolaryngol Head Neck Surg 2012; 64: 305-11.
21. Sharma R, Mathur DR. Diagnostic accuracy of fine needle aspiration cytology (FNAC) of the thyroid gland lesions. Int J Health Sci Res 2012; 2: 1-7.
22. Patel MM, Patel K, Kaptan KR, Italiya SL, Saini G. Fine needle aspiration cytology as a first line investigation in thyroid lesions. Natl J Med Res 2013; 3: 106-10.
23. Mondal SK, Sinha S, Basak B, Roy DN, Sinha SK. The Bethesda system for reporting thyroid fine needle aspirates: a cytologic study with histologic follow-up. J Cytol 2013; 30: 94-9.
24. Kukar N, Malhotra V, Saluja M. Analysis of fine needle aspiration cytology of thyroid lesions. Internet J Pathol 2013; 15: 1-9.
25. Bhasin TS, Mannan R, Manjari M, *et al*. Reproducibility of 'The Bethesda System for Reporting Thyroid Cytopathology': a multicenter study with review of the literature. J Clin Diagn Res 2013; 7: 1051-4.
26. Borgohain R, Lal RK, Chatterjee P, Brahma N, Khanna S. A study of cyto-histological correlation in the diagnosis of thyroid swelling. IOSR J Dent Med Sci 2014; 13: 46-9.
27. Mangshetty SS, Jewargikar R, Andola SK. Fine needle aspiration cytology of 220 thyroid lesions with histopathological correlation. Int J Res Health Sci 2014; 2: 243-53.
28. Panchal MG, Deshpande SA, Noone RB, Suvernakar SV, Meshram DP. Diagnostic accuracy fine needle aspiration cytology of thyroid gland lesions. Int J Pharm Sci Invent 2014; 3: 5-10.
29. Pathak P, Srivastava R, Singh N, Arora VK, Bhatia A. Implementation of the Bethesda System for Reporting Thyroid Cytopathology: interobserver concordance and reclassification of previously inconclusive aspirates. Diagn Cytopathol 2014; 42: 944-9.
30. Sukumaran R, Kattoor J, Pillai KR, *et al*. Fine needle aspiration cytology of thyroid lesions and its correlation with histopathology in a series of 248 patients. Indian J Surg Oncol 2014; 5: 237-41.
31. Arul P, Masilamani S. A correlative study of solitary thyroid nodules using the Bethesda System for Reporting Thyroid Cytopathology. J Cancer Res Ther 2015; 11: 617-22.
32. Arul P, Akshatha C, Masilamani S. A study of malignancy rates in different diagnostic categories of the Bethesda System for Reporting Thyroid Cytopathology: an institutional experience. Biomed J 2015; 38: 517-22.
33. Sekhar A, Inamdae SS, Dombale VD, Prabhu MH. Fine needle aspiration cytology study of thyroid lesions: a 2 year prospective study in a tertiary centre. Int J Pharm Biol Sci Arch 2015; 3: 15-9.
34. Mehra P, Verma AK. Thyroid cytopathology reporting by the Bethesda system: a two-year prospective study in an academic institution. Patholog Res Int 2015; 2015: 240505.
35. Agrawal R, Saxena M, Kumar P. A study of fine needle aspiration cytology of thyroid lesions with histopathological correlation. Indian J Pathol Oncol 2015; 2: 277-83.
36. Sharma C. Diagnostic accuracy of fine needle aspiration cytology of thyroid and evaluation of discordant cases. J Egypt Natl Canc Inst 2015; 27: 147-53.
37. Thakkar B, Patel PJ, Mangar U, Shiladariya P, Suthar NB, Patel P. Retrospective study of fine needle aspiration cytology of thyroid lesions according to the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC). Int J Res Med 2015; 4: 132-6.
38. Garg S, Desai NJ, Mehta D, Vaishnav M. To establish Bethesda system for diagnosis of thyroid nodules on the basis of FNAC with histopathological correlation. J Clin Diagn Res 2015; 9: EC17-21.
39. Kathirvel C. Criteria for cytological diagnosis of thyroid lesions. Otolaryngol Online J 2015; 5(Suppl 2): 10-9.
40. Alagarsamy J, Sivaraman R, Kadhirvel V. Accuracy of fine needle aspiration cytology and incidence of benign/malignant tumours in solitary thyroid nodules. Int J Allied Med Sci Clin Res 2015; 3: 193-9.

41. Hathila R, Patel S, Vaghela P, Makwana G, Parmar P. Cytology findings of the thyroid lesions with the histopathology findings correlation. Int J Med Sci Public Health 2016; 5: 642-6.
42. Mamatha M, Sekhar SC, Rani HS, Anil SS, Vandana G. A comparative study between conventional system and the Bethesda system applied for reporting thyroid cytopathology. Int Arch Integr Med 2015; 2: 87-95.
43. Gupta V, Bhake A, Dayal S. Pattern and frequency of thyroid pathologies among thyroid cytology specimen in rural part of central India: a retrospective secondary data analysis. Thyroid Res Pract 2015; 12: 93-5.
44. Shankar SP, Meenakshisundaram K, Rajalakshmi V, Selvakumar SA, Giridharan B. The Bethesda System for Reporting Thyroid Cytopathology: a two year retrospective review in a tertiary care hospital. Indian J Pathol Oncol 2016; 3: 48-54.
45. Prathima S, Suresh TN, Harendra Kumar ML, Bhaskaran A. Impact of the Bethesda system in reporting thyroid cytopathology. Thyroid Res Pract 2016; 13: 9-14.
46. Mehrotra D, Anita AM, Andola SK, Patil AG. Thyroid cytology evaluation based on the Bethesda system with clinico-morphological correlation. Ann Pathol Lab Med 2016; 3: A347-355.
47. Tagore S, Jayaprakash HT, Sasidharan A, Nagaraj T, Santosh HN, Balraj L. Cytological study of thyroid lesions by fine-needle aspiration cytology. J Med Radiol Pathol Surg 2016; 2: 12-5.
48. Kalita DJ, Das B. A three years study of FNAC of thyroid lesions in a tertiary care hospital. Indian J Appl Res 2016; 6: 559-61.
49. Bhartiya R, Mallik M, Kumari N, Prasad BN. Evaluation of thyroid lesions by fine-needle aspiration cytology based on Bethesda System for Reporting Thyroid Cytopathology classification among the population of South Bihar. Indian J Med Paediatr Oncol 2016; 37: 265-70.
50. Kulkarni CV, Mittal M, Nema M, Verma R. Diagnostic role of the Bethesda System for Reporting Thyroid Cytopathology in an academic institute of Central India: one year experience. Indian J Basic Appl Med Res 2016; 5: 157-66.
51. Lohiya V, Kalla AR, Bharadwaj V. Utilisation of Bethesda System for Reporting Thyroid Cytopathology: a study of 250 cases. Int J Appl Res 2016; 2: 103-7.
52. Kasliwal N, Tanwar S, Pachori G, Gupta N, Maheshwari N, Jain D. Usefulness of preoperative FNAC of thyroid swelling along with application of Bethesda system of reporting. Int J Med Res Prof 2016; 2: 204-9.
53. Khatib Y, Mulla A, Patel RD, Momin E, Gite V, Khade A. Classification of thyroid FNA smears into Bethesda categories and their correlation with thyroid function tests. Sch J Appl Med Sci 2016; 4: 916-23.
54. Pantola C, Kala S, Khan L, Pantola S, Singh M, Verma S. Cytological diagnosis of pediatric thyroid nodule in perspective of the Bethesda System for Reporting Thyroid Cytopathology. J Cytol 2016; 33: 220-3.
55. Babu SB, Raju R, Radhakrishnan S. Correlation of fine needle aspiration cytology with histopathology in the diagnosis of thyroid swellings. Int Surg J 2016; 3: 1437-41.
56. Solanki R, Chaudhary VK, Sharma M, Ansari M. Validity assessment of 'the Bethesda System for Reporting Thyroid Cytopathology'. Int J Health Sci Res 2016; 6: 126-32.
57. Aramani SS, Gururajaprasad C. A cytohistopathological correlation of thyroid lesions with critical evaluation of discordant cases: an experience at a tertiary care hospital. Ann Pathol Lab Med 2017; 4: A243-8.
58. Sunder KS, Khan MI. Role of fine needle aspiration cytology (FNAC) in diagnosis of thyroid lesions. J Contemp Med Dent 2017; 5: 30-4.
59. Garg S, Naik LP, Kothari KS, Fernandes GC, Agnihotri MA, Gokhale JC. Evaluation of thyroid nodules classified as Bethesda category III on FNAC. J Cytol 2017; 34: 5-9.
60. Kannan S, Raju N, Kekatpure V, *et al*. Improving Bethesda reporting in thyroid cytology: a team effort goes a long way and still miles to go. Indian J Endocrinol Metab 2017; 21: 277-81.
61. Mahajan S, Srinivasan R, Rajwanshi A, *et al*. Risk of malignancy and risk of neoplasia in the Bethesda indeterminate categories: study on 4,532 thyroid fine-needle aspirations from a single institution in India. Acta Cytol 2017; 61: 103-10.
62. Chandra S, Chandra H, Bisht SS. Malignancy rate in thyroid nodules categorized as atypia of undetermined significance or follicular lesion of undetermined significance: an institutional experience. J Cytol 2017; 34: 144-8.
63. Laishram RS, Zothanmawii T, Joute Z, Yasung P, Debnath K. The Bethesda system of reporting thyroid fine needle aspirates: a 2-year cytologic study in a tertiary care institute. J Med Soc 2017; 31: 3-7.
64. Witt BL, Schmidt RL. Rapid onsite evaluation improves the adequacy of fine-needle aspiration for thyroid lesions: a systematic review and meta-analysis. Thyroid 2013; 23: 428-35.
65. Shield PW, Cosier J, Ellerby G, Gartrell M, Papadimos D. Rapid onsite evaluation of fine needle aspiration specimens by cytology scientists: a review of 3032 specimens. Cytopathology 2014; 25: 322-9.
66. Pitman MB, Abele J, Ali SZ, *et al*. Techniques for thyroid FNA: a synopsis of the National Cancer Institute Thyroid Fine-Needle Aspiration State of the Science Conference. Diagn Cytopathol 2008; 36: 407-24.
67. Keyhani E, Sharghi SA, Amini R, *et al*. Liquid base cytology in evaluation of thyroid nodules. J Diabetes Metab Disord 2014; 13: 82.
68. Prasad D, Bhaskar V, Kumar B. Study of utility of manual liquid-based cytology and conventional smears in the evaluation of various fine-needle aspiration samples. Paripex Indian J Res 2017; 6: 609-10.
69. Tripathy K, Misra A, Ghosh JK. Efficacy of liquid-based cytology

versus conventional smears in FNA samples. J Cytol 2015; 32: 17-20.
70. Gupta V, Bhake A, Dayal S. Better thyroid cytopathology reporting and interpretation using different classification systems. Thyroid Res Pract 2016; 13: 110-4.
71. Bose S, Kapila K, Verma K. Medullary carcinoma of the thyroid: a cytological, immunocytochemical, and ultrastructural study. Diagn Cytopathol 1992; 8: 28-32.
72. Gupta A, Jain S, Khurana N, Kakar AK. Expression of p63 and Bcl-2 in malignant thyroid tumors and their correlation with other diagnostic immunocytochemical markers. J Clin Diagn Res 2016; 10: EC04-8.
73. Tandon A, Paul TR, Singh R, Narendra AM. Synchronous thyroid involvement in plasma cell leukemia masquerading as Hashimoto's thyroiditis: role of ancillary cytology techniques in diagnostic workup. Endocr Pathol 2015; 26: 324-7.
74. Goel MM, Budhwar P. Fine needle aspiration cytology and immunocytochemistry in tuberculous thyroiditis: a case report. Acta Cytol 2008; 52: 602-6.
75. Aron M, Kapila K, Verma K. Utility of galectin 3 expression in thyroid aspirates as a diagnostic marker in differentiating benign from malignant thyroid neoplasms. Indian J Pathol Microbiol 2006; 49: 376-80.
76. Jain D, Mathur SR, Guleria R, Iyer VK. Utility and pattern of positivity of p40 in the diagnosis of squamous cell carcinoma of the lung by cytology: the first study on fine needle aspiration smears. Cytopathology 2014; 25: 330-5.
77. Jain D, Mathur SR, Sharma MC, Iyer VK. Cytomorphology of sebaceous carcinoma with analysis of p40 antibody expression. Diagn Cytopathol 2015; 43: 456-61.
78. Vallonthaiel AG, Jain D, Singh V, *et al*. c-Myb overexpression in cytology smears of tracheobronchial and pulmonary adenoid cystic carcinomas. Acta Cytol 2017; 61: 77-83.
79. Roy M, Jain D, Bakhshi S, Mathur S, Iyer VK. Primary Langerhans cell histiocytosis of the thyroid gland: role of langerin in FNA cytological diagnosis. Cytopathology 2015; 26: 128-30.
80. Jain D, Mathur SR, Iyer VK. Cell blocks in cytopathology: a review of preparative methods, utility in diagnosis and role in ancillary studies. Cytopathology 2014; 25: 356-71.
81. Bhandari V, Yadav YK, Khanna G, Sharma M, Singh M, Rajni. Efficacy of cytokeratin 19 expression on fine needle aspiration cell blocks in pre-operative diagnosis of malignant thyroid neoplasms. Clin Cancer Invest J 2012; 1: 212-6.
82. Haugen BR, Alexander EK, Bible KC, *et al*. 2015 American Thyroid Association management guidelines for adult patients with thyroid nodules and differentiated thyroid cancer: the American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer. Thyroid 2016; 26: 1-133.
83. Reddy KH, Kumar DR, Punnoose AM, *et al*. Detection of *BRAF* mutations in different thyroid cancers of archival FNAC samples. Indian J Endocrinol Metab 2013; 17(Suppl 1): S373-394.
84. Bychkov A. Prevalence of *BRAF*V600E mutation in Asian patients with thyroid cancer. Malays J Pathol 2017; 39: 95-6.
85. Katna R, Shet T, Sengar M, *et al*. Clinicopathologic study and outcome analysis of thyroid lymphomas: experience from a tertiary cancer center. Head Neck 2013; 35: 165-71.
86. Usha M, Kamath S, Sridhar M, Soman S. Primary thyroid lymphoma in the background of Hashimoto thyroiditis. Clin Cancer Invest J 2015; 4: 362-4.
87. Yeshvanth SK, Lakshminarayana KP, Upadhyaya VS, Shetty JK. Primary thyroid lymphoma arising from Hashimotos thyroiditis diagnosed by fine needle aspiration cytology. J Cancer Res Ther 2012; 8: 159-61.
88. Somani S, Kotwal N. Core needle biopsy: an additional diagnostic armamentarium. Thyroid Res Pract 2016; 13: 52-6.
89. Straccia P, Rossi ED, Bizzarro T, *et al*. A meta-analytic review of the Bethesda System for Reporting Thyroid Cytopathology: has the rate of malignancy in indeterminate lesions been underestimated? Cancer Cytopathol 2015; 123: 713-22.
90. Olson MT, Clark DP, Erozan YS, Ali SZ. Spectrum of risk of malignancy in subcategories of 'atypia of undetermined significance'. Acta Cytol 2011; 55: 518-25.
91. Horne MJ, Chhieng DC, Theoharis C, *et al*. Thyroid follicular lesion of undetermined significance: evaluation of the risk of malignancy using the two-tier subclassification. Diagn Cytopathol 2012; 40: 410-5.
92. Kim SD, Han SH, Jeong WJ, Kim H, Ahn SH. Differences in clinical features between subcategories of "atypia/follicular lesion of undetermined significance". Endocr Pathol 2017 May 9 [Epub]. https://doi.org/10.1007/s12022-017-9486-3.
93. Rosario PW, Calsolari MR. Importance of cytological subclassification of thyroid nodules with Bethesda category III cytology (AUS/FLUS) into architectural atypia only and nuclear atypia: a prospective study. Diagn Cytopathol 2017; 45: 604-7.
94. Layfield LJ, Baloch ZW, Esebua M, Kannuswamy R, Schmidt RL. Impact of the reclassification of the non-invasive follicular variant of papillary carcinoma as benign on the malignancy risk of the Bethesda System for Reporting Thyroid Cytopathology: a meta-analysis study. Acta Cytol 2017; 61: 187-93.
95. Bychkov A, Hirokawa M, Jung CK, *et al*. Low rate of noninvasive follicular thyroid neoplasm with papillary-like nuclear features in Asian practice. Thyroid 2017; 27: 983-4.

Journal of Pathology and Translational Medicine 2017; 51: 548-554
https://doi.org/10.4132/jptm.2017.09.29

■ REVIEW ■

Thyroid Cytology: The Japanese System and Experience at Yamashita Thyroid Hospital

Shinya Satoh · Hiroyuki Yamashita
Kennichi Kakudo[1]

Department of Endocrine Surgery, Yamashita Thyroid Hospital, Fukuoka; [1]Department of Pathology, Kindai University Faculty of Medicine, Nara Hospital, Ikoma, Japan

Received: August 25, 2017
Revised: September 23, 2017
Accepted: September 29, 2017

Corresponding Author
Shinya Satoh, MD
Department of Endocrine Surgery, Yamashita Thyroid Hospital, 1-8 Shimogofuku-machi, Fukuoka City, Fukuoka 812-0034, Japan
Tel: +81-92-281-1300
Fax: +81-92-281-1301
E-mail: shinya.s.48128@kojosen.com

In Japan, fine-needle aspiration (FNA) cytology is the most important diagnostic modality for triaging patients with thyroid nodules. A clinician (endocrinologist, endocrine surgeon, or head and neck surgeon) generally performs FNA cytology at the outpatient clinic, and ultrasound (US)-guided FNA is widespread because US is extremely common and most clinicians are familiar with it. Although almost all FNA thyroid samples are examined by certified cytopathologists and pathologists, some clinicians assess cytological specimens themselves. In Japan, there are two clinical guidelines regarding the management of thyroid nodules. One is the General Rules for the Description of Thyroid Cancer (GRDTC) published by the Japanese Society of Thyroid Surgery (JSTS) in 2005, and the other is the national reporting system for thyroid FNA cytology published by the Japan Thyroid Association in 2013 (Japanese system). Although the Bethesda System for Reporting Thyroid Cytopathology (Bethesda system) is rarely used in Japan, both the GRDTC and Japanese system tried to incorporate the Bethesda system so that the cytological diagnoses would be compatible with each other. The essential point of the Japanese system is stratification of follicular neoplasm (FN) into three subgroups based on cytological features in order to reduce unnecessary diagnostic thyroidectomy, and this system has been successful in stratifying the risk of malignancy in FN patients at several high-volume thyroid surgery centers. In Japan, the measurement of thyroglobulin and/or calcitonin in FNA needle washings is often used as an adjunct for diagnosis of possible cervical lymph node metastasis when FNA cytology is performed.

Key Words: Thyroid; Fine needle aspiration cytology; Indeterminate; The Bethesda System for Reporting Thyroid Cytopathology; Japan; Risk stratification; Risk of malignancy

BRIEF HISTORY OF THYROID FINE-NEEDLE ASPIRATION

Shortly after Söderström in Sweden described thyroid fine-needle aspiration (FNA) cytology in 1952, it was introduced to Japan.[1,2] In as early as 1972, Toriya published his first report on thyroid FNA cytology (in Japanese with an English abstract) from Ito Hospital (a thyroid center in Tokyo, Japan)[3] where more than 1,000 surgical procedures for thyroid disease were carried out every year. His cytological diagnostic criteria were descriptive and similar to those for pathological diagnosis of surgical specimens. In 2005, Toriya[4] reported his results and cytological-histological correlations in 1,702 surgically treated Japanese patients. He found 68 patients (4.0% of those receiving surgical treatment) in the indeterminate category, including 39 with benign lesions and 29 with malignancies, and the risk of malignancy (ROM) was 42.6% in the indeterminate category.[4] In that report, he divided the indeterminate category into two subcategories (favor benign and favor malignant). Among 28 patients in the favor malignant sub-category, 16 had malignancy (4 papillary thyroid carcinomas [PTCs] and 12 follicular thyroid carcinomas [FTCs]) and the ROM was 57.1%. Among 40 patients in the favor benign sub-category, 13 had malignancy (3 PTCs and 10 FTCs) and the ROM was 32.5%.[4] This principle to sub-classify indeterminate nodules into low-risk and high-risk categories was similar to the Bethesda System, but the actual ROMs obtained by Toriya[4] in surgically treated nodules were higher than the implied ROMs for the atypia of undetermined significance (AUS)/follicular lesions of uncertain significance (FLUS) (5%–15%) and follicular neoplasm (FN)/suspicious for follicular neoplasm (15%–30%) categories estimated by the Bethesda system textbook.[5]

FINE-NEEDLE ASPIRATION THYROID CYTOLOGY: SAMPLING AND INTERPRETATION

In Japan, the clinician (endocrinologist, endocrine surgeon,

pISSN 2383-7837
eISSN 2383-7845

or head and neck surgeon) generally performs FNA cytology at an outpatient clinic and FNA sampling by cytopathologists is unusual. Ultrasonography (US) is widely available and inexpensive (it costs only 35 U.S. dollars, about 10 U.S. dollars [30%] paid by patients and 25 U.S. dollars covered by the Japanese national health insurance system). Thus, US-guided FNA is preferred by most clinicians. Pathologists in Japan usually have dual certification; they are certified for anatomical pathology by the Japanese Society of Pathology and also for cytopathology by the Japanese Society of Clinical Cytology (JSCC). One of the characteristics of Japanese cytology practice is that a significant number of cytopathologists have a second clinical speciality, such as gynecology, endocrine surgery, breast surgery, respiratory medicine, respiratory surgery, urology, dental surgery, etc. They usually make a cytological diagnosis after collecting the specimens themselves.[6] Approximately half of all thyroid FNA samples are diagnosed at commercial laboratories by board-certified pathologists and the other half are assessed at hospitals by pathologists and doctors who are board-certified cytologists.

CYTOTECHNOLOGIST TRAINING PROGRAM AND JAPANESE SOCIETY OF CLINICAL CYTOLOGY CERTIFICATION

The JSCC holds examinations for qualification of cytotechnologists and those who pass are registered as JSCC certified cytotechnologists. There are two pathways to obtaining national board recognition as a medical technologist in Japan; candidates must complete either a 3-year course at a vocational school or a 4-year course at a medical technology school, after which graduates can sit the national examination for medical technologists. Those who obtain a national medical technology license can apply to sit the JSCC examination for cytotechnologists after working for one year at the cytology laboratory of a teaching hospital. More than 800 teaching hospitals in Japan provide education on cytological diagnosis and technical skills, along with cytology laboratory experience. There are also college level (4-year) schools whose graduates are eligible to sit the JSCC cytotechnologist examination without 1 year of practical experience, but these schools are fewer than 10 and they remain a minor pathway. JSCC certified cytotechnologists provide both technical assistance and cytology screening services in the cytology laboratories of most institutions in Japan and often play a leading role in a central laboratory.

PREPARATION AND STAINING OF THYROID CYTOLOGY SAMPLES

A 22- or 23-gauge needle is often used instead of a 26- or 27-gauge needle, in combination with a 10-mL disposable syringe and a pistol-grip mechanical syringe holder. Local anesthesia is not provided. Two smear slides are usually prepared from one FNA specimen, and are fixed in 95% ethanol or an equivalent fixative. Wet-fixed smears with Papanicolaou stain are favored by most laboratories, although some cytopathologists prefer air-dried smears with Giemsa stain or Diff-Quik stain. Liquid-based cytology is used in some laboratories, but is not widely available. Examination of FNA needle washings (hormonal assays and other diagnostic markers) has recently become popular for confirmation of the cytological diagnosis.

Thyroid FNA cytology reporting systems in Japan

There are two sets of clinical guidelines for handling thyroid nodules. One is the *General Rules for the Description of Thyroid Cancer* (GRDTC) published by the Japanese Society of Thyroid Surgery (JSTS) in 2005 and updated in 2016.[7] It includes a reporting system for thyroid FNA cytology. The other is a national reporting system (the Japanese system) for thyroid FNA cytology that was included in the clinical guidelines published by the Japan Thyroid Association (JTA) in 2013.[8,9] The GRDTC reporting system was adapted from the Papanicoloau Society recommendations published in 1996,[10] while the Japanese system was adapted from Toriya's diagnostic system developed at Ito Hospital in Japan, which is characterized by two or three subclassifications of the indeterminate category.[8,9] The Japanese system initially classifies the lesions in indeterminate category into indeterminate A (follicular neoplasms without PTC-type nuclear features [PTC-N]) and indeterminate B (nodules with nuclear atypia). The indeterminate A is further classified into three subcategories: A1 (favor benign), A2 (borderline), and A3 (favor malignant) (Table 1).[9] Although the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) itself is rarely used in Japan, the GRDTC and JTA system tried to incorporate TBSRTC so that cytological diagnoses made with the GRDTC or Japanese system could be related to TBSRTC diagnoses, and even the JSTS recommended TBSRTC after significant modifications. The GRDTC system is widely used in Japan, but several high-volume thyroid surgery centers prefer the Japanese system because they want to stratify the ROM in patients with FN. The indication for surgery is judged comprehensively by risk assessment on the basis of cytological diagnosis, ultrasound findings, and clinical findings. The prototype of the

Japanese system has been used to reduce unnecessary diagnostic thyroidectomy in patients with FN cytology at several thyroid centers, including Ito Hospital (Tokyo, Japan) and Kuma Hospital (Kobe, Japan). As a result, it is not mandatory for Japanese patients with FN to undergo diagnostic thyroidectomy, which is completely different from the management in Western countries.

Thyroid cytology classification according to the Japanese system

In the Japanese system, thyroid nodules are classified into six categories by cytological diagnosis (inadequate, benign, indeterminate A, indeterminate B, suspicious for malignancy, and malignant) (Table 1, Fig. 1). Indeterminate A is a category for FN (equivalent to Bethesda IV, but excluding cases with PTC-N), which is divided into three subcategories (A1, A2, and A3) based on cytological findings and ROM. These cytological subcategories have an impact on making decisions about clinical management. The cytological features of indeterminate A are no colloid background, microfollicular growth, enlarged nuclei, 3-dimensional clusters, and high cellularity. Nodules that demonstrate prominent trabecular clusters, cellular atypia, loss of cellular polarity, loss of cohesiveness, and nuclear overlapping are classified as group A3 (Fig. 2), while those with sheet-like follicles, medium-sized follicles, and a low nucleus/cytoplasm ratio are classified as group A1 (Fig. 3). Group A2 is intermediate between groups A1 and A3. Oxyphilic follicular tumors are handled as a separate group from indeterminate A and do not undergo subclassification in the Japanese system. Indeterminate B consists of lesions with nuclear abnormalities that are difficult to distinguish between benign and malignant, and are differentiated from FN by the presence of worrisome PTC-N. Lesions with extremely equivocal findings of PTC or medullary thyroid carcinoma (MTC) may be classified into this category. Indeterminate

Table 1. Cytological reporting system recommended in the 2013 Japanese guideline for management of thyroid nodules

Diagnostic category	Risk of malignancy (%)
Inadequate (non-diagnostic)	10
Normal or benign	<1
Indeterminate	
Indeterminate A (foliicular neoplasm)	
A-1: favor benign	<15
A-2: borderline	15–30
A-3: favor malignant	40–60
Indeterminate B (others: atypia in non-follicular pattern lesions)	40–60
Suspicious for malignancy (not conclusive for malignancy)	>80
Malignancy	>99

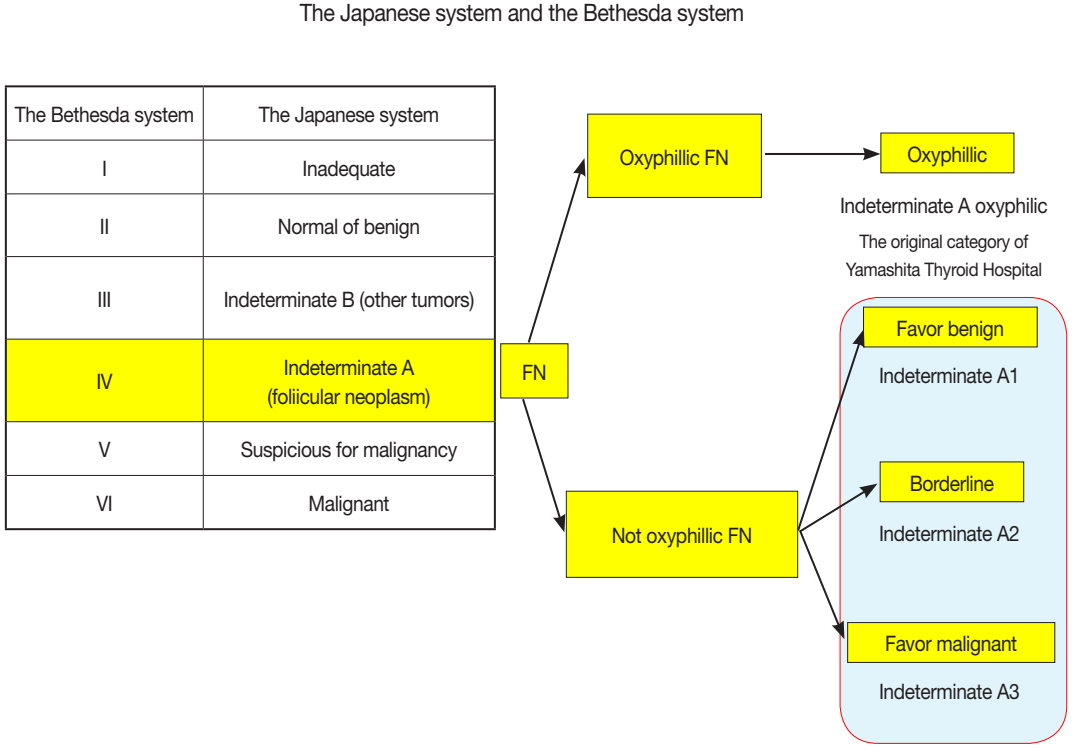


Fig. 1. Correlation between the Japan Thyroid Association reporting system (the Japanese system) and the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC). FN, follicular neoplasm.

https://doi.org/10.4132/jptm.2017.09.29

B is almost equivalent to the Bethesda III (AUS/FLUS) category, but FLUS without PTC-N are excluded and classified as indeterminate A1 by the Japanese system. The other categories of the Japanese system (inadequate, benign, suspicious for malignancy, and malignant) are equivalent to those of TBSRTC (Fig. 1). A further modification of the Japanese system is classifying samples that only contain cyst fluid as benign because of an extremely low ROM.[11]

The Japanese system in clinical practice at Yamashita Thyroid Hospital

There are differences in surgical indications between Western and Asian practice, and the indications vary even among the institutions in Japan.

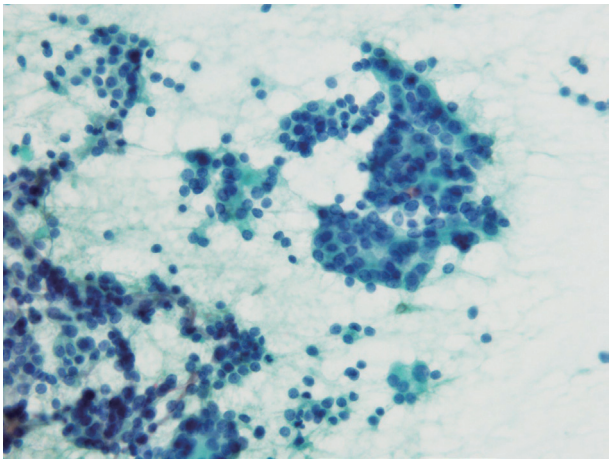


Fig. 2. Cytological findings of indeterminate A3. Cellular atypia, loss of cellular polarity, nuclear enlargement, and nuclear over-lapping are noted. The specimen was aspirated from a minimally invasive follicular carcinoma.

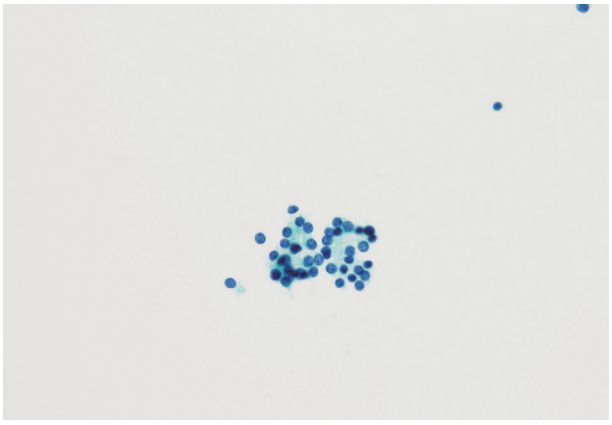


Fig. 3. Cytological findings of indeterminate A1. Microfollicular clusters are seen. The nuclei are round and slightly small. The specimen was aspirated from a follicular adenoma.

At Yamashita Thyroid Hospital, the indications for surgery in patients with thyroid nodules are as follows. Patients with inadequate cytology are recommended to undergo repeat FNA cytology or follow-up examination at intervals of 6 to 12 months. US is routinely performed at each follow-up visit in these patients. If a follicular tumor or cancer is suspected by US during the follow-up, repeat FNA cytology is usually performed. Patients with benign cytology are usually recommended to have follow-up examination, but diagnostic thyroidectomy may be recommended when the tumor is larger than 4 cm in diameter. Patients with indeterminate A1 cytology receive either diagnostic thyroidectomy or observation based on US findings and tumor size. Patients with indeterminate A2 or A3 cytology are recommended to undergo diagnostic thyroidectomy unless the tumor is small (e.g. < 1 cm in diameter). Although repeat FNA is recommended for patients with indeterminate B cytology, most patients with suspected malignancy according to US findings immediately undergo diagnostic thyroidectomy without repeat FNA. Patients with suspicious for malignancy or malignant cytology are recommended to undergo therapeutic thyroidectomy, but some patients with papillary microcarcinoma may be managed by observation, a so-called active surveillance.[12-14]

At Yamashita Thyroid Hospital (Fukuoka, Japan), about 1,000 surgical procedures for thyroid disease are carried out every year, and the results of US-guided FNA cytology for 1,600 nodules from January 2015 to April 2016 are summarized in Tables 2 and 3. These tables show the correlation between cytological and histological diagnoses categorized according to either TBSRTC or the Japanese system. Because liquid-based cytology was not performed during this period, the frequency of Bethesda I (inadequate category) was high at 23.9% (382/1,600). The resection rate (RR), ROM in resected patients, and ROM in all patients receiving FNA are presented in Table 2. Among 154 nodules classified as Bethesda IV (Indeterminate A), 75 nodules were A1, 29 nodules were A2, and 11 nodules were A3. Thirty-nine lesions were suspected to be oxyphilic follicular tumors and were separately classified as indeterminate A oxyphilic according to our original classification. The RR and ROM in each of these four subcategories are also listed in Table 3. We were able to successfully stratify category A into subcategories A1, A2, and A3 according to the increase of ROM and RR, thus reducing unnecessary thyroidectomies in patients with indeterminate A (FN).

The correlation between cytological and histological diagnoses in patients who underwent FNA cytology and surgical follow-up at Yamashita Thyroid Hospital is presented in Table 2. While PTC accounted for 98% of Bethesda VI, only two MTCs, one

Table 2. Correlation between cytological and histological diagnoses in patients undergoing FNA cytology and surgical follow-up at Yamashita Thyroid Hospital from January 2015 to April 2016

The Bethesda system			Surgery	Resection rate (%)	Histological diagnosis																ROM at surgery (%)	Overall ROM (%)
	Cytological diagnosis	No. (%)			AN	H	FA	FA oxy	Others	FTC	FTC oxy	PTC	PTC fol	PTC macrofol	PTC others	WDC-NOS	MTC	ATC	MALT	DLBCL		
I	Inadequate	382 (24)	63	16.5	35	0	11	2	2	1	0	8	2	0	2	0	0	0	0	0	20.6	3.4
II	Benign	626 (39)	89	14.2	67	4	5	0	2	1	0	8	0	0	0	0	0	0	2	0	12.4	1.8
III	AUS	171 (11)	47	27.5	22	0	16	2	0	0	0	0	2	0	0	0	0	0	4	1	14.9	4.1
IV	Follicular neoplasm	154 (9.6)	85	55.2	21	0	26	17	0	12	2	0	2	2	0	2	1	0	0	0	24.7	13.6
V	Suspicious for malignancy	52 (3.3)	39	75	3	0	0	0	0	1	0	27	4	0	3	0	0	0	1	0	92.3	69.2
VI	Malignant	215 (13)	199	92.6	0	0	0	0	0	0	0	188	5	0	2	0	2	1	0	1	100	92.6
		1,600 (100)	522	32.6	148	4	58	21	4	15	2	231	15	2	7	2	3	1	7	2	55	17.9
					Benign 235							Malignant 287										

FNA, fine-needle aspiration; AN, adenomatous nodule; H, Hashimoto's thyroiditis; FA, follicular adenoma; FA oxy, oxyphilic follicular adenoma; FTC, follicular thyroid carcinoma; FTC oxy, follicular thyroid carcinoma, oxyphilic variant; PTC, papillary thyroid carcinoma; PTC fol, papillary thyroid carcinoma, follicular variant; PTC macrofol, papillary thyroid carcinoma, macrofollicular variant; WDC-NOS, well-differentiated carcinoma, not otherwise specified; MTC, medullary thyroid carcinoma; ATC, anaplastic thyroid carcinoma; MALT, mucosa-associated lymphoid tissue; DLBCL, diffuse large B-cell lymphoma; ROM, risk of malignancy; AUS, atypia of undetermined significance.

Table 3. Resection rate and risk of malignancy in patients with indeterminate A cytology (including oxyphilic follicular neoplasm)

Cytological classification	No. (%)		Malignancy	ROM at histology (%)	Overall ROM (%)	Histological classification (malignancy)					
	FNA-cytology	Resection				FTC	FTC oxy	PTC fol	PTC macrofol	WDC-NOS	MTC
Indeterminate A1	75 (48.7)	33 (44)	4	12.1	5.3	2	1	-	1	-	-
Indeterminate A2	29 (18.8)	20 (68.9)	10	50	34.5	7	-	-	1	2	-
Indeterminate A3	11 (7.1)	10 (90.9)	6	60	54.5	3	-	2	-	-	1
Indeterminate A oxyphilic	39 (25.3)	22 (56.4)	1	4.5	2.6	-	1	-	-	-	-
	154 (100)	85 (55.2)	21	24.7	13.6	12	2	2	2	2	1

FNA, fine-needle aspiration; ROM, risk of malignancy; FTC, follicular thyroid carcinoma; FTC oxy, follicular thyroid carcinoma, oxyphilic variant; PTC fol, papillary thyroid carcinoma, follicular variant; PTC macrofol, papillary thyroid carcinoma, macrofollicular variant; WDC-NOS, well-differentiated carcinoma, not otherwise specified; MTC, medullary thyroid carcinoma.

anaplastic thyroid carcinoma and one diffuse large B-cell lymphoma were included in this category. Similarly, PTC accounted for more than 90% of the malignant tumors in Bethesda V. FTC accounted for about 70% of the malignant tumors in Bethesda IV (indeterminate A), while follicular variant PTC accounted for 20%. In each indeterminate A subcategory (A1, A2, and A3), more than half of the malignant tumors had a histological diagnosis of FTC. In our hospital, the final histological diagnosis was FTC for the majority of malignancies in indeterminate A.

In 2015, Sugino *et al*.[15] reported on the correlation between cytological and histological diagnosis in indeterminate A patients according to the Japanese system. They assessed 1,553 indeterminate A patients (4.3%) among 36,066 patients who underwent US-guided FNA thyroid cytology between 2005 and 2011 at Ito Hospital (Tokyo, Japan). Histological diagnosis of the primary thyroid tumor was available in 779 of the 1,553 patients with indeterminate A lesions. The overall RR was 50.2%, with the RR being 46.9% in A1, 55.4% in A2, and 65.1% in A3. The overall ROM was 34.5% in resected cases, while the ROM was 30.0% in resected A1 cases, 40.3% in A2, and 50.0% in A3. In addition, FTC accounted for about 75.5% of malignant tumors in indeterminate A patients, and 11 follicular variant PTCs (4.1%) were included in indeterminate A category. Their results are fairly consistent with our findings.

In conclusion, the Japanese system seems to direct high-risk patients with FN to surgery, while low-risk patients with FN are recommended for follow-up. FTC accounted for the majority of malignancies in Japanese patients with indeterminate A lesions who underwent diagnostic thyroidectomy, which is different from reports on Western patients.[16,17]

ANCILLARY TESTING, INCLUDING CORE BIOPSY

Core needle biopsy is rarely performed in thyroid patients in Japan; it may be used instead of open biopsy to confirm malignant lymphoma of the thyroid or undifferentiated carcinoma in inoperable cases. Molecular testing for *BRAF* mutation and other molecular alterations, as suggested in the 2015 guidelines of the American Thyroid Association,[18] may be used to triage patients with indeterminate nodules, but is not a common practice in Japan. As of 2017, it is only done for academic research[19] because its use is not supported by the Japanese national health insurance system. On the other hand, measurement of thyroglobulin (Tg) and/or calcitonin in FNA needle washings is often used as a helpful diagnostic adjunct in patients with thyroid nodules or cervical tumors, as are serum tumor markers. When the needle aspirate is immediately fixed and submitted for cytological examination, the needle and syringe are washed with 0.5 mL of saline and the washings are also submitted for measurement of biochemical markers. The presence of Tg in the FNA needle washings suggests that the tumor originated from thyroid follicular cells. This method is useful for establishing the diagnosis of lymph node metastasis when samples are obtained from possible cervical metastases of well-differentiated thyroid carcinoma. In 1983, Miyauchi *et al*.[20] (one of the leaders in the field of thyroid oncology in Japan and Asia) reported that the detection of very high thyroglobulin content in aspirates from cystic lymph nodes of the neck supported the diagnosis of metastatic PTC. This method is also applied to the diagnosis of MTC. Kudo (a colleague of Miyauchi) *et al*.[21] reported that the measurement of calcitonin in FNA needle washings could identify MTC with a high sensitivity and specificity. This is the historical background that explains why the measurement of Tg and/or calcitonin in aspirates is preferred by endocrinologists and endocrine surgeons in Japan.

CONCLUSION

In Japan, the clinician (endocrinologist, endocrine surgeon, or head and neck surgeon) usually performs FNA cytology at an

outpatient clinic, while it is rarely done by pathologists, and US-guided FNA is common.

A 22- or 23-gauge needle is popular instead of a 26- or 27-gauge needle. Wet-fixed smears (95% ethanol) and Papanicolaou stain are favored by most laboratories in Japan.

The most popular reporting system for thyroid FNA cytology in Japan is the GRDTC adapted from the 1996 Papanicolaou Society recommendations. Although TBSRTC was introduced to replace the GRDTC, it has not yet become popular in Japan. The Japanese system is used at several high-volume thyroid surgery centers to stratify the ROM for FN nodules.

Cytological diagnoses made by the GRDTC or Japanese system correspond to those of TBSRTC.

At high-volume thyroid centers, the Japanese system has been useful for risk stratification in patients with FN nodules and it has reduced unnecessary diagnostic thyroidectomy.

The measurement of Tg and/or calcitonin in FNA needle washings is often used as a diagnostic adjunct in patients with possible cervical lymph node metastasis.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

REFERENCES

1. Soderstrom N. Puncture of goiters for aspiration biopsy. Acta Med Scand 1952; 144: 237-44.
2. Okui K, Lindberg LG. Aspiration cytology in Lund University Hospital, Sweden. J Jpn Soc Clin Cytol 1979; 18: 105-9.
3. Toriya K. Cytological aspiration biopsy of goiters, especially with reference to their histological and cytological examinations. J Jpn Soc Clin Cytol 1972; 11: 101-14.
4. Toriya K. Fine needle aspiration cytology of the thyroid. Shindan-tochiryou 2005; 93: 1051-5.
5. Ali SZ, Cibas ES. The Bethesda system for reporting thyroid cytopathology: definitions, criteria and explanatory notes. New York: Springer, 2010.
6. Kakudo K, Kameyama K, Miyauchi A. History of thyroid cytology in Japan and reporting system recommended by the Japan Thyroid Association. J Basic Clin Med 2013; 2: 10-5.
7. Japanese Society of Thyroid Surgery Editorial Committee. General rules for the description of thyroid cancer. 6th ed. Tokyo: Kanahara Publisher, 2005.
8. Japan Thyroid Association. Guidelines for clinical practice for the management of thyroid nodules in Japan 2013. Tokyo: Nankodo Co., Ltd., 2013.
9. Kakudo K, Kameyama K, Miyauchi A, Nakamura H. Introducing the reporting system for thyroid fine-needle aspiration cytology according to the new guidelines of the Japan Thyroid Association. Endocr J 2014; 61: 539-52.
10. Guidelines of the Papanicolaou Society of Cytopathology for the examination of fine-needle aspiration specimens from thyroid nodules. The Papanicolaou Society of Cytopathology Task Force on Standards of Practice. Diagn Cytopathol 1996; 15: 84-9.
11. Takada N, Hirokawa M, Suzuki A, Higuchi M, Kuma S, Miyauchi A. Reappraisal of "cyst fluid only" on thyroid fine-needle aspiration cytology. Endocr J 2017; 64: 759-65.
12. Miyauchi A. Clinical trials of active surveillance of papillary microcarcinoma of the thyroid. World J Surg 2016; 40: 516-22.
13. Leboulleux S, Tuttle RM, Pacini F, Schlumberger M. Papillary thyroid microcarcinoma: time to shift from surgery to active surveillance? Lancet Diabetes Endocrinol 2016; 4: 933-42.
14. Brito JP, Ito Y, Miyauchi A, Tuttle RM. A clinical framework to facilitate risk stratification when considering an active surveillance alternative to immediate biopsy and surgery in papillary microcarcinoma. Thyroid 2016; 26: 144-9.
15. Sugino K, Kameyama K, Ito K. Characteristics and outcome of thyroid cancer patients with indeterminate cytology. J Basic Clin Med 2015; 4: 92-8.
16. Yang J, Schnadig V, Logrono R, Wasserman PG. Fine-needle aspiration of thyroid nodules: a study of 4703 patients with histologic and clinical correlations. Cancer 2007; 111: 306-15.
17. Williams BA, Bullock MJ, Trites JR, Taylor SM, Hart RD. Rates of thyroid malignancy by FNA diagnostic category. J Otolaryngol Head Neck Surg 2013; 42: 61.
18. Haugen BR, Alexander EK, Bible KC, *et al*. 2015 American Thyroid Association management guidelines for adult patients with thyroid nodules and differentiated thyroid cancer: the American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer. Thyroid 2016; 26: 1-133.
19. Kumagai A, Namba H, Akanov Z, *et al*. Clinical implications of pre-operative rapid BRAF analysis for papillary thyroid cancer. Endocr J 2007; 54: 399-405.
20. Miyauchi A, Takai S, Morimoto S, *et al*. Fine needle aspiration of neck tumor. Application of cytological, bacteriological and hormonal examinations. Nihon Geka Gakkai Zasshi 1983; 84: 667-73.
21. Kudo T, Miyauchi A, Ito Y, Takamura Y, Amino N, Hirokawa M. Diagnosis of medullary thyroid carcinoma by calcitonin measurement in fine-needle aspiration biopsy specimens. Thyroid 2007; 17: 635-8.

Journal of Pathology and Translational Medicine 2017; 51: 555-559
https://doi.org/10.4132/jptm.2017.07.14

■ REVIEW ■

Thyroid Fine-Needle Aspiration Practice in the Philippines

Agustina D. Abelardo

Department of Pathology, College of Medicine, University of the Philippines Manila, Manila, Philippines

Received: July 4, 2017
Accepted: July 14, 2017

Corresponding Author
Agustina D. Abelardo, MD, MIAC
Department of Pathology, College of Medicine, University of the Philippines Manila, 547 Pedro Gil St Ermita, Manila 1000, Philippines
Tel: +63-2-526-45-50
Fax: +63-2-526-45-50
E-mail: abelardoagustina@yahoo.com

Fine-needle aspiration (FNA) is a well accepted initial approach in the management of thyroid lesions. It has come a long way since its introduction for nearly a century ago. In the Philippines, FNA of the thyroid was first introduced 30 years ago and has been utilized until now as a mainstay in the diagnosis of thyroid malignancy. The procedure is performed by pathologists, endocrinologists, surgeons, and radiologists. Most pathologists report the cytodiagnosis using a combination of the aspiration biopsy cytology method that closely resembles the histopathologic diagnosis of thyroid disorders and the six-tier nomenclature of The Bethesda System for Reporting Thyroid Cytopathology. Local endocrinologists and surgeons follow the guidelines of the 2015 American Thyroid Association in the management of thyroid disorders. There is still a paucity of local research studies but available data deal with cytohistologic correlations, sensitivity, specificity, and accuracy rates as well as usefulness of ultrasound-guided FNA. Cytohistologic correlations have a wide range of sensitivity from 30.7% to 73% and specificity from 83% to 100%. The low sensitivity can be attributed to poor tissue sampling since a majority of the thyroid FNA is done by palpation only. The reliability can be improved if FNA is guided by ultrasound as attested in both international and local studies. Overall, FNA of the thyroid has enabled the diagnosis of thyroid disorders with an accuracy of 72.8% to 87.2% and it correlates well with histopathology.

Key Words: Fine-needle aspiration cytology; Thyroid fine-needle aspiration; The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC)

Fine-needle aspiration (FNA) biopsy has proved to be an accurate, safe, and cost-effective method in the initial management of thyroid nodules. It is a widely accepted diagnostic tool all over the world with a history spanning from the reports of Mannheim followed by the works of Martin & Ellis on needle aspirates way back in the 1930s.[1-3] In the 1950s to the 1970s, Scandinavian pioneers from Sweden continued to develop the method including aspiration biopsy cytology of practically all organs.[4-6] To date, voluminous works on accuracy data, cytohistologic correlations, the usefulness of six-tier Bethesda System for Reporting Thyroid Cytopathology (TBSRTC), and a wide array of ancillary studies on thyroid FNA are available.

HISTORY OF FINE-NEEDLE ASPIRATION IN THE PHILIPPINES

FNA of the thyroid started in the late 1980s at the Philippine General Hospital, a 1,500-bed tertiary government hospital that serves as the training facility for health sciences students enrolled in the University of the Philippines. Consultants and resident physician trainees from the departments of pathology and surgery started doing FNA of various palpable lesions mainly in the neck, 40% to 50% of which were thyroid aspirates. Thereafter, an aspiration cytology unit in the Department of Pathology housed at the College of Medicine of the University was established upon the return of a faculty member of the department from a 2-month training in FNA at the Cytology Unit of Karolinska Hospital in Stockholm, Sweden, in 1987. Two years later, another pathologist with interest in cytopathology established an FNA clinic in a private university hospital in Metro Manila.

Resident physicians in the Department of Pathology used the technique prescribed by the Scandinavian pioneers utilizing a 10-mL syringe with 23G to 25G needles attached to a syringe holder. Some surgeons and endocrinologists performed the procedure without a syringe holder. Smears were prepared by the aspirationist and cytotechnicians stained the smears using either Papanicolaou stain or modified Wright-Giemsa stain (Diff-Quik), or both. All interpretations were rendered by the pathologist on duty.

In the 1990s, private hospitals in the national capital region (Metro Manila) likewise engaged in the practice of thyroid FNA with pathologists, surgeons, and endocrinologists performing the procedure. Pathologists who were trained from the university

pISSN 2383-7837
eISSN 2383-7845

hospital performed thyroid FNA in their affiliated hospitals when they started their practice in various regions of the country.

In 2010, interventional radiologists, endocrinologists, and cytopathologists started doing thyroid FNA under ultrasound-guidance, especially for small lesions measuring from 1.0 to 1.5 cm.

Local endocrinologists follow the 2015 guidelines of the American Thyroid Association.[7] In 2011, a revisit of the clinical practice guidelines at the Philippine General Hospital for patients with thyroid cancer maintained the original 2008 recommendation of total or near total thyroidectomy for all patients with well differentiated thyroid carcinoma with nodule size of greater than 1.0 cm in diameter, and lobectomy with an isolated nodule size of less than 1.0 cm in diameter, and without lymph node metastasis on preoperative ultrasound.[8] There is no specific recommendation on preoperative FNA including terminology referable to TBSRTC. Intra-operative frozen section for non-diagnostic FNA is controversial as no consensus was reached by the multispecialty members of the panel. Likewise, international guidelines do not recommend the use of frozen section due to its limited role with high frequency of false-negative results and lack of consistent agreement between frozen section and final histological diagnosis.[9]

TECHNIQUE OF FINE-NEEDLE ASPIRATION

Most practicing pathologists performing thyroid FNA use a syringe holder made of aluminum or plastic. The procedure starts with proper positioning of the patient, application of 70% alcohol at the puncture site, and localization of the lesion by immobilizing the target with one hand followed by needle puncture of the skin into the target lesion. Negative pressure or suction is applied by back and forth cutting motions within the thyroid nodule and releasing this negative pressure prior to withdrawal of the needle once the material is obtained. Some surgeons and endocrinologists use only a syringe without a syringe holder while others use the non-aspirating technique.[10]

PREPARATION OF SAMPLE

The physician-aspirationist prepares the smears by placing a small drop of the aspirated material onto a glass slide. Smear is made by laying another glass on top of the sample material and pulling the slides apart to spread it. Wet smears are fixed in 95% ethyl alcohol or air dried for submission to the laboratory. Wet smears are subjected to Papanicolaou stain while air dried smears are prepared for modified Wright-Giemsa stain (Diff-Quik). The latter stain is also used in cases where there is an on-site request for rapid evaluation of the aspirates. All smears and liquid samples obtained from cyst fluids are processed in the laboratory by cytotechnicians. Smears are routinely stained with Papanicolaou stain and the remains of the samples are processed as cell block and stained with hematoxylin and eosin. Liquid based cytology is not used for thyroid aspirates.

CYTOPATHOLOGY AND CYTOTECHNOLOGY TRAINING

Formal cytopathology and cytotechnology training programs are not offered in the country at present. However, cytopathology is included as a rotation in anatomic pathology training programs. In both government and private hospitals with training in pathology, all aspirates are screened by pathology resident physician trainees and final cytodiagnosis is signed out by the pathologist on duty. In the absence of a training program, the pathologist on duty at the cytology unit screens and issues the final cytodiagnosis.

Pathologists interested in cytopathology have to study abroad for fellowship in cytopathology, after which they can apply to the local pathology society for recognition. To date, there are ten cytopathologists in the whole country, two of whom are certified by the local pathology society. Majority of the practice is in the national capital region.

There is no formal subspecialty society for cytopathology within the national organization of pathologists. Lectures in cytopathology are incorporated in the scientific program during the annual convention of the Philippine Society of Pathologists. Within individual training institutions, conferences in cytopathology are embedded in the training program of anatomic pathology.

THYROID CYTOLOGY REPORTING SYSTEM

Prior to the 2009 publication of TBSRTC,[11] which is a standardized six-tier nomenclature, the aspiration biopsy cytology method was used.[4,5] This method provides a spectrum of diagnosis that closely resembles the histopathologic diagnosis of thyroid disorders. In most instances, it is cytologically possible to categorize a thyroid lesion into one of the three main entities: hyperplasia/adenomatous colloid nodule, thyroiditis, and neoplasm. There are also indeterminate cases in which the presence of cellular atypia cannot totally rule out a malignancy. For this, "atypia suspicious for malignancy" cytodiagnosis is given and followed by a recommendation for clinical correlation or for fur-

 https://doi.org/10.4132/jptm.2017.07.14

ther investigation if warranted. Cytodiagnosis is based on the assessment of cellularity, architectural or group patterns of diagnostic cells, individual cell cytology including nuclear and cytoplasmic characteristics, and the presence of other cells and materials in the background. Criteria for non-diagnostic aspirates (category I) follow the criteria prescribed by TBSRTC, and these include bloody samples, presence of blood and colloid only, paucicellular smears with less than six follicular cell groups of ten cells each, and poorly prepared smears.[11] Aspiration performed with dissolution of the nodule is compatible with a thyroid cyst and is not considered as non-diagnostic if pathologist has a clear knowledge of the patient's biopsy findings. With the introduction of TBSRTC in 2009, a majority of the thyroid FNA reports utilized both descriptive diagnosis and its corresponding equivalent category number in the six-tier nomenclature for a more effective understanding by the referring physicians.

Difficult thyroid aspirates are handled in various ways by pathologists. Self-review is done with subsequent intradepartmental referral to another pathologist if a cytopathologist is not part of the hospital staff. Presently, the Philippines is still in the process of developing an external quality assurance program in cytopathology.

Based on a recent survey of FNA practice in 16 hospitals in the country, thyroid FNA comprised 46% to 85% of the total FNA in five major hospitals at the national capital region, and 30% to 85% in 11 hospitals in various regions of the country. At the Philippine General Hospital, 60% of aspirates were from the thyroid. Being a training hospital, it has the highest number of non-diagnostic aspirates, ranging from 37% to 46% with an average of 42%. The high number is unique since different resident physician trainees perform the procedure in the course of their rotation in the departments engaged in FNA. Four private hospitals from the national capital region with a bed capacity from 300 to almost 1,000 had a non-diagnostic yield ranging from 5% to 7%. In the various regions of the country from north to south, category I registered 0 to 19% from a survey of four governments and seven private hospitals with a 300–400 bed capacity in the former and 90–680 bed capacity in the latter. Category I was 0% in one private hospital in southern Philippines since the physician-aspirationist is a pathologist who always makes a rapid on-site evaluation as part of FNA. Benign aspirates (category II) comprised 50% to 85%. Aspirates in the category III accounted for 1% to 20% and were re-aspirated following the recommendation of TBSRTC. Category IV aspirates ranged from 1% to 14%, category V from 1% to 10%, and category VI from 2% to 20%.

THYROID CYTOLOGY AUDIT PROGRAM

Patients nowadays are highly mobile and may seek medical services in different institutions. It often happens that FNA is done in one hospital and thyroid surgery with histopathologic diagnosis is performed in another center. However, all thyroid surgeries with previous FNA done in the same hospital are reviewed as part of the hospitals' monitoring program. No data has been released but may soon be forthcoming.

STATUS OF ANCILLARY TESTING

Ancillary studies of thyroid FNA are infrequently used. Immunohistochemistry in the diagnosis of thyroid tumors such as galectin-3, cytokeratin 19, and HBME-1 are only rarely utilized in a few hospitals at the national capital region for thyroid surgical pathology specimens and not in aspirates. Molecular testing for somatic mutations is yet to be developed because most of the patients opt thyroid surgery with just regular follow-up.[12,13] Health care insurance does not cover molecular testing for somatic mutations. If requested, molecular testing is mainly physician driven. If the patient agrees to pay for the molecular testing, the test is referred to neighboring Asian countries or to North America.

REVIEW OF PUBLICATIONS

Researches on thyroid cytology in the country are mainly cytohistologic correlations with sensitivity, specificity, and accuracy rates. Cytohistologic correlation from three hospitals in the national capital region yielded a wide range of sensitivity from 30.7% to 73%, specificity from 83% to 100%, and accuracy from 72.8% to 87.2%.[14-16] The low sensitivity can be attributed to poor tissue sampling since most thyroid FNAs are done by palpation only. A majority (85%) of the discordant cases were due to sampling errors attributable to dual pathology with a dominant benign lesion, missing a small malignant focus which is usually a papillary microcarcinoma.[14] The reliability can be improved if FNA is guided by ultrasound as attested in both international and local studies.[16-18] Solid nodules with microcalcifications and irregular margins which were found to be significant predictors of malignancy can be readily detected.[19,20] Improvement in diagnostic yield and accuracy was observed from 82.6% to 86.2% with ultrasound guidance.[16] When combined with frozen section examination, ultrasound-guided FNA for concordant cases approached an accuracy of 97.2% in one study.[21]

The incidence of each diagnostic category of the Bethesda

Table 1. Distribution of thyroid fine-needle aspiration diagnoses by the Bethesda system

Study	Hospital	Year	Distribution by the Bethesda categories (%)						Nodule
			I	II	III	IV	V	VI	
Young *et al*. (2011)[24]	St. Luke's Medical Center, Quezon City	2007–2009	23.1	64.7		9.4[a]		2.8	2,239
Salillas and Almocera (2016)[25]	Private practice, Cebu City and Bohol	2010–2014	1.3	56.2	5.4	11.1	13.2	12.8	3,799

[a]Sum of indeterminate categories III to V: classified as "indeterminate."

Table 2. Cytohistopathologic correlations in operated thyroid nodules

Study	Hospital	Year	Risk of malignancy for the Bethesda categories (%)						Nodule
			I	II	III	IV	V	VI	
Young *et al*. (2011)[24]	St. Luke's Medical Center, Quezon City	2007–2009	17.2	9.6		36.5[a]		76.5	251
Abelardo *et al*. (2011)[15]	Philippine General Hospital, Manila	2010	44.4	18.9	40.0	44.4	60.0	100.0	99
Canete *et al*. (2014)[19]	Philippine General Hospital, Manila	2008–2011	n/a	32.0	n/a	41.4	89.7	100.0	837
Carlos *et al*. (2014)[22]	St. Luke's Medical Center, Quezon City	2012–2013	n/a	n/a	35.3	n/a	n/a	n/a	68
Ramos and Mirasol (2014)[16]	St. Luke's Medical Center, Quezon City	2012–2013	33.3	13.5	22.2	33.3	72.2	100.0	175
Salillas *et al*. (2015)[23]	SWU-MHAM College of Medicine, Cebu City	2009–2012	n/a	2.6	50.0	50.0	78.0	100.0	80
Salillas and Almocera (2016)[25]	Private practice, Cebu City and Bohol	2010–2014	n/a	n/a	39.4	n/a	n/a	n/a	33
Abelardo and Abesamis (2016)[14]	The Medical City, Pasig City	2010–2015	n/a	26.5	n/a	n/a	71.4[b]		448
The Bethesda system reference rate[11]			1–4	0–3	5–15	15–30	60–75	97–99	

n/a, not available.
[a]Sum of indeterminate categories III to V: classified as "indeterminate"; [b]Overall risk of malignancy for suspicious for malignancy and malignant cases.

system is rarely addressed in local studies (Table 1). Summary of available publications on the malignancy risk in the Bethesda categories is presented in Table 2. An overall risk of malignancy for malignant and suspicious for malignant cytodiagnosis is 71.4% while the overall risk of malignancy for benign cytodiagnosis is 26.5%.[14] The risk of malignancy for category III ranges from 35.3% to 50%,[15,22,23] which is higher than TBSRTC reference rate. Conveying these rates to our clinical colleagues with the recommendation to do a repeat FNA should be reconsidered for optimal patient care.[22,23]

FUTURE CHALLENGES

Although thyroid FNA was introduced more than 30 years ago, a centralized database may be difficult to construct because the FNA practice in the Philippines is highly individualistic. If teaching and training of FNA are structured into the major training programs of physicians, a standardized FNA protocol, particularly in specimen sampling, can be improved. Looking forward, there is a need to establish a national registry from various hospitals and training institutions. Quality assurance and other related issues can be addressed through the establishment of a local society of cytopathologists in order to develop and improve the art and science of FNA. Finally, collaborative networking amongst pathologists, endocrinologists and surgeons is a strong impetus toward unified practice in the diagnosis of thyroid disorders using FNA.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

Acknowledgments

The author wishes to acknowledge the contribution of the following pathologists from various regions of the Philippines who responded to the survey of FNA practice in the Philippines: Elizabeth Ann Alcazaren, Ma.Lourdes Goco, and Rolando Lopez (National Capital Region); Brenda Rosuman (Luzon); Jude Martin Cimafranca, Marvi Dulnuan, David Pedroza, Ansarie Salpin, Dianne Singson, and Walter Villanueva (Visayas); Rachel Alegata, Kenneth Crisostomo, Sherjan Kalim, and Gerard Lamayra (Mindanao).

REFERENCES

1. Diamantis A, Magiorkinis E, Koutselini H. Fine-needle aspiration (FNA) biopsy: historical aspects. Folia Histochem Cytobiol 2009; 47: 191-7.
2. Martin HE, Ellis EB. Biopsy by needle puncture and aspiration. Ann Surg 1930; 92: 169-81.
3. Wu M, Burstein DE. Fine needle aspiration. Cancer Invest 2004; 22: 620-8.

4. Linsk JA, Franzen S. Clinical aspiration cytology. Philadelphia: Lippincott, 1983.
5. Zajicek J. Aspiration biopsy cytology, Part I. Cytology of supradiaphragmatic organs. Basel: S. Karger, 1974.
6. Zajicek J. Aspiration biopsy cytology, Part II. Cytology of infradiaphragmatic organs. Basel: S. Karger, 1979.
7. Haugen BR, Alexander EK, Bible KC, *et al*. 2015 American Thyroid Association management guidelines for adult patients with thyroid nodules and differentiated thyroid cancer: the American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer. Thyroid 2016; 26: 1-133.
8. Sison CM, Obaldo J, Matsuo J, Uy GL, Jaring C. University of the Philippines - Philippine General Hospital revised clinical practice guidelines for the management of well-differentiated thyroid carcinoma of follicular cell origin. J Asean Fed Endocr Soc 2012; 27: 49-61.
9. Mulcahy MM, Cohen JI, Anderson PE, Ditamasso J, Schmidt W. Relative accuracy of fine-needle aspiration and frozen section in the diagnosis of well-differentiated thyroid cancer. Laryngoscope 1998; 108(4 Pt 1): 494-6.
10. Santos JE, Leiman G. Nonaspiration fine needle cytology: application of a new technique to nodular thyroid disease. Acta Cytol 1988; 32: 353-6.
11. Cibas ES, Ali SZ; NCI Thyroid FNA State of the Science Conference. The Bethesda System For Reporting Thyroid Cytopathology. Am J Clin Pathol 2009; 132: 658-65.
12. Bychkov A. Prevalence of *BRAF*V600E mutation in Asian patients with thyroid cancer. Malays J Pathol 2017; 39: 95-6.
13. Navarro-Locsin CG, Chang AM, Daroy ML, Alfon AC, Andal JJ, Padua PF. Clinical and histopathological profile of *BRAF* V600E mutation in conventional papillary thyroid carcinoma in a Filipino population. Malays J Pathol 2016; 38: 141-8.
14. Abelardo AD, Abesamis RG. Cytohistologic correlation of thyroid lesions: a six year review at the Medical City, Philippines. Acta Cytol 2016; 60(Suppl 1): 220.
15. Abelardo AD, Ang MA, Enriquez AM. Thyroid aspirates using the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) in the assessment of risk malignancy: a comparison with conventional system of cytodiagnosis. Proc Taiwan Soc Pathol 2011; 2: 148.
16. Ramos CO, Mirasol RC. Ultrasound guidance improves the diagnostic yield and accuracy of fine needle aspiration biopsy of thyroid nodules in detecting malignancy. J Thyroid Disorders Ther 2014; 3: 148.
17. Baloch ZW, LiVolsi VA. Fine-needle aspiration of thyroid nodules: past, present, and future. Endocr Pract 2004; 10: 234-41.
18. Lo TE, Uy AT, Maningat PD. Well-differentiated thyroid cancer: the Philippine General Hospital experience. Endocrinol Metab (Seoul) 2016; 31: 72-9.
19. Canete EJ, Sison-Pena CM, Jimeno CA. Clinicopathological, biochemical, and sonographic features of thyroid nodule predictive of malignancy among adult Filipino patients in a tertiary hospital in the Philippines. Endocrinol Metab (Seoul) 2014; 29: 489-97.
20. Puno-Ramos MP, Villa ML, Kasal RG, Arzadon J, Alcazaren EA. Ultrasound features of thyroid nodules predictive of thyroid malignancy as determined by fine needle aspiration biopsy. Philipp J Intern Med 2015; 53: 1-8.
21. Young J, Lumapas-Gonzalez CG, Mirasol R. The diagnostic accuracy of ultrasound guided fine-needle aspiration biopsy and intraoperative frozen section examination in nodular thyroid disease. J Asean Fed Endocr Soc 2011; 26: 44-50.
22. Carlos AD, Mirasol R, Thomas Aquino E, Lourdes Goco M, Rizelle Toledo P, Carl Santos K. Management and malignancy rate of thyroid nodules with a cytologic diagnosis of atypia or follicular lesion of undetermined significance. J Asean Fed Endocr Soc 2014; 29: 78-84.
23. Salillas AL, Sun FC, Almocera EG. Review of the Bethesda System for Reporting Thyroid Cytopathology: a local study in Bohol Island, Philippines. Acta Cytol 2015; 59: 77-82.
24. Young JK, Lumapas CG, Mirasol R. Sonographically guided fine-needle aspiration biopsy of thyroid nodules: correlation between cytologic and histopathologic findings. Philipp J Intern Med 2011; 49: 8-14.
25. Salillas AL, Almocera EG. The implication of atypia of undetermined significance (AUS) category to a pathologist in a third world country. Acta Cytol 2016; 60(Suppl 1): 50-1.

Journal of Pathology and Translational Medicine 2017; 51: 560-564
https://doi.org/10.4132/jptm.2017.09.20

REVIEW

Thyroid Fine-Needle Aspiration in Taiwan: The History and Current Practice

Jen-Fan Hang[1,2] · Chih-Yi Hsu[1,2]
Chiung-Ru Lai[1,2]

[1]Department of Pathology and Laboratory Medicine, Taipei Veterans General Hospital, Taipei; [2]School of Medicine, National Yang-Ming University, Taipei, Taiwan

Received: September 12, 2017
Accepted: September 20, 2017

Corresponding Author
Chiung-Ru Lai, MD
Department of Pathology and Laboratory Medicine, Taipei Veterans General Hospital, No. 201, Section 2, Shipai Road, Taipei 11217, Taiwan
Tel: +886-2-2875-7451
Fax: +886-2-2873-7052
E-mail: crlai@vghtpe.gov.tw

In Taiwan, thyroid cancer is the most common endocrine gland malignancy and the incidence of thyroid cancer has increased four-fold in the past two decades. Fine-needle aspiration is an accurate and cost-effective method of evaluating thyroid nodules and has been the gold-standard diagnostic tool for thyroid tumors in Taiwan since the 1980s. This article reviews the history, current practice, reporting systems, training, and quality assurance for thyroid fine-needle aspiration cytology in Taiwan.

Key Words: Thyroid; Biopsy, fine-needle; Cytology; Taiwan

Thyroid cancer is the most common endocrine gland malignancy, accounting for 3.26% of all cancers in Taiwan.[1] The incidence of thyroid cancer is 14.34 per 100,000 persons per year. It has a female predominance at 21.6 per 100,000 women per year (representing the fifth most common malignancy in women) versus 7.06 per 100,000 men per year (the 15th most common malignancy in men). The incidence has increased four-fold in the past two decades (from 3.3 per 100,000 persons per year in 1995). Unlike in South Korea,[2] there is no thyroid cancer screening program in Taiwan. However, the general population is increasingly aware of cancer-related issues and health checkups are growing in popularity. Detection of small thyroid nodules via ultrasonography largely accounts for the increasing incidence of thyroid cancer in Taiwan. Papillary thyroid carcinoma, which comprises 91% of all newly diagnosed thyroid cancers, has contributed to the majority of this increase (Fig. 1). Nevertheless, the mortality rate of thyroid cancer remains relatively low (0.86 per 100,000 women per year and 0.47 per 100,000 men per year) and has increased little since 1995 (0.73 per 100,000 women per year and 0.28 per 100,000 men per year).

Fine-needle aspiration (FNA) is an accurate and cost-effective method of evaluating thyroid nodules. It has been the gold-standard diagnostic tool for thyroid tumors in Taiwan since the 1980s. This article reviews the history, current practice, reporting systems, training, and quality assurance for thyroid FNA cytology in Taiwan.

THE HISTORY OF THYROID FINE-NEEDLE ASPIRATION IN TAIWAN

Before the 1980s, the application of diagnostic cytology in Taiwan was limited to exfoliative cytology, such as sputum, body fluid, and Pap test. The utilization of interventional cytology began with thyroid FNA. Prof. Tien-Chun Chang, an endocrinologist at National Taiwan University Hospital, is considered to be the pioneer of thyroid FNA in Taiwan. In 1979, he performed the first thyroid FNA on a patient with follicular thyroid carcinoma. After this initial attempt, which demonstrated promising diagnostic value, he began an on-site aspiration and cytologic diagnosis service at bedsides and in his thyroid clinic. Instead of using standard May-Grünwald-Giemsa staining, which usually takes less than 10 minutes, he applied Liu staining

pISSN 2383-7837
eISSN 2383-7845

on cytologic smears, which produced similar staining quality for rapid diagnosis.[3] The Liu stain is a modified Romanowsky stain invented by Prof. Chen-Hui Liu in 1953 for hematologic smears. The entire staining procedure is simple and takes only 2 minutes.[3] Therefore, it is suitable for on-site diagnostic services and has been widely used in Taiwan and China.

In 1981, Prof. Chang published the first article on thyroid FNA in a Taiwanese medical journal and attracted attention among clinicians.[4] Later on, he published a series of articles on the cytologic presentation of non-neoplastic thyroid diseases, such as Hashimoto's thyroiditis,[5] acute suppurative thyroiditis,[6] and granulomatous thyroiditis,[7] as well as thyroid carcinomas, such as papillary carcinoma[8] and anaplastic carcinoma[9] in the same journal. With these experiences from local practice, he started to publish articles in international journals in 1989.[10-22] An anecdote about him is that he performs thyroid FNA while holding the needle like a Chinese brush pen.[10] He published this unique FNA method in Acta Cytologica.[10] Prof. Chang was also the pioneer of ultrasound-guided thyroid FNA,[23] cytomorphometry analysis of thyroid tumors,[11,14] and immunoperoxidsase staining of thyroglobulin and parathyroid hormone on cytologic smears.[12,15] In 1995, he published the first color atlas of thyroid and parathyroid cytology using Liu staining, which was written in Chinese. This book has been the textbook of choice for thyroid FNA in Taiwan for many years and a second edition was published in 2015 (Fig. 2).[24]

CURRENT THYROID FINE-NEEDLE ASPIRATION PRACTICE IN TAIWAN

Clinicians, especially endocrinologists, used to be both the performer and the interpreter of thyroid FNA. However, after the establishment of Taiwan's National Health Insurance in 1995, specialized medicine has become the mainstay of healthcare. Physicians from different specialties started taking over the multidisciplinary tasks. Currently, radiologists are the main performers of FNA and pathologists are the main cytologic diagnosticians in Taiwan. Ultrasonography is used in the majority of thyroid FNA cases to guide the procedure.

When clinicians were the main performer of thyroid FNA and interpreter of corresponding cytology, air-dried direct smears

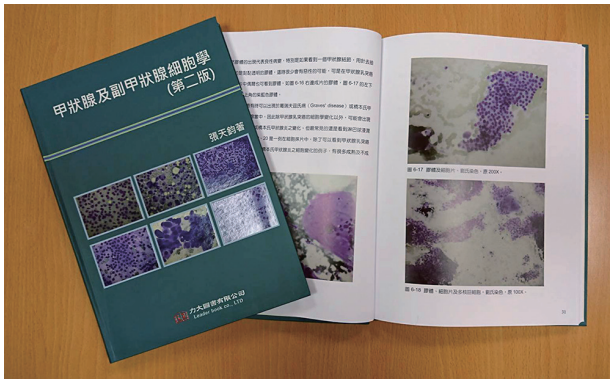
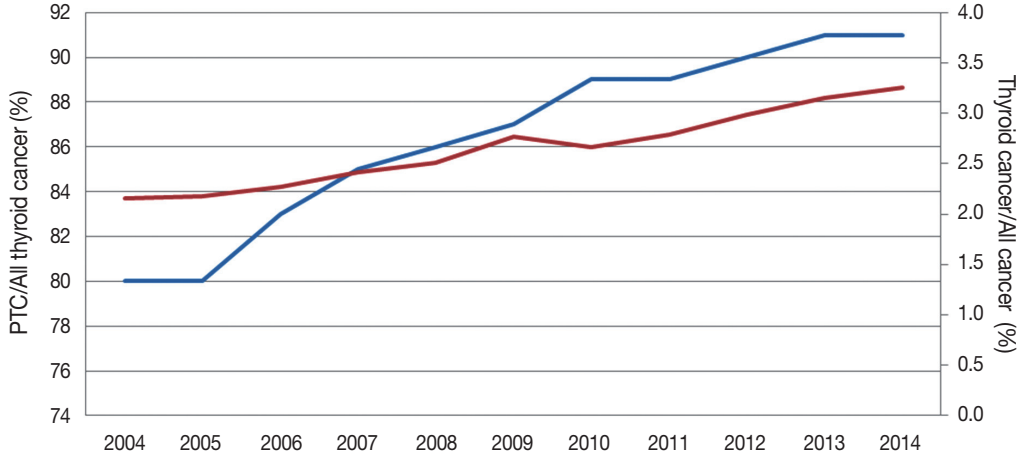


Fig. 2. The second edition of *Thyroid and Parathyroid Cytology* (Leader Book Company, 2015)[24] by Prof. Tien-Chung Chang.



	2004	2005	2006	2007	2008	2009	2010	2011	2012	2013	2014
PTC/All thyroid cancer	80%	80%	83%	85%	86%	87%	89%	89%	90%	91%	91%
Thyroid cancer/all cancer	2.16%	2.18%	2.27%	2.42%	2.51%	2.77%	2.66%	2.79%	2.99%	3.15%	3.26%

Fig. 1. Thyroid cancer statistics from 2004 to 2014. The proportion of papillary thyroid carcinoma (PTC) among thyroid cancers has increased 11% (blue line) and the proportion of thyroid cancer among all cancers has increased 1.1% (red line). Data from the Taiwan Cancer Registry Annual Report.[1]

and Liu staining were the method of choice for slide preparation. Papanicolaou stain has been commonly applied since pathologists took on thyroid cytology diagnosis, due to its superiority in demonstrating nuclear features that are important in papillary carcinoma diagnosis. Currently, conventional smears for both Papanicolaou staining (on alcohol-fixed slides) and Liu staining (on air-dried smears) are the most common preparations for thyroid FNA cytology in Taiwan. Liquid-based preparation was first introduced and applied in thyroid FNA in 2014, and is becoming more popular due to its high cell yield and standardized quality. In some institutions, immunocytochemical staining and molecular testing are also utilized on thyroid cytologic specimens to further facilitate diagnosis. Thyroid core-needle biopsy is a rare practice in Taiwan.

REVIEW OF THE LITERATURE AND REPORTING SYSTEM FOR THYROID CYTOLOGY IN TAIWAN

Given the history of thyroid FNA in Taiwan, most studies on the diagnostic accuracy and rate of malignancy were published by endocrinologists before the introduction of the Bethesda System. The major issue in previous studies is that investigators used different diagnostic categories, making it difficult to compare the findings of different studies. For example, Prof. Jen-Der Lin from Chang Gung Memorial Hospital used a four-tier system (benign, follicular neoplasm, malignant, and unsatisfactory) to emphasize the follicular pattern and this approach resulted in 16.6% of resected papillary carcinomas being diagnosed as follicular neoplasm.[25,26] In contrast, other groups used different four-tier systems without further specifying the follicular pattern.[27,28] Among all hospitals in Taiwan, only three branches of the Veterans General Hospital, in which American System of thyroid FNA practice was adopted initially, use the Bethesda System or a diagnostic system compatible with the Bethesda System.[29]

In Taiwan, the diagnostic criteria for inadequate specimen vary between different individuals. The "more than six groups and each group more than 10 follicular cells" rule proposed by the Bethesda System is usually applied.[30] Most pathologists from Taiwan still consider the specimen negative instead of non-diagnostic in the following situations: a specimen with less than six groups but more than 50 follicular cells in total, or a degenerative hemorrhagic cyst with scant benign follicular cells.

Comparing the diagnostic system used at our hospital with the corresponding categories in the Bethesda System and the British System,[31] the frequency of atypical diagnosis is lower in Taiwan (Table 1). However, the rate of malignancy in these categories is compatible to data collected in Western countries. A study from Taichung Veterans General Hospital, Taichung showed a similar trend.[29] We consider microcarcinomas larger than 0.5 cm found on resection specimens, which are usually detectable on ultrasonography, as malignant on follow-up. This could explain the higher rate of malignancy in our negative categories.

CYTOLOGY TRAINING AND QUALITY ASSURANCE IN TAIWAN

Cytology training for medical graduates is integrated into the Anatomic Pathology Residency Training Program. In addition to at least 3 months of cytology practice training, pathology residents are required to take a 2-week intensive course. This course covers various topics in cytology from Pap smear to thyroid FNA. A microscopic exam is held on the last day of the course. Residents need to pass the exam to be qualified. On the anatomic pathology board exam, in addition to surgical pathology, there is a separate cytopathology section. Residents need to get passing scores in both parts in order to pass the anatomic pathology board exam. For board-certified pathologists, annual cytology education activities are required.

The training program for cytotechnologists consists of one

Table 1. Comparison of diagnostic categories used at the Taipei Veterans General Hospital with the Bethesda System and the British System

Taiwan[a]			The Bethesda System[32]			The British System[31]		
Category	Prevalence (%)	Malignancy (%)[b]	Category	Prevalence (%)	Malignancy (%)[b]	Category	Prevalence (%)	Malignancy (%)[b]
Inadequate	8	16.7	I	13	16.8	Thy 1/1c	18	4
Negative	81	6.5	II	59	3.7	Thy 2/2c	42	1.4
Atypical	2	41.7	III	9.6	15.9	Thy 3a	5	17
Suspicious for follicular tumor	2	21.1	IV	10.1	26.1	Thy 3f	14	Up to 40
Suspicious for malignancy	2	75.0	V	2.6	75.2	Thy 4	2	Up to 68
Positive	5	97.2	VI	5.4	98.6	Thy 5	5	Up to 100

[a]Data from Taipei Veterans General Hospital, Taipei from 2010 to 2011; [b]Rate of malignancy on resected nodules.

 https://doi.org/10.4132/jptm.2017.09.20

year of on-site training at a qualified training institution and a final exam. The training institution must routinely provide both surgical pathology and cytology services and process more than 8,000 Pap smears annually. The cytology lab director must be a qualified cytopathologist (certified by either the International Academy of Cytology or the Taiwan Society of Clinical Cytology) and there must be at least two senior cytotechnologists on site with more than 3 years of work experience. Currently, there are 12 qualified institutions for cytotechnologist training in Taiwan. After training and exams, each qualified cytotechnologist is allowed to screen up to 10,000 cytology cases (Pap smears and non-gynecologic specimens) per year. Currently, there are about 550 practicing cytotechnologists in Taiwan. There have been no systematic surveys of the number of cytotechnologists working with thyroid specimens in Taiwan. Most work at hospitals where thyroid FNA is performed. Cytotechnologists usually perform specimen preparation and screening under the supervision of pathologists. Since 2017, the Taiwan Society of Clinical Cytology has started an inter-laboratory diagnosis comparison program for non-gynecologic cytology, including thyroid FNA. So far, a total of 81 labs have participated. The results are still under analysis and will be given to individual lab.

FUTURE CHALLENGES

Thyroid FNA cytology is being performed more frequently and has become the most common FNA specimen in Taiwan. Therefore, it is important to apply a uniform diagnostic system for consistent communication and management. Newly developed thyroid cytology technologies, such as liquid-based preparation, immunocytochemistry and molecular testing, may facilitate more accurate diagnosis.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

REFERENCES

1. Taiwan Cancer Registry annual report [Internet]. Taipei: Health Promotion Administration, Ministry of Health and Welfare, 2014 [cited 2017 Jun 1]. Available from: http://www.hpa.gov.tw/Pages/List.aspx? nodeid=269.
2. Ahn HS, Kim HJ, Welch HG. Korea's thyroid-cancer "epidemic": screening and overdiagnosis. N Engl J Med 2014; 371: 1765-7.
3. Chang TC, Kou SH, Chen FW, *et al*. Cytopathology of thyroid cancer by fine needle aspiration with Liu's stain. Taiwan Yi Xue Hui Za Zhi 1982; 81: 1404-12.
4. Huang TC, Chang TC, Lin CC, Chang CC, Chen FW. Fine needle aspiration cytology in thyroid disease. Taiwan Yi Xue Hui Za Zhi 1981; 80: 902-8.
5. Chang TC, Chen FW, Chang CC, Huang TS, Lin CC, Kuo SH. The role of fine needle aspiration cytology compared with thyroid antibody determination in the diagnosis of Hashimoto's thyroiditis. Taiwan Yi Xue Hui Za Zhi 1981; 80: 1152-9.
6. Chang TC, Chen FW, Kou SH, Chang CC, Fan JS, Liaw KY. The application of fine needle aspiration cytology in the diagnosis of acute suppurative thyroiditis: a case report. Taiwan Yi Xue Hui Za Zhi 1983; 82: 155-60.
7. Chang TC, Chen FW, Kou SH, Chang CC, Huang SM. Diagnostic criterion of granulomatous thyroiditis by the needle aspiration cytopathology. Taiwan Yi Xue Hui Za Zhi 1983; 82: 496-502.
8. Chang TC, Kou SH, Chen FW, *et al*. Cytologic presentation and its correlation with clinical stage in papillary carcinoma of the thyroid. Taiwan Yi Xue Hui Za Zhi 1987; 86: 941-6.
9. Chang TC, Liaw KY, Kuo SH, Chang CC, Chen FW. Anaplastic thyroid carcinoma: review of 24 cases, with emphasis on cytodiagnosis and leukocytosis. Taiwan Yi Xue Hui Za Zhi 1989; 88: 551-6.
10. Chang TC. Fine needle aspiration biopsy of the thyroid using the Chinese brush-holding technique. Acta Cytol 1989; 33: 939-40.
11. Chang TC, Kuo SH, How SW. Coefficient of variation of nuclear diameters as a prognostic factor in papillary thyroid carcinoma. Anal Quant Cytol Histol 1991; 13: 403-6.
12. Tung CC, Chang TC, Hsieh HC. Value of immunoperoxidase staining of thyroglobulin in fine needle aspiration cytology of thyroid diseases. Acta Cytol 1995; 39: 396-401.
13. Wang CY, Chang TC. Preoperative thyroid ultrasonography and fine-needle aspiration cytology in ectopic thyroid. Am Surg 1995; 61: 1029-31.
14. Tsai TH, Chang TC, Chiang CP. Nuclear area measurements of parathyroid adenoma, parathyroid hyperplasia and thyroid follicular adenoma: a comparison. Anal Quant Cytol Histol 1997; 19: 45-8.
15. Chang TC, Tung CC, Hsiao YL, Chen MH. Immunoperoxidase staining in the differential diagnosis of parathyroid from thyroid origin in fine needle aspirates of suspected parathyroid lesions. Acta Cytol 1998; 42: 619-24.
16. Wang CY, Chang TJ, Chang TC, Hsiao YL, Chen MH, Huang SH. Thyroidectomy or radioiodine? The value of ultrasonography and cytology in the assessment of nodular lesions in Graves' hyperthyroidism. Am Surg 2001; 67: 721-6.
17. Tseng FY, Hsiao YL, Chang TC. Ultrasound-guided fine needle

aspiration cytology of parathyroid lesions: a review of 72 cases. Acta Cytol 2002; 46: 1029-36.
18. Tseng FY, Hsiao YL, Chang TC. Cytologic features of metastatic papillary thyroid carcinoma in cervical lymph nodes. Acta Cytol 2002; 46: 1043-8.
19. Tsou PL, Hsiao YL, Chang TC. Multinucleated giant cells in fine needle aspirates: can they help differentiate papillary thyroid cancer from benign nodular goiter? Acta Cytol 2002; 46: 823-7.
20. Su DH, Liao KM, Hsiao YL, Chang TC. Determining when to operate on patients with Hashimoto's thyroiditis with nodular lesions: the role of ultrasound-guided fine needle aspiration cytology. Acta Cytol 2004; 48: 622-9.
21. Chang TC, Wu SL, Hsiao YL. Medullary thyroid carcinoma: pitfalls in diagnosis by fine needle aspiration cytology and relationship of cytomorphology to *RET* proto-oncogene mutations. Acta Cytol 2005; 49: 477-82.
22. Shih SR, Shun CT, Su DH, Hsiao YL, Chang TC. Follicular variant of papillary thyroid carcinoma: diagnostic limitations of fine needle aspiration cytology. Acta Cytol 2005; 49: 383-6.
23. Chang TC, Hong CT, Chang SL, Hsieh HC, Liaw KY, How SW. Correlation between sonography and pathology in thyroid diseases. J Formos Med Assoc 1990; 89: 777-83.
24. Chang TC. Thyroid and parathyroid cytology. 2nd ed. Taipei: Leader Book Company, 2015.
25. Lin JD, Chao TC, Huang BY, Chen ST, Chang HY, Hsueh C. Thyroid cancer in the thyroid nodules evaluated by ultrasonography and fine-needle aspiration cytology. Thyroid 2005; 15: 708-17.
26. Liu FH, Liou MJ, Hsueh C, Chao TC, Lin JD. Thyroid follicular neoplasm: analysis by fine needle aspiration cytology, frozen section, and histopathology. Diagn Cytopathol 2010; 38: 801-5.
27. Chen PY, Chiou SC, Yeh HY, *et al*. Correlation of ultrasonography with fine needle aspiration cytology and final pathological diagnoses in patients with thyroid nodules. Chin J Radiol 2010; 35: 1-7.
28. Tseng CE, Wei CK, Kuo CS, *et al*. Fine needle aspiration cytology of thyroid nodules: evaluation of diagnostic accuracy. Tzu Chi Med J 2008; 20: 296-303.
29. Kuo YT, Chang MC, Song YM, *et al*. Diagnosis and management of thyroid nodules with atypia: a three-year study at an institution in Taiwan. Diagn Pathol Open 2016; 1: 105.
30. Ali SZ, Cibas E. The Bethesda System for Reporting Thyroid Cytopathology: definitions, criteria and explanatory notes. New York: Springer, 2010.
31. Cross P, Chandra A, Giles T, *et al*. Guidance on the reporting of thyroid cytology specimens [Internet]. London: Royal College of Pathologists, 2016 [cited 2017 Jun 1]. Available from: http://ukeps.com/docs/thyroidfna.pdf.
32. Bongiovanni M, Spitale A, Faquin WC, Mazzucchelli L, Baloch ZW. The Bethesda System for Reporting Thyroid Cytopathology: a meta-analysis. Acta Cytol 2012; 56: 333-9.

 https://doi.org/10.4132/jptm.2017.09.20

Journal of Pathology and Translational Medicine 2017; 51: 565-570
https://doi.org/10.4132/jptm.2017.08.12

■ REVIEW ■

Current Status of Thyroid Fine-Needle Aspiration Practice in Thailand

Somboon Keelawat
Samreung Rangdaeng[1]
Supinda Koonmee[2]
Tikamporn Jitpasutham
Andrey Bychkov

Department of Pathology, Faculty of Medicine, Chulalongkorn University, Bangkok; [1]Department of Pathology, Faculty of Medicine, Chiang Mai University, Chiang Mai; [2]Department of Pathology, Faculty of Medicine, Khon Kaen University, Khon Kaen, Thailand

Received: August 3, 2017
Revised: August 10, 2017
Accepted: August 11, 2017

Corresponding Author
Andrey Bychkov, MD, PhD
Department of Pathology, Faculty of Medicine, Chulalongkorn University, Rama IV Rd., Pathumwan, Bangkok 10330, Thailand
Tel: +66-2-256-4235
Fax: +66-2-652-4208
E-mail: andrey.b@chula.ac.th

Thyroid carcinoma is one of the leading malignancies in Thailand increasingly prevalent in the female population. Fine-needle aspiration (FNA) cytology is a widely used diagnostic tool for evaluation of thyroid nodules and thyroid cancer. Thyroid FNA is a routine procedure universally performed in Thai hospitals by a variety of clinical specialists. Manual guidance is the first-line choice complemented by ultrasound assistance in selected cases. Despite national guidelines recommendations, the diagnostic criteria and terminology of the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) was slowly adopted in the local settings. Currently, the Bethesda system is actively promoted by the local professional societies as a uniform reporting system. Experience with thyroid FNA has been rarely reported to date—only a handful of publications are available in local journals. Our review, in addition to presenting various aspects of thyroid FNA in Thailand, established for the first time national references for a certain statistical outputs of TBSRTC based on the original multi-institutional cohort. The risk of malignancy in 2,017 operated thyroid nodules collected from three tertiary thyroid cancer centers was 21.7%, 14.7%, 35.9%, 44.4%, 76.7%, and 92.6% for categories I to VI, respectively. The malignancy risk in several diagnostic categories (II to IV) was higher than the risk estimated by TBSRTC and recent meta-analysis studies. We endorse the use of uniform terminology of the Bethesda system in Thailand, which will help facilitate communication among diverse medical professionals involved in the management of patients with thyroid nodules, to share local experience with the international audience.

Key Words: Fine-needle aspiration cytology; Thyroid fine-needle aspiration; The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC); Review; Thailand

Fine-needle aspiration (FNA) entered the clinical practice around 1930s, which was highlighted by the landmark paper by Martin and Ellis.[1] Further elaboration of FNA by the Swedish school in the mid-century was crucial for the establishment of the technique and its worldwide acceptance.[2] Thyroid, owing to its easy access, was one of the first organs to be practiced.[3] Since that time, the technique has gained popularity and is now commonly used as a standard method for initial diagnosis of lesions in various anatomic sites/organs.

Thyroid nodules are common, with a prevalence ranging 20%–60% depending on the screening approach, age, sex, iodine status, and other factors.[4] Thyroid cancer is the most serious clinical condition behind thyroid nodule. Thyroid cancer is one of the most common malignancies in Thai women; its prevalence is ranked fourth and its incidence is ranked seventh.[5] Recent reports found that papillary carcinoma incidence increased and follicular carcinoma incidence decreased in Thailand due to higher recognition of microcarcinomas, iodination policy, and changes in histological classification.[6,7] Thyroid FNA is the most widely used diagnostic test for the management of thyroid nodules.

Thyroid cytology is considered as an effective screening and diagnostic tool for rendering a management decision. Thyroid FNA significantly reduces unnecessary surgery, which, in turn, can prevent associated complications and save excessive costs.[8] The procedure can be performed by different practitioners including endocrinologists, surgeons, otolaryngologists, radiologists, and pathologists. Thyroid FNA can be guided manually or with the help of ultrasound imaging.[8] Aspiration of thyroid nodules is safe and rarely causes serious outcomes.[9] The complications may include local infection, hematoma, and vasovagal reflex.[10]

This article represents an overview of the thyroid FNA practice in Thailand. Information was provided by cytopathologists from 10 different institutions around the country via direct interviews and supplemented by available publications from the Thai series and original data.

pISSN 2383-7837
eISSN 2383-7845

HISTORY OF THYROID FINE-NEEDLE ASPIRATION PRACTICE IN THAILAND

It is unclear in exactly what year thyroid FNA practice has started in Thailand. It is known that endocrinologists were the pioneers to experience thyroid FNA in Thailand. Aside from performing the procedure, they were also responsible for interpretation of the cytologic findings. Pathologists started to play a role in this area as interpreters of thyroid cytology after 1986. Initially, it was started by senior pathologists from the Institute of Pathology, a referral pathology laboratory center, and Ramathibodi Hospital, a large university based hospital. At present, almost all cases of thyroid FNA cytology are signed out by certified pathologists. General cytopathology and thyroid cytology are the essential parts of the training program for anatomic pathology residents. In addition, there are few endocrinologists signing out thyroid FNA in academic centers. Currently, there is no specific law forbidding medical doctors from practicing pathology or cytology in Thailand; hence, officially it is not illegal for clinicians to sign out pathology cases. Cytotechnologists are not involved in thyroid FNA since their major role is confined to performing cervical screening on Pap smears.

FINE-NEEDLE ASPIRATION PERFORMERS

FNA procedure is universally performed by clinicians, including endocrinologists, surgeons, radiologists, otolaryngologists, and general practitioners. In academic environment, inexperienced trainees (i.e., residents, young fellows) are frequently responsible to perform FNA, which may contribute to relatively high rates of non-diagnostic results (see below). There are very few pathologists performing this procedure where rapid onsite evaluation can be carried out. The eminent benefit is that, in case of unsatisfactory results, repeat aspiration can be done while the patients are waiting in the aspiration venue, and that preliminary diagnosis may sometimes be provided. Attending clinicians will be able to commence appropriate therapeutic approaches. Thus, patients are more satisfied with the procedure and the waiting time is cut short. However, the disadvantage is that it is time consuming for the pathologists who have to perform the procedure. In general, there is no financial incentive for those who perform FNA in public hospitals. To our knowledge, such services are available only in university based centers by a few dedicated cytopathologists. Core needle biopsy of thyroid nodules is an alternative diagnostic approach performed by some clinicians; however, this practice is very uncommon in Thailand.

PREPARATION OF THYROID CYTOLOGY SAMPLES

The technique of a smear preparation is rather standard. Briefly, after a smear is placed on a glass slide, another slide is laid on top of the sample material, and then each slide is pulled apart to spread the sample into a thin layer. Both slides are submitted for further staining. Wet smears are fixed in 95% ethyl alcohol or with spray fixatives while air-dried smears are left unfixed. The staining methods for thyroid cytology are not much different between each institute. Conventional Papanicolaou stain is used for wet smears, and air dried samples are stained with Diff-Quik method. Liquid-based preparation for thyroid FNA cytology is rarely used in Thailand, because most cytopathologists are not familiar with the technique and are not trained to interpret these cytologic smears. To our knowledge, this method is used only in a few institutions, which are basically private hospitals.

THYROID CYTOLOGY REPORTING SYSTEM

Currently, there is no standard format used by all cytopathologists across Thailand, but the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) is supposed to be the most widely accepted method for reporting thyroid cytopathology. In addition, the Bethesda system is acclaimed by most clinicians and has recently been incorporated into the residency and fellowship training programs in the relevant clinical fields, including otolaryngology, endocrinology, and general surgery. It is also credited in the national guidelines for the diagnosis and treatment of thyroid cancer released by the National Cancer Institute of Thailand.[11]

The Bethesda system has started to be widely recognized locally since 2010 after the well-illustrated explanatory notes were published.[12] Prior to the era of TBSRTC, the cytologic reports of thyroid nodules in Thailand were not standardized and varied significantly among institutions and pathologists. At that time, cytology reporting of thyroid lesions was based on specific diagnosis of the lesions, e.g., hyperplasia, thyroiditis, equivocal, goiter, etc. Nowadays, the Bethesda terminology is consistently employed for indeterminate nodules from the categories III, IV, and V. At the same time, benign and malignant lesions are often signed out with a specific diagnosis. Another different issue is that, contrary to TBSRTC recommendations, aspirates with cystic fluid only were considered as benign by some readers and were signed out accordingly. Otherwise, the Bethesda criteria of adequacy that require six groups of 10 well-visualized follicular cells are widely accepted.[13]

https://doi.org/10.4132/jptm.2017.08.12

QUALITY ASSURANCE

External quality assurance program for Thai pathologists is aimed to standardize their competencies in cytology interpretation. The program is set up and supported by the Thai Society of Cytology (http://www.thaicytology.org/). All enrolled participants are granted certificates upon successful completion. The cases, which are distributed to all participants, consist of cytology cases from different organs/systems including the thyroid gland. So far, there has been no nationwide thyroid FNA cytologic-histological correlation program similar to the recent Korean multi-institutional project.[14] This issue, as a measure of the quality control, is partially resolved in our original series described below.

ANCILLARY TESTS

Ancillary methods for thyroid FNA, e.g., immunocytochemistry and molecular studies, are not routinely used in Thailand. Immunostaining in thyroid pathology, either surgical or cytopathology, is not much used because of its insufficient accuracy in distinguishing benign from malignant follicular-patterned lesions and other limitations.[15,16] Immunocytochemistry on cell blocks is infrequently employed for differential diagnosis of certain malignancies, such as papillary carcinoma (thyroid transcription factor 1 [TTF-1], thyroglobulin), medullary carcinoma (TTF-1, calcitonin, carcinoembryonic antigen), and metastatic adenocarcinoma (cytokeratin 7, CDX2).[17]

Molecular techniques for preoperative evaluation of indeterminate thyroid nodules are currently being increasingly adopted in the United States.[18] Mutation testing is one of the approaches, which potentially can be implemented in our local settings. For example, a presence of the *BRAF*^{V600E} mutation is highly concordant with malignant thyroid nodules.[18] However, several limitations including high cost, limited access to equipment, uncertain regulations, and lack of local studies interfere with potential implementation of the molecular testing in Thailand.[19]

SUMMARY ON FINE-NEEDLE ASPIRATION STUDIES AND UTILITY OF THE BETHESDA SYSTEM IN THAILAND

Three major national academic centers from geographically different parts of the country (King Chulalongkorn Memorial Hospital, Bangkok; Srinagarind Hospital, Khon Kaen; Maharaj Nakorn Hospital, Chiang Mai), where the leading authors are affiliated, were queried to provide their data on thyroid FNA over the applicable time period, 5–6 years. Since the terminology of TBSRTC was not uniformly used in local settings, especially in the early 2010s, we converted all the cytological diagnoses to the Bethesda system categories.[13,20] Category I (non-diagnostic), in addition to initially unsatisfactory smears, included samples signed out as hemorrhagic content, old hemorrhagic content, cyst with hemorrhage, cystic fluid, and no cells. Category II (benign) included FNA diagnosed as nodular goiter, multinodular goiter, adenomatous goiter/nodule, Hashimoto's/lymphocytic thyroiditis, and colloid fluid/cyst. Categories III–V were usually signed out according to the Bethesda terminology. Category VI (malignant) referred to various malignancies according to their histological type, e.g. papillary carcinoma, anaplastic carcinoma, lymphoma, etc. All the patients who underwent thyroid FNA were matched with thyroid surgical pathology database to find their histological follow-up. Surgical pathology reports were analyzed with a special emphasis on malignancy.

In addition, PubMed and Google Scholar were searched for a combination of keywords: "Thailand," "thyroid," "FNA," and "cytology". All available publications in English and Thai languages released since 1990 were reviewed to obtain information on cytological diagnosis and cytological-histological correlations. The same adjustment of cytological diagnosis to the Bethesda terminology as described above was done, where possible. We found 11 publications (including eight in Thai language) from nine institutions published in local journals from 1991 to 2017.[20-30] Currently, there are no published Thai data on thyroid FNA in international journals.

There are several statistical outputs of the Bethesda system. The most acknowledged is a risk of malignancy (ROM), and additional are distribution of fine-needle aspiration cytology samples by the Bethesda category and operation/resection rate, i.e. ratio of surgically excised nodules to all nodules sampled by FNA within a category. Estimated ranges of ROM for the diagnostic categories were provided in the original Bethesda publication.[13] These ranges were further adjusted to the so-called actual ROMs as per numerous original studies and meta-analysis (Table 1).[31-33] Statistical outputs of the Bethesda system serve as a quality control tool for individual institution and demonstrate concordance with or deviation from national and international standards.

Distribution of thyroid FNA diagnoses by the Bethesda system has been rarely addressed in the Thai publications.[20] Our own data based on 7,447 samples from two institutions are presented in Table 2. There was a high variability not only between the hospitals, but also between departments of the same hospital where the smears were signed out. A notable finding was a high

Table 1. Implied and actual ROM according to the Bethesda diagnostic categories

Source	Parameter	Mean % (95% CI)					
		I	II	III	IV	V	VI
Cibas *et al*. (2009)[13]	Estimated ROM	1–4	0–3	5–15	15–30	60–75	97–99
Bongiovanni *et al*. (2012)[31]	Actual ROM, meta-analysis (n=6,362)	16.8	3.7	15.9	26.1	75.2	98.6
Sheffield *et al*. (2014)[32]	Actual ROM, meta-analysis (n=8,044)	18.7 (11–26)	6.5 (5–9)	28.3 (19–38)	33.1 (27–40)	65 (50–79)	98.6 (98–100)
Krauss *et al*. (2016)[33]	Actual ROM, meta-analysis (n=8,214)	12 (9–14)	5 (3–7)	17 (11–23)	25 (20–29)	72 (61–84)	98 (97–99)

ROM, risk of malignancy; CI, confidence interval.

Table 2. Distribution of thyroid FNA diagnoses by the Bethesda system

Hospital	Period years	Thyroid nodules	No. (%)					
			I	II	III	IV	V	VI
KCMH (Department of Pathology), Bangkok[20]	2010–2015	2,762	1,314 (47.6)	1,127 (40.8)	108 (3.9)	72 (2.6)	53 (1.9)	88 (3.2)
KCMH (Department of Endocrinology), Bangkok	2015–2017	1,542	196 (12.7)	1,274 (82.6)	4 (0.3)	37 (2.4)	23 (1.5)	8 (0.5)
Chiang Mai University	2011–2015	3,143	672 (21.4)	2,141 (68.1)	47 (1.5)	87 (2.8)	43 (1.4)	153 (4.9)

FNA, fine-needle aspiration; KCMH, King Chulalongkorn Memorial Hospital.

Table 3. Malignancy rates of the adjusted Bethesda categories by cytological-histological correlation

Hospital	Study cohort	Operated nodules	Risk of malignancy in the Bethesda category (%)					
			I	II	III	IV	V	VI
Srinagarind Hospital, Khon Kaen[21]	1984–1990	101	n/a	11.4	n/a	12.5	n/a	100
Ramathibodi Hospital, Bangkok[22]	1988–1990	77	n/a	8.6	31.3	n/a	n/a	75
Chiang Mai University[23]	1996–1999	230	n/a	2.5	n/a	22.6	n/a	90.2
Chiang Mai University[24]	1992–2000	174	n/a	5.1	n/a	30.6	50	88.2
Chonburi Hospital[25]	1999–2003	97	17.4	9.1	n/a	15.4	33.3	100
Songklanagarind Hospital, Hat Yai[26]	1999–2003	341	n/a	8.5	n/a	23.2	68	73.9
Ratchaburi Hospital[27]	2001–2005	101	n/a	11.4	n/a	n/a	71.4	100
Buddhachinaraj Phitsanulok Hospital[28]	2005–2009	234	14.3	6.8	57.1	6.1	92.9	100
Ramathibodi Hospital, Bangkok[29]	2005–2008	469	28.4	9.5	n/a	42.4	86.7	100
Siriraj Hospital, Bangkok[30]	2002–2004	60	n/a	5.6	n/a	37.5	n/a	100

Modified from Limlunjakorn *et al*. J Med Assoc Thai 2017;100:783-92, with permission of Medical Association of Thailand.[20]
n/a, not available.

rate of non-diagnostic category, which approached almost 50% at the King Chulalongkorn Memorial Hospital (KCMH), Bangkok. This extremely high rate was addressed in our previous study.[20] Briefly, it was attributed to several factors such as unskilled operators (resident physician trainees), absence of ultrasound guidance, and high prevalence of cystic nodules in the local population.[20] Currently, a special training program for residents is implemented to fix this issue. It is interesting that another department at the same hospital (Department of Endocrinology), where the sampling is performed by experienced staff and often with ultrasound guiding and rapid onsite evaluation, had much more acceptable level of non-diagnostic aspirates, 12.7% (Table 2). Cyst fluid only is another contributing factor to the non-diagnostic category.[13,34] Based on our experience at the KCMH, 10% (Department of Pathology) to 40% (Department of Endocrinology) of all non-diagnostic smears were cyst fluid only aspirates.

Benign thyroid FNA samples were the most prevalent among all the diagnostic categories (Table 2), which is concordant with the international experience.[31-33] Collectively, indeterminate categories III–V were 4.2% to 8.4% (Table 2). Resection rates for each diagnostic category based on 5,905 FNA from the KCMH, Bangkok and Chiang Mai University were as follows: 13.3% (category I), 22.0% (category II), 30.3% (category III), 68.6% (category IV), 56.3% (category V), and 50.2% (category VI).

Local experience with thyroid FNA in the pre-Bethesda era is summarized in Table 3. There were only 10 publications in 23 years, which basically had obvious limitations including small sample size, poor design, poor data presentation, etc. There was a high diversity in the terminology used. None of the recent studies published in 2010–2013 adopted the Bethesda system. After adjustment according to TBSRTC criteria, we found that ROM in each category was generally comparable to the range described by the Bethesda system (Table 3). The issue of indeterminate nodules was not well explored, partially due to diverse terminology. To

 https://doi.org/10.4132/jptm.2017.08.12

Table 4. Risk of malignancy per the Bethesda diagnostic category

Hospital	Year	Operated nodules	Risk of malignancy in the Bethesda category, n (%)					
			I	II	III	IV	V	VI
KCMH, Bangkok[20]	2010–2015	457	20/104 (19.2)	29/207 (14)	11/29 (37.9)	9/43 (20.9)	22/27 (81.5)	44/47 (93.6)
Srinagarind Hospital, Khon Kaen	2011–2015	701	60/265 (22.6)	37/243 (15.2)	23/70 (32.9)	14/24 (58.3)	25/32 (78.1)	62/67 (92.5)
Chiang Mai University	2011–2015	859	35/161 (21.7)	76/513 (14.8)	8/18 (44.4)	36/66 (54.5)	19/27 (70.4)	68/74 (91.9)
Mean/Total		2,017	115/530 (21.7)	142/963 (14.7)	42/117 (35.9)	59/133 (44.4)	66/86 (76.7)	174/188 (92.6)

conclude, a majority of existing publications do not adequately represent the Thai experience with thyroid FNA.

To address cytologic-histologic correlations and the ROM for the Bethesda diagnostic categories, we collected a cohort of 2,017 thyroid nodules with surgical follow-up from three institutions (Table 4). Malignant diagnoses on histopathology were mainly papillary carcinoma, followed by follicular and Hurthle cell carcinomas, as well as rare cases of anaplastic, insular, and medullary thyroid carcinomas, thyroid lymphomas and secondary malignancies. Mean ROM for all the categories except for malignant in our cohort was higher than the ROM estimated by TBSRTC or provided by the meta-analysis studies (Table 1). We found particularly high malignancy rate for the categories II (14.7%), III (35.9%), and IV (44.4%) whereas a ROM in malignant category (92.6%) was lower than expected. There may be several reasons which contribute to these deviations, such as interpretation errors, co-existence of dominant benign nodule and cancer, tertiary center bias, etc. In addition, there was a variation among the three institutions. For instance, the low ROM for category IV and high ROM for category V was noted in the KMCH (Table 4). Nevertheless, we believe that our multi-institutional study has established for the first time a reference range of the ROM for the Bethesda diagnostic categories in Thailand.

CONCLUSION

Thyroid FNA is a routine procedure universally performed in Thai hospitals by a variety of clinical specialists. The number of thyroid aspirations is growing, which is reflected by the increasing workload of general and head-neck pathologists and also cytopathologists who basically evaluate cytologic smears. TBSRTC was slowly adopted in local settings, but currently it is actively promoted as a uniform reporting system by the local professional societies. This review, in addition to presenting various aspects of thyroid FNA in Thailand, provides for the first time national references for several statistical outputs of the Bethesda system based on our original multi-institutional cohort. A notable finding was that the ROM in several diagnostic categories (II to IV) was higher than the malignancy risk established by TBSRTC. We endorse the use of uniform terminology of the Bethesda system in local settings, which will help facilitate communication among diverse medical professionals involved in the management of patients with thyroid nodules, and with the international audience.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

Acknowledgments

We wish to acknowledge the contribution of Dr. Panudda Srichomkwun, MD (Division of Endocrinology and Metabolism, Department of Medicine, Faculty of Medicine, Chulalongkorn University).

REFERENCES

1. Martin HE, Ellis EB. Biopsy by needle puncture and aspiration. Ann Surg 1930; 92: 169-81.
2. Diamantis A, Magiorkinis E, Koutselini H. Fine-needle aspiration (FNA) biopsy: historical aspects. Folia Histochem Cytobiol 2009; 47: 191-7.
3. Bibbo M. Comprehensive cytopathology. Philadelphia: Saunders, 1991.
4. Dean DS, Gharib H. Epidemiology of thyroid nodules. Best Pract Res Clin Endocrinol Metab 2008; 22: 901-11.
5. Tangjaturonrasme N, Vatanasapt P, Bychkov A. Epidemiology of head and neck cancer in Thailand. Asia Pac J Clin Oncol 2017 Aug 16 [Epub]. https://doi.org/10.1111/ajco.12757.
6. Bychkov A. A pathologist's perspective on thyroid cancer trends in Thailand. Cancer Epidemiol 2017; 47: 133-4.
7. Bychkov A, Hirokawa M, Jung CK, *et al*. Low rate of noninvasive follicular thyroid neoplasm with papillary-like nuclear features in Asian practice. Thyroid 2017; 27: 983-4.
8. Pisharodi L, Guiter G, Layfield L. Fine-needle aspiration biopsy. In:

Gnepp DR, ed. Diagnostic surgical pathology of the head and neck. 2nd ed. Philadelphia: Saunders Elsevier, 2001; 1069-130.
9. Sanchez MA, Stahl RE. The thyroid, parathyroid, and neck masses other than lymph nodes. In: Koss LG, Melamed MR, eds. Koss' diagnostic cytology and its histopathologic bases. 5th ed. Philadelphia: Lippincott Williams & Wilkins, 2006; 1148-85.
10. Busseniers AE, Silver SA. Fine-needle aspiration cytology of the thyroid. In: Oertli D, Udelsman R, eds. Surgery of the thyroid and parathyroid glands. Berlin Heidelberg: Springer-Verlag, 2007; 61-80.
11. Chindavijak S, Panchan V, Chaiwerawattana A, Imsamran W. Guidelines for the diagnosis and treatment of thyroid cancer. Bangkok: Kosit Press Co., Ltd., 2015; 114.
12. Ali SZ, Cibas E. The Bethesda System for Reporting Thyroid Cytopathology: definitions, criteria, and explanatory notes. New York: Springer, 2010.
13. Cibas ES, Ali SZ; NCI Thyroid FNA State of the Science Conference. The Bethesda System For Reporting Thyroid Cytopathology. Am J Clin Pathol 2009; 132: 658-65.
14. Kim M, Park HJ, Min HS, *et al*. The use of the Bethesda System for Reporting Thyroid Cytopathology in Korea: a nationwide multicenter survey by the Korean Society of Endocrine Pathologists. J Pathol Transl Med 2017; 51: 410-7.
15. Bychkov A, Sampatanukul P, Shuangshoti S, Keelawat S. TROP-2 immunohistochemistry: a highly accurate method in the differential diagnosis of papillary thyroid carcinoma. Pathology 2016; 48: 425-33.
16. Rosai J, Kuhn E, Carcangiu ML. Pitfalls in thyroid tumour pathology. Histopathology 2006; 49: 107-20.
17. Rottuntikarn W, Wangsiricharoen S, Rangdaeng S. Cytomorphology and immunocytochemistry of columnar cell variant of papillary thyroid carcinoma. Cytopathology 2017; 28: 338-41.
18. Nikiforov YE. Role of molecular markers in thyroid nodule management: then and now. Endocr Pract 2017; 23: 979-88.
19. Bychkov A. Prevalence of *BRAF*V600E mutation in Asian patients with thyroid cancer. Malays J Pathol 2017; 39: 95-6.
20. Limlunjakorn P, Keelawat S, Bychkov A. Evaluation of thyroid fine needle aspiration cytology by the Bethesda reporting system: a retrospective analysis of rates and outcomes from the King Chulalongkorn Memorial Hospital. J Med Assoc Thai 2017; 100: 783-92.
21. Himakoun L, Kanjanavirojkul N, Vatanasapt V, Jaroensiri DJ, Aranyasen O. Fine needle aspiration cytologic diagnosis of the thyroid nodule in Srinagarind Hospital. Srinagarind Hosp Med J 1991; 6: 243-51.
22. Issarapan P, Kulapaditharom B, Chiemchanya S. Diagnostic value of fine needle aspiration cytology in thyroid nodule: experience in the ENT department, Ramathibodi Hospital. Thai J Otolaryngol Head Neck Surg 1991; 6: 133-9.
23. Settakorn J, Chaiwun B, Thamprasert K, Wisedmongkol W, Rangdaeng S. Fine needle aspiration of the thyroid gland. J Med Assoc Thai 2001; 84: 1401-6.
24. Pattarasakulchai T, Sittitrai P, Mahanupab P. Fine needle aspiration biopsy (FNAB) in management of thyroid nodules. Chiang Mai Med Bull 2002; 41: 51-7.
25. Pradittaphonlert V. Fine needle aspiration biopsy (FNAB) of thyroid nodule at Chonburi Hospital. Chonburi Hosp J 2004; 29: 75-82.
26. Sinkijcharoenchai W, Pruegsanusak K, Jantharapattana K. Management of thyroid nodules in Department of Otolaryngology. Songklanagarind Med J 2005; 23: 303-11.
27. Nopkunwijai A. Fine needle aspiration cytology (FNAC) of thyroid nodule in Ratchaburi Hospital. Region 6-7 Med J 2006; 25: 211-7.
28. Eamchan P. Diagnosis of thyroid nodules by fine needle aspiration biopsy in Buddhachinaraj Phitsanulok Hospital. Buddhachinaraj Med J 2010; 27: 170-6.
29. Himakhun W, Chansom R, Aroonroch R, Pongtippan A. Fine needle aspiration of thyroid: a cyto-histopathological correlation in Ramathibodi Hospital. J Med Assoc Thai 2012; 95 Suppl 1: S74-8.
30. Boonyaarunnate T, Amornpichetkul K. Advantage of frozen section in management of thyroid nodule with preoperative fine needle aspiration in Siriraj Hospital, Bangkok, Thailand. Asian Arch Pathol 2013; 9: 57-63.
31. Bongiovanni M, Spitale A, Faquin WC, Mazzucchelli L, Baloch ZW. The Bethesda System for Reporting Thyroid Cytopathology: a meta-analysis. Acta Cytol 2012; 56: 333-9.
32. Sheffield BS, Masoudi H, Walker B, Wiseman SM. Preoperative diagnosis of thyroid nodules using the Bethesda System for Reporting Thyroid Cytopathology: a comprehensive review and meta-analysis. Expert Rev Endocrinol Metab 2014; 9: 97-110.
33. Krauss EA, Mahon M, Fede JM, Zhang L. Application of the Bethesda classification for thyroid fine-needle aspiration: institutional experience and meta-analysis. Arch Pathol Lab Med 2016; 140: 1121-31.
34. Takada N, Hirokawa M, Suzuki A, Higuchi M, Kuma S, Miyauchi A. Reappraisal of "cyst fluid only" on thyroid fine-needle aspiration cytology. Endocr J 2017; 64: 759-65.

Journal of Pathology and Translational Medicine 2017; 51: 571-578
https://doi.org/10.4132/jptm.2017.10.19

■ REVIEW ■

The Use of Fine-Needle Aspiration (FNA) Cytology in Patients with Thyroid Nodules in Asia: A Brief Overview of Studies from the Working Group of Asian Thyroid FNA Cytology

Chan Kwon Jung · SoonWon Hong[1]
Andrey Bychkov[2] · Kennichi Kakudo[3]

Department of Hospital Pathology, Seoul St. Mary's Hospital, College of Medicine, The Catholic University of Korea, Seoul; [1]Department of Pathology, Gangnam Severance Hospital, Yonsei University College of Medicine, Seoul, Korea; [2]Department of Pathology, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand; [3]Department of Pathology, Nara Hospital, Kindai University Faculty of Medicine, Nara, Japan

Received: October 12, 2017
Accepted: October 16, 2017

Corresponding Author
Chan Kwon Jung, MD
Department of Hospital Pathology, Seoul St. Mary's Hospital, College of Medicine, The Catholic University of Korea, 222 Banpo-daero, Seocho-gu, Seoul 06591, Korea
Tel: +82-2-2258-1622
Fax: +82-2-2258-1627
E-mail: ckjung@catholic.ac.kr

Ultrasound-guided fine-needle aspiration (FNA) cytology is the most widely used screening and diagnostic method for thyroid nodules. Although Western guidelines for managing thyroid nodules and the Bethesda System for Reporting Thyroid Cytopathology are widely available throughout Asia, the clinical practices in Asia vary from those of Western countries. Accordingly, the Working Group of Asian Thyroid FNA Cytology encouraged group members to publish their works jointly with the same topic. The articles in this special issue focused on the history of thyroid FNA, FNA performers and interpreters, training programs of cytopathologists and cytotechnicians, staining methods, the reporting system of thyroid FNA, quality assurance programs, ancillary testing, and literature review of their own country's products. Herein, we provide a brief overview of thyroid FNA practices in China, India, Japan, Korea, the Philippines, Taiwan, and Thailand.

Key Words: Thyroid; Cytology; Fine-needle aspiration; Asia; History; Methods; Survey

Fine-needle aspiration (FNA) cytology has been widely accepted as a safe, cost-effective, and accurate tool for the preoperative diagnosis of thyroid nodules. In the past, aspirations were performed only with the manual aid. Since FNA under ultrasound guidance proved to be more accurate for the detection of thyroid cancer, FNA should be performed under ultrasound guidance using a 23-, 25-, or 27-gauge needle for cytological evaluation.[1,2] The wide use of FNA cytology for thyroid nodules has significantly decreased the rate of unnecessary surgery for benign thyroid nodules over the last three decades.[1,3]

Although North American and European guidelines for managing thyroid nodules and the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) are available throughout Asia, the clinical practices in Asia vary from those of Western countries in terms of disease incidence, diagnostic methods, availability of diagnostic tests, conservative management approach, national health insurance system, and governmental regulations on health care. Moreover, there is considerable variation among Asian countries due to the different rates of economic development and kinds of healthcare systems. Asian countries have increasingly reported their experiences of FNA of thyroid nodules using TBSRTC. Despite these efforts, Asian data on thyroid FNA have not been very well-organized so far. Accordingly, the Working Group of Asian Thyroid FNA Cytology established in 2016 has encouraged group members to publish their work jointly.[4]

In this special issue, seven articles from China, India, Japan, Korea, the Philippines, Taiwan, and Thailand jointly focused on the same topic regarding the history of thyroid FNA, FNA performers and interpreters, the training programs of cytopathologists and cytotechnicians, staining methods, the reporting system of thyroid FNA, quality assurance programs, and ancillary testing, added by the comprehensive review of publications released

pISSN 2383-7837
eISSN 2383-7845

http://jpatholtm.org/

Table 1. A brief overview of the history of thyroid FNA cytology in Asian countries

	China	India	Japan	Korea	Philippines	Taiwan	Thailand
1950s	1950s: Introduction of cytology	-	1952: Introduction of thyroid FNA	-	-	-	-
1960s	-	1965: First attempt of needle biopsy of thyroid	1962: Japanese Society of Clinical Cytology was founded.	-	-	-	-
1970s	1970-1980s: FNA was applied to thyroid. 1972: First Chinese FNA book-Atlas of clinical cytology	1970: Indian Academy of Cytologists 1975: First publication on FNA by Gupta *et al*.	1972: First report of thyroid FNA cytology by Toriya, Ito Hospital	1977: Thyroid FNA was introduced by a physician. Korean Thyroid Study group was founded.	-	1979: Tien-Chun Chang, an endocrinologist at National Taiwan University Hospital, started thyroid FNA	-
1980s	1985: Chinese Academy of Cytology was founded and the first National Clinical Cytology Conference was held.	1987: First paper on thyroid FNA cytology by Rege *et al*.	-	1981: Cytology training program for pathologists and cytotechnicians 1986: The Korean Society for Cytopathology was founded.	Late 1980s: Thyroid FNA started at the Philippine General Hospital. 1987: Aspiration cytology unit was established in the Department of Pathology, University of the Philippines.	1981: First article on thyroid FNA was published in a local journal by Tien-Chun Chang. 1988: Taiwan Society of Clinical Cytology was founded. 1989: Articles on thyroid FNA were published in international journals.	Endocrinologists started experience with thyroid. 1986: Pathologists started to interpret thyroid FNA.
1990s	1990s: Some of the hospitals started US-guided thyroid FNA.	-	1990s: Wide introduction of US-guidance for thyroid FNA	1996: Cytology proficiency testing has been performed since 1996.	1990s: Private hospitals in the Metro Manila started practice of thyroid FNA.	1995: First color atlas of thyroid and parathyroid cytology	-
2000s	2007: Cytology Operational Manual and Quality Control Standards were proposed by the Cytology Section of the Chinese Pathology Association.	-	-	2006: Korean management guidelines for patients with thyroid nodules and thyroid cancer 2007: Korean Endocrine Pathology Study Group was founded. 2008: Korean Thyroid Association (KTA) was founded.	-	-	-
2010s	-	2011: Endocrine Society of India management guidelines for patients with thyroid nodules	2013: The Japan Thyroid Association Guidelines for the management of thyroid nodules	2010: Revised KTA management guidelines 2016: 2016 Revised KTA management guidelines	2010: Radiologists, endocrinologists, and cytopathologists started US-guided thyroid FNA.	-	2000: Thai Society of Cytology was founded. 2015: Guidelines for the diagnosis and treatment of thyroid cancer

FNA, fine-needle aspiration; US, ultrasound.

https://doi.org/10.4132/jptm.2017.10.19

from the individual countries.[5-11] Herein, we provide a brief overview of contemporary thyroid FNA practices based on the review articles from seven Asian countries.

HISTORICAL ASPECTS OF THYROID FINE-NEEDLE ASPIRATION

In the late 1920s, Hayes Martin and Edward Ellis performed aspiration biopsies using an 18-gauge needle for the cytological evaluation of thyroid lesions in the Memorial Hospital of New York.[12] In 1952, thyroid aspiration cytology using a fine needle (diameter of 0.4–0.8 mm) was introduced by Nils Söderström in Sweden.[13] Thyroid FNA was used in routine practice as an accurate test for distinguishing between benign and malignant thyroid nodules in Sweden since the 1950s. In the United States, FNA was not successfully used for the diagnosis of thyroid nodules before 1970s because of the clinician's preference for surgical biopsies, a lack of familiarity with the FNA procedure, and concerns about tumor seeding along the needle tract.[14,15] After that time, thyroid FNA was reintroduced in the United States and became widely available in the 1980s.[14]

In Asia, thyroid FNA was introduced in China and Japan in the 1950s.[9,10] In Korea, India, and Taiwan, thyroid FNA was introduced in the 1970s.[5-7,11] Table 1 summarizes the brief history of thyroid FNA in seven Asian countries.[5-11]

PERFORMERS AND INTERPRETERS OF THYROID FINE-NEEDLE ASPIRATION CYTOLOGY

Since thyroid FNA was initially introduced by clinicians in most countries, interpretation of FNA cytology were mostly done by clinicians in the past, including endocrinologists, surgeons, and radiologists. In recent years, thyroid FNA has been performed under ultrasound guidance by clinicians in Japan, Korea, the Philippines, Taiwan, and Thailand. Chinese clinicians prefer an intraoperative frozen section rather than FNA for the diagnosis of thyroid nodules.[9] Thyroid FNA is more frequently performed through palpation rather than ultrasound in India, Thailand, and the Philippines because of limited or delayed access to sonography.[5,8,11] Table 2 summarizes the current practices of thyroid FNA performers and interpreters in seven Asian countries.[5-11]

STAINING METHODS OF THYROID FINE-NEEDLE ASPIRATION CYTOLOGY SAMPLES

The most widely used staining method for thyroid FNA specimens was Papanicolaou stain. Hematoxylin and eosin stain was favored by most Chinese pathologists.[9] In India and Thailand, thyroid FNA samples were stained with a combination of two classical stains: alcohol-fixed smears were stained with Papani-

Table 2. Thyroid FNA performer and interpreter

Country	Sampling	Interpretation
China	Primarily performed in endocrinology department in 1970–80s. After 1987, thyroid FNA began to be popular in the pathology department. Thyroid FNA is not yet well accepted in China. Most general hospital use frozen section as a diagnostic method instead of thyroid FNA.	Primarily performed in endocrinology department in 1970–80s. After 1987, thyroid FNA began to be interpreted mainly by pathologists.
India	Blind, palpation-guided FNAs performed by cytopathologists US-guided FNA performed by clinicians or radiologists Palpation-guided FNA appears to be the most commonly used technique.	Interpretation done by pathologists Rapid on-site evaluation done in few academic institutions
Japan	US-guided FNA usually performed by clinicians	Pathologists and clinicians with a board certification in cytopathology
Korea	US-guided FNA usually performed by clinicians	Pathologists (cytopathologists) only interpret the thyroid FNA.
Philippines	Thyroid FNA procedure under US-guidance is performed by pathologists and clinicians.	Majority of pathologists report the diagnosis of cytology. All interpretations are rendered by the pathologist.
Taiwan	Radiologists are the major performer of thyroid FNA. US is used in most cases.	Pathologists are the main diagnostician. Before 1995, clinicians used to be both the performer and the interpreter of thyroid FNA.
Thailand	FNA procedure is universally performed by clinicians. In academic environment, trainees are frequently responsible to perform FNA.	Almost all cases of thyroid FNA cytology are signed out by certified pathologists. Cytotechnologists are not involved in thyroid FNA. Few endocrinologists sign out thyroid FNA in academic centers. Rapid on-site evaluation is rarely performed.

FNA, fine-needle aspiration; US, ultrasound.

colaou stain and air-dried smears were stained with modified Giemsa stain (e.g., May-Grünwald-Giemsa stain or Diff-Quik stain).[5,8] Table 3 summarizes the staining methods for thyroid FNA cytology specimens.[5-11]

REPORTING SYSTEM OF THYROID FINE-NEEDLE ASPIRATION CYTOLOGY

The reporting system of thyroid FNA cytology has improved significantly over the past 10 years with the introduction of TBSRTC.[1,3] TBSRTC consists of six diagnostic categories in order to facilitate communication among cytopathologists and their clinical colleagues and to provide the risk of malignancy for each diagnostic category.[3] After the introduction of TBSRTC, the system has been most widely accepted in China, India, Korea, the Philippines, and Thailand.[5,6,8,9] Other reporting systems for thyroid FNA cytology used in Asia were the General Rules for the Description of Thyroid Cancer by the Japanese Society of Thyroid Surgery, the Japanese System for Thyroid FNA Cytology by the Japan Thyroid Association, and the 6-tier System of Taiwan.[7,10] Table 4 summarized the reporting system of thyroid FNA cytology before and after the introduction of TBSRTC.[5-11]

Table 3. Staining methods for thyroid fine-needle aspiration cytology specimens

Country	Staining method
China	Wrights staining is popular in endocrine and clinical laboratory department. H&E stain is common in pathology department.
India	Combination of Romanowsky (May-Grünwald-Giemsa stain) and Papanicolaou stains is most widely used. H&E stain in few institutions
Japan	Papanicolaou stain is the most widely used. Giemsa stain or Diff-Quik stain
Korea	Papanicolaou stain is the most widely used. H&E or Giemsa stain are used in some institutions.
Philippines	Papanicolaou stain Diff-Quik stain H&E stain in cell blocks
Taiwan	Papanicolaou stain Liu stain
Thailand	Combination of Papanicolaou and Diff-Quik stains are most widely used.

H&E, hematoxylin and eosin.

NON-DIAGNOSTIC THYROID FINE-NEEDLE ASPIRATION

Although ultrasound-guided thyroid FNA has high sensitivity and specificity in distinguishing benign from malignant thyroid nodules, in 1%–40% of cases, thyroid FNA is insufficient for diagnosis and is categorized as non-diagnostic according to TBSRTC.[16] The non-diagnostic FNA cytology by TBSRTC includes virtually acellular specimens (requiring the presence of at least six groups of well-visualized follicular cells with each group containing at least 10 well-preserved epithelial cells), cystic fluid only, and other specimens (obscuring blood, crushed artifacts, poor clotting artifacts, air drying artifacts, overly thick smears, etc.).[3] In the Japanese system, thyroid FNA with "cystic fluid only" is classified as benign rather than non-diagnostic.[10,17] In Taiwan, a paucicellular specimen with fewer than six groups of ten benign follicular cells is considered benign if it contains more than 50 follicular cells in total or consists of degenerative hemorrhagic cyst fluid and scant benign follicular cells.[7]

Table 5 summarizes the criteria and rate of non-diagnostic thyroid FNA.[5-11] A high rate of non-diagnostic aspirates was reported from several teaching hospitals in the Philippines and Thailand, which was linked to the training activity of unskilled residents and limited access to ultrasound guidance.[8,11]

Table 4. Reporting system of thyroid FNA cytology

Country	Before TBSRTC	After TBSRTC
China	No data	TBSRTC is the most widely accepted.
India	No data	TBSRTC is the most widely used.
Japan	General Rules for the Description of Thyroid Cancer (GRDTC): adapted from the 1996 Papanicolaou Society recommendations; published by the Japanese Society of Thyroid Surgery in 2005 and updated in 2006	GRDTC system is widely used. Japanese system for thyroid FNA cytology published by the Japan Thyroid Association (JTA) in 2013: used in several high-volume thyroid surgery centers TBSRTC is rarely used.
Korea	Not standardized and varied, but mostly followed guidelines of the Papanicolaou Society of Cytopathology	TBSRTC is the most widely accepted.
Philippines	Based on histopathologic terminology of thyroid disorder	TBSRTC is the most widely used.
Taiwan	All investigators used different diagnostic categories.	TBSRTC or the 6-tier system corresponding to each Bethesda category
Thailand	Not standardized and varied, e.g., thyroid FNA reporting was based on specific diagnosis of the lesions.	TBSRTC is the most widely accepted.

FNA, fine-needle aspiration; TBSRTC, The Bethesda System for Reporting Thyroid Cytopathology.

https://doi.org/10.4132/jptm.2017.10.19

LIQUID-BASED CYTOLOGY AND ANCILLARY TESTS

In Korea, the use of liquid-based cytology in thyroid FNA was adopted in 2008 and became widely used since 2010.[6,18] In Taiwan, liquid-based cytology in thyroid FNA was first introduced in 2014 and then became commonly used.[7] However, liquid-based cytology has not been made widely available for thyroid FNA in other countries.[5,8-11]

Core needle biopsy as an alternative to thyroid FNA has been used mainly in Korea whereas in other countries, this biopsy is performed only in a few institutions.[5-10]

Immunocytochemistry generally has limited applications for the diagnosis of thyroid FNA in Asian countries. Although molecular testing has been useful for the diagnosis of indeterminate thyroid FNA, it is often not practical for most clinical laboratories and is generally not covered by health insurance in Asia.[5-11]

Table 6 summarizes the ancillary tests in thyroid FNA cytology.[5-11]

Table 5. Non-diagnostic thyroid FNA

Country	Criteria for non-diagnostic FNA	Incidence of non-diagnostic FNA
China	TBSRTC	3.6% at one institution
India	TBSRTC Different criteria in a study: 10 clusters are needed with each having more than 20 cells; in case of presence of tissue fragments, minimum number of fragments required is 8. Royal College of Pathologists guidelines in one study	7.4% (0.5%–25.7%) from 38 studies
Japan	General Rules for the Description of Thyroid Cancer system Japanese system	10% according to the Japanese system
Korea	TBSRTC	12.4% (0%–32.6%) from 12 institutions
Philippines	TBRSTC	1.3% and 23.1% from 2 studies
Taiwan	Variable but different from TBRSTC[a]	8% at one institution
Thailand	TBSRTC	12.7%–47.6% from three institutions

FNA, fine-needle aspiration; TBSRTC, the Bethesda System for Reporting Thyroid Cytopathology.
[a]Most Taiwan pathologists consider that the specimen is negative, but not non-diagnostic when there are less than six groups but more than 50 follicular cells in total or a degenerative hemorrhagic cyst with scant benign follicular cells.

Table 6. Ancillary tests in thyroid FNA cytology

Country	Liquid-based cytology	Core needle biopsy	Immunocytochemistry	Molecular or other testing
China	Not commonly used	No data	No data	Amplification refractory mutation system for *BRAF* V600E is the most popular technique. Next generation sequencing is not well accepted.
India	Has been used in some institutions as addition to conventional smears	Limited applicability and acceptability	Limited applicability and acceptability	Limited applicability and acceptability
Japan	Used in some laboratories, but is not widely available	Rarely performed	No data	*BRAF* testing is uncommon practice and not covered by national health insurance system. Thyroglobulin and/or calcitonin in FNA needle washings is often used in thyroid or lymph node aspirates.
Korea	Became popular since 2010 Used in 68% institutions in 2016	Widely used	Not routinely used but can be applied in specific cases	*BRAF* testing is used. Thyroglobulin and/or calcitonin in FNA needle washings is often used in thyroid or lymph node aspirates.
Philippines	Not used	No data	Rarely performed	Not covered by health care insurance Referred to outsource/abroad facilities if patients agree to pay
Taiwan	Became popular since 2014	Rarely performed	In some institutions, immunocytochemical staining is used.	In some institutions, molecular testing is used.
Thailand	Rarely used	Very uncommon	Available, but rarely performed	Rarely used due to limited availability

FNA, fine-needle aspiration.

TRAINING PROGRAM

Asian pathologists receive a certification in pathology and cytopathology after completing residency training and passing board examinations, and usually practice both surgical pathology and cytopathology. Training programs for cytotechnicians have been well organized in Japan, Korea, and Taiwan. Certified cytotechnicians screen thyroid FNA cytology, but are not eligible to declare a final diagnosis without supervision by a pathologist. Table 7 summarizes the training programs for cytopathologists and cytotechnicians.[5-11]

QUALITY CONTROL AND QUALITY ASSURANCE PRACTICES

Quality control in cytology includes all activities to improve the performance of the test from the time of specimen collection until the cytology report is completed. Quality assurance defined by the College of American Pathologists includes quality review activities and systematic monitoring of quality control results to provide confidence that all quality control systems are functioning properly and quality requirements are fulfilled.[19] Quality control materials in thyroid FNA should include the distribution of each diagnostic category, histologic outcomes of FNA diagnostic categories, rate of surgical follow-up, and risk of malignancy calculated using the total number of each diagnostic category with and without surgical follow-up.

Quality improvement programs in Asian countries are organized by local societies of cytology and/or pathology. Table 8 summarizes the quality assurance and quality control programs in thyroid FNA cytology.[5-11]

CONCLUSION

The purpose of the recently established Working Group of Asian Thyroid FNA Cytology is to promote communication and share practices among pathologists, cytopathologists, and clinicians dealing with thyroid FNA in Asia. In this special issue, we presented for the first time a single volume collection of contemporary reviews on Asian practices of thyroid FNA. Despite

Table 7. Training programs for cytopathologists and cytotechnicians

Country	Cytopathologist	Cytotechnician
China	No data	No data
India	No data	Indian Academy of Cytologists conducts exam for cytotechnicians and cytotechnologists. Few centers run cytotechnician and cytotechnologist training programs for certification. Only limited institutions have cytoscreeners.
Japan	Pathologists have dual boards of anatomical pathology and cytopathology. Clinicians also have board of cytopathology.	JSCC certification Candidate 1: medical technologist after 3-year course at a vocational school or 4-year course at a medical technology school, 1-year work experience requirements at the cytology laboratory of a teaching hospital Candidate 2: 4-year college graduate
Korea	Pathology residents must pass the exam in both fields of surgical pathology and cytolopathology to get the pathology board. For board certificated pathologists, there is annual requirement for continuing cytology education activities.	Nationwide cytotechnician education program began under the auspices of the World Health Organization in 1981. After 2-year pathology or cytology laboratory practice as a technician, 1-year training program at a National Cancer Center and certification exam
Philippines	Formal cytopathology training programs are not offered. Conferences in cytopathology are embedded in the training program of anatomic pathology.	No training programs
Taiwan	At least 3 months of cytology screening and sign-out practice Pathology residents are required to attend a 2-week intensive course. Pathology residents should pass both exams of surgical pathology and cytolopathology to get the pathology board. For board certificated pathologists, there is annual requirement for continuing cytology education activities.	One-year on-site training at a qualified training institution and a final exam There are 12 qualified institutions for cytotechnologist training in Taiwan.
Thailand	General cytopathology and thyroid cytology are the essential parts of the training program for anatomic pathology residents.	No data

JSCC, Japanese Society of Clinical Cytology.

https://doi.org/10.4132/jptm.2017.10.19

Table 8. Quality assurance and quality control programs in thyroid FNA cytology

Country	Internal program	Nationwide external program
China	No data	No data
India	No data	External Quality Assurance Programme of the Indian Academy of Cytologists Only straightforward diagnose are assessed for thyroid FNA quality control.
Japan	No data	No data
Korea	Accuracy assessment by cyto-histological correlations Annual reports on quality control of thyroid FNA have been published since 1996.	Cytology proficiency testing in the Korean Society for Cytopathology has been performed since 1996. National quality control program in the Korean Society of Pathologists began in 1999.
Philippines	Self-review Intradepartmental referral to another pathologist All thyroid surgeries with previous FNA done in the same hospital are reviewed.	In the process of developing an external quality assurance program in cytopathology
Taiwan	No data	Currently there is no authoritative quality assurance program for external evaluation.
Thailand	No data	External quality assurance program for Thai pathologists is set up and supported by the Thai Society of Cytology. No nationwide thyroid FNA cytologic-histological correlation program

FNA, fine-needle aspiration.

most countries adopting Western systems and guidelines and incorporated them into their national systems early, there remains local variation which should be considered when doing comparisons between Asian and Western countries, and among Asian countries. We hope that these reports from Asia will encourage further studies on thyroid FNA cytology to improve the diagnosis of thyroid nodules and subsequently provide optimal care for patients with thyroid nodules in Asia.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

Acknowledgments

We would like to acknowledge all the members of the Working Group of Asian Thyroid FNA Cytology. This work was supported by a grant (HI16C2013) from the Korean Health Technology R&D Project, Ministry of Health & Welfare, Republic of Korea.

REFERENCES

1. Haugen BR, Alexander EK, Bible KC, *et al*. 2015 American Thyroid Association management guidelines for adult patients with thyroid nodules and differentiated thyroid cancer: the American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer. Thyroid 2016; 26: 1-133.
2. Paschke R, Cantara S, Crescenzi A, Jarzab B, Musholt TJ, Sobrinho Simoes M. European Thyroid Association guidelines regarding thyroid nodule molecular fine-needle aspiration cytology diagnostics. Eur Thyroid J 2017; 6: 115-29.
3. Cibas ES, Ali SZ; NCI Thyroid FNA State of the Science Conference. The Bethesda System For Reporting Thyroid Cytopathology. Am J Clin Pathol 2009; 132: 658-65.
4. Bychkov A, Kakudo K, Hong S. Current practices of thyroid fine-needle aspiration in Asia: a missing voice. J Pathol Transl Med 2017; 51: 517-20.
5. Agarwal S, Jain D. Thyroid cytology in India: contemporary review and meta-analysis. J Pathol Transl Med 2017; 51: 533-47.
6. Cha YJ, Pyo JY, Hong S, *et al*. Thyroid fine-needle aspiration cytology practice in Korea. J Pathol Transl Med 2017; 51: 521-7.
7. Hang JF, Hsu CY, Lai CR. Thyroid fine-needle aspiration in Taiwan: the history and current practice. J Pathol Transl Med 2017; 51: 560-4.
8. Keelawat S, Rangdaeng S, Koonmee S, Jitpasutham T, Bychkov A. Current status of thyroid fine-needle aspiration practice in Thailand. J Pathol Transl Med 2017; 51: 565-70.
9. Liu Z, Liu D, Ma B, *et al*. History and practice of thyroid fine-needle aspiration in China, based on retrospective study of the practice in Shandong University Qilu Hospital. J Pathol Transl Med 2017; 51: 528-32.
10. Satoh S, Yamashita H, Kakudo K. Thyroid cytology: the Japanese system and experience at Yamashita Thyroid Hospital. J Pathol Transl Med 2017; 51: 548-54.
11. Abelardo AD. Thyroid fine-needle aspiration practice in the Philippines. J Pathol Transl Med 2017; 51: 555-9.
12. Martin HE, Ellis EB. Biopsy by needle puncture and aspiration. Ann Surg 1930; 92: 169-81.
13. Soderstrom N. Puncture of goiters for aspiration biopsy. Acta Med

Scand 1952; 144: 237-44.
14. Cramer H. Fine-needle aspiration cytology of the thyroid: an appraisal. Cancer 2000; 90: 325-9.
15. Diamantis A, Magiorkinis E, Koutselini H. Fine-needle aspiration (FNA) biopsy: historical aspects. Folia Histochem Cytobiol 2009; 47: 191-7.
16. Lee SH, Kim MH, Bae JS, Lim DJ, Jung SL, Jung CK. Clinical outcomes in patients with non-diagnostic thyroid fine needle aspiration cytology: usefulness of the thyroid core needle biopsy. Ann Surg Oncol 2014; 21: 1870-7.
17. Takada N, Hirokawa M, Suzuki A, Higuchi M, Kuma S, Miyauchi A. Reappraisal of "cyst fluid only" on thyroid fine-needle aspiration cytology. Endocr J 2017; 64: 759-65.
18. Jung CK, Lee A, Jung ES, Choi YJ, Jung SL, Lee KY. Split sample comparison of a liquid-based method and conventional smears in thyroid fine needle aspiration. Acta Cytol 2008; 52: 313-9.
19. Medicare, Medicaid and CLIA programs: regulations implementing the Clinical Laboratory Improvement Amendments of 1988 (CLIA)-HCFA: final rule with comment period. Fed Regist 1992; 57: 7002-186.

https://doi.org/10.4132/jptm.2017.10.19

Journal of Pathology and Translational Medicine 2017; 51: 579-587
https://doi.org/10.4132/jptm.2017.08.11

■ ORIGINAL ARTICLE ■

Current Cytology Practices in Korea: A Nationwide Survey by the Korean Society for Cytopathology

Eun Ji Oh[1] · Chan Kwon Jung[1,2] Dong-Hoon Kim[3] · Han Kyeom Kim[4] Wan Seop Kim[5] · So-Young Jin[6] Hye Kyoung Yoon[7] · The Fellowship Council and Committee of Quality Improvement of the Korean Society for Cytopathology

[1]Department of Hospital Pathology, [2]Cancer Research Institute, College of Medicine, The Catholic University of Korea, Seoul; [3]Department of Pathology, Kangbuk Samsung Hospital, Sungkyunkwan University School of Medicine, Seoul; [4]Department of Pathology, Korea University Guro Hospital, Korea University College of Medicine, Seoul; [5]Department of Pathology, Konkuk University School of Medicine, Seoul; [6]Department of Pathology, Soonchunhyang University Seoul Hospital, Soonchunhyang University College of Medicine, Seoul; [7]Department of Pathology, Inje University Busan Paik Hospital, Inje University College of Medicine, Busan, Korea

Received: July 4, 2017
Revised: August 7, 2017
Accepted: August 10, 2017

Corresponding Author
Chan Kwon Jung, MD, PhD
Department of Hospital Pathology, Seoul St. Mary's Hospital, College of Medicine, The Catholic University of Korea, 222 Banpo-daero, Seocho-gu, Seoul 06591, Korea
Tel: +82-2-2258-1622
Fax: +82-2-2258-1627
E-mail: ckjung@catholic.ac.kr

Background: Limited data are available on the current status of cytology practices in Korea. This nationwide study presents Korean cytology statistics from 2015. **Methods:** A nationwide survey was conducted in 2016 as a part of the mandatory quality-control program by the Korean Society for Cytopathology. The questionnaire was sent to 208 medical institutions performing cytopathologic examinations in Korea. Individual institutions were asked to submit their annual cytology statistical reports and gynecologic cytology-histology correlation data for 2015. **Results:** Responses were obtained from 206 medical institutions including 83 university hospitals, 87 general hospitals, and 36 commercial laboratories. A total of 8,284,952 cytologic examinations were performed in 2015, primarily in commercial laboratories (74.9%). The most common cytology specimens were gynecologic samples (81.3%). Conventional smears and liquid-based cytology were performed in 6,190,526 (74.7%) and 2,094,426 (25.3%) cases, respectively. The overall diagnostic concordance rate between cytologic and histologic diagnoses of uterine cervical samples was 70.5%. Discordant cases were classified into three categories: category A (minimal clinical impact, 17.4%), category B (moderate clinical impact, 10.2%), and category C (major clinical impact, 1.9%). The ratio of atypical squamous cells of undetermined significance to squamous intraepithelial lesion was 1.6 in university hospitals, 2.9 in general hospitals, and 4.9 in commercial laboratories. **Conclusions:** This survey reveals the current status and trend of cytology practices in Korea. The results of this study can serve as basic data for the establishment of nationwide cytopathology policies and quality improvement guidelines in Korean medical institutions.

Key Words: Cytology; Statistics; Surveys; Quality; Accuracy

The number of cytology cases has been increasing over time in Korea.[1] In 1988, the Papanicolaou (Pap) smear was first introduced to screen for cervical cancer during regular health check-ups for Korean industrial workers and their family members.[2] The National Cancer Screening Program for stomach, breast and cervical cancer began in 1999. Over time, participation in this program has increased as both the target population and the cancer types included have expanded.[2] Recent guidelines for cervical cancer screening in Korea recommend that all asymptomatic women over the age of 20 begin cervical cancer screening with a Pap smear or liquid-based cytology (LBC) and continue every 3 years until the age of 74 (if three-consecutive cytologic examinations have been negative within the previous 10 years).[3]

The Korean Society for Cytopathology is committed to improving the quality of cytologic diagnosis and guiding the administration and management of cytology laboratories. The

pISSN 2383-7837
eISSN 2383-7845

Table 1. Assessment of diagnostic accuracy in cervical cytology

Histologic diagnosis	Cytologic diagnosis	Category
Squamous cell		
Negative	Negative	O
	ASCUS	A
	ASC-H	B
	LSIL	B
	HSIL	C
	Squamous cell carcinoma	C
LSIL	Negative	B
	ASCUS	A
	ASC-H	A
	LSIL	O
	HSIL	B
	Squamous cell carcinoma	B
HSIL	Negative	C
	ASCUS	B
	ASC-H	A
	LSIL	B
	HSIL	O
	Squamous cell carcinoma	A
Squamous cell carcinoma	Negative	C
	ASCUS	C
	ASC-H	B
	LSIL	B
	HSIL	A
	Squamous cell carcinoma	O
Glandular cell		
Negative	Negative	O
	Atypical glandular cells	A
	Atypical glandular cells favor neoplastic	B
	Endocervical adenocarcinoma *in situ*	C
	Adenocarcinoma	C
Adenocarcinoma in situ	Negative	B
	Atypical glandular cells	A
	Atypical glandular cells favor neoplastic	A
	Endocervical adenocarcinoma *in situ*	O
	Adenocarcinoma	A
Adenocarcinoma	Negative	C
	Atypical glandular cells	B
	Atypical glandular cells favor neoplastic	B
	Endocervical adenocarcinoma *in situ*	A
	Adenocarcinoma	O
Other		
Other malignant neoplasm	Negative	C
	ASCUS	B
	ASC-H	B
	LSIL	B
	HSIL	A
	Squamous cell carcinoma	A
	Atypical glandular cells	B
	Atypical glandular cells favor neoplastic	A
	Endocervical adenocarcinoma *in situ*	A
	Adenocarcinoma	A

ASCUS, atypical squamous cells of undetermined significance; ASC-H, atypical squamous cells ca nnot exclude high-grade squamous intraepithelial lesion; LSIL, low-grade squamous intraepithelial lesion; HSIL, high-grade squamous intraepithelial lesion.

Committee for Quality Improvement (QI), founded in 1992, monitors and evaluates cytopathologic outcomes on a regular basis. This committee has generated standard operating procedures and documents for quality assurance/quality control since 1995; this is the same year that the first nationwide survey for quality control in cytopathology was conducted.[1] Cytology proficiency testing has been performed as a part of QI programs since 1996. Every year since 2003, all cytopathology laboratories have been required to submit the previous year's quality control statistics and participate in two cytology proficiency testing programs as provided by the QI committee.[1] The QI program has contributed to a reduction in cervical cancer mortality, reducing the odds ratio to 0.36 (95% confidence interval, 0.31 to 0.43).[3]

In this study, we present nationwide cytology statistics from 2015, including the number of cytology cases, specimen type, case volume by diagnosis, specimen preparation methods, and cytology-histology correlations.

MATERIALS AND METHODS

The QI Committee of the Korean Society for Cytopathology conducted a nationwide quality control survey in cytopathology from February 2, 2016, to February 22, 2016. The questionnaire was sent to 208 medical institutions performing cytopathologic examinations in Korea in order to gather statistical data on cytology and gynecologic (GYN) cytology-histology correlation results from 2015. Written informed consent was obtained from each institution. This study was approved by the Institutional Review Board of The Catholic University of Korea, Seoul St. Mary's Hospital (KC13SISI0198).

For the purposes of this study, medical institutions were categorized into three groups: university hospitals, general hospitals, and commercial laboratories. All cytology samples were also classified into three groups: GYN, fine needle aspiration (FNA), and non-GYN/non-FNA samples including body fluids, urine, bronchial washing/brushing samples, cerebrospinal fluid, etc.

https://doi.org/10.4132/jptm.2017.08.11

The diagnostic concordance between cytologic and corresponding histologic examinations of the uterine cervix was categorized as either concordant (category O) or into one of three discordant categories: category A (minimal clinical impact), category B (moderate clinical impact), and category C (major clinical impact). The criteria for the assessment of diagnostic accuracy are shown in Table 1.

Statistical analysis was performed using GraphPad Prism software ver. 6.05 (GraphPad Software, La Jolla, CA). p-values less than .05 were considered statistically significant. Graphs were generated using GraphPad Prism and Microsoft Excel.

RESULTS

Responses were obtained from 206 out of 208 medical institutions (99.0%) including 83 university hospitals, 87 general hospitals, and 36 commercial laboratories.

Statistics of all cytopathology cases in 2015

The total number of cytopathologic examinations performed in 2015 was 8,284,952, of which 5,717,336 (69.0%) were performed in commercial laboratories, 1,603,591 (19.4%) in university hospitals, and 964,025 (11.6%) in general hospitals (Fig. 1A).

Out of a total of 8,284,952 samples examined, 6,734,465 (81.3%) were classified as GYN, 314,893 (3.8%) as FNA, and 1,235,594 (14.9%) as non-GYN/non-FNA (Fig. 1B).

In all, 6,190,526 (74.7%) cytology samples were prepared for conventional smear (CS) and 2,094,426 (25.3%) were prepared for LBC. Among the CS samples, 81.2% were GYN, 3.5% were FNA, and 15.3% were non-GYN/non-FNA. Among the LBC samples, 81.5% were GYN, 4.7% were FNA, and 13.9% were non-GYN/non-FNA (Fig. 1C).

Statistics of GYN cytology

The number of GYN cytology samples was 6,734,465. GYN cytology samples were examined mainly at commercial labora-

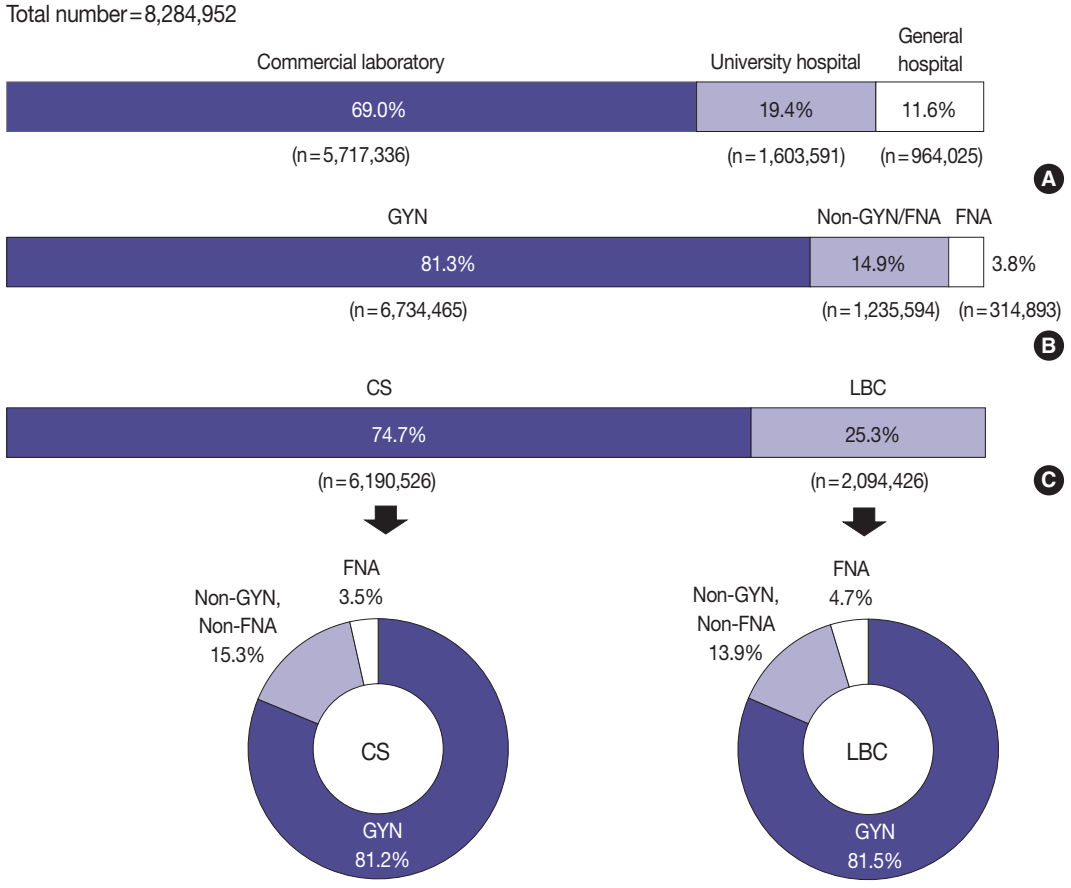


Fig. 1. Number of cytopathology cases in 2015 according to the type of medical institution (A), specimen (B), and preparation method (C). GYN, gynecologic; FNA, fine needle aspiration; CS, conventional smear; LBC, liquid-based cytology.

tories (74.9%, n=5,043,280) and followed by university hospitals (13.9%, n=935,590) and general hospitals (11.2%, n=755,595) (Fig. 2A).

GYN cytology samples in commercial laboratories and general hospitals were more frequently examined by CS than LBC, whereas university hospitals preferred LBC over CS (Fig. 2B).

Statistics of FNA cytology

A total of 314,893 FNA cytology samples were examined. Among them, 133,849 (42%) were examined in commercial laboratories, 125,059 (40%) in university hospitals, and 55,985 (18%) in general hospitals (Fig. 3A).

The most common specimens were thyroid followed by lung (Fig. 3B). FNA Samples in university hospitals showed the highest ratio of LBC to CS followed by general hospitals and commercial laboratories in descending order (Fig. 3C).

Statistics of non-GYN, non-FNA cytology

A total of 1,235,594 non-GYN/non-FNA cytology samples were examined, consisting of 542,942 (43.9%) from university hospitals, 540,207 (43.7%) from commercial laboratories, and 152,445 (12.3%) from general hospitals (Fig. 4A). The proportion of each specimen type (respiratory, body fluid, urine, cerebrospinal fluid, etc.) is shown in Fig. 4B. The most common type of non-GYN/non-FNA specimen was respiratory cytology, regardless of the type of institution. The ratio of LBC to CS was higher in university and general hospitals than in commercial laboratories (Fig. 4C).

Methods of LBC preparation

A total of 11 methods for LBC preparation were used: ThinPrep (Hologic, Marlborough, MA, USA), Cell Prep (Biodyne, Seongnam, Korea), SurePath (BD Diagnostics, Sparks, MD, USA), Huro Path (CelltraZone, Seoul, Korea), MonoPrep (MonoGen, Arlington Heights, IL, USA), Prex-Prep, EASY Prep (YD Diagnostics, Yongin, Korea), Cell Scan (Cell & Tech Bio, Seoul, Korea), CYTO-fast (Hospitex Diagnostics, Firenze, Italy), Liqui-PREP (LGM International, Melbourne, FL, USA), and Max-Prep (Fig. 5). The number of medical institutions using each type of LBC preparation method is listed here in descending order: ThinPrep (68), SurePath (44), Cell Prep (17), EASY Prep (9), Huro Path (5), Cell Scan (4), and Prex-Prep (2). Only one institution used a preparation other than the aforementioned methods. The most commonly used LBC method was ThinPrep (39.7%) followed by Cell Prep (26.3%) and SurePath (23.7%); the others methods accounted for less than 5% of LBC cases. In GYN cytology specimens, ThinPrep (38.8%) was most commonly used followed by Cell Prep (30.0%) and SurePath (19.4%). In non-GYN/non-FNA cytology specimens, SurePath (49.9%) was most commonly used followed by ThinPrep (44.7%) and Cell Prep (2.6%). In

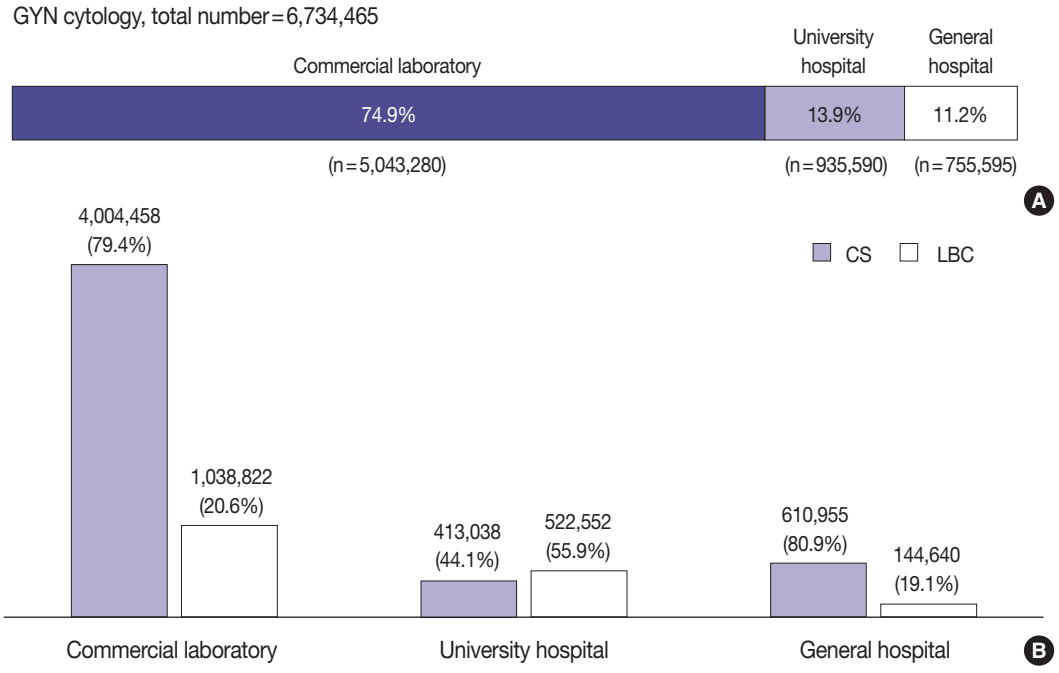


Fig. 2. Number of gynecologic (GYN) cytology cases in 2105 according to the type of medical institution (A) and sample preparation method (B). CS, conventional smear; LBC, liquid-based cytology.

https://doi.org/10.4132/jptm.2017.08.11

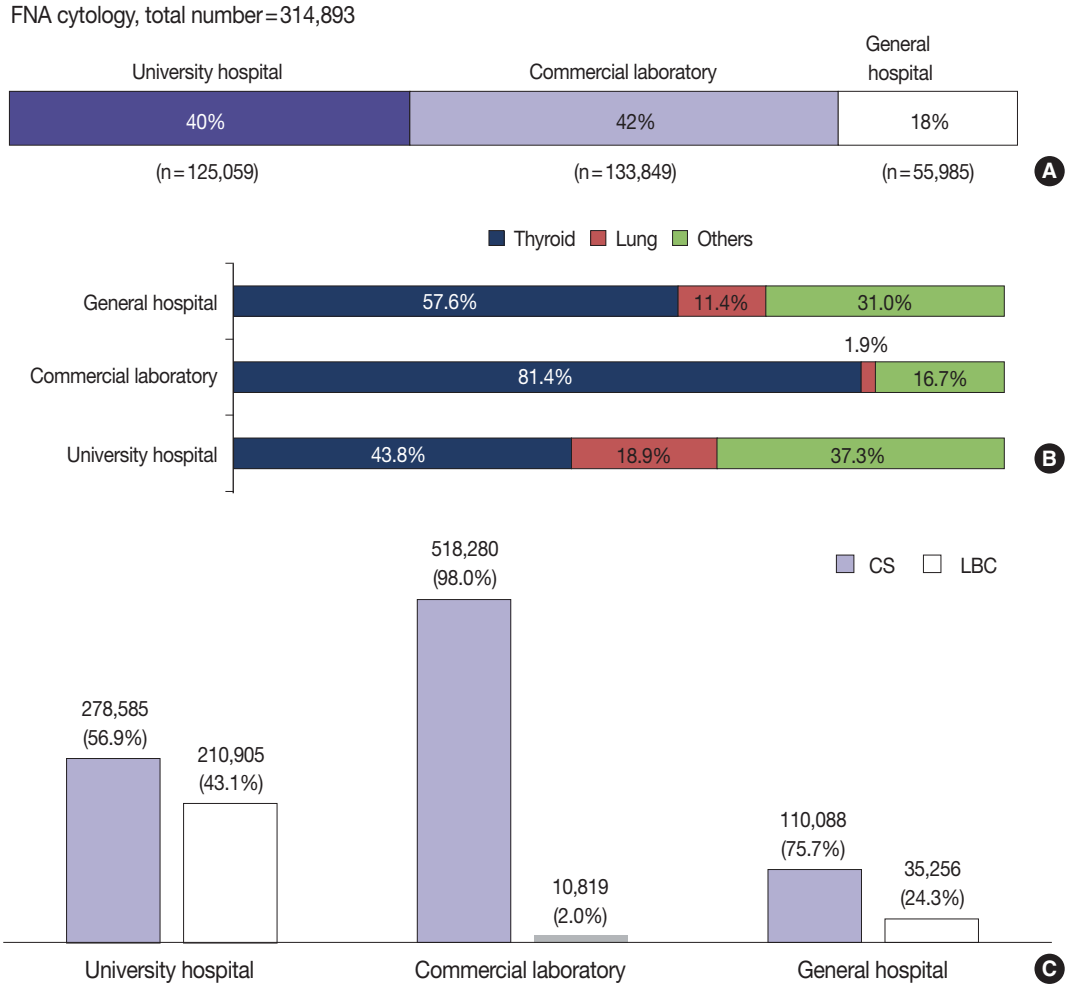


Fig. 3. Number of fine needle aspiration (FNA) cytology cases in 2105 according to the type of medical institution (A), specimen (B), and sample preparation method (C). CS, conventional smear; LBC, liquid-based cytology.

FNA cytology specimens, ThinPrep (45.0%) was most commonly used followed by SurePath (33.6%), EASY Prep (12.9%), and Huro Path (4.5%).

Prevalence of cytologic diagnoses of the uterine cervix

Of the GYN cytology samples, 95% were diagnosed as negative, 3% as atypical squamous cells of undetermined significance (ASCUS), and 1% as low-grade squamous intraepithelial lesion (LSIL). In each type of institution, 1% of samples were considered to be unsatisfactory. The frequencies of unsatisfactory, negative, ASCUS, atypical squamous cells-cannot exclude high-grade squamous intraepithelial lesion, atypical glandular cells (AGC), AGC-favor neoplastic, LSIL, high-grade squamous intraepithelial lesion, adenocarcinoma *in situ*, squamous cell carcinoma, adenocarcinoma, etc. are shown in Table 2.

The ratio of atypical squamous cells of undetermined significance to squamous intraepithelial lesion (ASCUS:SIL), representing the screening sensitivity, was significantly higher in commercial laboratories (6.0 ± 9.2) than in university hospitals (1.9 ± 2.0) and general hospitals (2.8 ± 3.0) ($p < .001$) (Fig. 6).

Diagnostic accuracy of GYN cytology

Statistical data on diagnostic accuracy were obtained from 77 university hospitals and 54 general hospitals (Fig. 7). In university hospitals, a total of 44,044 GYN cytology cases were compared with matching histologic specimens with the following results: 9.1% in category A, 4.0% in category B, 0.6% in category C, and 86.3% in category O. In general hospitals, a total of 3,898 GYN cytology cases were compared with matching histologic specimens with the following results: 17.4% in category A, 10.2% in category B, 1.9% in category C, and 69.5% in category O. Accuracy data from commercial laboratories was not available.

Table 2. Prevalence of cytologic diagnosis of uterine cervix according to the type of medical institution

Cytology	University hospital	Commercial laboratory	General hospital	Total
Unsatisfactory	5,010 (0.5)	30,271 (0.7)	5,421 (0.7)	40,702 (0.6)
Negative	872,873 (93.6)	4,382,832 (94.7)	741,720 (95.5)	5,997,425 (94.6)
ASCUS	29,421 (3.2)	171,549 (3.7)	20,224 (2.6)	221,194 (3.5)
ASC-H	3,792 (0.4)	6,640 (0.1)	1,504 (0.2)	11,936 (0.2)
LSIL	13,262 (1.4)	28,667 (0.6)	5,253 (0.7)	47,182 (0.7)
HSIL	5,243 (0.6)	6,010 (0.1)	1,833 (0.2)	13,086 (0.2)
Squamous cell carcinoma	890 (0.1)	500 (<0.1)	246 (<0.1)	1,636 (<0.1)
AGC	1,055 (0.1)	2,350 (0.1)	507 (0.1)	3,912 (0.1)
AGC, favor neoplastic	195 (<0.1)	292 (<0.1)	74 (<0.1)	561 (<0.1)
Adenocarcinoma *in situ*	51 (<0.1)	4 (<0.1)	19 (<0.1)	74 (<0.1)
Adenocarcinoma	348 (<0.1)	65 (<0.1)	64 (<0.1)	477 (<0.1)
Others	56 (<0.1)	628 (<0.1)	13 (<0.1)	697 (<0.1)
Total	932,196 (100)	4,629,808 (100)	776,878 (100)	6,338,882 (100)

Values are presented as number (%).
ASCUS, atypical squamous cells of undetermined significance; ASC-H, atypical squamous cells cannot exclude high-grade squamous intraepithelial lesion; LSIL, low-grade squamous intraepithelial lesion; HSIL, high-grade squamous intraepithelial lesion; AGC, atypical glandular cells.

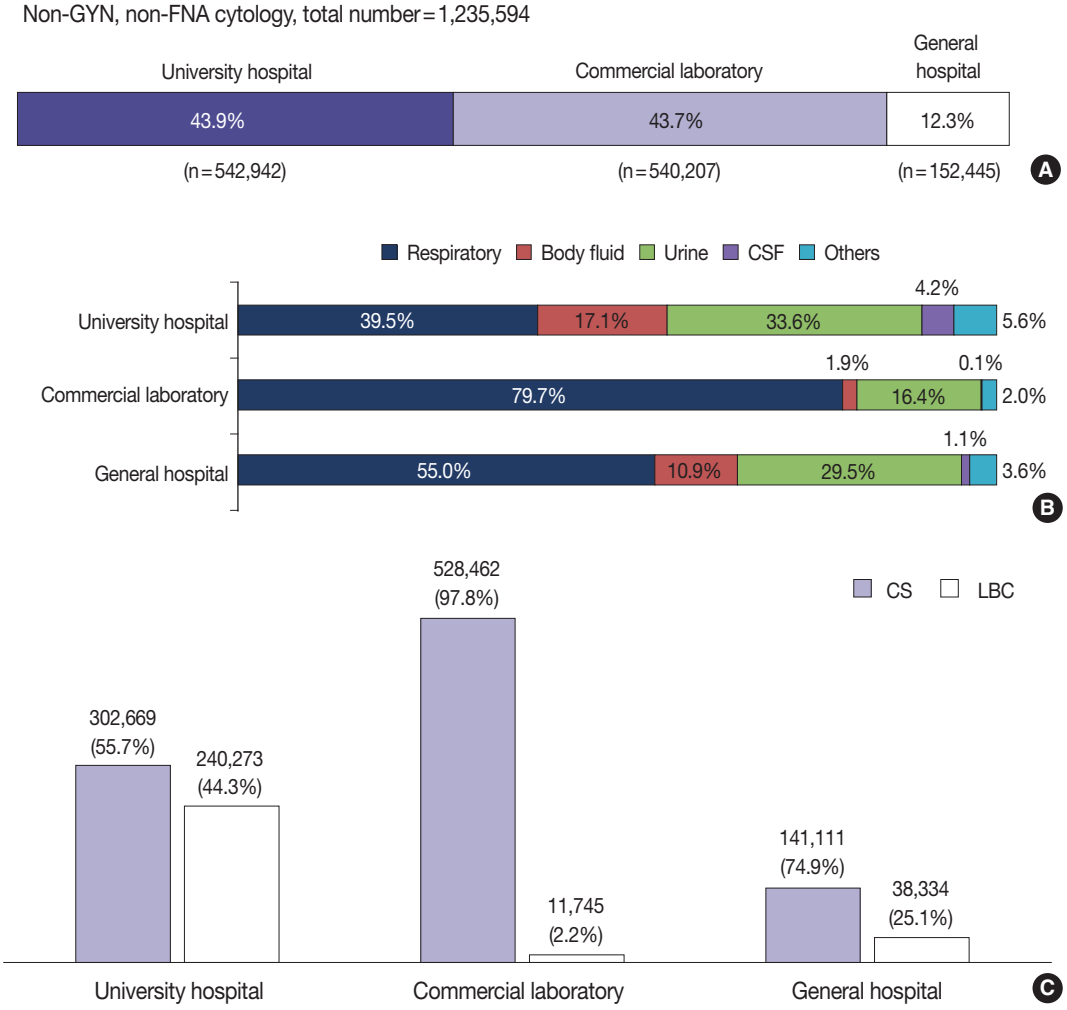


Fig. 4. Number of non-gynecologic (GYN), non-fine needle aspiration (FNA) cytology cases in 2105 according to the type of medical institution (A), specimen (B), and sample preparation method (C). CSF, cerebrospinal fluid; CS, conventional smear; LBC, liquid-based cytology.

https://doi.org/10.4132/jptm.2017.08.11

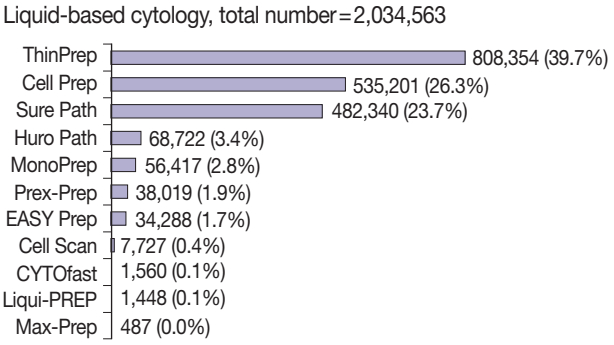


Fig. 5. Usage of liquid-based cytology in 2015 according to number of institutions, number of specimens, and type of specimen.

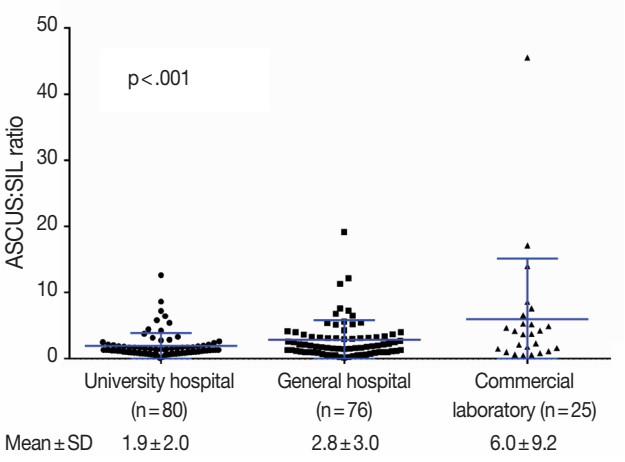


Fig. 6. Ratio of atypical squamous cells of undetermined significance (ASCUS) to squamous intraepithelial lesions (SIL) according to the type of medical institution in 2015.

DISCUSSION

The survey response rate for this study was quite high. In all, 99.0% of medical institutions providing cytopathology service responded to this survey as mandated by the QI program of the Korean Society for Cytopathology.

GYN cytology comprised 81.3% of all cases, which is due to the national cervical cancer screening program. Of the GYN cytology samples, 74.9% were examined at commercial laboratories, 13.9% at university hospitals, and 11.2% at general hospitals. In Korea, Pap smears are generally performed in primary care clinics and health promotion centers, and most of those specimens are then sent to commercial laboratories.

The conventional Pap smear is a simple and effective method for screening cervical cancer. LBC was initially introduced in GYN cytology, and its use has increased continuously over the past two decades. According to a previous nationwide study in Korea, the use of LBC comprised 7.6% and 20.5% of all cytology

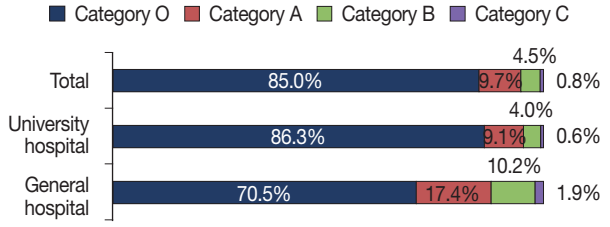


Fig. 7. Diagnostic accuracy of gynecologic cytology in 2015 according to the type of medical institution.

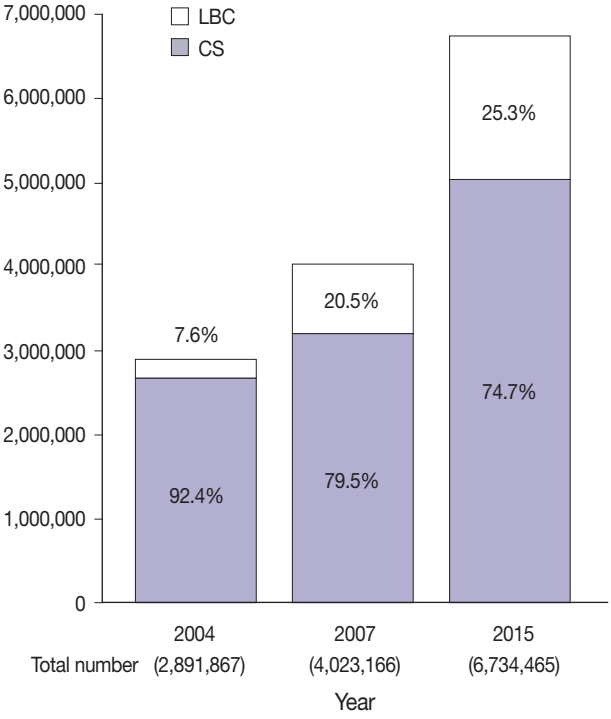


Fig. 8. Total number of gynecologic cytology cases according to the method of sample preparation at three different times. The number of cytology cases and the proportion of liquid-based cytology (LBC) have increased over time. CS, conventional smear.

cases in 2004 and 2007, respectively (Fig. 8).[1] In 2015, LBC was used in 25.3% of all GYN cytology cases. In university hospitals, 55.9% of all GYN cytology were examined by LBC, whereas commercial laboratories used LBC less frequently (Fig. 2B).

In the field of non-GYN cytology, LBC is only used for body fluid and thyroid aspiration samples by the National Health Insurance System in Korea. The proportion of LBC in non-GYN cytology cases was higher in university hospitals than in other types of institutions (Figs. 3C, 4C). The thyroid gland was the highest source of FNA samples (Fig. 3B). The high number of thyroid FNA cases in Korea closely correlates with the high rate of thyroid cancer screening, as Korea has the highest incidence of thyroid cancer in the world.[4] Although 11 methods of LBC

were introduced in Korea, more than 95% of cases used either ThinPrep, Cell Prep, or SurePath. This is the first report to investigate the current status of the LBC methods used in Korea.

The overall rates of unsatisfactory, negative, and ASCUS for GYN cytology cases were 0.6%, 94.6%, and 3.5% respectively. These results did not significantly differ among the three types of medical institutions. The ASCUS rate may have varied according to the cytopathologist performing the exam. The ASCUS:SIL ratio was less affected than the ASCUS rate by patient population, prevalence of disease, and the effect of screening.[5-10] Thus, the ASCUS:SIL ratio was used as a quality control reference.[10] In one study, cytotechnologists with an ASCUS:SIL ratio < 1.5 showed a significantly lower sensitivity than those with a ratio > 3.0.[5] In our study, the mean ASCUS:SIL ratio was 1.9 in university hospitals, 2.8 in general hospitals, and 6.0 in commercial laboratories (Fig. 6). Thus, the mean screening sensitivity of commercial laboratories may be higher when compared with referral hospitals.

Quality control of diagnostic accuracy was assessed by correlating cervical cytology with histologic results as mandated in university hospitals and general hospitals by the QI program of the Korean Society for Cytopathology. However, this correlation was not performed in most commercial laboratories. The concordance rate of cytologic and histologic diagnoses may vary depending upon the time difference between the two examinations.[11-13] Diagnostic correlations between cytology and histology are lower when the two examinations are performed simultaneously than when cytology precedes histologic examination.[11,12] During this study, we recognized that there was no standardization of cytologic-histologic correlation methods, including the type of histologic specimens, the intervals for correlation, and the statistical metrics used. In our study, cytologic-histologic correlation was performed in real time and/or retrospectively. The tissue samples for histologic correlation were obtained from either cervical biopsy, conization, or hysterectomy.

We found that the volume of cytology cases, sample type, preparation method, access to paired tissue specimens, practice variables, and screening sensitivity all differed from institution to institution. In general, both the total volume of cytology cases and the proportion of cases using LBC have significantly increased. The screening sensitivity of GYN cytology was higher and more variable in commercial laboratories when compared with university and general hospitals. The results of this study can serve as basic data for the establishment of nationwide cytopathology policies and QI guidelines in Korean medical institutions.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

Acknowledgments

This research was supported by The Korean Society for Cytopathology Grant No. 2015-02. The authors would like to acknowledge the Korean Society for Cytopathology and BD Korea for providing the funding necessary to carry out this study. We would also like to thank the Fellowship Council of the Korean Society for Cytopathology for its guidance in preparing and conducting this project and the Committee of Quality Improvement of the Korean Society for Cytopathology for providing the data.

REFERENCES

1. Lee HK, Kim SN, Khang SK, Kang CS, Yoon HK. Quality control program and its results of Korean Society for Cytopathologists. Korean J Cytopathol 2008; 19: 65-71.
2. Kim Y, Jun JK, Choi KS, Lee HY, Park EC. Overview of the National Cancer screening programme and the cancer screening status in Korea. Asian Pac J Cancer Prev 2011; 12: 725-30.
3. Min KJ, Lee YJ, Suh M, et al. The Korean guideline for cervical cancer screening. J Gynecol Oncol 2015; 26: 232-9.
4. Ahn HS, Kim HJ, Welch HG. Korea's thyroid-cancer "epidemic": screening and overdiagnosis. N Engl J Med 2014; 371: 1765-7.
5. Renshaw AA, Deschênes M, Auger M. ASC/SIL ratio for cytotechnologists: a surrogate marker of screening sensitivity. Am J Clin Pathol 2009; 131: 776-81.
6. Nascimento AF, Cibas ES. The ASC/SIL ratio for cytopathologists as a quality control measure: a follow-up study. Am J Clin Pathol 2007; 128: 653-6.
7. Juskevicius R, Zou KH, Cibas ES. An analysis of factors that influence the ASCUS/SIL ratio of pathologists. Am J Clin Pathol 2001; 116: 331-5.
8. Davey DD, Naryshkin S, Nielsen ML, Kline TS. Atypical squamous cells of undetermined significance: interlaboratory comparison and quality assurance monitors. Diagn Cytopathol 1994; 11: 390-6.
9. Davey DD, Nielsen ML, Naryshkin S, Robb JA, Cohen T, Kline TS. Atypical squamous cells of undetermined significance: current laboratory practices of participants in the College of American Pathologists Interlaboratory: comparison program in cervicovaginal cytology. Arch Pathol Lab Med 1996; 120: 440-4.
10. Clary KM, Davey DD, Naryshkin S, et al. The role of monitoring

interpretive rates, concordance between cytotechnologist and pathologist interpretations before sign-out, and turnaround time in gynecologic cytology quality assurance: findings from the College of American Pathologists Gynecologic Cytopathology Quality Consensus Conference working group 1. Arch Pathol Lab Med 2013; 137: 164-74.
11. Jones BA, Novis DA. Cervical biopsy-cytology correlation. A College of American Pathologists Q-Probes study of 22 439 correlations in 348 laboratories. Arch Pathol Lab Med 1996; 120: 523-31.
12. Joste NE, Crum CP, Cibas ES. Cytologic/histologic correlation for quality control in cervicovaginal cytology: experience with 1,582 paired cases. Am J Clin Pathol 1995; 103: 32-4.
13. Crothers BA, Jones BA, Cahill LA, et al. Quality improvement opportunities in gynecologic cytologic-histologic correlations: findings from the College of American Pathologists Gynecologic Cytopathology Quality Consensus Conference working group 4. Arch Pathol Lab Med 2013; 137: 199-213.

Journal of Pathology and Translational Medicine 2017; 51: 588-593
https://doi.org/10.4132/jptm.2017.08.10

■ CASE STUDY ■

Aggressive Supratentorial Ependymoma, *RELA* Fusion-Positive with Extracranial Metastasis: A Case Report

Seong-Ik Kim · Yoojin Lee
Seung Ki Kim[1] · Hyoung Jin Kang[2]
Sung-Hye Park

Departments of Pathology, [1]Neurosurgery, and [2]Pediatrics, Seoul National University Children's Hospital, Seoul, Korea

Received: July 31, 2017
Revised: August 8, 2017
Accepted: August 10, 2017

Corresponding Author
Sung-Hye Park, MD
Department of Pathology, Seoul National University College of Medicine, 101 Daehak-ro, Jongno-gu, Seoul 03080, Korea
Tel: +82-2-740-8278
Fax: +82-2-765-5600
E-mail: shparknp@snu.ac.kr

Ependymoma is the third most common pediatric primary brain tumor. Ependymomas are categorized according to their locations and genetic abnormalities, and these two parameters are important prognostic factors for patient outcome. For supratentorial (ST) ependymomas, *RELA* fusion-positive ependymomas show a more aggressive behavior than *YAP1* fusion-positive ependymomas. Extracranial metastases of intra-axial neuroepithelial tumors are extremely rare. In this paper, we report a case of aggressive anaplastic ependymoma arising in the right frontoparietal lobe, which had genetically 1q25 gain, *CDKN2A* homozygous deletion, and L1CAM overexpression. The patient was a 10-year-old boy who underwent four times of tumor removal and seven times of gamma knife surgery. Metastatic loci were scalp and temporalis muscle overlying primary operation site, lung, liver, buttock, bone, and mediastinal lymph nodes. He had the malignancy for 10 years and died. This tumor is a representative case of *RELA* fusion-positive ST ependymoma, showing aggressive behavior.

Key Words: Ependymoma; Transcription factor RelA; Neoplasm metastasis; Supratentorial neoplasms; Genetics

Ependymoma is the third most common neuroepithelial tumor of the childhood.[1] It is a circumscribed glioma consisting of uniform small cells with ependymal differentiation, commonly originates from the walls of the cerebral ventricles or spinal canal, and is mostly manifest in children and young adults.[2] Ependymomas account for 9% and 8% of intracranial and intraspinal neoplasms of all primary brain and central nervous system (CNS) tumors in ages 0–14 and 0–19, respectively.[3] Like other primary glioneuronal tumors, extracranial metastasis of ependymoma is extremely rare.[4-11] Korshunov *et al*.[12] reported that the incidence of extracranial metastasis in their cohort was 2% (5/258 cases). In this paper, we present a rare case of anaplastic ependymoma with widespread extraneural metastasis. This study abides by the World Medical Association Declaration of Helsinki recommendations and was approved by the Institutional Review Board of Seoul National University Hospital (IRB No. 1507-040-690). The patient agreed and signed the agreement.

CASE REPORT

A 10-year-old boy visited the pediatric neurosurgery outpatient clinic due to headache and vomiting for 1 week. On physical examination, grade IV+ motor weakness in the right upper extremity and limping gait of the right leg were noted. Initial magnetic resonance imaging (MRI) revealed a large cystic mass (6.2×4.5 cm) in the left frontoparietal lobe (Fig. 1A). Craniotomy and gross total removal (GTR) of the tumor were performed in May 2007. Both solid and cystic portions were dissected from the normal-appearing brain parenchyma for GTR of the tumor. Histological diagnosis was World Health Organization (WHO) grade II ependymoma. Postoperatively, the patient's neurological status was significantly improved, and right hemi-weakness gradually normalized.

Follow-up (F/U) MRI 3 months after initial surgery showed a small enhancing lesion with peripheral edema at the superolateral side of the operation bed compared with immediate postoperative MRI. At that time, clinicians could not rule out tumor recurrence. Therefore, the patient was treated with 8 weeks of adjuvant radiotherapy with 61.2 Gy. Five months after surgery, when the dose of antiepileptic drug was decreased, focal seizure developed as twitching in the right arm. After reverting to the full dosage of antiepileptic drugs, the symptoms disappeared. By that time, the

pISSN 2383-7837
eISSN 2383-7845

right upper extremity weakness had almost normalized. However, 6 months after surgery, F/U MRI (Fig. 1B) revealed an increased extent of the recurrent tumor and peritumoral edema at the operation site compared with the 3-month F/U MRI finding (1×0.8×1.9 cm vs 1.5×1.6×3.1 cm). On reoperation 8 months after initial surgery, intraoperative ultrasound was used for GTR, and anterior, posterior, and both lateral margins were checked through frozen section biopsy. The second reoperation revealed WHO grade III anaplastic ependymoma. Subsequent F/U MRI was performed, and GTR was suggested.

Two months after the second operation, F/U MRI showed no definite enhancing recurrent mass; however, rim enhancement was noted along the resection margin. Tumor recurrence was suspected, and chemotherapy was considered; however, no recommendable chemotherapy regimen was available. Therefore, gamma knife surgery (GKS) (No. 1) was performed (volume, 3.7 cc; dose, 18 Gy at 52%; shot, 8×). However, the lesion enlarged again one month after the GKS. Subsequent GKS was then performed twice (GKS No. 2: volume, 1 cc; dose, 16 Gy at 50%; shot, 6×; GKS No. 3: volume, 1.8 cc; dose, 19 Gy; shot, 6×). Approximately a year after the second surgery, the tumor showed a rapid growth (February 2009, 13.8×18.6×13.8 cm; April 2009, 20.6×21.5×15.8 cm), and reoperation for GTR was considered but eventually could not be carried out. Without reoperation, there was a gradual decrease in tumor size for a year.

However, findings of the 2-year and 6-month F/U MRI worsened. Several newly appearing enhancements were found in the left parietal and temporal lobes, left cranial nerves (CN) VII and VIII, and right CN V. Leptomeningeal seeding was also noted (Fig. 1C). GKS No. 4 was performed again. A third operation was again considered due to increased enhancing mass, but was refused by the guardian. Fractionated GKS No. 5 was performed instead. Numbness of the right arm and myoclonic movement accompanied by limb weakness and sensory change in taste developed. F/U MRI showed increased enhancing mass and increased edema compared with the MRI findings a month ago. Fractionated GKS No. 6 was then performed. Right hemiparesis worsened. A third craniotomy and tumor removal was performed, and the tumor along the gliotic plane was totally removed. However, after surgery, right hemiparesis worsened; as such, the patient started to receive rehabilitation management. Two-month F/U MRI showed an increase in the size of the enhancing dura-based mass. Fractionated GKS No. 7 was done, which was preferred by the guardian. The patient then went to the emergency room due to right-sided tonic clonic seizure. On computed tomography scan, focal nodular enhancing lesion appeared again at the operation site. Tumor invasion to the incision scar and temporalis muscle was observed. Additionally, new enhancing nodule in the left parietal lobe was found (Fig. 1D). A fourth craniotomy and excision of the tumor, which showed enhancement on MRI, was subsequently performed.

Seven years after initial presentation, several new tumors were found in the leptomeninges, scalp, temporalis muscle, lung, mediastinal lymph node (LN), liver, and bones on positron emission tomography imaging study (Fig. 2A). Excision of the scalp masses and needle biopsy of the liver were performed for diagnostic and palliative purpose (Fig. 3). The tumors were pathologically confirmed as mestastatic anaplastic ependymoma. The patient received chemotherapy, seven cycles of VIP (etoposide, ifosfamide and cisplatin).

However, the disease progressed and his condition worsened. He died after 9 years of having anaplastic ependymoma.

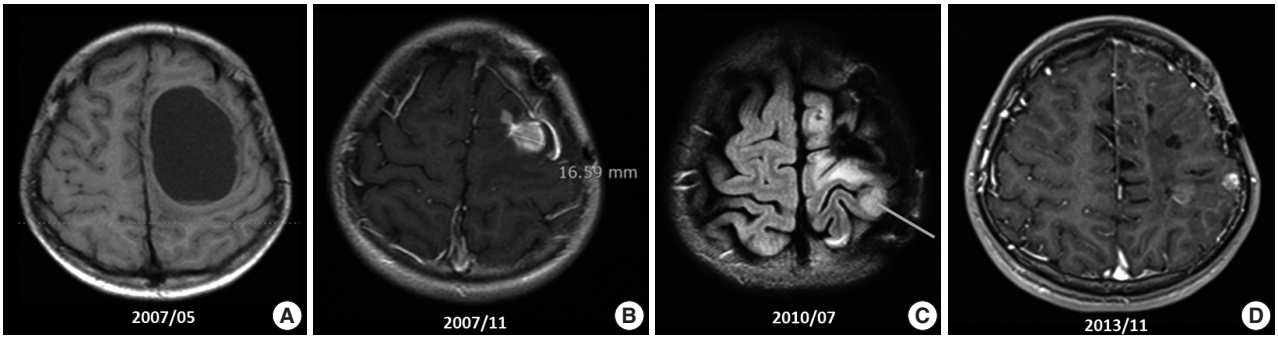


Fig. 1. Magnetic resonance imaging (MRI) findings. (A) Initial MRI scan shows a 6.2-cm large cystic mass in the left frontoparietal lobe (May 2007). First craniotomy and tumor removal was done. (B) An increase in the extent of the recurred tumor and surrounding peritumoral edema at the superior-lateral side of the postoperative defect was noted on several follow-up MRI scans (November 2007). (C) Clinical leptomeningeal seeding was first detected via MRI, which also shows a new ill-defined lesion with contrast enhancement in the left parietal lobe (July 2010). (D) Extracranial scalp metastasis was suspected clinically. It was detected via MRI (November 2013) and showed focal nodular enhancing lesion overlying the left temporalis muscle.

Pathological findings

Pathologic diagnosis of the initially resected tumor in 2007 was grade II ependymoma (Fig. 2B), with low mitotic rate (4/10 high-power field) and low Ki-67 labeling index (1%). However, the pathologic diagnosis of the second recurrence at 5 months after the initial GTR, which was removed 8 months after the initial GTR, was WHO grade III anaplastic ependymoma, with high mitotic rates, extensive necrosis, microvascular proliferation, and high Ki-67 (1:1,000, DAKO, Glostrup, Denmark) labeling index (Fig. 2C, G). Moreover, Ki-67 labeling indices in the specimen from leptomeningeal seeding were 24.5% to 37.3%, suggesting rapid malignant transformation. After GKS, the tumor showed extensive necrosis (more than 50%), suggesting that radiotherapy and GKS were effective to a certain level. However, the tumor eventually metastasized systemically. The tumor cells were robustly positive for glial fibrillary acidic protein (GFAP; 1:300, DAKO), and showed extensive dot-like positivity for epithelial membrane antigen (1:100, DAKO) (Fig. 2D, E). Results of the ultrastructural study showed microvilli and cilia in the intercellular and intracytoplasmic microrosettes. In addition, long intermediate junctions were present. Collectively, these ultrastructural findings suggested anaplastic ependymoma. In all specimens, L1CAM (1:200, Abcam, Cambridge, UK) was robustly positive in the entire tumor cell cytoplasm, suggesting *RELA* fusion-positive ependymoma (Fig. 2F).

Results of the liver biopsy revealed metastatic anaplastic ependymoma, which showed robust GFAP positivity and high Ki-67 index of 92.47% (Fig. 3A–C).

Using formalin-fixed paraffin embedded tissue, fluorescence *in situ* hybridization (FISH) was performed to examine genetic characteristics. Locus-specific 1p36 (Spectrum Orange)/LSI 1q25 (Spectrum Green) dual-color FISH Probe (Vysis, Downers Grove, IL, USA) and 9p21.3 (*CDKN2A*) (Spectrum Orange)/CEP9 (Spectrum Green) dual color FISH Probe (Vysis) were used. A total of 100 nuclei were counted. The 1q25/1p36 ratio was 1.55, which revealed 1 copy gain of chromosome 1q25,

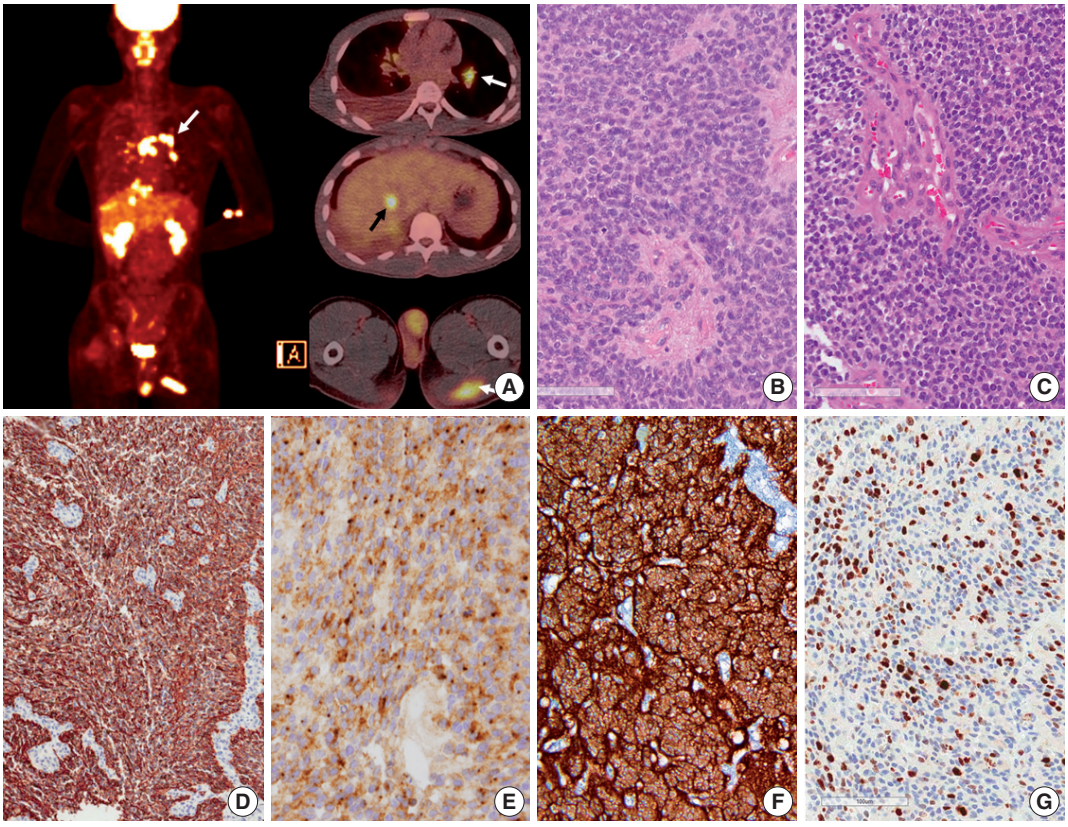


Fig. 2. Positron emission tomography image of the patient and microscopic and immunohistochemical findings of the primary and recurrent tumors. (A) Positron emission tomography scan shows metastasis to the muscle, lung, mediastinal lymph node (white arrows), liver (black arrow), buttocks, and bones. (B, C) Initial (in 2007) and recurrent tumors (in 2013) show sheets of monotonous cells with oval nuclei with salt-and-pepper chromatin pattern. The recurrent tumor in 2013 shows microvascular proliferation. (D) Glial fibrillary acidic protein is robustly positive in tumor cells. (E) Epithelial membrane antigen shows dot-like positivity, suggesting ependymal tumor. (F) L1CAM shows diffuse strong positivity in the tumor cells, suggesting *RELA* fusion-positive ependymoma. (G) Ki-67 labeling index in the recurrent tumor was high (37.3%).

https://doi.org/10.4132/jptm.2017.08.10

and 9p21.3 homozygous deletion was also found (Fig. 3D, E).

DISCUSSION

Ependymomas are tumors arising from ependymal lining cells, which are classified according to the location and age of the patient because different genetic abnormalities and biological behaviors are found according to these two parameters.[13,14] Ependymomas are more common in children than in adults and are the third most frequent neuroepithelial tumors of childhood.[1] Ependymomas in children tend to occur in the posterior fossa (PF), whereas they tend to occur in the supratentorium and spinal cord (SC) in adults. The tumors can be graded as WHO grade II and III according to histopathological parameters, such as mitotic rate, microvascular proliferation, and nuclear pleomorphism; however, it is difficult to apply and the clinical utility is questionable.[15]

By far, no effective anticancer regimen has been found against ependymomas. Surgical resection and radiotherapy is the treatment of choice. Therefore, unlike other CNS gliomas, the outcome of ependymomas does not follow grade but the location and resectability.

Ependymomas are divided into three groups according to their location: supratentorial (ST), PF, and SC. ST ependymoma is further categorized as *RELA-C11* or *f95* fusion and *YAP1-MAMDL1* or *YAP1-FAM118B* fusion.[16] *RELA* fusion subtype comprises 70% of ST ependymomas, and the rest is *YAP1* fusion.[16] The *RELA* fusion group has remarkably worse survival than that of *YAP1* fusion group. *RELA* fusion can be detected via reverse transcriptase–polymerase chain reaction or immunohistochemical marker L1CAM.[15] In case of PF ependymomas, LAMA2-expressing ependymomas (group A) has worse outcome than NELL2-expressing ependymomas (group B).[17]

At initial presentation, the tumor is located in the left frontoparietal area. In general, patients with ST ependymomas are believed to have better survival rates than those with infratentorial ependymomas because GTR is possible.[18] However, several contradictory reports regarding patients' outcomes have been published.[19]

The most important prognostic factor for ependymomas is

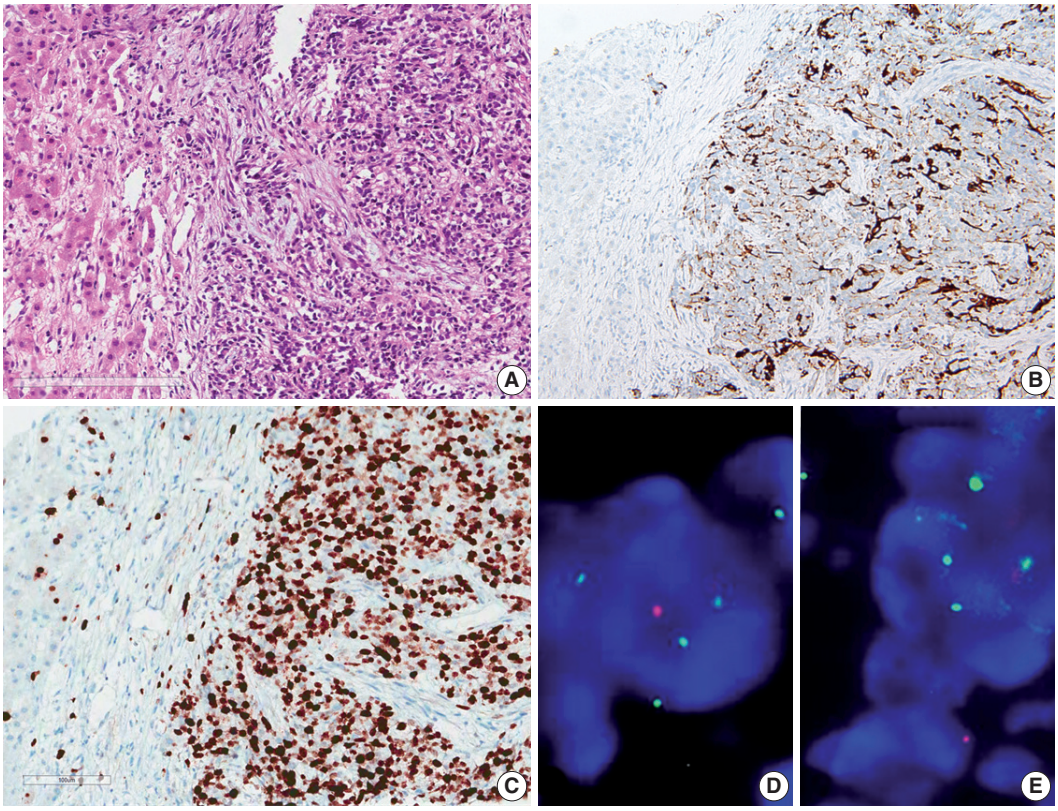


Fig. 3. Microscopic finding and immuohistochemical and fluorescence *in-situ* hybridization studies of metastatic ependymoma to the liver. (A) Results of the liver biopsy reveal metastatic anaplastic ependymoma. The metastatic tumor shows robust glial fibrillary acidic protein positivity (B) and high MIB-1 (Ki-67) labeling index of 92.47% (C). Fluorescence *in-situ* hybridization reveals low copy gain of 1q25 (1q25 [spectrum green]/1p36 [spectrum orange] ratio = 1/55) (D) and CDKN2A (9p21.3) homozygous deletion (E).

genomic aberration. In cases of ST ependymomas, *RELA* fusion-positive ependymomas have worse prognosis than *YAP1* fusion-positive ependymomas.[8] Additionally, gain of 1q25 and homozygous deletion of *CDKN2A* (9p21.3) are powerful independent indicators of unfavorable prognosis.[13] Our case is L1CAM-expressing ST ependymoma with poor clinical course and extracranial metastases to the skull, temporalis muscle, lung, mediastinal LN, liver, and bone.

Rickert published a review article on extracranial metastasis of pediatric brain tumors that included six cases of ependymoma, not otherwise specified, and two cases of anaplastic ependymoma.[20] Among them, one had metastasis without prior surgical intervention, including biopsy, and seven had metastasis after surgical intervention. The mean latency, which is the latency between brain surgery and extracranial metastasis, for metastasis was 25.7 months for non-shunt related metastasis.

In our case, although three surgeries had been done before extracranial metastasis, the latency for extracranial metastasis was 78 months from the first surgery. We assume that the surgery might have played a major role in metastasis to the skull and temporalis muscle; however, the tumor recurrence in the primary site despite GTR in a short duration (in 8 months) and repeated recurrences suggest its aggressive nature, which might be due to its genetic alteration of *RELA* fusion positivity and 1q25 gain. In general, cerebrospinal dissemination indicates poor prognosis in brain tumors.[15] Our case showed leptomeningeal seeding after tumor recurrence in the primary site.

In this paper, we report a case of aggressive anaplastic ependymoma with extracranial metastasis, positive for *RELA* fusion, 1q25 gain, and *CDKN2A* homozygous deletion.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

Acknowledgments

This study was supported by a grant of the Korea Health Technology R&D Project through the Korea Health Industry Development Institute (KHIDI), funded by the Ministry of Health & Welfare, Republic of Korea (grant number: HI14C 1277).

REFERENCES

1. Chhabda S, Carney O, D'Arco F, Jacques TS, Mankad K. The 2016 World Health Organization Classification of tumours of the central nervous system: what the paediatric neuroradiologist needs to know. Quant Imaging Med Surg 2016; 6: 486-9.
2. Perry A, Wesseling P. Histologic classification of gliomas. Handb Clin Neurol 2016; 134: 71-95.
3. Ostrom QT, Gittleman H, Xu J, *et al*. CBTRUS statistical report: primary brain and other central nervous system tumors diagnosed in the United States in 2009-2013. Neuro Oncol 2016; 18(Suppl_5): v1-v75.
4. Pachella LA, Kamiya-Matsuoka C, Lee EL, Olar A, Yung WK. Supratentorial extraventricular anaplastic ependymoma with extracranial metastasis. J Clin Neurosci 2015; 22: 605-7.
5. Davis MJ, Hasan F, Weinreb I, Wallace MC, Kiehl TR. Extraventricular anaplastic ependymoma with metastasis to scalp and neck. J Neurooncol 2011; 104: 599-604.
6. Chao MM, Packer RJ, Myseros JS, Rood BR. Isolated extracranial recurrence of anaplastic ependymoma. Pediatr Blood Cancer 2011; 56: 317-8.
7. Kumar P, Rastogi N, Jain M, Chhabra P. Extraneural metastases in anaplastic ependymoma. J Cancer Res Ther 2007; 3: 102-4.
8. Itoh J, Usui K, Itoh M, Hashizume Y. Extracranial metastases of malignant ependymoma: case report. Neurol Med Chir (Tokyo) 1990; 30: 339-45.
9. Ferracini R, Manetto V, Poletti V, Giangasparo F. A cerebral ependymoma with extracranial metastases. Tumori 1984; 70: 389-92.
10. Wentworth P, Birdsell DC. Intracranial ependymoma with extracranial metastases. J Neurosurg 1966; 25: 648-51.
11. Maass L. Occipital ependymoma with extracranial metastases. J Neurosurg 1954; 11: 413-21.
12. Korshunov A, Golanov A, Sycheva R, Timirgaz V. The histologic grade is a main prognostic factor for patients with intracranial ependymomas treated in the microneurosurgical era: an analysis of 258 patients. Cancer 2004; 100: 1230-7.
13. Korshunov A, Witt H, Hielscher T, *et al*. Molecular staging of intracranial ependymoma in children and adults. J Clin Oncol 2010; 28: 3182-90.
14. Pajtler KW, Witt H, Sill M, *et al*. Molecular classification of ependymal tumors across all CNS compartments, histopathological grades, and age groups. Cancer Cell 2015; 27: 728-43.
15. Louis DN, Perry A, Reifenberger G, *et al*. The 2016 World Health Organization classification of tumors of the central nervous system: a summary. Acta Neuropathol 2016; 131: 803-20.
16. Gajjar A, Bowers DC, Karajannis MA, Leary S, Witt H, Gottardo NG. Pediatric brain tumors: innovative genomic information is transforming the diagnostic and clinical landscape. J Clin Oncol 2015; 33: 2986-98.
17. Nobusawa S, Hirato J, Yokoo H. Molecular genetics of ependymomas

and pediatric diffuse gliomas: a short review. Brain Tumor Pathol 2014; 31: 229-33.
18. Ernestus RI, Schroder R, Stutzer H, Klug N. Prognostic relevance of localization and grading in intracranial ependymomas of childhood. Childs Nerv Syst 1996; 12: 522-6.
19. McGuire CS, Sainani KL, Fisher PG. Both location and age predict survival in ependymoma: a SEER study. Pediatr Blood Cancer 2009; 52: 65-9.
20. Rickert CH. Extraneural metastases of paediatric brain tumours. Acta Neuropathol 2003; 105: 309-27.


JPTM
Journal of Pathology and
Translational Medicine