Universidade Federal do Rio Grande do Sul

Faculdade de Medicina

Programa de Pós Graduação em Ciências Médicas: Endocrinologia

Mestrado

Características Ultrassonográficas e Risco de Câncer de Tireoide: revisão sistemática e meta-análise de estudos observacionais

Aluna: Luciana Loss Reck Remonti

Orientador: Prof. Dr. Jorge Luiz Gross

Co-orientadora: Prof. Dra. Cristiane Bauermann Leitão

Porto Alegre, abril de 2013

Universidade Federal do Rio Grande do Sul

Faculdade de Medicina

Programa de Pós Graduação em Ciências Médicas: Endocrinologia

Mestrado

Características Ultrassonográficas e Risco de Câncer de Tireoide: revisão sistemática e meta-análise de estudos observacionais

Aluna: Luciana Loss Reck Remonti

Orientador: Prof. Dr. Jorge Luiz Gross

Co-orientadora: Prof. Dra. Cristiane Bauermann Leitão

Dissertação apresentada ao Programa de Pós-Graduação em Ciências Médicas: Endocrinologia como requisito parcial para a obtenção do título de mestre

Porto Alegre, abril de 2013

CIP - Catalogação na Publicação

Remonti, Luciana Reck
Características Ultrassonográficas e Risco de Câncer de Tireoide: revisão sistemática e meta-análise de estudos observacionais / Luciana Reck Remonti. -- 2013.
52 f.

Orientador: Jorge Luiz Gross.
Coorientador: Cristiane Bauermann Leitão.

Dissertação (Mestrado) -- Universidade Federal do Rio Grande do Sul, Faculdade de Medicina, Programa de Pós-Graduação em Ciências Médicas: Endocrinologia, Porto Alegre, BR-RS, 2013.

1. Nódulo de tireóide. 2. Câncer de tireóide. 3. Ultrassonografia. 4. Meta-análise. I. Gross, Jorge Luiz, orient. II. Leitão, Cristiane Bauermann, coorient. III. Título.

Elaborada pelo Sistema de Geração Automática de Ficha Catalográfica da UFRGS com os dados fornecidos pelo(a) autor(a).

Dedicatória

Gostaria de dedicar essa dissertação ao meu amado e esperado filho que, em breve, estará fisicamente presente iluminando cada dia da minha vida.

Agradecimentos

Em primeiro lugar, agradeço aos meus pais e ao meu esposo por todo o apoio, incentivo, amor e compreensão durante todos os meus dias e, em especial, durante os períodos mais difíceis dessa trajetória até a conclusão dessa fase.

Ao Prof. Dr. Jorge Luiz Gross, meu orientador, por todo o apoio, incentivo e todos ensinamentos, não só durante os últimos dois anos, mas desde o início de minha formação no Serviço de Endocrinologia do Hospital de Clínicas de Porto Alegre.

À Prof. Dra. Cristiane Bauermann Leitão, não só minha co-orientadora, mas também uma amiga muito especial, por todo carinho, compreensão, ajuda, aprendizado e incentivo.

Aos meus colegas e amigos, Dr. Fernando Gerchman, Dra. Ticiana Rodrigues e Dra. Ângela Reichelt pelo auxílio em tantos momentos durante esses dois anos.

Às minhas colegas Dra. Caroline Karcher Kramer e Lana Catani Ferreira Pinto pela ajuda no desenvolvimento do projeto.

A todos os demais professores do Programa de Pós-Graduação em Endocrinologia e Serviço de Endocrinologia do Hospital de Clínicas de Porto Alegre pelos ensinamentos.

Formato da Dissertação

Essa dissertação de Mestrado segue o formato proposto pelo Programa de Pós-Graduação em Ciências Médicas: Endocrinologia da Universidade de Federal do Rio Grande do Sul, sendo apresentada através de dois capítulos referentes ao tema estudado:

Capítulo 1: Introdução.

Capítulo 2: Artigo original referente ao trabalho realizado, a ser submetido para publicação no periódico *British Medical Journal* e redigido conforme as normas do periódico.

Conteúdo

Lista de Abreviaturas

Capítulo 1 e Capítulo 2

US – Ultrasound / Ultrassonografia

FNA – fine needle aspiration

LHR – Likelihood Ratio

Se – Sensitivity / sensibilidade

Sp - Specificity

OR – Odds Ratio

PAAF – Punção Aspirativa com Agulha Fina

RR – Risco Relativo

RC – Razão de chances

RV – Razão de Verossimilhança

Lista de Tabelas e Figuras

Capítulo 1

Introdução: Nódulos de tireoide – risco de malignidade e avaliação diagnóstica

Capítulo 2

Ultrasound features and risk of thyroid carcinoma: a systematic review and meta-analysis of observational studies

Resumo da dissertação

Introdução: nódulos de tireoide são uma anormalidade comum podendo ser encontrados em até 70% da população com o uso de ultrassonografia de alta resolução. As características ecográficas dos nódulos são tradicionalmente utilizadas como critérios de risco para malignidade, porém sua acurácia diagnóstica é bastante variável na literatura.

Objetivo: avaliar a performance diagnóstica das características ultrassonográficas para carcinoma de tireoide em nódulos não selecionados e naqueles com citologia não diagnóstica na punção aspirativa com agulha fina.

Desenho do estudo: revisão sistemática e meta-análise de estudos observacionais

Bases de dados: bancos de dados eletrônicos (Medline, Embase) até Julho de 2012 além de busca manual nas referências dos principais estudos, artigos de revisão e meta-análises prévias

Critérios de Elegibilidade: estudos que tenham avaliado características ultrassonográficas de nódulos de tireoide e que reportem o exame histopatológico como diagnóstico final. Uma meta-análise em separado foi realizada com os estudos que incluíram apenas nódulos com citologia não diagnóstica (insatisfatória ou indeterminada). As características analisadas foram: ser sólido, hipoecogenicidade, margens irregulares, ausência de halo, presença de microcalcificações, vascularização central, ser solitário, heterogeneidade, formato mais alto do que largo e ausência de elasticidade.

Resultados: 52 estudos observacionais (12786 nódulos) foram incluídos. Onze estudos avaliaram apenas nódulos com citologia não diagnóstica, compreendendo 2338 nódulos. Todas as características avaliadas foram significativamente associadas a malignidade com Razão de Chances (RC) variando de 1,78 a 35,7. Presença de microcalcificações, margens irregulares e uma formato mais alto do que largo apresentaram alta especificidade (87,8%, 83,1%, 96,6%) e Razão de Verossimilhança (RV) (3,26, 2,99, 8,07) nos nódulos não

selecionados. A presença de microcalcificações foi a característica ultrassonográfica com melhor especificidade (84,4%) e RV positiva (3,01) nos nódulos com citologia não diagnóstica. A ausência de elasticidade foi a característica que, isoladamente, mostrou a melhor performance diagnóstica (sensibilidade de 87,9%, especificidade de 86,2% e RV positiva de 6,39).

Conclusão: As características ultrassonográficas avaliadas, isoladamente, não fornecem informações suficientes para selecionar os pacientes que devem ser submetidos a punção aspirativa com agulha fina de tireoide. Nódulos com microcalcificações, margens irregulares, uma formato mais alto do que largo ou ausência de elasticidade demonstraram uma maior probabilidade pós teste para câncer, sugerindo que devem receber manejo mais agressivo. A elastografia é uma técnica promissora, porém mais estudos ainda são necessárias correlacionando seus resultados com o diagnóstico histopatológico, especialmente em nódulos com citologia não diagnóstica.

Capítulo 1

Introdução: Nódulos de tireoide – risco de malignidade e avaliação diagnóstica

Nódulos de tireoide constituem-se em uma anormalidade comum na população em geral. Estima-se que sejam diagnosticados em 5% da população pela simples palpação da glândula. [1] Com o uso mais disseminado de ultrassonografia (US), a prevalência de nódulos de tireoide tem aumentado chegando a perto de 70% da população.[1-5] A importância do diagnóstico e correta avaliação dos nódulos de tireoide baseia-se no risco de carcinoma que pode estar presente em 5 – 15% dos nódulos.[5-10] Tanto a prevalência de nódulos como o risco de malignidade são influenciados por fatores tais como sexo, idade, deficiência de iodo e exposição à radiação ionizante.[11]

A prevalência de nódulos de tireoide é maior em mulheres e aumenta com a idade. Um estudo de triagem populacional demonstrou que em mulheres não tratadas para doenças de tireoide, a prevalência de nódulos aumenta de 9% naquelas abaixo de 25 anos para 45% nas acima de 55 anos. Já em homens, essa prevalência aumentou de 5% para 32% nessas mesmas faixas etárias.[12]

A prevalência de malignidade nos nódulos de tireoide também varia com a idade. Pacientes jovens (abaixo de 18-25 anos) no momento do diagnóstico do nódulo apresentam um risco 1,5 a 2 vezes maior de câncer em comparação com faixas etárias mais avançadas.[10] Pacientes do sexo masculino também apresentam risco aumentado de câncer de tireoide em 1,5 a 2 vezes em relação às mulheres.[10] A exposição prévia, em especial na infância, a radiação ionizante também constitui fator de risco importante para o desenvolvimento de câncer de tireoide. Uma recente análise combinada de dados de 4 coortes de crianças sobreviventes de câncer, incluindo 12930 crianças expostas à radiação, demonstrou que o risco câncer de tireoide aumenta progressivamente com a dose de exposição de radiação até 10Gy, havendo um platô entre 10 e 30Gy e diminuindo a partir de doses maiores que 30Gy.[13] Esse estudo também demonstrou que o RR para câncer após exposição a 10Gy de

radiação foi de 17,5 para crianças expostas antes dos 5 anos de idade e de 3,9 para aquelas expostas após os 15 anos de idade.[13]

A ultrassonografia é indicada em todos os pacientes com nódulos identificados ou suspeitos a palpação.[5] Algumas características ultrassonográficas são consistentemente associadas a malignidade de tireoide, tais como nódulos com ausência de halo, hipoecogenicidade, aumento de vascularização intranodular, formato do nódulo, presença de microcalcificações ou margens irregulares.[14] Entretanto, a acurácia diagnóstica da ultrassonografia é muito variável na literatura e, isoladamente, nenhuma dessas características parece apresentar uma relação entre sensibilidade e especificidade satisfatória para corretamente diagnosticar o câncer de tireoide. As sensibilidades e especificidades dos principais critérios ultrassonográficos utilizados estão descritos na tabela 1.[14-15]

Mais recentemente, a avaliação da elasticidade do nódulo de tireoide tem sido estudada como parâmetro para auxiliar no diagnóstico diferencial de benignidade e malignidade. A elastografia foi inicialmente descrita na avaliação de nódulos de mama e próstata, mas vários estudos avaliaram sua performance em nódulos de tireoide revelando altas sensibilidade e especificidade (81,8 – 97% e 81,1 – 100%, respectivamente).[16-18] Geralmente, a elasticidade do tecido é descrita em uma escala de 1 a 4 sendo que escores de 1-2 representam nódulos com elasticidade em todo ou na maior parte do nódulo (sugestivo de benignidade) e, escores de 3-4 nódulos representam nódulos com pouca ou nenhuma elasticidade em todo ou em sua maior parte (sugestivo de malignidade). Alguns estudos classificam o escore de elasticidade em 1-3 como sugestivo de benignidade e 4-5 de malignidade.[16,17,19,20] Lyshchik e colaboradores avaliaram a acurácia diagnóstica da elastografia em nódulos de tireoide através do índice de elasticidade (calculado a partir da área do nódulo) e demonstraram que um índice maior do que 4 apresentava uma sensibilidade de 82% com especificidade de 96% para o diagnóstico de câncer de tireoide.[21] Uma meta-

análise incluindo 8 estudos e 639 nódulos encontrou uma sensibilidade de 92% e especificidade de 90% para a ausência de elasticidade como preditor de malignidade em nódulos de tireoide. Essa meta-análise, entretanto, incluiu estudos cujo diagnóstico final do nódulo era baseado na avaliação histopatológica ou citopatológica.[19] Um estudo recente demonstrou que a associação da análise de elasticidade aos parâmetros normalmente avaliados na ecografia aumenta a sensibilidade da ecografia no diagnóstico de malignidade em nódulos de tireoide de 85% para 97%.[22]

A punção aspirativa com agulha fina (PAAF) é considerada o método mais fidedigno para a diferenciação entre nódulos benignos e malignos de tireoide e está indicada na maioria dos pacientes com nódulos maiores do que 1 cm.[5] O resultado da análise citopatológica da PAAF é classificada, segundo a classificação de Bethesda em: [23]

- I - Insatisfatória
- II - Benigna
- III - Lesão folicular de significado indeterminado
- IV - Suspeito para neoplasia (neoplasia folicular ou de células de Hürthle)
- V - Suspeito para malignidade
- VI - Maligno

Entretanto, até 10% das PAAF são insatisfatórias e de 15 a 30% apresentam resultado indeterminado (classes III-V), cujo risco de malignidade é de até 30%.[4,5,23] Além disso, realizar a PAAF em todos os pacientes que apresentam nódulo de tireoide não parece factível e gera um número grande de exames desnecessários uma vez que a maioria dos nódulos de tireoide são benignos. Geralmente, a cirurgia é recomendada naqueles casos de nódulos com exame citopatológico confirmadamente insatisfatório ou indeterminado [5], o que leva, também, a uma grande quantidade de cirurgias desnecessárias.

Tentativas têm sido realizadas para melhorar a seleção pré-operatória dos pacientes com PAAFs indeterminadas ou não-diagnósticas em especial a análise citogenética do material da PAAF. Rossi e colaboradores avaliaram a performance diagnóstica da análise da mutação no gene *BRAF* no material da PAAF de 123 nódulos com PAAF indeterminada e encontraram uma sensibilidade de 32,5% com especificidade de 100% para o diagnóstico de malignidade.[24] Nikiforov e colaboradores avaliaram um painel de pesquisa de mutações dos genes BRAF V600E, códon 61 do NRAS, HRAS e códons 12/13 do gene KRAS além de rearranjos no RET/PTC1, RET/PTC3 e PAX8/PPARγ em pacientes com nódulos de tireoide com citologia indeterminada (classes III – V de Bethesda). Nesse estudo, foram incluídos 513 amostras de PAAF e foi encontrada uma sensibilidade global de 61% e especificidade de 97,7%.[25] Outro grupo avaliou o uso de um classificador de expressão gênica que mede a expressão de 167 genes em nódulos de tireoide com citologia indeterminada. A análise incluiu 265 nódulos e foi encontrada uma sensibilidade de 92% e especificidade de 51,6%.[26] Esses testes citogenéticos parecem promissores na seleção pré-operatória de pacientes com citologia indeterminada, mas ainda apresentam alto custo e nenhum dos testes apresenta ainda um relação sensibilidade e especificidade adequada. Nacamulli e colaboradores compararam a acurácia diagnóstica da elastografia associada à pesquisa de mutação no gene BRAF com a associação de ultrassonografia e citologia e encontraram sensibilidades e especificidades de 95% e 74% e, 99% e 28%, respectivamente.[27]

Devido a elevada prevalência de nódulos de tireoide na população em geral e as incertezas quanto a acurácia diagnóstica da ecografia na detecção de nódulos malignos, a seleção correta dos pacientes que devem ser submetidos a PAAF e/ou cirurgia é difícil na prática clínica. Pelo exposto, decidimos conduzir uma revisão sistemática com meta-análise de estudos observacionais que avaliaram a performance diagnóstica da ecografia para a detecção de câncer de tireoide em pacientes com nódulos não selecionados e naqueles com

PAAF não diagnóstica, considerando-se somente o exame histopatológico como método diagnóstico de referência.

Tabela 1: Sensibilidade e especificidade das principais características ultrassonográficas para câncer de tireoide

Característica	Sensibilidade (%)	Especificidade (%)
Presença de microcalcificações	26,1 – 59,1	85,8 – 95
Hipoecogenicidade	26,5 – 87,1	43,4 – 94,3
Vascularização central	54,3 – 74,2	78,6 – 80,8
Formato mais alto que largo	32,7	92,5

Adaptado de Frates et al (9)

Referências

1. Dean DS, Gharib H. Epidemiology of thyroid nodules. *Best Pract Res Clin Endocrinol Metab* 2008;**22**:901-11.

2. Ezzat S, Sarti DA, Cain DR, Braunstein GD. Thyroid incidentalomas. Prevalence by palpation and ultrasonography. *Arch Intern Med* 1994;;**154:**1838-40.

3. Brander A, Viikinkoski P, Nickels J, Kivisaari L. Thyroid gland: US screening in a random adult population. *Radiology* 1991;**181**:683-7.

4. Hegedus L. Clinical practice. The thyroid nodule. *N Engl J Med* 2004;**351**:1764-71.

5. Cooper DS, Doherty GM, Haugen BR, Kloos RT, Lee SL, Mandel SJ, et al. Revised American Thyroid Association management guidelines for patients with thyroid nodules and differentiated thyroid cancer. *Thyroid* 2009;**19**:1167-214.

6. Werk EE, Jr., Vernon BM, Gonzalez JJ, Ungaro PC, McCoy RC. Cancer in thyroid nodules. A community hospital survey. *Arch Intern Med* 1984;**144**:474-6.

7. Frates MC, Benson CB, Doubilet PM, Kunreuther E, Contreras M, Cibas ES, et al. Prevalence and distribution of carcinoma in patients with solitary and multiple thyroid nodules on sonography. *J Clin Endocrinol Metab* 2006;**91**:3411-7.

8. Jin J, McHenry CR. Thyroid incidentaloma. *Best Pract Res Clin Endocrinol Metab* 2012;**26**:83-96.

9. Mazzaferri EL. Management of a solitary thyroid nodule. *N Engl J Med* 1993;**328**:553-59.

10. Alexander EK. Approach to the Patient with a Cytologically Indeterminate Thyroid Nodule. *J Clin Endocrinol Metab* 2008;**93**:4175–82.

11. Tan, GH, Gharib H. Thyroid Incidentalomas: Management Approaches to Nonpalpable Nodules Discovered Incidentally on Thyroid Imaging. *Ann Intern Med* 1997;**126**:226-31.

12. Reiners C, Wegscheider K, Schicha H, Theissen P, Vaupel R, Wrbitzky R et al. Prevalence of thyroid disorders in the working population of Germany: ultrasonography screening in 96,278 unselected employees. *Thyroid* 2004;**14**:926-32.

13. Veiga LH, Lubin JH, Anderson H, de Vathaire F, Tucker M, Bhatti P et al. A pooled analysis of thyroid cancer incidence following radiotherapy for childhood cancer. *Radiat Res* 2012;**178**:365-76.

14. Frates MC, Benson CB, Charboneau JW, Cibas ES, Clark OH, Coleman BG, et al. Management of thyroid nodules detected at US: Society of Radiologists in Ultrasound consensus conference statement. *Radiology* 2005;**237**:794-800.

15. Rago T, Vitti P. Role of thyroid ultrasound in the diagnostic evaluation of thyroid nodules. *Best Pract Res Clin Endocrinol Metab* 2008;**22**:913-28.

16. Rago T, Santini F, Scutari M, Pinchera A, Vitti P. Elastography: new developments in ultrasound for predicting malignancy in thyroid nodules. J Clin Endocrinol Metab 2007;92:2917-22.

17. Asteria C, Giovanardi A, Pizzocaro A, Cozzaglio L, Morabito A, Somalvico F, et al. US-elastography in the differential diagnosis of benign and malignant thyroid nodules. *Thyroid* 2008;**18**:523-31.

18. Rubaltelli L, Corradin S, Dorigo A, Stabilito M, Tregnaghi A, Borsato S, et al. Differential diagnosis of benign and malignant thyroid nodules at elastosonography. *Ultraschall Med* 2009;**30**:175-9.

19. Bojunga J, Herrmann E, Meyer G, Weber S, Zeuzem S, Friedrich-Rust M. Real-time elastography for the differentiation of benign and malignant thyroid nodules: a meta-analysis. *Thyroid* 2010;**20**:1145-50.

20. Ueno E, Ito A. Diagnosis of breast cancer by elasticity imaging. *Eizo Joho Medical*. 2004;36:2-6.

21. Lyshchik A, Higashi T, Asato R, Tanaka S, Ito J, Mai JJ et al. Thyroid gland tumor diagnosis at US elastography. *Radiology* 2005;**237**:202-11.

22. Trimboli P, Guglielmi R, Monti S, Misischi I, Graziano F, Nasrollah N, et al. Ultrasound sensitivity for thyroid malignancy is increased by real-time elastography: a prospective multicenter study. *J Clin Endocrinol Metab* 2012;**97**:4524-30.

23. Cibas ES, Ali SZ. The Bethesda System for Reporting Thyroid Cytopathology. *Thyroid* 2009;**19:**1159-65.

24. Rossi M, Buratto M, Bruni S, Filieri C, Tagliati F, Trasforini G et al. Role of ultrassonographic/clinical profile, cytology, and BRAF V600E mutation evaluation in thyroid nodules screening for malignancy: a prospective study. *J Clin Endocrinol Metab* 2012;**97**:2354-61.

25. Nikiforov YE, Ohori NP, Hodak SP, Carty SE, LeBeau SO, Ferris RL et al. Impact of mutational testing on the diagnosis and management of patients with cytologically indeterminate thyroid nodules: a prospective analysis of 1056 FNA samples. *J Clin Endocrinol Metab* 2011;**96:**3390-97.

26. Alexander EK, Kennedy GC, Baloch ZW, Cibas ES, Chudova D, Diggans J, et al. Preoperative diagnosis of benign thyroid nodules with indeterminate cytology. *N Engl J Med* 2012;**367**:705-15.

27. Nacamulli D, Nico L, Barollo S, Zambonin L, Pennelli G, Girelli ME et al. Comparison of the diagnostic accuracy of combined elastosonography and BRAF analysis vs cytology and ultrasonography for thyroid nodule suspected of malignancy. *Clin Endocrinol (Oxf)* 2012;**77:**608-14.

Capítulo 2

Ultrasound features and risk of thyroid carcinoma: a systematic review and meta-analysis of observational studies *

* Artigo a ser submetido para publicação no periódico *British Medical Journal*

Abstract

Objective: to evaluate the diagnostic performance of ultrasound (US) features for thyroid malignancy in patients with unselected thyroid nodules or nodules with nondiagnostic fine needle aspiration (FNA) cytology submitted to thyroid surgery and having a histopathologic diagnosis.

Design: Systematic review and meta-analysis.

Data sources: Electronic databases (Medline, Embase) to July, 2012 and manual search of references of review articles, previous meta-analyses, and key articles.

Eligibility criteria: Studies evaluating US features of thyroid nodules and reporting histopathologic diagnosis were selected. A separate meta-analysis was performed with studies including only nodules with nondiagnostic cytology. The following features were analyzed: solidity, hypoechogenicity, irregular margins, absence of a halo, microcalcifications, central vascularization, being solitary, heterogeneity, taller than wide shape, and absence of elasticity.

Results: A total of 52 observational studies (12786 nodules) were included. Eleven studies included only patients with nondiagnostic cytology aspirates, comprising 2338 nodules All US features were significantly associated with malignancy, with OR varying from 1.78 to 35.7. Microcalcifications, irregular margins and a taller than wide shape had high specificities (Sp) (87.8%, 83.1%, 96.6%) and positive likelihood ratio (LHR) (3.26, 2.99, 8.07) in unselected nodules. Presence of microcalcifications was the most specific US feature in nodules with a nondiagnostic cytology (Sp 84.4% and positive LHR 3.01). Absence of elasticity was the single feature with the best diagnostic performance (sensitivity 87.9%, Sp 86.2% and positive LHR 6.39).

Conclusions: US features evaluated do not provide reliable information to select nodules that should have a fine-needle aspiration performed. Nodules with microcalcifications, or a taller than wide shape, or irregular margins, or absence of elasticity showed higher likelihood ratios

and, consequently, post test probabilities of malignancy and should be more aggressively managed. Elastography is a promising technique, but further studies are still required to compare it to the reference method, especially in nodules with nondiagnostic cytology.

Introduction

Thyroid nodules are a common finding in the general population and their detection is increasing with the widespread use of ultrasound (US). The prevalence of thyroid nodularity varies from 19 to 67% and increases with age, affecting about 50% of the population over 40 years.(1-5) The clinical significance of thyroid nodules relates to the need to exclude thyroid cancer, which is found in 5-15% of cases, depending on sex, age, and exposure to other risk factors.(5-8)

Some US parameters, such as microcalcifications, hypoechogenicity, absence of a halo, increased intranodular vascularity, nodule shape or irregular margins have been traditionally associated with increased risk of malignancies.(9) However, none of these characteristics seems sufficiently reliable in isolation to diagnose malignancy. Diagnostic sensitivity ranges from 26.5% to 87.1% for hypoechogenicity, 54.3 to 74.32% for intranodular vascularity, 26.1 to 59.1 for microcalcifications, whereas specificity ranges from 43.4 to 94.3%, 78.6 to 80.8% and 85.8 to 95% respectively.(4, 9, 10) More recently, US determination of tissue elasticity (elastography) has been suggested to detect malignancy in thyroid nodules. A recent meta-analysis found sensitivity of 92% and specificity of 90% using this technique. However, only a few studies were included, and only three used histopathology of surgical specimens for final diagnosis .(11) Fine-needle aspiration (FNA) is considered the most accurate procedure to differentiate benign and malignant nodules and it is indicated in most patients with nodules larger than 1 centimeter.(5) However, the performance of biopsies in all patients harboring a thyroid nodule is too burdensome. Moreover up to 10% of FNA specimens are insufficient for diagnosis and 15-30% are indeterminate, carrying a 20-30% risk of malignancy.(4, 5) Surgery has been usually recommended in patients with confirmed unsatisfactory or indeterminate cytology, but this may lead to a high rate of

unnecessary thyroidectomy.(5) Although cytogenetic studies may improve diagnostic accuracy, the reported specificity is still low, around 50%.(12)

Therefore the aim of this study was to conduct a systematic review and meta-analysis of observational studies evaluating the diagnostic performance of US features for thyroid malignancy in patients with unselected thyroid nodules or nodules with non diagnostic FNA cytology, considering only histopathologic diagnosis of surgical specimens as the final diagnosis.

Methods

Identification of studies

We searched MEDLINE using the following Medical Subject Heading terms: "Thyroid Nodule"[MeSH] AND ("Ultrasonography"[MeSH] OR "ultrasonography"[Subheading] OR "Ultrasonography, Doppler"[MeSH]). EMBASE was searched using EmTree terms “Thyroid nodule” and “Ultrasonography.” The search period ended in July, 2012. A manual search of the references of review articles, previous meta-analyses and key articles was also performed. We also searched on abstracts of main endocrine mettings. All potentially eligible studies were considered for review regardless of the primary outcome or language.

Selection criteria

Observational studies of patients with thyroid nodules evaluated by US and submitted to thyroidectomy regardless of the reason for surgery considered for inclusion. Only studies with histopathologic diagnosis of surgical specimens were considered.

Study Selection, Data Extraction and Quality Assessment

Two independent investigators (LLRR and CKK) selected potentially eligible studies based on titles and abstracts. All the studies selected were retrieved for full-text evaluation. Disagreements were solved by a third investigator (CBL). Two investigators reviewed the

studies selected to extract data on patient characteristics, US features and histopathologic results. Any discrepancies between the data extracted were discussed until a consensus was reached. We extracted the absolute number of patients with and without the evaluated features and with and without malignancy. These data were entered into a computerized spreadsheet considering true positives, true negatives, false positives and false negatives.

We evaluated the diagnostic ability to diagnose thyroid malignancy of the following US features: solidity, hypoechogenicity, irregular margins, absence of halo, microcalcifications, central vascularization, being solitary, heterogeneity, taller than wide shape, and absence of elasticity.

Two independent investigators (LRR and LCFP) evaluated the quality of the included studies was assessed using QUADAS-2 tool. (13) Any disagreements were solved by a third investigator (CBL).

Statistical analysis

Overall odds ratio (OR) was calculated to assess the predictive value of each US feature for malignancy. The Cochran χ^2 and the I^2 tests were used to evaluate heterogeneity among studies and a threshold value of P = 0.10 was considered significant. Risk estimates were obtained with a random effects meta-analysis if significant heterogeneity was found among the studies in preliminary models.

The pooled positive and negative likelihood ratios were calculated, as well as post-test probabilities,(14) using a mean pretest probability of 10% based on the average of malignancy found in thyroid nodules in general.(5-8) Likelihood ratios above 10 or below 0.1 are considered strong evidence to, respectively, confirm or rule out the diagnosis of interest.(14)

The possibility of publication bias was evaluated using a funnel plot of a trial's effect size against the SE. Funnel plot asymmetry was analyzed by the Begg and Egger tests. Trim-and-fill computation was used to estimate the effect of publication bias.

We performed a separate prespecified meta-analysis including only studies that selected patients with nodules with a nondiagnostic cytology. We defined as nondiagnostic cytology those reported as unsatisfactory or indeterminate.

All statistical analyses were performed using Stata 11.0 software.

Results

The initial search retrieved 1917 articles, of which 1766 were excluded based on title and abstract. Full text assessment was performed in 151 and, of these, 53 were selected for the present study (Figure 1). One additional study (Kim et al) evaluating only nodules with egg-shell calcifications was excluded. Therefore, 52 studies comprising 12786 nodules were included in the analysis.(15-66) Eleven studies including only patients with nondiagnostic cytology aspirates, comprising 2338 nodules, were included in the separate meta-analysis. The characteristics of the included studies are described in Table 1.

High heterogeneity was identified in the analysis of all but two features (heterogeneity and having a taller than wide shape); therefore, the random effects model was used. Funnel plot and Egger test suggested a publication bias on analysis of heterogeneity, hypoechogenicity, solidity and central vascularization features when considering all unselected nodules. However, trim and fill computation revealed that publication bias did not interfere with the interpretation of results.

Quality of studies

Included studies had, in general, low risk of bias. The most concerning issue was the lack of description if the US assessor was blinded for the histopathologic diagnosis. As US has to be performed prior to surgery, we believe that, in most cases, this person was not aware of the histopathologic diagnosis. However, as US results may have been reviewed after surgery, we considered as a risk of bias unclear in this item if there was no description about

blindness. We considered that some studies may have concerns about the applicability due to patient selection, in most cases, because studies included only patients with cold nodules.

Diagnostic performance of US features in all nodules

All the features evaluated were significantly associated with malignancy, with an overall OR ranging from 1.77 to 35.7 (figure 2). However, the sensitivity of US features traditionally associated with malignancy was somewhat low, ranging from 26.7 to 63%. Four of these features – microcalcifications, central vascularization, irregular margins, and a taller than wide shape – showed better specificity than the other features: 87.8%, 78%, 83.1% and 96.6% respectively. The positive likelihood ratio ranged from 1.33 to 8.07, and the negative likelihood ratio from 0.13 to 0.77 (table 2). Considering a pre-test probability of 10%, the post-test probability of malignancy ranged from 12.8 to 47.0% after a positive test and 1.4 to 7.8% with a negative test result. Absence of elasticity was the US feature that showed the best diagnostic accuracy, with sensitivity of 87.9%, specificity of 86.2%, and positive and negative LHR of 6.39 and 0.13 respectively (table 2).

Diagnostic performance of US features in nodules with nondiagnostic cytology

Only a few of the studies reported histopathologic diagnosis specifically for nodules with a nondiagnostic cytology. Because of that, only the following features were analyzed: absence of elasticity, hypoechogenicity, solidity, presence of microcalcifications, being solitary, and central vascularization. Of these, only hypoechogenicity and presence of microcalfcifications were significantly associated with malignancy (figure 3). Still in this subgroup of nodules, any of the US features was able to determinate the risk of malignancy with an acceptable sensitivity. Presence of microcalcifications was the feature with the best specificity, 84.4%. Positive likelihood ratio ranged from 1.06 to 3.01 and negative likelihood ratio from 0.63 to 0.96. Considering a pre-test probability of 10%, the post-test probability of malignancy ranged from 10.4 to 24.9% after a positive test and 6.5 to 9.6% with a negative

test result. We were not able to determinate the diagnostic accuracy of elastography or central vascularization in this group because the number of studies was not sufficient.

Meta-regression

In the analysis of several features, we have included less than ten studies making meta-regression not suitable. For analysis of hypoechogenicity, irregular margins, microcalcifications, being solid and central vascularization we performed meta-regression using the year of publication and/or the prevalence of cancer in the study sample as covariates. However none of these variables was able to significantly explain the high heterogeneity found.

Discussion

In the present meta-analysis, none of the analyzed US features had reliable diagnostic performance to identify malignancy in unselected thyroid nodules or in nodules with nondiagnostic cytology. None of the US features analyzed had a relevant positive likelihood ratio (above 10) and post-test probabilities, so they did not provide strong evidence to diagnose malignancy. However, some US features were associated with higher post-test probability: taller than wide shape (47.0%), absence of elasticity (41.3%), presence of microcalcifications (26.4%), and irregular margins (24.7%).

The strengths of our meta-analysis are the large number of nodules evaluated and the fact that all the nodules included had a histopathologic diagnosis, which is the reference method for the definite diagnosis of thyroid nodules. Moreover, we also analyzed the performance of US in nodules with nondiagnostic cytology, which constitutes the most challenging group of patients for clinical decision-making To the best of our knowledge, there are no previous systematic reviews and meta-analyses that focus on histopathology only instead of cytopathology results obtained by FNA as the final diagnosis. Our study has some limitations. First, we had no information on individual characteristics of patients regarding

risk factors for malignancy, and on the reason for surgery. Also, the number of studies was insufficient for the analysis of some US features in patients with nondiagnostic cytology, possibly the subgroup of patients that would most benefit from the use of US as a tool to help in clinical management decision.

Our results confirm the findings of previous isolated studies. Moon et al.(67) evaluated 831 patients with thyroid nodules and found low sensitivity values for most of the US features. Hypoechogenicity was the only finding that showed a sensitivity of 87.2%. In the same study, taller than wide shape, speculated margins, marked hypoechogenicity, micro and macrocalcifications features demonstrated a high specificity for malignancy, ranging from 90.8 to 96.1%. In one of the largest series comprising 672 patients and 1141 nodules, Popovicz et al. also found low sensitivity values of US features for malignancy and high specificity for microcalcifications and taller than wide shape features.(48) Moreover, in another study including 550 patients with multinodular goiter, Salmaslioglu found that the presence of microcalcifications had a high sensitivity of 89.3% for malignancy.(55) The best diagnostic performance in our meta-analysis was seen for absence of elasticity. Usually, elasticity is described in a scale ranging from 1 to 4 (1-2 being suggestive of a benign nodule and 3-4 of malignancy) or 1 to 5 (where 1-3 is suggestive of a benign lesion and 4-5 of malignancy).(11,51,68,69) This US feature was initially described for breast or prostate cancer, but several studies have evaluated its performance to differentiate between malignant and benign thyroid nodules, revealing high sensitivity and specificity (81.8 - 97% and 81.1 – 100%).(51,69,70) A recent meta-analysis including 8 studies with a total of 639 nodules diagnosed by FNA cytopathology or histopathology reported a sensitivity of 90% and specificity of 92% for elasticity, however not all studies included had a final histopathologic diagnosis of the nodule.(11) A recent study, which included 498 thyroid nodules evaluated by

US, color flux Doppler, and real-time elastography concluded that the combination of elastography with US parameters increased the sensitivity for malignancy to 97%.(71)

Our findings have important clinical implications. They reinforce that US results on their own are not reliable for selecting which patients with thyroid nodules should be submitted to fine-needle aspiration. Our findings also suggest that more accurate criteria are needed to recommend surgery in patients with nondiagnostic cytology. This is an important practical matter since we need to be able to better select which patients should be submitted to FNA and, specially, when surgery should be indicated in those nodules with nondiagnostic cytology.

Attempts have been made to improve patient selection in evaluation of thyroid nodules. Moon et al. evaluated a classification that considered as suspicious for malignancy a nodule that was solid plus having two additional risk features. Those authors found sensitivity of 87.7%, specificity of 97.8%, and overall accuracy of 96.2%.(72) Recently, molecular analysis of fine-needle aspirates has been evaluated. Nikiforov et al. have demonstrated a specificity of 96-99% for malignancy in cytologically indeterminate thyroid nodules using a panel of mutations that includes the most common mutations found in thyroid cancer. However, in that study, the panel sensitivity was only 57-68%, producing a false-negative rate of 6 to 28%.(73) In another study, a gene-expression classifier including 167 transcript cluster identification number was tested in thyroid nodules with indeterminate cytology reaching a sensitivity of 92%, but specificity of only 52%.(12)

Conclusions

Our results show that there is no isolated US feature capable of predicting malignancy in thyroid nodules with acceptable diagnostic accuracy, which limits the value of the US for decision-making regarding the need for FNA or surgery. However, the presence of some US features, such as a taller than wide shape and microcalcifications is suggestive of malignancy,

favoring a more aggressive management of nodules with these characteristics. Ideally, meta-analyses should be performed with individual patient data, which could enable the creation of a risk classification for malignancy in thyroid nodules considering all US features and other risk factors to better define which patients should be submitted to FNA and surgery. Elastography is a new technique with promising results and may be a good tool to select patients at increased risk for thyroid malignancy. Nevertheless, more studies are still required to standardize the technique and confirm its usefulness. .

Ethical approval: Not needed.

What is already known on this topic
Thyroid nodules are a common finding n the general population which carry a 5–15% risk of malignancy
Some US features are associated with malignancy, but their diagnostic accuracy to diagnose or exclude malignancy are low
Many unnecessary thyroidectomy are still performed
What this study adds
US features in isolation have no sufficient diagnostic performance to be used as a criteria to select patients for FNA or surgery
Microcalcifications and a taller than wide shape are more specific for malignancy suggesting that nodules whit these characteristics should be more aggressively managed
Elastography is a promising technique with a good diagnostic performance but more studies are still required

References

1. Brander A, Viikinkoski P, Nickels J, Kivisaari L. Thyroid gland: US screening in a random adult population. *Radiology* 1991;**181**:683-7.

2. Ezzat S, Sarti DA, Cain DR, Braunstein GD. Thyroid incidentalomas. Prevalence by palpation and ultrasonography. *Arch Intern Med* 1994;;**154:**1838-40.

3. Dean DS, Gharib H. Epidemiology of thyroid nodules. *Best Pract Res Clin Endocrinol Metab* 2008;**22**:901-11.

4. Hegedus L. Clinical practice. The thyroid nodule. *N Engl J Med* 2004;**351**:1764-71.

5. Cooper DS, Doherty GM, Haugen BR, Kloos RT, Lee SL, Mandel SJ, et al. Revised American Thyroid Association management guidelines for patients with thyroid nodules and differentiated thyroid cancer. *Thyroid* 2009;**19**:1167-214.

6. Werk EE, Jr., Vernon BM, Gonzalez JJ, Ungaro PC, McCoy RC. Cancer in thyroid nodules. A community hospital survey. *Arch Intern Med* 1984;**144**:474-6.

7. Frates MC, Benson CB, Doubilet PM, Kunreuther E, Contreras M, Cibas ES, et al. Prevalence and distribution of carcinoma in patients with solitary and multiple thyroid nodules on sonography. *J Clin Endocrinol Metab* 2006;**91**:3411-7.

8. Jin J, McHenry CR. Thyroid incidentaloma. *Best Pract Res Clin Endocrinol Metab* 2012;**26**:83-96.

9. Frates MC, Benson CB, Charboneau JW, Cibas ES, Clark OH, Coleman BG, et al. Management of thyroid nodules detected at US: Society of Radiologists in Ultrasound consensus conference statement. *Radiology* 2005;**237**:794-800.

10. Rago T, Vitti P. Role of thyroid ultrasound in the diagnostic evaluation of thyroid nodules. *Best Pract Res Clin Endocrinol Metab* 2008;**22**:913-28.

11. Bojunga J, Herrmann E, Meyer G, Weber S, Zeuzem S, Friedrich-Rust M. Real-time elastography for the differentiation of benign and malignant thyroid nodules: a meta-analysis. *Thyroid* 2010;**20**:1145-50.

12. Alexander EK, Kennedy GC, Baloch ZW, Cibas ES, Chudova D, Diggans J, et al. Preoperative diagnosis of benign thyroid nodules with indeterminate cytology. *N Engl J Med* 2012;**367**:705-15.

13. Whiting PF, Rutjes AWS, Westwood ME, Mallett S, Deeks JJ, Reitsma JB et al. QUADAS-2: a revised tool for the quality assessment of diagnostic accuracy studies. *Ann Intern Med* 2011;**155:**529-36.

14. Deeks JJ, Altman DG. Diagnostic tests 4: likelihood ratios. *BMJ* 2004;**329**:168-9.

15. Aggarwal SK, Jayaram G, Kakar A, Goel GD, Prakash R, Pant CS. Fine needle aspiration cytologic diagnosis of the solitary cold thyroid nodule. Comparison with ultrasonography, radionuclide perfusion study and xeroradiography. *Acta Cytol* 1989;**33**:41-7.

16. Alexopoulou O, Beguin C, Buysschaert M, Squifflet JP, De Burbure C, De Nayer P, et al. Predictive factors of thyroid carcinoma in non-toxic multinodular goitre. *Acta Clin Bel* 2004;**59**:84-9.

17. Bakhshaee M, Davoudi Y, Mehrabi M, Layegh P, Mirsadaee S, Rad MP, et al. Vascular pattern and spectral parameters of power Doppler ultrasound as predictors of malignancy risk in thyroid nodules. *Laryngoscope* 2008;**118**:2182-6.

18. Bozbora A, Erbil Y, Ozarmagan S, Barbaros U, Sari S, Degirmenci B. Color Doppler sonography in cold thyroid nodules for malignancy prediction. *Acta Chir Belg* 2002;**102**:259-62.

19. Brkljacic B, Cuk V, Tomic-Brzac H, Bence-Zigman Z, Delic-Brkljacic D, Drinkovic I. Ultrasonic evaluation of benign and malignant nodules in echographically multinodular thyroids. *J Clin Ultrasound* 1994;**22**:71-6.

20. Cakir B, Aydin C, Korukluoglu B, Ozdemir D, Sisman IC, Tuzun D, et al. Diagnostic value of elastosonographically determined strain index in the differential diagnosis of benign and malignant thyroid nodules. *Endocrine* 2011;**39**:89-98.

21. Cantisani V, D'Andrea V, Biancari F, Medvedyeva O, Di Segni M, Olive M, et al. Prospective evaluation of multiparametric ultrasound and quantitative elastosonography in the differential diagnosis of benign and malignant thyroid nodules: Preliminary experience. *Eur J Radiol* 2012;**81**:2678-83.

22. Cappelli C, Castellano M, Pirola I, Cumetti D, Agosti B, Gandossi E, et al. The predictive value of ultrasound findings in the management of thyroid nodules. *QJM* 2007;**100**:29-35.

23. Casella C, Talarico C, La Pinta M, Nascimbeni R, Di Fabio F, Salerni B. The role of color flow-Doppler ultrasonography in the diagnosis of nodular goiter. *Ann Ital Chir* 2003;**74**:495-9.

24. Castro MR, Espiritu RP, Bahn RS, Henry MR, Gharib H, Caraballo PJ, et al. Predictors of malignancy in patients with cytologically suspicious thyroid nodules. *Thyroid* 2011;**21**:1191-8.

25. Chen G, Zhu XQ, Zou X, Yao J, Liang JX, Huang HB, et al. Retrospective analysis of thyroid nodules by clinical and pathological characteristics, and ultrasonographically detected calcification correlated to thyroid carcinoma in South China. *Eur Surg Res* 2009;**42**:137-42.

26. Choi YJ, Kim SM, Choi SI. Diagnostic accuracy of ultrasound features in thyroid microcarcinomas. *Endocr J* 2008;**55**:931-8.

27. Cox MR, Marshall SG, Spence RAJ. Solitary thyroid nodule: A prospective evaluation of nuclear scanning and ultrasonography. *Br J Surg* 1991;**78**:90-3.

28. De Nicola H, Szejnfeld J, Logullo AF, Wolosker AMB, Souza LRMF, Chiferi Jr V. Flow pattern and vascular resistive index as predictors of malignancy risk in thyroid follicular neoplasms. *J Ultrasound Med* 2005;**24**:897-904.

29. Fukunari N, Nagahama M, Sugino K, Mimura T, Ito K. Clinical evaluation of color doppler imaging for the differential diagnosis of thyroid follicular lesions. *World JSurg*. 2004;**28**:1261-5.

30. Ghervan CMV, Dumitriu D, Botar-Jid C, Dudea SM, Muntean V, Duncea IM. Ultrasound elastography a valuable method to exclude thyroid malignancy. *Endocr Rev* 2011;**32**(3):P1-702.

31. Giammanco M, Di Gesu G, Massenti MF, Di Trapani B, Vetri G. Role of color flow Doppler sonography in pre-operative diagnostics of the thyroid pathology. *Minerva Endocrinol* 2002;**27**:1-10.

32. Gulcelik NE, Gulcelik MA, Kuru B. Risk of malignancy in patients with follicular neoplasm: predictive value of clinical and ultrasonographic features. *Arch Otolaryngol Head Neck Surg* 2008;**134**:1312-5.

33. Hong Y, Liu X, Li Z, Zhang X, Chen M, Luo Z. Real-time ultrasound elastography in the differential diagnosis of benign and malignant thyroid nodules. *J Ultrasound Med* 2009;**28**:861-7.

34. Hubsch P, Niederle B, Barton P, Pesau B, Knittel M, Schratter M, et al. Color-coded Doppler sonography of the thyroid: an advance in carcinoma diagnosis? *Rofo* 1992;**156**:125-9.

35. Kakkos SK, Scopa CD, Chalmoukis AK, Karachalios DA, Spiliotis JD, Harkoftakis JG, et al. Relative risk of cancer in sonographically detected thyroid nodules with calcifications. *J Clin Ultrasound* 2000;**28**:347-52.

36. Khoo ML, Asa SL, Witterick IJ, Freeman JL. Thyroid calcification and its association with thyroid carcinoma. *Head Neck* 2002;**24**:651-5.

37. Kobayashi K, Fukata S, Miyauchi A. Diagnosis of follicular carcinoma of the thyroid: Role of sonography in preoperative diagnosis of follicular nodules. *J Med Ultrasonics* 2005;**32**:153-8.

38. Kountakis SE, Skoulas IG, Maillard AAJ. The radiologic work-up in thyroid surgery: Fine-needle biopsy versus scintigraphy and ultrasound. *Ear, Nose Throat J* 2002;**81**:151-4.

39. Leenhardt L, Menegaux F, Franc B, Delbot T, Mansour G, Hoang C, et al. Selection of patients with solitary thyroid nodules for operation. *Eur J Surg* 2002;**168**:236-41.

40. Lippolis PV, Tognini S, Materazzi G, Polini A, Mancini R, Ambrosini CE, et al. Is elastography actually useful in the presurgical selection of thyroid nodules with indeterminate cytology? *J Clin Endocrinol Metab* 2011;**96**:E1826-E30.

41. Liu FH, Hsueh C, Chang HY, Liou MJ, Huang BY, Lin JD. Sonography and fine-needle aspiration biopsy in the diagnosis of benign versus malignant nodules in patients with autoimmune thyroiditis. *J Clin Ultrasound* 2009;**37**:487-92.

42. Maia FFR, Matos PS, Silva BP, Pallone AT, Pavin EJ, Vassallo J, et al. Role of ultrasound, clinical and scintigraphyc parameters to predict malignancy in thyroid nodule. *Head Neck Oncol* 2011;**3**: doi: 10.1186/1758-3284-3-17 .

43. Mendelson AA, Tamilia M, Rivera J, Hier MP, Sherman M, Garfield N, et al. Predictors of malignancy in preoperative nondiagnostic biopsies of the thyroid. *J Otolaryngol Head Neck Surg* 2009;**38**:395-400.

44. Ousehal A, Abdelouafi A, Essodegui F, Ouzidane L, Moumen M, Kadiri R. Contribution of ultrasonography in thyroid diseases. Apropos of 100 cases. *Ann Radiol (Paris)* 1996;**39**:146-52.

45. Peccin S, De Castro JAS, Furlanetto TW, Furtado APA, Brasil BA, Czepielewski MA. Ultrasonography: Is it useful in the diagnosis of cancer in thyroid nodules? *J Endocrinol Invest* 2002;**25**:39-43.

46. Penfold A, Vargas Perez C, Chipolla A, Civeriatti O, Macagno G, Lopez Vinuesa F, et al. The ultrasonography of high resolution and color Doppler in the nodular pathology thyroid - Correlato pathological anatomo. Rev Argent Endocrinol Metab 2004;**41**:131-42.

47. Phuttharak W, Somboonporn C, Hongdomnern G. Diagnostic performance of gray-scale versus combined gray-scale with colour doppler ultrasonography in the diagnosis of malignancy in thyroid nodules. *Asian Pac J Cancer Prev* 2009;**10**:759-64.

48. Popowicz B, Klencki M, Lewinski A, Slowinska-Klencka D. The usefulness of sonographic features in selection of thyroid nodules for biopsy in relation to the nodule's size. *Eur J Endocrinol* 2009;**161**:103-11.

49. Popowicz B, Slowinska-Klencka D, Sporny S, Klencki M, Lewinski A. Small lesions of the thyroid gland--the significance of ultrasound examination in the selection of lesions for biopsy. *Endokrynol Pol* 2006;**57**:292-8.

50. Rago T, Di Coscio G, Basolo F, Scutari M, Elisei R, Berti P, et al. Combined clinical, thyroid ultrasound and cytological features help to predict thyroid malignancy in follicular and Hurthle cell thyroid lesions: Results from a series of 505 consecutive patients. *Clin Endocrinol (Oxf)* 2007;**66**:13-20.

51. Rago T, Santini F, Scutari M, Pinchera A, Vitti P. Elastography: new developments in ultrasound for predicting malignancy in thyroid nodules. *J Clin Endocrinol Metab* 2007;**92**:2917-22.

52. Rago T, Scutari M, Santini F, Loiacono V, Piaggi P, Di Coscio G, et al. Real-time elastosonography: useful tool for refining the presurgical diagnosis in thyroid nodules with indeterminate or nondiagnostic cytology. *J Clin Endocrinol Metab* 2010;**95**:5274-80.

53. Rago T, Vitti P, Chiovato L, Mazzeo S, De Liperi A, Miccoli P, et al. Role of conventional ultrasonography and color flow-doppler sonography in predicting malignancy in 'cold' thyroid nodules. *Eur J Endocrinol* 1998;**138**:41-6.

54. Sahin M, Sengul A, Berki Z, Tutuncu NB, Guvener ND. Ultrasound-guided fine-needle aspiration biopsy and ultrasonographic features of infracentimetric nodules in patients with nodular goiter: Correlation with pathological findings. *Endocr Pathol* 2006;**17**:67-74.

55. Salmaslioglu A, Erbil Y, Dural C, Issever H, Kapran Y, Ozarmagan S, et al. Predictive value of sonographic features in preoperative evaluation of malignant thyroid nodules in a multinodular goiter. *World J Surg* 2008;**32**:1948-54.

56. Schueller-Weidekamm C, Schueller G, Kaserer K, Scheuba C, Ringl H, Weber M, et al. Diagnostic value of sonography, ultrasound-guided fine-needle aspiration cytology, and diffusion-weighted MRI in the characterization of cold thyroid nodules. *Eur J Radiol* 2010;**73**:538-44.

57. Seiberling KA, Dutra JC, Grant T, Bajramovic S. Role of intrathyroidal calcifications detected on ultrasound as a marker of malignancy. *Laryngoscope* 2004;**114**:1753-7.

58. Sharma R, Chakravarty KL, Tripathi M, Kaushik A, Bharti P, Sahoo M, et al. Role of 99mTc-Tetrofosmin delayed scintigraphy and color Doppler sonography in characterization of solitary thyroid nodules. *Nucl Med Commun* 2007;**28**:847-51.

59. Shuzhen C. Comparison analysis between conventional ultrasonography and ultrasound elastography of thyroid nodules. *Eur J Radiol* 2012**;81**:1806-11**.**

60. Sillery JC, Reading CC, Charboneau JW, Henrichsen TL, Hay ID, Mandrekar JN. Thyroid follicular carcinoma: Sonographic features of 50 cases. *Am J Roentgenol* 2010;**194**:44-54.

61. Sippel RS, Elaraj DM, Khanafshar E, Kebebew E, Duh QY, Clark OH. Does the presence of additional thyroid nodules on ultrasound alter the risk of malignancy in patients with a follicular neoplasm of the thyroid? *Surgery* 2007;**142**:851-7.

62. Varverakis E, Neonakis E, Tzardi M, Chrysos E. Role of color Doppler ultrasonography in the preoperative management of cold thyroid nodules. *Hormones (Athens)* 2007;**6**:44-51.

63. Walker J, Findlay D, Amar SS. A prospective study of thyroid ultrasound scan in the clinically solitary thyroid nodule. *Br J Radiol* 1985;**58**:617-9.

64. Wang N, Xu Y, Ge C, Guo R, Guo K. Association of sonographically detected calcification with thyroid carcinoma. *Head Neck* 2006;**28**:1077-83.

65. Wang Y, Dan HJ, Dan HY, Li T, Hu B. Differential diagnosis of small single solid thyroid nodules using real-time ultrasound elastography. *J Int Med Res* 2010;**38**:466-72.

66. Yoon JH, Kwak JY, Kim EK, Moon HJ, Kim MJ, Kim JY, et al. How to approach thyroid nodules with indeterminate cytology. *Ann Surg Oncol* 2010;**17**:2147-55.

67. Moon WJ, Jung SL, Lee JH, Na DG, Baek JH, Lee YH, et al. Benign and malignant thyroid nodules: US differentiation--multicenter retrospective study. *Radiology* 2008;**247**:762-70.

68. Ueno E, Ito A. Diagnosis of breast cancer by elasticity imaging. *Eizo Joho Medical*. 2004;36:2-6.

69. Asteria C, Giovanardi A, Pizzocaro A, Cozzaglio L, Morabito A, Somalvico F, et al. US-elastography in the differential diagnosis of benign and malignant thyroid nodules. *Thyroid* 2008;**18**:523-31.

70. Rubaltelli L, Corradin S, Dorigo A, Stabilito M, Tregnaghi A, Borsato S, et al. Differential diagnosis of benign and malignant thyroid nodules at elastosonography. *Ultraschall Med* 2009;**30**:175-9.

71. Trimboli P, Guglielmi R, Monti S, Misischi I, Graziano F, Nasrollah N, et al. Ultrasound sensitivity for thyroid malignancy is increased by real-time elastography: a prospective multicenter study. *J Clin Endocrinol Metab* 2012;**97**:4524-30.

72. Moon HG, Jung EJ, Park ST, Ha WS, Choi SK, Hong SC, et al. Role of ultrasonography in predicting malignancy in patients with thyroid nodules. *World J Surg* 2007;**31**:1410-6.

73. Nikiforov YE, Ohori NP, Hodak SP, Carty SE, LeBeau SO, Ferris RL, et al. Impact of mutational testing on the diagnosis and management of patients with cytologically indeterminate thyroid nodules: a prospective analysis of 1056 FNA samples. *J Clin Endocrinol Metab* 2011;**96**:3390-7.

Appendix - Tables

Table 1: Characteristics of studies included in meta-analysis

Author	Year	Number of nodules	Male sex (%)	Age (mean)	Clinical background	US features evaluated
Walker J (62)	1985	94	14.8		All patients submitted to surgery	Solid
Aggarwal S (14)	1989	36			Patients with cold nodules	Solid
Cox M (26)	1991	68	10.0	48.6	Patients submitted to surgery with no compressive symptoms or obvious malignancy	Solitary, solid
Hübsch V (33)	1992	65	30.0	42.0	Patients with cold nodules, compressive symptoms, hyperthyroidism or malignancy suspected or confirmed	Hypoechogenicity, microcalcifications, central vascularization, solitary, solid, heterogeneity and irregular margins
Brkljacic B (18)	1994	426	13.3	46.5	Patients with multinodular goiter	Hypoechogenicity, microcalcifications
Ousehal A (43)	1996	100		39.0	Unselected patients submitted to surgery	Hypoechogenicity, irregular margins
Rago T (52)	1998	104	33.0	42.3	Patients with single nodule with compressive symptoms or suspicion of malignancy	Hypoechogenicity, absence of halo, microcalcifications
Kakkos S (34)	2000	188			All patients submitted to surgery	Microcalcifications
Bozbora A (17)	2002	81	25.0	33.0	Patients with cold solitary or dominant nodule	Solitary
Giammanco M (30)	2002	125	21.6	57.0	All patients submitted to surgery	Central vascularization
Khoo M (35)	2002	361			Consecutive patients submitted to surgery	Microcalcifications
Kountakis S (37)	2002	83			All patients submitted to surgery	Solitary
Leenhardt L (38)	2002	155	25.1		Patients submitted to surgery with no hormonal dysfunction	Hypoechogenicity, microcalcifications, solid, irregular margins
Peccin S (44)	2002	80	20.0	45.3	Patients with compressive symptoms or suspicion of malignancy	Hypoechogenicity, absence of halo, microcalcifications
Casella C (22)	2003	66	15.1	44.6	Patients submitted to surgery	Central vascularization
Alexopoulou O (15)	2004	109			Patients with nontoxic multinodular goiter	Hypoechogenicity, microcalcifications
Fukunari N (28)	2004	310	14.1	47.0	Patients with cold solitary nodules	Central vascularization
Penfold A (45)	2004	83	13.2		Patients submitted to surgery	Hypoechogenicity, absence of halo, microcalcifications, central vascularization, irregular margins

Seiberling K (56)	2004	159	23.2	46.0	Patients submitted to surgery	Microcalcifications
Kobayashi K (36)	2005	910		49.0	Patients submitted to surgery with diagnosis of follicular nodule	Solitary, solid, irregular margins
Nicola H (27)	2005	86			Patients with follicular neoplasms on FNA	Central vascularization
Popowicz B (48)	2006	356			Patients submitted to surgery	Microcalcifications, solitary
Sahin M (53)	2006	472	17.8	51.5	Patients submitted to surgery	Hypoechogenicity, absence of halo, microcalcifications, solid, heterogeneity, irregular margins
Wang N (63)	2006	322	18.4	44.0	Patients submitted to surgery	Microcalcifications
Cappelli C (21)	2006	349			Nodules with malignant or suspicious cytology	Hypoechogenicity, microcalcifications, central vascularization, irregular margins
Rago T (49)	2007	505	21.1	45.0	Cold nodules with cytological diagnosis of follicular or Hürthle cell lesion	Hypoechogenicity, microcalcifications, irregular margins
Rago T (50)	2007	92	31.5	41.6	Patients submitted to surgery for compressive symptons or FNA suspicious	Hypoechogenicity, absence of halo, microcalcifications, central vascularization, elasticity
Sharma R (57)	2007	52	26.9		Patients with cold solitary nodules	Central vascularization
Sippel R (60)	2007	325	18.5	47.0	Patient with FNA diagnosis of follicular or Hürthle cell neoplasm or indeterminate	Hypochogenicity, central vascularization, solid, heterogeneity
Varverakis E (61)	2007	85	20.0		Patients submitted to surgery due to risk of malignancy or compressive symptoms	Central vascularization
Bakhshaee M (16)	2008	85	13.0	36.8	Patients submitted to surgery due to |FNA diagnosis or obstructive or cosmetic reasons	Central vascularization, solid
Choi Y (25)	2008	175			Patients submitted to surgery	Hypoechogenicity, microcalcifications, taller than wide shape, solid, irregular margins
Gulcelik N (31)	2008	98	16.3	46.7	Patients with cytology reporting follicular neoplasm	Hypoechogenicity, microcalcifications, solitary, solid
Salmaslioglu A (54)	2008	1926	19.0	46.9	Patients with multinodular goiter submitted to surgery	Hypoechogenicity, microcalcifications, solid, irregular margins
Chen G (24)	2009	758	23.0		Patients submitted to surgery	Microcalcifications
Hong Y (32)	2009	145	17.7	46.0	Consecutive patients submitted to surgery	Hypoechogenicity, microcalcifications, central vascularization, elasticity, taller than wide shape, irregular margins
Liu F (40)	2009	40	7.5	43.7	Patients with lymphocytic thyroiditis and	Hypoechogenicity, absence of halo,

					nodules with malignant or indeterminate	microcalcifications, irregular margins
Mendelson A (42)	2009	77	16.8		FNA reporting follicular, Hürthle cell or nondiagnostic	Microcalcifications, solid
Phuttharak W (46)	2009	31	3.3	41.8	Patients with risk of malignancy after US and FNA	Hypoechogenicity, microcalcifications, central vascularization, taller than wide shape
Popowicz B (47)	2009	1141		49.5	Patients submitted to surgery	Hypoechogenicity, microcalcifications, central vascularization, solitary, taller than wide shape
Rago T (51)	2010	195	26.1	44.0	Patients with indeterminate or nondiagnostic cytology	Hypoechogenicity, absence of halo, microcalcifications, central vascularization, elasticity
Schueller-Weidekamm C (55)	2010	31	31.4	55.2	Patients with cold nodules	Hypoechogenicity, microcalcifications, central vascularization, irregular margins
Sillery J (59)	2010	102	35.0	53.0	Patients with diagnosis of follicular carcinoma and adenoma	Hypoechogenicity, absence of halo, microcalcifications, central vascularization, heterogeneity
Wang Y (64)	2010	51	25.5	48.6	Patients with single nodules submitted to surgery	Microcalcifications, central vascularizaion, elasticity, irregular margins
Yoon J (66)	2010	99	13.1	43.7	Patients with indeterminate cytology	Hypoechogenicity, microcalcifications, solitary, taller than wide shape, solid, irregular margins
Cakir B (19)	2011	391	17.1	46.0	Patients with compressive symptoms or malignant or suspicious cytology	Elasticity
Maia F (41)	2011	143	15.4	47.2	Patients submitted to surgery	Hypoechogenicity, microcalcifications, central vascularization, irregular margins
Castro M (23)	2011	462	53.7	31.0	Patients with suspicious cytology	Solitary
Ghervan (29)	2011	99			Patients with suspicious nodules	Elasticity
Lippolis P (39)	2011	102	32.3	46.5	Patients with indeterminate cytology	Hypoechogenicity, microcalcifications, central vascularization, elasticity
Shuzhen C (58)	2011	291	25.0	43.4	Patients submitted to surgery	Elasticity
Cantisani V (20)	2012	97	33.0	54.0	Patients submitted to surgery due to compressive symptoms or suspicious nodules	Hypoechogenicity, microcalcifications, central vascularization

Table 2: Diagnostic performance of each US feature in the differentiation of benign and malignant thyroid nodules in unselected nodules

Feature	Sensitivity (%)	Specificity (%)	Positive likelihood ratio	Post-test probability (%) *	Negative likelihood ratio	Post-test probability (%) §
Taller than wide	26.7	96.6	8.07	47.0	0.75	7.6
Halo absent	56.4	72.0	2.02	18.1	0.60	6.2
Absence of elasticity	87.9	86.2	6.39	41.3	0.13	1.4
Heterogeneity	47.5	70.0	1.58	14.8	0.74	7.5
Hypoechogenicity	62.7	62.3	1.66	15.4	0.62	6.3
Solid	72.7	53.2	1.55	14.6	0.51	5.3
Microcalcifications	39.5	87.8	3.26	26.4	0.68	7.0
Solitary	53.0	60.2	1.33	12.8	0.77	7.8
Central vascularization	45.9	78.0	2.09	18.7	0.69	7.1
Irregular margins	50.5	83.1	2.99	24.7	0.59	6.1

* probability of malignancy after having a positive test result

§ probability of malignancy after having a negative test result

Table 3: Diagnostic performance of each US feature in the differentiation of benign and malignant thyroid nodules in nodules with nondiagnostic cytology

Criterion	Sensitivity (%)	Specificity (%)	Positive likelihood ratio	Post-test probability (%) *	Negative likelihood ratio	Post-test probability (%) §
Hypoechogenicity	39.0	62.0	1.06	10.4	0.96	9.6
Solid	71.8	43.7	1.27	11.9	0.64	6.6
Microcalcifications	46.7	84.4	3.01	24.9	0.63	6.5

* probability of malignancy after having a positive test result

§ probability of malignancy after having a negative test result

Appendix - Figures

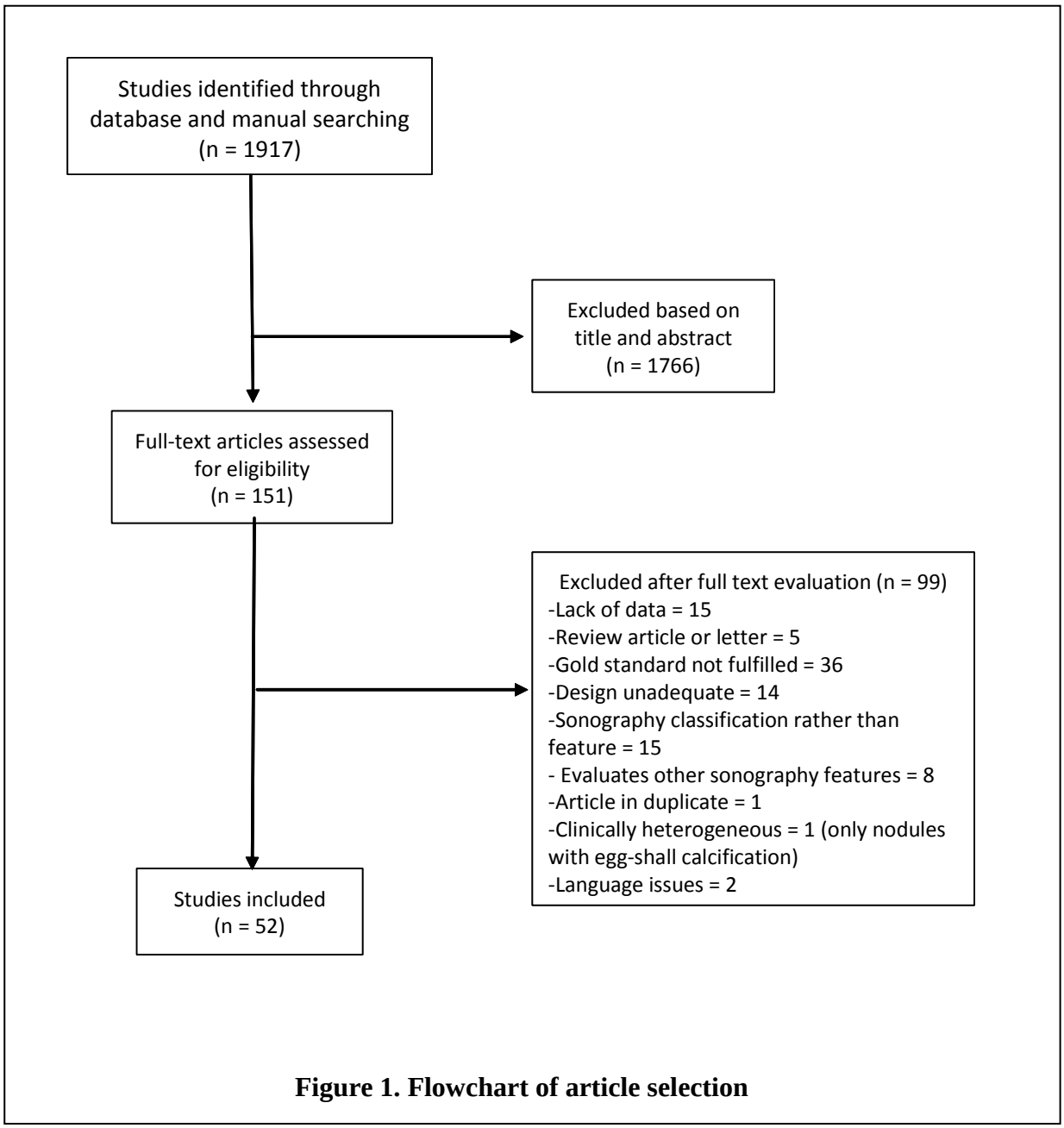
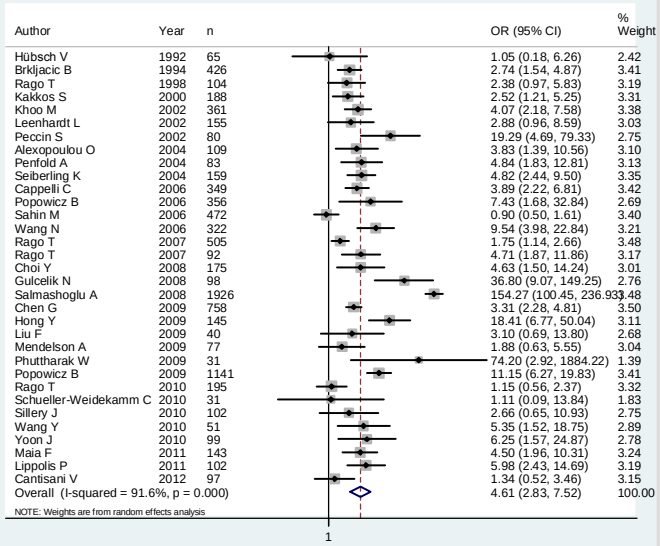
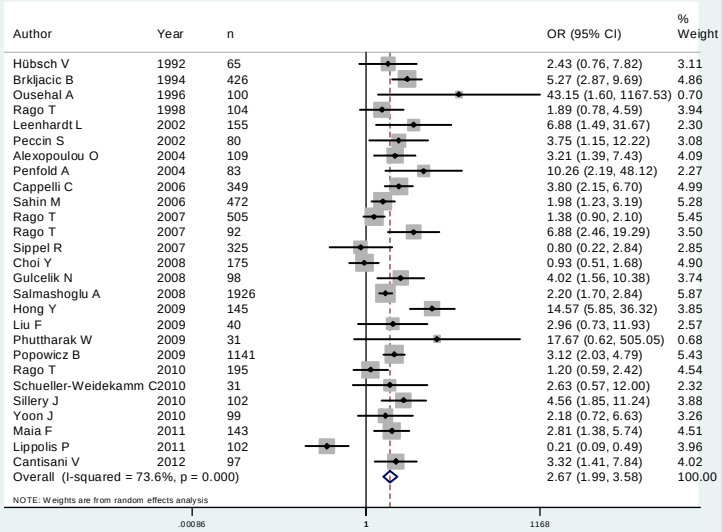


Figure 1. Flowchart of article selection

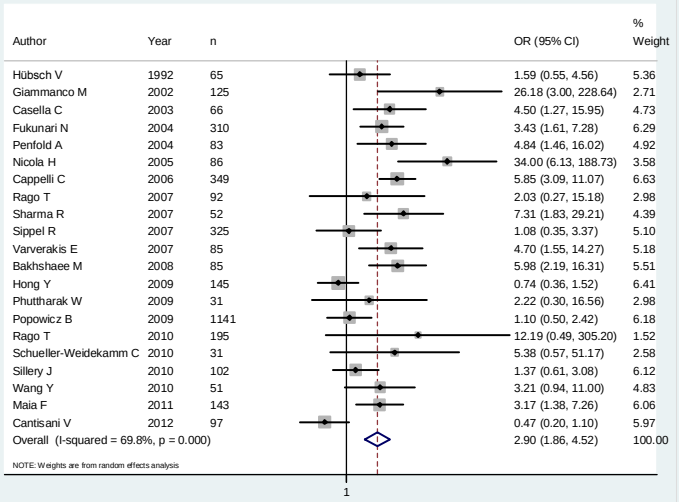
Microcalcifications



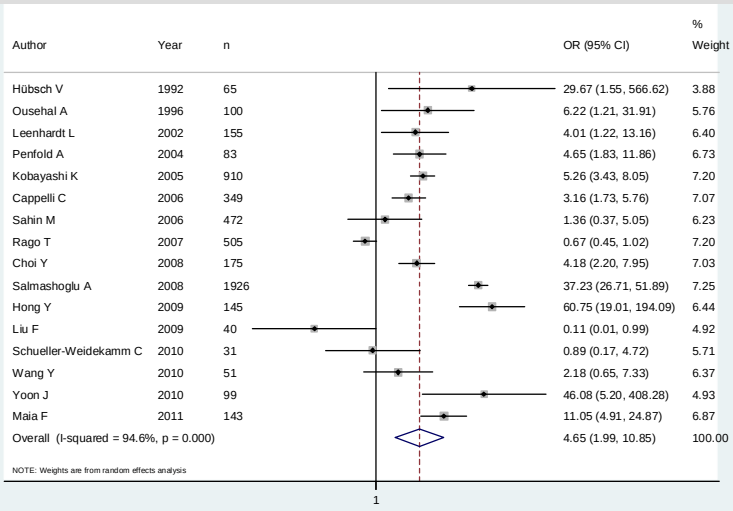
Hypoechogenicity



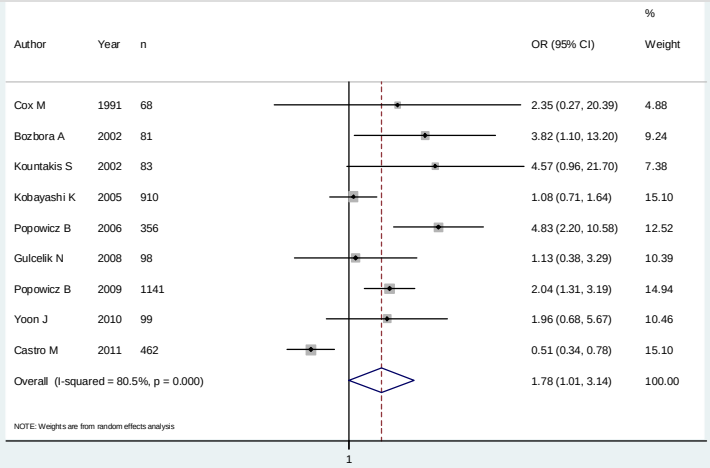
Central vascularization



Irregular margins



Solitary



Solid

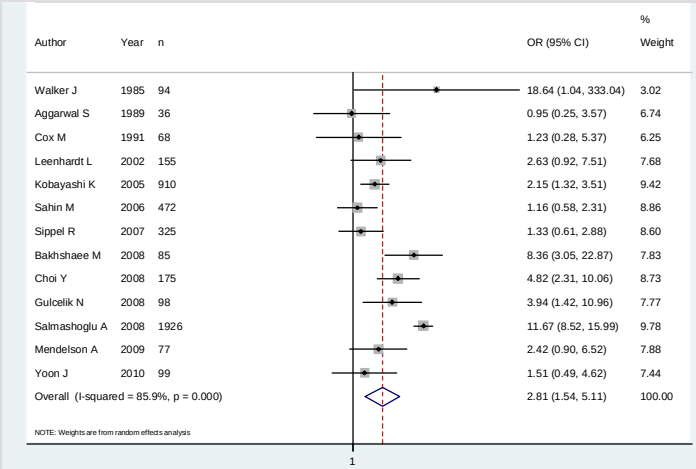


Figure 2. Forrest plot representing OR for malignancy of each ultrasound feature evaluated.

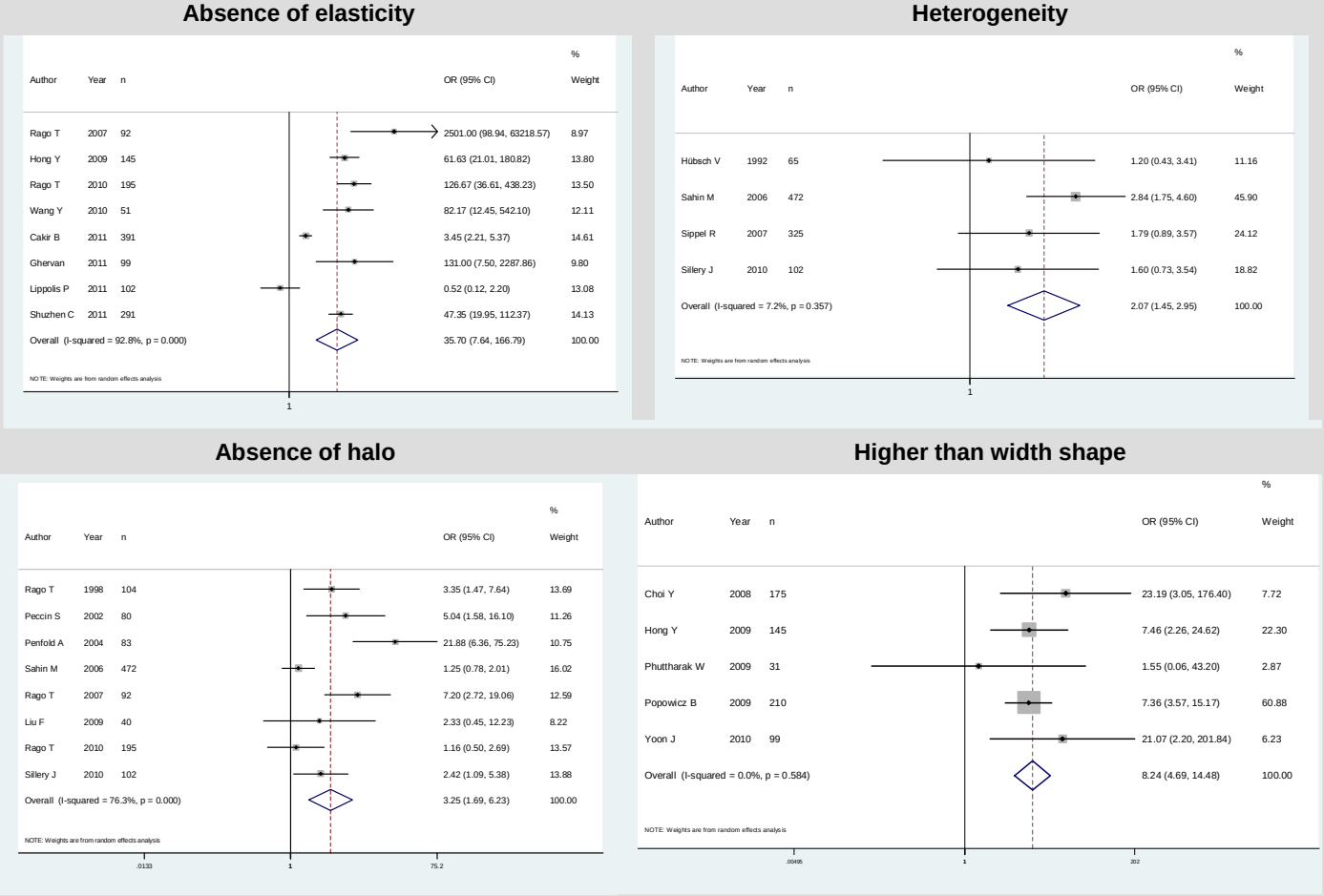


Figure 2. (cont) Forrest plot representing OR for malignancy of each ultrasound feature evaluated.

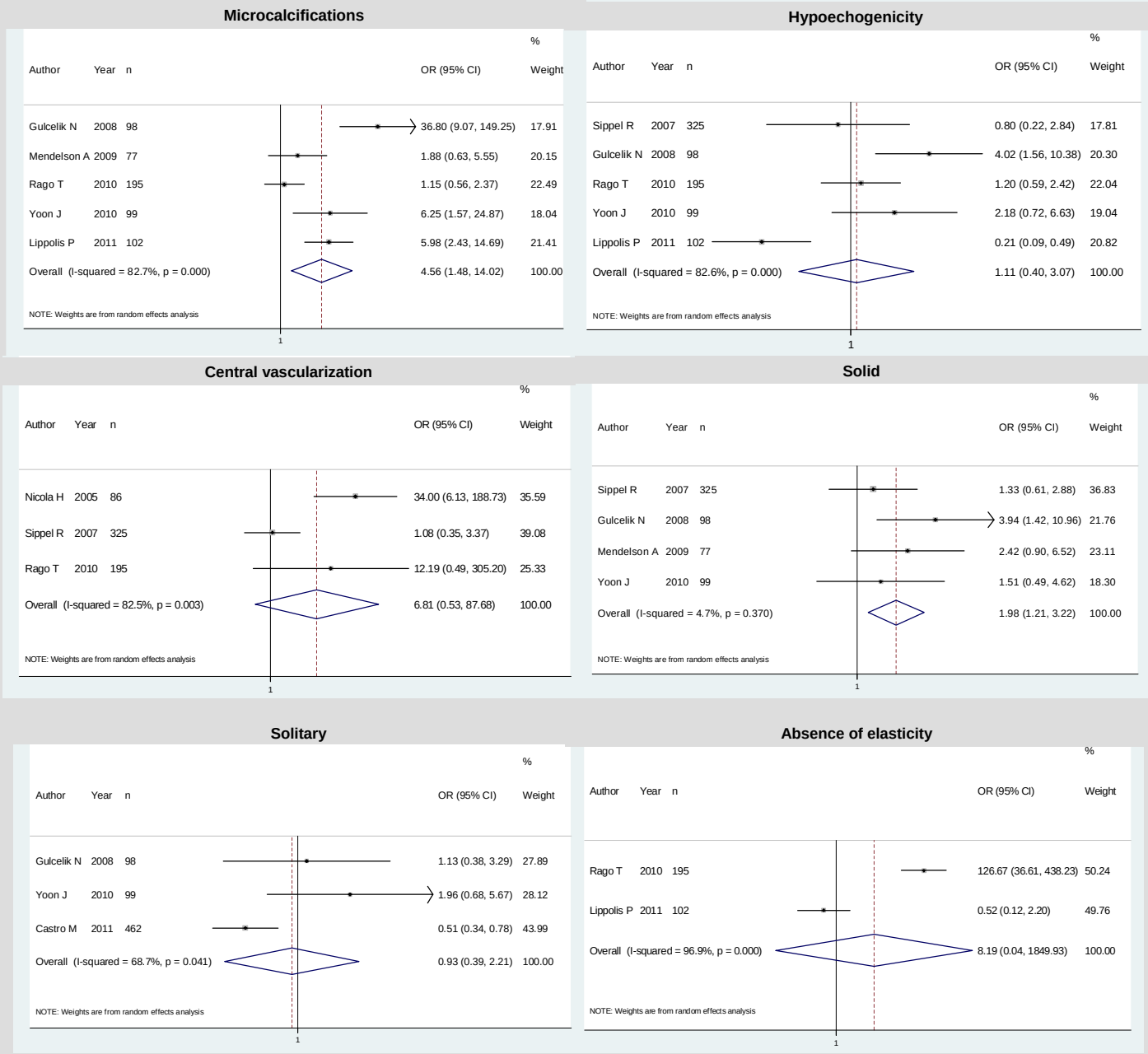


Figure 3. Forrest plot representing OR for malignancy of each ultrasound feature evaluated in nodules with nondiagnostic cytology

SUPPLEMENTAL FILES

1. Table 4: Quality evaluation of included studies

Author	Risk of bias				Applicability concerns		
	Patient selection	Index test	Reference standard	Flow and timing	Patient selection	Index test	Reference standard
Walker J	N	U	N	N	N	N	N
Aggarwal S	N	U	N	N	Y	N	N
Cox M	U	U	N	N	N	N	N
Hübsch V	U	N	N	N	N	N	N
Brkljacic B	N	U	N	N	N	N	N
Ousehal A	N	U	N	N	N	N	N
Rago T	N	U	N	N	Y	N	N
Kakkos S	N	U	N	N	N	N	N
Bozbora A	N	U	N	N	Y	N	N
Giammanco M	N	N	N	N	N	N	N
Khoo M	N	U	N	N	N	N	N
Kountakis S	N	U	N	N	N	N	N
Leenhardt L	N	U	N	N	N	N	N
Peccin S	N	N	N	N	N	N	N
Casella C	N	U	N	N	N	N	N
Alexopoulou O	Y	U	N	N	Y	N	N
Fukunari N	N	U	N	N	Y	N	N
Penfold A	N	U	N	N	N	N	N
Seiberling K	N	U	N	N	N	N	N
Kobayashi K	Y	U	N	N	Y	N	N
Nicola H	N	U	N	N	N	N	N
Popowicz B	N	U	N	N	N	N	N
Sahin M	N	U	N	N	N	N	N
Wang N	N	U	N	N	N	N	N
Cappelli C	N	U	N	N	N	N	N
Rago T	N	N	N	N	N	N	N
Rago T	N	U	N	N	Y	N	N
Sharma R	U	U	N	N	Y	N	N
Sippel R	N	N	N	N	N	N	N
Varverakis E	U	U	N	N	Y	N	N
Bakhshaee M	N	N	N	N	N	N	N
Choi Y	N	N	N	N	N	N	N
Gulcelik N	N	U	N	N	N	N	N
Salmaslioglu A	N	U	N	N	N	N	N

Chen G	N	U	N	N	N	N	N
Hong Y	N	U	N	N	N	N	N
Liu F	Y	U	N	N	Y	N	N
Mendelson A	N	U	N	N	N	N	N
Phuttharak W	N	N	N	N	N	N	N
Popowicz B	N	U	N	N	N	N	N
Rago T	N	N	N	N	N	N	N
Schueller-Weidekamm C	N	U	N	N	Y	N	N
Sillery J	Y	U	N	N	Y	N	N
Wang Y	N	U	N	N	Y	N	N
Yoon J	U	N	N	N	U	N	N
Cakir B	N	N	N	N	N	N	N
Maia F	N	U	N	N	N	N	N
Castro M	N	U	N	N	N	N	N
Ghervan	U	U	N	N	U	N	N
Lippolis P	N	U	N	N	N	N	N
Shuzhen C	N	U	N	N	N	N	N
Cantisani V	N	U	N	N	N	N	N

Y = yes, U = unclear, N = no