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Invited speakers
Cytology education in the Netherlands

1. Gynecological Cytology
Lia van Zuylen

Abstract: Since a couple of years the role of the cytotechnologist changes. On about 8-10
highschools/universities there was cytology education. But when the screening program changes, not
100-150 cytotechnologists are screening for the screening program, but only 35 cytotechnologists.
This means for the future there are not so many jobs, so the need for education goes down.

Now there are only 4 institutes for cytology education in the Netherlands. On these 4 institutes the
students can follow the same program for cervical cytology. In one year they learn the cervical
cytology. In 2 (of the 4) institutes they can learn the non-gyn cytology in almost the same program.

A big role of a cytotechnologist in the Netherlands is the support at EUS/EBUS. This subject is
therefore a big part in the new education.

In my presentation I give an update over the program of cytology education in the Netherlands.
Histopathologic correlation of atypical squamous cells, (ASC-US and ASC-H) a retrospective cross-sectional study

1. Gynecological Cytology
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**Objectives:** The aim of every screening test is to prematurely recognise the presence of cancer in its primary stages in order for timely treatment to commence, reducing incidence and mortality rates of cancer as a result. The Pap smear is a cytological screening test for the detection of carcinoma of the cervix and the primary stages associated with it.

Within the interpretation of cytological smears, the category Pap III (Bethesda: ASC-US, ASC-H) is somewhat of a grey area as it does not allow for accurate risk assessment. It is considered a dubious cytological diagnostic finding of which the malignancy and prognosis is unclear. Optimisation of this entity through a sub-classification steered by morphological characteristics would achieve more meaningful and effective patient management.

As far as the morphology of those cell profiles with a higher likelihood of ≥HSIL (≥CIN 2) differentiates from those with a higher likelihood of ≤LSIL (≤CIN1), the corresponding clinical management could also be ensured differently. For patients with higher risk, immediate histological intervention is advised, however for patients with lower risk, the clinical „wait and see“ approach is preferred.

**Material and methods:** This database study was executed retrospectively, by subtyping of 1281 cytological results according to the Bethesda nomenclature. Pap IIIK, utilised as a synonym for ASC-US, and Pap IIIH, used for ASC-H, were correlated with consecutive histological findings from biopsies and curettages respectively. The odds ratio for an obligate precancerous disease (≥HSIL) was calculated. To illustrate the discrepancies between cytological diagnoses and histological diagnoses a quality assessment grid was created. From the same sample 500 histological diagnoses and the diagnoses of the conisation process were correlated with a chi2-test. Divergent results were evaluated to show problems of over- and undertreatment.

**Results:** The age distribution of patients across the Pap III group ranged from 15 to 90 years, whereas a Pap IIIK finding was most commonly detected amongst patients between 20 and 29 years of age and a Pap IIIH result was more frequently observed amongst patients from 30 to 39. Within the Pap IIIK group, 66.8% of histological findings were classified as ≤LSIL and 33.2% as ≥HSIL. For Pap IIIH, 61.5% of histological diagnoses were categorized as ≥HSIL and 38.5% as ≤LSIL. A significant correlation could be shown between both the cytological diagnosis of a Pap IIIK result with a corresponding histological result of ≤LSIL and between that of a Pap IIIH and ≥HSIL result. Within this study, the probability for the presence of an obligate precancerous disease (≥HSIL) with a corresponding Pap IIIK diagnostic finding rather than a Pap IIIH result is 70% lower. The overall concordance rate between cytological findings and histological findings was 63.8%. A significant under-valuation of cytological diagnoses was made for 13.3% and a significant over-valuation for 0.4%.

In addition to these findings, a comparison of the histological diagnoses with the histological diagnoses of a conisation process could be made for 500 of the randomly selected patients. A
A concordance rate of 83.4% was identified using the WHO-classification and a concordance rate of 69.4% regarding the CIN-nomenclature. For 12.2% of patients who underwent a conisation process, no clinical indication was deductible, suggesting a heightened level of unnecessary treatment.

For 16.6% of ≥HSIL no high risk HPV was identified.

**Conclusions:** Women with a cytological diagnosis of Pap IIIK have a 70% lower risk of harboring a significant lesion (≥HSIL) than women with Pap IIIH. Pap IIIH showed a histological proven ≥HSIL in 61.5% and Pap IIIK a histological proven ≤LSIL in 66.8%. This indicates a reliable reproducibility of cytological subtype-diagnoses on the basis of specific morphological criteria. Nevertheless the comparison between cytological and histological diagnoses shows potential for quality assurance. Five histological carcinomas were diagnosed as Pap IIIK and should be evaluated in the context of quality-management. Referring to the WHO-classification the concordance rate between histological diagnoses and the diagnoses of the conisation processes was 14% higher than referring to the CIN-nomenclature. This is highlighting the better reproducibility of the two-tiered WHO-classification. High risk HPV-DNA was negative for 16.6% of ≥HSIL. This is consistent with available studies and shows that a negative HPV test provides no absolute assurance for not harboring a precancerous lesion!
Risk of high-grade lesions after atypical glandular cells in cervical screening

2. HPV Cervical Screening

Ingrid Norman

Karolinska Universitetssjukhuset, Huddinge

Abstract: Objectives: To determine how human papillomavirus (HPV) positivity of atypical glandular cells (AGC) affects the predictive values for presence of high-grade cervical lesions.


Participants: Between 2014-02-17 and 2016-06-30, there were 562 women with AGC in a cervical sample in the region. Registry linkages up to 2016-06-30 identified that 392 women had also had an associated HPV test and a histopathological follow-up.

Main outcome measure: Presence of high-grade cervical lesions in the cervical biopsies taken after the AGC diagnose, in relation to the HPV status of the AGC-containing index smear.

Results: The proportion of HPV-positive AGC was 56% (n=222). In this group, there were 6 cases of invasive cervical adenocarcinoma, 33 cases of cervical adenocarcinoma in situ and 93 cases of high grade squamous intraepithelial lesion (HSIL), giving a positive predictive value (PPV) for a cervical lesion to treat of 60% (132/222). Among the 170 women with HPV-negative AGC, there was 1 invasive cervical squamous cell cancer and 4 HSIL, giving a PPV for a cervical lesion to treat of 2.9% (5/170). This group also contained 5 endometrial cancers and 1 breast cancer.

Conclusions: HPV triaging of AGC will greatly increase the predictive ability for cervical lesions to treat [Odds Ratio: 48.4 (95% Confidence Interval: 19.1-122.6)] and the high sensitivity (96%; 132/137 women) implies safety of primary HPV screening strategies. The measurable risk for endometrial cancer among women with HPV-negative AGC (2.9%) suggests that research on screening for endometrial cancer is needed.
Utilit

Utility of cell blocks and immunocytochemistry in routine diagnostic fine needle aspiration cytology of thyroid gland

5. Thyroid Cytology

LJUBE IVKOVSKI

IRINA PRODANOVA, ZANETA BOCESKA, TAMARA IVKOVSKA

1 PHI HISTOLAB, Department of Cytology
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Objectives: FNAC is recommended as a routine prime investigation in the work-up of solitary thyroid nodules. Requirements for accurate diagnosis are adequate material, optimal smearing, proper staining and experience of the aspirator and diagnostic cytologist. Further, the importance of cell-block study in combination with FNAC in the diagnosis of different thyroid lesions is emphasized. The aim of the present study is to assess the values of FNAC in routine diagnostic thyroid cytology unit and to highlight the importance of its association with cell-block technique in order to avoid FNAC diagnostic pitfalls.

Material and methods: This study was a retrospective study of 85 patients with thyroid swellings, coming to cytopathology unit. FNAC and cell-blocks were performed for each case. Cytological results of FNAC were compared to the cell-block histopathological picture.

Results: FNAC findings were diagnosed as follows: 52 (61%) as colloid goiter, 5 (5.8%) as Hashimoto thyroiditis, 2 (2.3%) follicular neoplasm without atypia, 2 (2.3%) as follicular neoplasm with atypia, 2 (2.3%) as suspicious of malignancy, 10 (11.7%) papillary thyroid carcinoma, and 1.0 (1.1%) thyroid carcinoma of undefined category. Insufficient cases were 5 in number. The sensitivity of FNAC in comparison with histopathological results of cell-blocks was 91.6% and specificity was 97.2%.

Conclusions: Cell block technique is a simple, inexpensive procedure which allows processing of minute amounts of cellular material, facilitating better classification of thyroid lesions when reviewed along with cytological smears. This technique showed to be highly effective with high sensitivity and specificity and could be utilized as a cost effective diagnostic tool in the armamentarium of diagnostic work up in the patients with thyroid lesions. It is advised to perform cell-block for each case of FNAC of thyroid lesions whenever it is possible, to decrease the pitfalls and to improve the cytologic diagnosis.

Selected references
Nuclear grade in lung carcinoma

6. Lung Cytology and Mediastinum

Kosuke Inoue\(^1\)
Kyuichi Kadota\(^1\), Nachino Kimura\(^1\), Emi Ibuki\(^1\), Ryou Ishikawa\(^1\), Seiko Kagawa\(^1\), Yumi Miyai\(^1\), Reiji Haba\(^1\)

\(^1\) Departments of Diagnostic Pathology, Faculty of Medicine, Kagawa University

Abstract: For patients with resected lung adenocarcinoma, the predominant subtype according to the 2015 WHO lung tumor classification has been identified as a prognostic indicator. The prognostic usefulness of nuclear grade using cytological specimens still remains unknown in patients with advanced non-small cell lung cancer (NSCLC) although greater than one-half of the patients have advanced disease at the time of diagnosis.

The clinical utility of a nuclear grading system has already been established in other carcinomas, such as breast, kidney, and bladder. In NSCLC, although no nuclear grading system has been universally accepted to date, previous studies have demonstrated the prognostic value of nuclear features. Japanese group demonstrated that, using analytical computer software and the size of small lymphocytes as a standard, nuclear area and diameter could estimate the malignant potential of small-sized lung adenocarcinomas, and the interobserver agreement for this nuclear grading system was significantly higher than for histological classifications (Nakazato et al. Cancer 2010, J Thorac Oncol. 2013). A study from Memorial Sloan Kettering Cancer Center reported that large nuclear diameter was significant predictors of prognosis in resected stage I lung adenocarcinoma and in squamous cell carcinoma (Kadota et al. Mod Pathol 2012, J Thorac Oncol 2014). We have recently validated this finding in Japanese patients with lung squamous cell carcinoma. Moreover, we demonstrated that, based on transbronchial cytology, nuclear diameter assessed by an image analysis system and small lymphocytes was associated with prognosis in advanced NSCLC.

For NSCLC, to lead to a uniform grading system, data of architectural and nuclear grading system are needed. There can be significant interobserver variability, which is a criticism of the evaluation of nuclear features. To minimize interobserver variability, the recent studies suggest to use analytical computer software or the size of small lymphocyte to evaluate nuclear grade.
Cytologic Findings in Progressive Multifocal Leukoencephalopathy

13. CNS Cytology

Yuri Batoroev

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Objectives: Progressive multifocal leukoencephalopathy (PML) (1) is a fatal demyelinating disease of the central nervous system (CNS) characterized by destruction of cells that produce myelin. Clinical and radiological findings can indicate multifocal or solitary brain masses mimicking glioma, lymphoma, or some CNS infections (toxoplasmosis).

Material and methods: Five cases of PML diagnosed by stereotactic brain biopsy at the Irkutsk Oncology Hospital, were reviewed (2015-2018). Crush preparations prepared from portion of each sample at the time of biopsy were stained Diff-Quik. Additional smears of crush preparations were H&E stained.

Results: Smears from of the radiolucent area revealed foci of white matter demyelination and a few eosinophilic inclusions in oligodendrocytes plus abnormal giant astrocytes. Cytopathologic findings. In most cases smears showed moderate cellularity. Vascular proliferation was a feature of all cases. In some cases the vessels were cuffed by lymphocytes and plasma cells. Predominance of reactive astrocytic cells with enlargement size, surrounding thin-walled vessels. A conspicuous feature was elongated, radial cytoplasmic processes in reactive astrocytes. Nucleus were lobulated, enlargement, with smidgen chromatin. No mitotic figures no identified in the smears preparations. Histopathologic examination H&E stained material revealed glial proliferation. The astrocytes were bizarre, with abounded cytoplasm and enlarged, hypercromatic nuclei (sever degrees of nuclear pleomorphism). Gemistocytes were typically presented in foci of demyelinization containing lipid-laden histiocytes. Perivascular infiltrates of lymphocytes and plasma cells in some cases. In four cases oligodendrocytes exhibited nuclear changes typical of a JC viral infection (enlargement, hyperchromasia, chromatin effacement).

Conclusions: We concluded, that the accrurance stereotactic biopsy in the diagnosis of PML enhanced with combination of the histology and cytologic examination.

Selected references


EBUS and EUS ROSE assisted by cytotechnologist, experience from Akershus University hospital

18. Others
Mette Kristin
1 Akershus University Hospital

Abstract:

Uptil 2015 EBUS was sent to the patholoy department for staining and evaluation by a Pathologist.

In 2015 we introduced EBUS with ROSE (Raipd On Site Evaluation) assisted by a cytolotechnoloist .

This reduced waiting time on the operation table for the patient, reduced number of smear pr pasient and increased technical quality on the smears.

In the autum 2018 we also started up ROSE on EUS assisted by a cytotechnologist.

I will go through our experiences the EBUS and EUS ROSE assited by a cytotechnologist in my presentation.
Atypical Glandular Cells – A Diagnostic Challenge to the Cytopathologist

Invited Speakers

Do Minh Hoang Trong¹
¹ Laboratory of Anatomical Pathology, Hoan My Saigon Hospital, Vietnam.

Objectives: Unlike cervical squamous epithelium, the cytologic features of precursor glandular lesions of degrees less than adenocarcinoma in situ have not been well defined. This review shows the difficulty in interpretation of atypical glandular cells on cervical cytology.

Material and methods: Review of medical literature.

Results: The interpretation of atypical glandular cells on cervical cytology is not common and the cytologic criteria have been found to be poorly reproducible. The difficulty in identifying correctly glandular lesions lies not only in differentiating clinically significant lesions from benign reactive conditions but also in classifying accurately the cell of origin as being squamous, endocervical, or endometrial. Biopsy follow-up of a cervical smear interpretation of atypical glandular cells most often reveals either a benign reactive process or a squamous cervical intraepithelial neoplasia rather than a glandular one.

Conclusions: When overlapping cytologic features make accurate identification difficult, the interpretation of “atypical glandular cells” is rendered and the smears should be rechecked by an experienced gynecologic cytopathologist and then further clinical investigations, including colposcopy and histological evaluation, should be indicated.

Selected references
Babeș-Papanicolaou staining method

Invited Speakers
Paula Warnhag

1 Paula Warnhag, Labmedicin, Department of Clinical Genetics and Pathology, Lund, Sweden

Objectives: To bring light upon the importance of the linked name Babeș-Papanicolaou used for the Papanicolaou test in Romania, the names of two remarkable professors and pioneers in the medical field, Dr. Aurel Babeș and Dr. George Papanicolaou.

Material and methods: As material various published articles, about Dr. Babeș’ and Dr. Papanicolaou’s findings and choices of view on the staining method of the cervical cell, were searched and studied.

A deep search on the internet and at the Swedish medical libraries was made, especially for the original article of Dr. Babeș published in La Presse Medicale, in 1928.

Results: There are major differences between the two doctor’s choices to take the samples, to stain the cervical cells and even to interpret the results regarding cases of cervical cancer. Dr. Babeș’s method involves an air/dried fixation and seemed to be focused more on the whole cell, mostly found in the tissue than solitary, and its nucleus. On the other hand Dr. Papanicolaou’s wet fixation can provide important details even of the cytoplasm, which leads to a better diagnose. Besides this, Dr. Babeș had neither screened symptomless women nor named the hormonal significance, which actually Dr. Papanicolaou’s method is used for nowadays.

Conclusions: In one of the articles I found an interesting quote of Sir Francis Darwin, son of The Charles Darwin, saying: “In science, the credit goes to the man who convinces the world, not the man to whom the idea first occurs.” This summarises the unseen battle of who was the first to establish the amazing stain method of not only the cervical cells, but of materials containing exfoliative cells and from needle aspiration as well. A stain that saves lives every day.

Selected references
Cytologic diagnosis of mesothelioma – Update with use of ancillary assays based on genetical changes in mesothelioma

Invited Speakers
Kazuki Nabeshima1
Makoto Hamasaki1, Yoshiaki Kinoshita1, Shinji Matsumoto1, Ayuko Sato2, Tohru Tsujimura2, Kunimitsu Kawahara3, Kenzo Hiroshima4, Toshiaki Kamei5
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4 Department of Pathology, Tokyo Women's Medical University Yachiyo Medical Center, Yachiyo, Japan
5 Pathology and Cytology Center of the PCL Fukuoka, Fukuoka, Japan

Objectives: The diagnosis of malignant pleural mesothelioma (MPM) involves verification of the mesothelial origin of the tumor and the often-challenging discrimination of MPM from reactive mesothelial hyperplasia (RMH) or reactive mesothelial cells (RMC). Homozygous deletion (HD) of the 9p21 chromosomal region, as detected by fluorescence in situ hybridization (FISH), and loss of BRCA1-associated protein 1 (BAP1) expression, as detected by immunohistochemistry (IHC), are useful in distinguishing MPM from RMH/RMC. Furthermore, we reported that IHC of the protein product of methylthioadenosine phosphorylase (MTAP) gene located in the 9p21 locus correlated with the deletion status of 9p21 detected using FISH in MPM tissues and cell blocks. Therefore, MTAP IHC can serve as a surrogate assay for 9p21 FISH.

Material and methods: IHC and FISH were performed in tissues and cytologic preparations.

Results: For the differentiation of epithelioid MPM from RMH/RMC, a combination of 9p21 FISH and BAP1 IHC produced greater sensitivity than that observed with either assay independently, an observation also made in smear preparations. Combined IHC assays using BAP1 and MTAP antibodies also exhibited 100% specificity, with sensitivity that was lower than that of combined 9p21 FISH and BAP1 IHC, albeit higher than that of BAP1 IHC or 9p21 FISH alone in both tissue and cell blocks. However, the evaluation of IHC in cell blocks poses some unique technical challenges. For example, while loss of MTAP expression usually occurs in both the nucleus and cytoplasm, occasionally it is seen only in one, and in such cases, loss of cytoplasmic MTAP expression correlates with 9p21 HD.

Conclusions: Our study shows that the use of combined IHC detection of MTAP-BAP1 loss can assist in discriminating MPM from RMH/RMC in both tissues and cytological preparations, provided data is cautiously interpreted. Nevertheless, a combination of BAP1 IHC and 9p21 FISH assays remains the most accurate ancillary method in both sample types.
Cytology in Norway today and future perspectives

Invited Speakers
Maj Liv Eide¹
¹ Trondheim University Hospital and Norwegian University of Science and Technology, Trondheim, Norway

**Objectives:** Controlled implementation of HPV primary screening started in three counties in 2015. Non-gynaecologic cytology is for the most part a small portion of the cytology specimens in the laboratories. The aim of this presentation is to show how the implementation has changed the everyday work in the cytology laboratory, the present situation of non-gynaecologic cytology and thoughts on the future of cytology in diagnostic pathology.

**Material and methods:** The cytology laboratories at the University Hospitals in Trondheim, Bergen and Stavanger implemented HPV primary screening in women 34-69 years old. Cytotechnologists in these laboratories also perform the HPV-testing. Cytology remains the screening method of choice for women 25-33 years old. During this period, the screening algorithm changed once and test of cure was implemented. A survey on tasks besides cervical screening among cytotechnologists was conducted in 2015 and a survey on training programs in non-gynaecologic cytology in 2017.

**Results:** The number of cytology specimens has decreased approximately 40% since the start in 2015. Due to two different screening methods and three different algorithms, the cytotechnologists and cytopathologists spend more time than before on specimen logistics and control of the screening history to ensure proper follow-up. The surveys on different tasks and training programs in non-gynaecologic cytology showed great variations among laboratories.

**Conclusions:** The controlled implementation of HPV primary screening has been successful and made it possible for the laboratories to adjust. The experience of having cytotechnologists perform the HPV-testing is very good, but in other counties, the HPV-testing is done in departments of microbiology. Competence in cervical cytology is needed for several years still and this should not be forgotten in this age of primary HPV screening. The surveys showed that there is a need for structured training programs in non-gynaecologic cytology and ancillary techniques. This is important for the future of cytology in diagnostic pathology.
Cytology services in Iceland

Invited Speakers
Ingibjorg Guðmundsdottir

Objectives: 55 years of organised cervical screening in Iceland and approximately the same time for non-gynaecological services.

All this time the Cancer Society has been responsible for the screening with yearly financial contributions from the state. Women also pay a contribution for screening appointments. This arrangement could change in the near future.

Icelandic society and its members have changed a lot over the last 10 to 20 years. Figures from the National Registry show that immigrants comprise now more than 12% of the population and this is the same proportion as in Norway and Denmark. There have also been considerable changes when it comes to how screening is organised and in relation to techniques in screening. Today, steadily decreasing uptake of screening services, particularly by younger women, poses the biggest challenge. At the beginning of 2019 the official response to this was offering first screening for 23 year olds for free.

For the last 4 years, non-gynaecology services are offered at the National Hospital (Landspítalinn Iceland). The number of samples has been relatively stable over the last decades. Prior to services in Landspítalinn, these services were provided by private service providers. There is, however, diversity in the samples, for example there has been an overall increase in thyroid samples but a reduction in breast samples. There is considerable demand for assistance with EBUS and EUS and ultrasound assistance, samples in cellblock and flow.

Material and methods:

Results:

Conclusions:
Cytomorphology of blood smear

Invited Speakers
Veronika Anic Paradis¹
Ika Kardum Skelin¹
¹ Department of Clinical Cytology and Cytogenetics, Merkur University Hospital, Zagreb, Croatia

Objectives: Although new diagnostic tests are available, peripheral blood smear analysis remains a basic diagnostic procedure of outstanding importance in the detection and monitoring of many conditions and diseases.

Material and methods: Data were collected through long-term experience in routine, daily cytomorphological analysis and reporting of peripheral blood smears at the Department of Clinical Cytology and Cytogenetics of the Merkur University Hospital in Zagreb, Croatia.

Results: Cytomorphological analysis of peripheral smear is most commonly performed at the physician's request due to clinical suspicion or an abnormal finding of an automated blood counter. The automatic counter identifies and quantifies leukocytes, erythrocytes and platelets in the blood sample, but does not describe morphological changes, precisely identifies and classifies immature or abnormal cells. In the case of a changed finding, the counter indicates a sample for further microscopic analysis. Cytological analysis of blood cells, apart from morphology and quantification of the different types of leukocytes (differential count), detects and classifies immature or abnormal leukocytes. The erythrocyte analysis, apart from determining abnormal or immature forms, includes morphological evaluation of erythrocyte appearance. Platelet evaluation includes descriptive amount, morphology and thrombocyte appearance. The cytological findings can be complemented by cytochemical analysis, flow cytometry, cytogenetics or molecular techniques and are of great importance to clinicians in a diagnosis.

Conclusions: The diagnostic importance of cytomorphological analysis of peripheral blood cells has not been reduced or replaced by automation in haematological diagnostics or by new diagnostic procedures. Cytological finding of blood smear with a clinician understanding how to interpret such finding is a basic, highly informative diagnostic method of immense importance in diagnosing, monitoring the success of treatment or progression of the disease. Cytotechnologists who possess knowledge and skills, from sampling and processing to microscopic analysis, are the perfect choice for this task.
Cytotechnologists in Sweden today

Invited Speakers
Paula Warnhag1
1 Labmedicin, Department of Clinical Genetics and Pathology, Lund, Sweden

Objectives: To share detailed information about the work and the concept of cytotechnologist (CT) in Sweden nowadays.

Material and methods: An inquiry by the Swedish Society for Cytotechnologists sent to the laboratories, where cytotechnologists work.

Information gathered from senior CTs at the Department of Cytology in Lund, Sweden.

Results: Answers received from 22 of 26 laboratories where around 120 members in the Swedish Society for Cytotechnologists have different responsibilities in order to diagnose various types of the samples. The number of active Swedish CTs is higher.

In 41 % of laboratories (labs) CTs give out both benign and malign diagnosis in cervical cytology. CTs in 19 % labs send out only the benign diagnosis is.

The benign urine and respiratory cytology is answered out by CTs in 79 %, respective 82 % labs. One laboratory sends even the benign respiratory samples to the doctor for the final answer.

The benign exudates are answered out by CTs in 41% labs. In 59 % labs both benign and abnormal samples are sent forward to the doctor by a CT.

In Final Needle Aspiration, CTs from only 36 % labs give diagnostic suggestions to the doctor.

EBUS and EUS, when CT assists and diagnoses the samples, are performed at 8 of 22 labs. The doctor gives the final diagnosis for the abnormal cells.

Conclusions: Many other tasks are performed by CTs depending at which clinic they work. The semen analysis, assisting at the needle aspirations, being a part of the actual laboratory team and showing interest in the quality work are some of them. The administrative part of work can be vast.

Today CTs have far more responsibility and can work more independently in comparison with three decades ago. Back then the title was “cytoassistent”, meaning the cytology assistant. Nowadays no one assists anyone but CTs diagnose individually, giving even diagnostic suggestions to the doctor.

Selected references
The Swedish Society for Cytotechnologists
https://cytodiagnostiker.se/
Differential diagnosis in FNA: thyroid and parathyroid. Role of immunocytochemistry and washout fluid analysis

Invited Speakers
Elwira Bakuła-Zalewska

Department of Pathology, Maria Sklodowska-Curie Institute - Oncology Center, Warsaw, Poland

Objectives: Fine needle aspiration cytology (FNAC) has been traditionally of limited value in the preoperative assessment of patients with hyperparathyroidism. The majority of FNA parathyroid lesions is unintended, as intrathyroidal parathyroid adenomas and cysts are often mistaken for thyroid lesions. The US-guided FNA for examination of parathyroid has been recently increasingly used to distinguish abnormal parathyroid glands from lymph nodes and to localize atypical or intrathyroidal parathyroid gland location and parathyroid lesions in recurrent disease. Many investigators have stressed the difficulty to discerning parathyroid from thyroid in FNA smears.

Material and methods: 73 patients underwent parathyroid FNA either unintended (35 patients) during the FNA sampling of suspicious thyroid nodules or performed specifically to localize parathyroid lesions (38 patients). In addition to the examination of routinely stained smears, measurement of parathyroid hormone (PTH) in the needle washouts (FNA-PTH testing) were performed in all patients and immunocytochemical (IC) examination on 15 smears.

Results: All of the examined FNA samples were correctly diagnosed as parathyroid lesions in the examination of routinely stained smears or smears completed by FNA-PTH testing and/or IH.

Conclusions: The architectural pattern in the smears helps to distinguish between parathyroid and thyroid aspirates. Overall organoid and cribriform or trabecular architecture of three-dimensional and frequently crowded clusters with overlap of cells as well as loose sheets and clusters with acinar-follicular and/or rounded-pseudopapillary or papillary formations are suggestive of a parathyroid lesion. Cells with small, dark nuclei, somewhat smaller than nuclei in follicular thyroid cells, and with stippled chromatin and clean or bloody, colloid-free background are another sign suggestive of parathyroid origin. FNA-PTH testing of washout specimen helps to determine whether the specimen obtained by FNA represents parathyroid or thyroid lesions. IC on aspirated material is yet another technique which increases the diagnostic accuracy of FNAC of the parathyroid lesions.

Selected references
EACC recommendations for education of cytotechnologists

Invited Speakers

Veronika Anic¹
Maj Liv Eide²

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² Trondheim University Hospital and Norwegian University of Science and Technology, Trondheim, Norway

Objectives: The incomplete, inadequate and different cytotechnology educational programs in European countries are alarming rate with which we are increasingly faced. The shortcomings of the acquired knowledge are most pronounced in the changes that arise with the introduction of new diagnostic methods and technologies.

Material and methods: The European Advisory Committee of Cytotechnology (EACC), at the EFCS initiative, since 2010, collected and used relevant data from available cytotechnology education programs implemented in European countries. The collected data, analysis of results and data from the literature were used in the preparation of these recommendations.

Results: Newer additional diagnostic methods used in cytodiagnostics seek additional knowledge and skills and require a rapid change in the education of future generations in cytotechnology. Although of great importance, there are still no official recommendations for education of cytotechnologists in Europe that European cytological societies and educational institutions could use as a basic model for cytotechnological education. They are forced to create education programs according to their own criteria, which results in great differences and incompleteness in education. According to collected data, needs and capabilities, acceptable to most European countries, EACC propose one year of training and education program which should be divided into three modules: gynaecological cytology, non-gynaecological exfoliative and non-gynaecological fine needle aspiration cytology, with included education of new and additional methods, organized at an accredited university. After successful completion of the training programme, EACC propose official title: certified Cytotechnologist.

Conclusions: The European Advisory Committee of Cytotechnology (EACC) has set up guidelines for the education of cytotechnologists to assist in the harmonization of existing and development of appropriate programs in those countries that have incomplete or no education in cytotechnology. We believe that the recommendations for cytotechnology education in Europe need to be developed in the future as a result of EACC cooperation with EFCS.

Selected references

Education and work of cytotechnologists in Finland

Invited Speakers

Eeva Liikanen¹
¹ Tampere University of Applied Sciences

Objectives: The aim of this study was to survey the work that cytotechnologists carry out in Finland.

Material and methods: An electronic questionnaire was planned with the Board of the Finnish Association of Cytotechnologists and an email containing the link was sent to all 107 of its members in January 2018. It included 17 questions on their age and work experience, education and work. There was also space for them to add others comments.

Results: Just under half (45%) replied. Their average age was 51 (range 28-64), 41% had a Bachelor degree, 59% had college-level training and they had spent an average of 15 years screening cytology specimens. After basic professional education they had completed their cytology education in many ways and the most common routes were internship training (71%) and one-year cytology specialisation (38%). Most of the cytotechnologists (85%) had duties other than screening and they mostly included a combination of histotechnology and cytotechnology (37.5%) or just cytotechnology (17%) or histotechnology (9%). The other 15% only screened cytology specimens. All cytotechnologists screened Pap smears, 94% screened urinary and respiratory specimens, 82% screened effusions and 39% screened fine-needle aspirations.

Conclusions: We found that internship training provided essential training for Finnish cytotechnologists after they complete their basic professional studies. They reported many other duties in addition to microscopy screening.
Epithelioid hemangioendothelioma – lung and liver

Invited Speakers
Tajana Stoos-Veic¹
Cedna Tomasic Loncaric¹, Josip Curic², Danijel Cvetko², Marija Skoro¹
¹ University Hospital Dubrava, Department of pathology and cytology, Croatia
² University Hospital Dubrava, Department of diagnostic and interventional radiology, Croatia

Objectives: To describe cytomorphology of epithelioid haemangioendothelioma (EHE), a rare vascular tumour of borderline or low-grade malignancy prone to unusual presentations. The lungs and liver are the two common primary organs affected. This neoplasm usually presents as multinodular organ involvement sometimes simulating metastases. Microscopic features in FNA and small biopsies can be challenging and caution is advised as not to lead to erroneous diagnosis of metastatic carcinoma or even HCC or, on the other hand, of angiosarcoma.

Material and methods: Herein authors present two cases of EH diagnosed with FNA.

Results: A 51-year old female patient was referred to our hospital with multiple pulmonary nodules, largest measuring 6 mm. CT-guided FNA yielded several single cells with abundant polygonal cytoplasm, oval nuclei with reticular chromatin and slightly visible nucleoli. The descriptive diagnosis of possible tumour was rendered. The patient had a firm subcutaneous lesion of the right forearm and FNA revealed single cells and loose clusters of polygonal cells with round nuclei, often with intranuclear inclusions. Some cytoplasms contained hemosiderin pigment. The diagnosis of epithelioid hemangioendothelioma was made with a note that the cells found in the lung were of the same morphology. Subsequent surgery of forearm lesion and open lung biopsy confirmed the diagnosis. Two years later there is a slight progression in size and number of the lung lesions. Second patient was 57-years old male with multiple liver lesions described as multiple hemangiomas on CT scan. MR failed to clear the diagnostic dilemma. US-guided FNA of one lesion was done. Cellular sample revealed the same cytomorphology described previously for the first patient. The diagnosis of EHE was made. The patient is at the moment waiting for liver transplant.

Conclusions: As organ transplant for multiple nodules is sometimes a treatment option, accurate preoperative diagnosis is very important. Typical cytomorphologic features make definitive cytological diagnosis possible.

Selected references
FOXP3 POSITIVE LYMPHOCYTES IN CYTOLOGIC SMEARS OF LYMPH NODE ASPIRATES AND THEIR ASSOCIATION WITH PROGNOSTIC MARKERS IN PATIENTS WITH HODGKIN LYMPHOMA

Invited Speakers
Biljana Jelić Puškarić
Gordana Kaelić, Marina Pažur, Slobodanka Ostojić Kolonic, Delfa Radić-Krišto, Inga Mandac Rogulj, Veronika Anić, Ika Kardum-Skelin

1 Department of Cytology and Cytogenetic, University Hospital Merkur, Zagreb, Croatia.
2 Department of Medicine, University Hospital Merkur, Zagreb, Croatia.
3 University of Zagreb, School of Medicine, Zagreb, Croatia.

Objectives: The CD4, CD25 positive T regulatory (Treg) lymphocytes expressing Foxp3 transcription factor account for a relevant portion of tumour microenvironment in patients with solid tumours and haematologic malignancies. An increased number of tumour-infiltrating Treg cells has been associated with negative prognostic factors and an adverse clinical outcome in patients affected with some solid tumours. On the contrary, in patients with classical Hodgkin lymphoma (CHL), increased number of Treg cells have been associated with a better prognosis.

Material and methods: The objective of this investigation was to compare the number of immunocytochemically Foxp3 positive lymphocytes in fine needle aspirates of lymph nodes from patients with CHL (16 patients) with a control group of patients with benign reactive hyperplasia (26 patients). In CHL patients, the Foxp3 positive lymphocyte count was correlated with prognostic factors relevant for the clinical course of lymphoma (age, gender, clinical stage, size of the largest tumour, the presence of B symptoms, haemoglobin, lactate dehydrogenase, albumin and lymphocyte count). The number of Foxp3 positive cells per 100 lymphocytes (% Foxp3+ lymphocytes) was quantified, counting an average of a minimum of 10 counts per a cytological smear (1000 cells).

Results: In the group of patients with CHL a statistically higher number of Foxp3+ lymphocytes (19.61±9.80, p<0.01) was found compared to the control group of patients with reactive hyperplasia (11.15±3.82). The number of Foxp3+ lymphocytes was found to be statistically significant increased in the group of patients with early-stage CHL (I, II) (p<0.05) compared to the patients with CHL in advanced clinical stages (III, IV). There was no significant correlation between number of Foxp3+ lymphocytes and other prognostic parameters.

Conclusions: Our results show that Foxp3+ lymphocytes are increased in CHL and their detection may be a useful tool in differentiating CHL from other entities and can contribute to the prediction of outcome in CHL.
Genotyping and Subtyping based on LBC for Lung Adenocarcinoma

Invited Speakers
Ryota Tanaka¹
Norihiko Sakamoto², Hitomi Suzuki², Keisei Tachibana¹, Kouki Ohtsuka¹, Koji Kishimoto², Masachika Fujiwara², Hiroshi Kamma², Junji Shibahara², Haruhiko Kondo¹
¹ Department of Surgery, Kyorin University School of Medicine
² Department of Pathology, Kyorin University School of Medicine
³ Department of Clinical Laboratory, Kyorin University School of Medicine

Objectives: Liquid-based cytology (LBC) allows for analyses by immunohistochemistry, fluorescence in situ hybridization and molecular testing on nucleic acid from remaining fixed cells. Some studies have demonstrated that LBC systems provide excellent cell preservation for routine cytopathology and can used for DNA- and RNA-based molecular testing. The aim of this study was to determine if LBC samples were suitable for molecular testing techniques in relation to lung cancer treatments, and to examine the relationships among gene mutational status, cytomorphological features by analyzing LBC materials.

Material and methods: Forty consecutive lung cancer patients with primary lung adenocarcinoma (ADC) underwent surgical resection in our hospital. Cytological material was obtained by scraping the cut-surface of the lesion and samples were fixed and stored by CytoRich Red™ as LBC material. Epidermal growth factor receptor (EGFR), Kirsten rat sarcoma viral oncogene homologue (KRAS), anaplastic lymphoma kinase (ALK), and c-ros oncogene 1 (ROS1) mutations were detected and cytomorphological studies were performed.

Results: Twenty cases (50%) were positive for EGFR mutation and four (10%) were positive for KRAS mutation. ALK gene rearrangement was identified in one case (2.5%) by immunohistochemistry and fluorescence in situ hybridization; ROS-1 gene rearrangement was identified in one case (2.5%) by immunohistochemistry and real-time polymerase chain reaction. Higher proportions of cases with an inflammatory background (100%), papillary structure in terms of predominant architecture (75%) and papillary type of ADC pattern (75%) were detected in the KRAS-positive group compared with the EGFR-positive group and the other group which included ALK and ROS1 gene rearrangements.

Conclusions: LBC material is suitable for preparing materials for use in molecular testing, and the differences in major gene aberrations might show and predict some specific cytomorphological features. These findings might aid in differentiating ADC subtypes for predicting patient prognosis or in decision-making for treatment strategies in personalized medicine.
Histiocytes as predictors of endometrial pathology

Invited Speakers
Karmela Sentija1
Ines Krivak Bolanca1, Suzana Katalenic Simon1
1 Unit for gynecological cytology, Department for Clinical cytology and cytogenetics, Clinical hospital Merkur, Zajceva 19, Zagreb

Objectives: Exfoliated normal endometrial cells in pap smear are usually found in the proliferative phase of the cycle, often with fresh erytrocytes, while abnormal endometrium is frequently followed by histiocytes, deep and superficial stromal and inflammatory cells, with watery background.

Material and methods: Two cases of endometrial carcinoma in postmenopausal women with no findings of endometrial cells in pap smear are presented. Patients reported clinical symptoms, light watery discharge and spotted bleeding from the genital tract. Clinical examination showed trace bleeding and in other case, watery discharge in vagina. Ultrasound showed thickened endometrium over 10 mm, and in other case, 14 mm thick endometrium with presence of endocervical and endometrial polyps. MSCT didnt show any abnormality of uterus. The last pap smear made a year ago showed no abnormalities.

Results: In both cases, during clinical examination, the pap smear for cytological analysis was taken. No endometrial cells in pap smears were found. However, a large number of foamy histiocytic cells were noticed, followed by numerous inflammatory cells on the watery background. Epithelial cells indicated the presence of significant estrogen stimulation. Although in both cases no abnormal cells were found and the pap test was evaluated "without abnormalities", presence of endometrial abnormalities was suggested and further processing was recommended. In both cases, the fractional curettage was performed and the pathohistology analysis confirmed the malignancy, a well-differentiated endometrial adenocarcinoma with an initial invasion in myometrium.

Conclusions: Although some authors find that the presence of histiocytic cells in pap smear is not an indicator of pathological processes, whether it is the number or appearance of histiocytes, our cases indicate that the presence of abundance of endometrial histiocytes may follow endometrial pathology and requires further treatment.

Selected references
IDYLLA - automated PCR platform in the detection of EGFR mutations in cytological samples obtained from transthoracic biopsy of lung tumours.

Invited Speakers

Roman Monczak
Sylwia Magalas, Marcin Napierala, Marek Kowal, Piotr Wójcik

1 K. Marcinkowski University Hospital in Zielona Góra, Department of Pathology, Poland
2 K. Marcinkowski University Hospital of Zielona Góra, Department of Oncology, Poland
3 University of Zielona Góra, Institute of Control and Computation Engineering, Poland
4 Oncogene Diagnostics, Kraków, Poland

Objectives: In patients with certain types of lung cancer EGFR mutation testing is now standard practice. Recently the use of cytological samples for this purpose has become fully accepted.

Material and methods: The research was conducted at the K. Marcinkowski University Hospital in Zielona Góra, where 70 patients with lung tumours requiring EGFR mutation testing underwent transthoracic biopsy. From the samples obtained, cytological smears were produced and stained with H&E, the remaining part being preserved in 1ml of BD SurePath. After confirmation of the presence of cancer cells in the smears, cytospin slides were prepared and immunocytochemical tests conducted (TTF-1,p40/p63, NapsinA, CK5/6). On the basis of microscope images and the results of the immunocytochemical tests, the diagnosis was established in accordance with currently accepted WHO terminology. The cell suspensions were examined using automated platform IDYLLA and EGFR test (RUO-BIOCARTIS). The slides with the cytological smears were scanned and sent to the Oncogene Diagnostics Laboratory in Cracow. After DNA extraction using macro-dissection method, the samples were examined with “Cobas EGFR mutation test v2” (CE-IVD-ROCHE).

Results: The presence of the EGFR mutation was detected in samples from 8 (11.4%) of the patients. This represented 100% correlation in terms of the presence and type of mutation or its absence. In one case the presence of 2 mutations was detected in the Cobas test and 1 in the IDYLLA test. A valid result was obtained in 70/70 (100%) of cases using the IDYLLA platform and 68/70 (97.1%) using COBAS. Establishing the full diagnosis using the IDYLLA TEST took just 2 working days.

Conclusions: The IDYLLA system is a good alternative for EGFR mutation testing. It allows for quick testing of cytological samples and can be used in centres without experience in molecular biology testing.

This research was supported by National Science Center, Poland (2015/17/B/ST7/03704).
Long-term immunoreactivity of antigens detected by immunocytochemistry on cytological slides protected by polyethylene glycol (PEG)

Invited Speakers
Irena Srebotnik Kirbis
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¹ Institute of Pathology, Faculty of Medicine, University of Ljubljana, Slovenia
² Anatomic Pathology Service, Portuguese Institute of Oncology Francisco Gentil, Lisbon EPE, Portugal

Objectives: To evaluate long-term immunoreactivity (IR) of antigens detected by immunocytochemistry (ICC) on cytospins and direct smears protected with polyethylene glycol (PEG) used as a positive controls for diagnostic ICC.

Material and methods: Direct smears and cytospins prepared from different cytological samples in two independent laboratories were immediately fixed in cold methanol (4°C) for at least 30 minutes, protected with 5% PEG and stored at room temperature (RT). Immunocytochemical (ICC) reactions were performed first on the baseline methanol-fixed slides followed by several subsequent ICC on corresponding PEG protected slides stored at RT for a different time. Intensity of ICC reactions and the number of positive cells was semiquantitatively assessed using scores from zero to three. Immunoreactivity was considered reduced if two sequential scores were lower by at least one point.

Results: Immunocytochemical reactions for all together 40 antigens with membrane (10), cytoplasmic (22) and nuclear (8) localization were performed on 921 slides prepared from 183 cytological samples. For majority of antigens (29/40, 73 %) IR on PEG protected slides stored at RT remained unchanged in the first 12 months. Immunoreactivity for GFAP, p40 and Hepatocyte antigen was followed and remained unchanged for one, 8 and 7 months, respectively. Partial or complete loss of IR on slides stored for less than 12 months was found on a single sample out of total evaluated for CD3 (1/7), CD30 (1/4), CD45 (1/10), CK5/6 (1/7), MelanA (1/7) and Vimentin (1/7), while more frequent and earlier loss of IR was found for Ki67 (4/7) and p63 (2/7) with the median time to both events 5.5 months.

Conclusions: Immunoreactivity of majority of antigens detected by ICC on cytological slides protected with PEG and stored at RT is well preserved for at least 12 months, however further follow-up of IR for some antigens is suggested.
Molecular profiling of proteins and transcripts from breast cancer fine needle aspirates (FNA)

Invited Speakers
Bo Franzén
Rolf Lewensohn, Andrey Alexeyenko, Masood Kamali-Moghaddam, Ulf Landegren, Thomas Hatschek, Gert Auer

Karolinska Institutet

Objectives: There are increasing demands for informative cancer biomarkers, accessible via minimally invasive procedures, both for initial diagnostics and for follow-up of personalized cancer therapy, including immunotherapy.

Fine-needle aspiration (FNA) biopsy provides ready access to relevant tissue samples; however, the minute amounts of sample require sensitive multiplex molecular analysis to be of clinical biomarker utility.

The primary objective of our study was to explore recently developed ultra sensitive methods for molecular profiling of FNA leftover materials from breast lesions.

Material and methods: We have applied proximity extension assays (PEA, www.olink.com) and NanoString (NS, www.nanostring.com) technology for analyses of proteins and of RNA, respectively, in FNA samples from patients with breast cancer (BC, n = 25) or benign lesions (n = 33).

Results: We demonstrate that FNA-based molecular analyses (a) can offer high sensitivity and reproducibility, (b) correlate with results from routine analysis (i.e., benchmarking vs ER, PR, HER2, and Ki67), and (c) may also help identify new markers related to immunotherapy. A specific 11-protein signature distinguished all cancer patient samples from all benign lesions in our main cohort and in smaller replication cohort. In addition, FNA BC samples could be divided into two main clusters. This clustering corresponded to some extent to established BC subtypes. Our analysis also revealed several proteins whose expression levels differed between BC and benign lesions (e.g., CA9, GZMB, IL-6, VEGFA, CXCL11, PDL1, and PCD1), as well as several chemokines correlating with ER and Ki67 status.

Conclusions: We conclude that the combined proteomics and transcriptomic methodology is promising for diagnostics and evaluation of treatment efficacy in BC. Our pilot study also supports the emerging role of chemokines in BC progression. Due to the minimally traumatic sampling and clinically important molecular information for therapeutic decisions, this methodology is promising for future immunoscoring and monitoring of treatment efficacy in BC.

Selected references

Normal cytology before cervical cancer and HSIL – large scale rescreening project in Sweden.

Invited Speakers
Henrik Edvardsson

1 Henrik Edvardsson, MD, Consultant Pathologist and Cytopathologist, Central Hospital Karlstad Sweden

Objectives: In recent years cervical cancer has increased in Sweden. This increase has mainly been seen in women aged 50 years and younger. Rescreening of normal cytology before histologically confirmed cancer, high grade intraepithelial squamous lesion (HSIL) and adenocarcinoma in situ (AIS) was done to investigate if there also has been an increase in the number of false negative cytology samples and to find ways to improved quality assurance work.

Material and methods: Files with cases to rescreen were sent to all 27 laboratories in Sweden from the National Cervical Screening Registry. Only women 50 years or younger were included. Reevaluation was done on samples taken between 2001 and 2016 (normal cytology before cancer) and between 2009 and 2017 (normal cytology before HSIL and AIS). Reviewing was done at the local hospitals and only on in-house cases. No central rescreening was performed. Results were collected and analyzed by the National Cervical Screening Registry.

Results: 26 out of 27 laboratories reported results on normal cytology before cancer. Diagnosis was changed from normal to non-normal in 400 out of 1500 cases.
19 out of 27 laboratories reported results on normal cytology before HSIL and AIS. Diagnosis was changed from normal to non-normal in 1460 out of 5980 cases.
In recent years the number of changed diagnoses has been increased on a national level.

Conclusions: In the future rescreening results will be followed more closely on individual, laboratory and national levels.
Reporting rescreening results will be mandatory and standardized.
Large scale rescreening is time consuming and stressful for individuals and laboratories.

Invited Speakers

Lea Isabell Shari van der Linde¹, ²
Christiane Kümpers³, Markus Reischl⁴, Wenzel Vogel¹, ³, Martin Reck¹, ², David Heigener⁵, Klaus Friedrich Rabe¹, ², Jutta Kirfel³, Sven Perner¹, ³, Lutz Welker¹, ²

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⁵ Helios Klinikum Schleswig, Schleswig, Germany

Objectives: The expression of programmed cell death ligand-1 (PD-L1) of non-small cell lung cancer (NSCLC) is a promising feature for immunotherapy [1]. Actually, the PD-L1 expression rate is solely detected by immunohistochemistry with histological sections [2]. The suitability of cytological samples for determination of PD-L1 expression rate is not clear.

Material and methods: The study retrospectively examined 247 (153 adenocarcinomas, 94 squamous cell carcinomas) paired cytological and histological samples of surgical resected specimens of 234 patients (130 male, average age 71.06 years; 104 females, average age 65.24 years) with NSCLC. Samples were stained with 22C3pharmDX and PD-L1 expression was evaluated by three observers by determining the amount of membranous positive tumor cells. Expression rate was rated as exact values (<1%, 10%, 20%...100%) and as tumor proportion score (TPS) (<1%, 1-49%, ≥50%). Mean values of detected expression rates were built and the concordances calculated. The interobserver variability was determined as standard deviation of mean.

Results: Considering the exact values, expression rate of paired cytological and histological samples was concordant in 53% within a delta of 0% and concordant in 82% within a delta of 10%. Regarding the TPS, category for histology and cytology was the same in 74.1%, at which most samples (68%) were scored as <1% and no corresponding samples were scored as ≥50%. Interobserver variability was 0.44 for cytological and 0.20 for histological samples (p = 0.145). In 14.5% cases a PD-L1 Expression was detected by cytology, which could not be determined in histology and 8.9% vice versa.

Conclusions: Determination of PD-L1-expression rate of paired cytological and histological samples is comparable within a delta of 10% discrepancy. Evaluation of cytological samples is more challenging than histological ones demonstrating the requirement of training. Cytological and histological methods are reliable to determine the PD-L1 expression rate with a natural variation due to intratumoral heterogeneity.

Selected references

Pemphigus-like changes – vulvar smears

Invited Speakers
Ines Krivak Bolanca
Suzana Katalenic Simon
1 Department of Clinical Cytology and Cytogenetics, Merkur University Hospital, Zagreb, Croatia

Objectives: Pemphigus vulgaris is an autoimmune blistering disease of the skin and mucous membranes. Genital involvement occurs when most other common sites are concurrently affected or are in remission.

Material and methods: We report a case of a 39-year-old-patient who came to the Department of Gynaecology & Obstetrics for the second opinion of vulvar smear result. First one showed high grade vulvar lesion with abundant acantholytic cells. HPV status revealed positive type 6. After the examination two vulvar smears were taken: one for the immunocytochemical staining with p16INK4A antibody and another for the morphological analysis, stained by PAP staining.

Results: Morphological analysis showed acantholytic cells, and a rather small, almost uniform cells with coarse chromatin with centrally nucleolus but no dyskariotic squamous cells. Immunostaining with p16INK4a antibody was negative. After a talk with a patients and a considerate examination, similar dermatologic changes were noticed under her underarms presented with erythema and macerated plaques and erosions. She said that she had Hailey-Hailey disease (familial benign chronic pemphigus) which affects multiple family members. Additional smears were taken from that lesion and the results were the same. Data reports of squamous cell carcinoma, or high grade lesion of vulva developing in the setting of Hailey-Hailey disease are extremely rare, but one must keep in mind the role of human papilloma virus infection. In this case infection is with low oncogenic type of HPV, and no presence of actually vulvar disease was found. Histologic finding reveal epidermal acanthosis and incipient acanthosis of many of the spinous keratinocytes of the epidermis and foci of dyskeratosis which is in favour of Hailey-Hailey disease and not a vulvar condition.

Conclusions: A case like this confronts the gynaecologists, cytologists and pathologist with the necessity of including a rare disease into particular consideration and investigation.
Population-based primary HPV mRNA cervical screening compared with cytology screening.

Invited Speakers
Ylva Lindroth 1
Christer Borgfeldt 2, Gunilla Thorn 3, Gunilla Bodelsson 3, Ola Forslund 1

1 Department of Laboratory Medicine, Medical Microbiology, Lund University, Sweden
2 Department of Obstetrics and Gynaecology at Skåne University Hospital, Lund University, Sweden
3 Department of Laboratory Medicine, Clinical Genetics and Pathology, Lund, Sweden

Objectives: To audit a primary HPV screening program, by comparing its cytology results 2017 to that of corresponding women screened with conventional cytology during 2016.

Material and methods: Primary HPV screening for cervical cancer by HPV mRNA testing (Aptima HPV assay, Hologic) was implemented in January 2017, for women ≥30-70 years, in the Region of Skåne, Sweden. HPV positive samples underwent cytology assessment, and women with any degree of abnormal cytology were referred for colposcopy. Women not attending invitation at 65 years receive re-invitations up 70 years.

During 2017 63,055 women were analysed for presence of HPV within the primary HPV screening. To ensure that the primary HPV screening also detects the few cases of cellular changes that may occur without an active HPV infection 5,039 women aged 40-42 years were co-tested by cytology and for presence of HPV.

We compared proportions of abnormal cytology or worse (ASCUS+) between cytology screening 2016 (N=45,906) and primary HPV screening 2017 (N=49,842) of women aged 30-65 years.

Results: Among women ≥30-65 years, the proportion ASCUS+ was similar for cytology screening (3.52%) and for primary HPV screening (3.70%). Only the proportion of ASC-H changed by the use of primary HPV screening, with an increase from 0.13% to 0.23% (p<0.001). The colposcopy referral rate increased by 54% (from 2.41% to 3.70%) when primary HPV screening was introduced.

HPV was detected among 7.0% (4,433/63,055) of the screened women ≥30-70 years. The HPV prevalence decreased with increasing age, from 11.2% to 3.95% among women aged 30-39 and 66-70 years, respectively (Figure 1).

Among the co-tested, HPV was detected in 100% (28/28) of HSIL and ASC-H (9/9), and in 80% (4/5) of atypical glandular cells (AGC).

Conclusions: Primary HPV screening demonstrated similar prevalence of ASCUS+ cytology as conventional screening. Primary HPV screening decreased cytology assessments by 86% of women 30 - 70 years.
Figure 1. Prevalence of HPV-mRNA stratified by age groups (orange), and prevalence of ASCUS+ cytology by the use of cytology screening year 2016 (blue) and by primary HPV screening 2017 (grey).
Primary pleomorphic rhabdomyosarcoma of the submandibular gland - a case report

Invited Speakers

Iris Fabijanić
1
1 Department of Pathology and Cytology, University Hospital Centre Zagreb, Zagreb, Croatia

Objectives: Rhabdomyosarcoma (RMS) accounts approximately for 2-3% of all adult sarcomas. RMS mostly appears as embryonal and alveolar subtype, while pleomorphic subtype (PRMS) is exceedingly rare. Due to controversial diagnostic criteria and similarities in clinical and imaging features between PRMS and other soft tumors, PRMS is often misdiagnosed. Hence, we present the case of pleomorphic rhabdomyosarcoma arising in the submandibular gland diagnosed by fine needle aspiration cytology (FNAC) and ancillary methods.

Material and methods: A 40-year-old woman with an unremarkable past medical history presented with a non-tender, palpable mass in the left submandibular region of 2 months duration. There was no evidence of fever, lymphadenopathy or hepatosplenomegaly. Routine laboratory tests were within normal limits. Ultrasound (US) showed a hypoechoic mass in the left submandibular gland and US-guided FNA was performed. Cytologic examination revealed pleomorphic population of atypical cells with abundant eosinophilic cytoplasm and prominent mitotic activity.

Results: Immunocytochemistry showed these cells to be MyoD1, Myogenin and Desmin positive and EA, EMA, ERA, AE1/AE3 negative. A diagnosis of malignant mesenchymal tumor, probably pleomorphic rhabdomyosarcoma was made. Molecular analysis was negative for EWSR1 and SS18(SYT) rearrangements. Imaging revealed multiple cystic lesions of varying sizes of high-signal intensity. Subsequently, a histologic diagnosis of PRMS was confirmed. A 1-year later, the patient had palpable mass near the post-operative scar and US-guided FNAC revealed recurrence.

Conclusions: US-guided FNAC is currently regarded as minimally invasive and effective diagnostic procedure with an accuracy of 96% in the diagnosis of salivary glands tumors. Furthermore, US-guided FNAC may be useful for the follow-up and monitoring of the disease. A multidisciplinary approach is necessary in diagnosis and sub classification of malignant soft tissue tumors.

Selected references

Reactive atypical cells mimicking adenocarcinoma in respiratory cytology: a review

Invited Speakers

Kunimitsu Kawahara1
1 Department of Pathology, Osaka Habikino Medical Center, Osaka, Japan

Objectives: All cellular components of the respiratory tract, including squamous metaplastic cells, respiratory columnar cells, terminal bronchial or alveolar lining cells, and pulmonary macrophages can exhibit potentially concerning atypia. The presence of these reactive atypical cells can sometimes be alarming even for highly experienced cytomorphologists.

Material and methods: In this review, I analyze and illustrate cellular changes and associated benign clinical conditions that can mimic pulmonary adenocarcinomas in respiratory cytology.

Results: Among the cellular components of the respiratory tract, reactive type II pneumocytes and reactive bronchial respiratory epithelial cells can occasionally be highly atypical, leading to a false-positive diagnosis of adenocarcinomas. Atypical type II pneumocytes aggregate in a three-dimensional cluster consisting of cells with high nuclear-to-cytoplasmic ratio, hyperchromatic nuclei, coarse chromatin, prominent nucleoli, and nuclear membrane irregularities. They have been reported in cytology from samples of organizing pneumonia, pulmonary infarcts, adult respiratory distress syndrome, acquired immunodeficiency syndrome, oxygen therapy, and pulmonary fibrosis. An atypical bronchial respiratory epithelium also shows three-dimensional clusters with a compact papillary configuration consisting of cells with coarse chromatin, and macronucleoli, but with smooth and regular membranes. It can be seen in samples from patients with pneumonia, pulmonary infarct, and chronic obstructive pulmonary diseases (asthma, chronic bronchitis and bronchiectasis), and from those undergoing radiation therapy and chemotherapy.

Conclusions: Careful attention to cellular details and consideration of potential false-positive results are the first step towards avoiding false-positive diagnosis of adenocarcinoma. Ensuring the correlation of clinical and radiological findings with the cytologic features is very important to avoiding such mistakes. Caution should be exercised in rendering positive diagnoses, especially if all cytologic criteria for pulmonary adenocarcinoma are not fulfilled.
Results of HPV screening in the Netherlands

Invited Speakers
Lia van Zuylen
1 Lia. van Zuylen-Manders

Objectives: -
Material and methods: -
Results: -
Conclusions: "Primary HPV Screening, results of the first two years."

First I will give a quick review how we started in the Netherlands with primary HPV screening. In 2011 the Dutch Health Council recommended Primary HPV Screening above Primary Cytology Screening. In 2017 after an European Tender we started with primary HPV screening in 5 labs throughout 5 regions in the Netherlands. Each region has a population of 100,000-120,000 women who were invited for HPV screening.

Following are the results of the first two years of primary HPV screening based on data of cohort 2017-2018. Over 9% of all the participants were HPV positive. The HPV positivity decreases, as expected, by age. From 20 % in the youngest age group to 5 % in the oldest.

In 32% of the hrHPV positive women, abnormal cells (pap2+ / ASCUS+) were found. In the renewed population screening for cervical cancer, participants can also order a self-sampling device. Nearly 7% of the participants used this self-sampling device. Of the women who used the self-sampling device, more than 7% were hrHPV positive, and in 37% of them, abnormal cells were found.

Selected references
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SPINDLE EPITHELIAL TUMOR WITH THYMUS-LIKE ELEMENTS (SETTLE): A Case Report

Invited Speakers
Alex Anton Bruno Lozić
Neven Mateša, Žana Besser Silconi, Loredana Labinac Peteh, Petra Peruško Kozina, Tamara Mićić, Claudio Primc, Elvira Mustać

1 Pula General Hospital, Pula, Department of clinical cytology, Croatia
2 Sestre milosrdnice University Hospital Center Zagreb, Department of oncology and nuclear medicine, Croatia
3 Pula General Hospital, Pula, Department of pathology, Croatia
4 Pula General Hospital, Pula, Department of nuclear medicine, Croatia
5 Rijeka University Hospital Center, Department of pathology and cytology, Rijeka, Croatia

Objectives: Spindle epithelial tumor with thymus-like elements is a rare, biphasic and initially indolent tumor of thyroid with a low malignant potential in young patients (median age 18 yrs). Delayed metastases can occur in lymph nodes, mediastinum, vertebrae or lungs even many years after the initial diagnosis. There are less than 50 cases reported in the literature and few reported metastases. The cytologic descriptions of SETTLE are rare and tend to focus on a hypercellular biphasic appearance. Macroscopic description is a well-circumscribed encapsulated or infiltrative tumor with white tan cut surface separated by sclerotic stroma. Microscopically, it has a biphasic pattern made by spindle cells and epithelial structures (cords, tubules, papillae or glandular appearance), but there are also rare monomorphic variants which have spindle or glandular appearance only. SETTLE may have a good prognosis if appropriately treated at the initial presentation and if a patient undergo long-term monitoring with a regular clinical and morphological evaluation. The golden standard is surgery and in advanced stages of disease also chemotherapy and radiotherapy.

Material and methods: We report findings in a 17-year-old girl through ultrasound and cytologic findings, surgical procedure and histopathological findings.

Results: After finding a small, hypoechogenic node 5 mm in diameter on ultrasound examine by a specialist of nuclear medicine, we made a FNAB. The node was interpreted morphologically by a cytologist as “probably SETTLE” although the basic immunocytochemistry was partially done. The patient underwent a recommended left lobectomy of the thyroid and the diagnosis was confirmed by pathologist (morphology and immunohistochemistry). The follow-up ultrasound almost 4 years later revealed no local recurrence or metastatic disease.

Conclusions: Fine needle aspiration cytology can be a useful diagnostic tool to consider the SETTLE of thyroid as a possible differential diagnosis due to its specific cytomorphological features and further on leads to the right treatment.

Selected references
Cytologic findings of spindle epithelial tumor with thymus-like elements: Baste Subia MN et al., Laryngoscope, 2018 Feb; 128(2)
Spindle epithelial tumor with thymus-like differentiation of the thyroid in a 70-year-old man: Lee S et al., Ann Surg Treat Res, 2018 Jun; 94(6)
Spindle epithelial tumor with thymus-like differentiation (SETTLE): clinical-pathological features, differential pathological diagnosis and therapy: Ippolito et al., Endokrine, 2016 Mar; 51(3)
Thyroid gland Other carcinoma SETTLE Wei S et al., PathologyOutlines.com, Inc., 2003, rev.12th Sep 2018
Swedish Society for Cytotechnologists and what we do

Invited Speakers

**Kamilla Backlund Arvidsson**¹
¹ Swedish Society for Cytotechnologists

**Objectives:** The Swedish Society for Cytotechnologists was founded 1979 as a non-profit society. In 2015 the society became a so called “profession”-society, within the Swedish union Naturvetarna, still as a non-profit society. The board is made up of a president and six to eight members. The board is elected at the annual meeting, which takes place at a yearly conference. Every other year this conference is hosted solely by the Swedish Society for Cytotechnologists, every other year it is cohosted with other Swedish societies in the pathology field. Since the beginning the aim has been to represent the interests of cytotechnologists in Sweden, and to promote and develop the profession. One of the most important tasks has been, and still is, the goal to become a licensed profession.

**Material and methods:** N/A

**Results:** N/A

**Conclusions:** N/A
The changing role of Cytotechnologists after implementation of HPV primary screening in Norway

Invited Speakers
Maj Liv Eide¹
¹ Trondheim University Hospital and Norwegian University of Science and Technology, Trondheim, Norway

Objectives: The Cancer Registry administer the cervical screening program in Norway. In 2015, three counties implemented HPV primary screening. The aim of this presentation is to show how HPV primary screening has changed the everyday work for the Cytotechnologists in these counties.

Material and methods: The cytology laboratories at the University Hospitals in Trondheim, Bergen and Stavanger implemented HPV primary screening in women 34-69 years old, with five years screening intervals. The follow up after positive HPV result is reflex cytology. Cytotechnologists in these laboratories also perform the HPV-testing. From 2015-2018 the laboratories performed primary HPV screening on women aged 34-69 who were born on an even birth date, and from the first half of 2018 on all women aged 34-69. Cytology remains the screening method of choice for women 25-33 years old with three years screening intervals. During this period, the screening algorithm changed once and test of cure was implemented.

Results: Seven percent of women 34-69 years are HPV positive. Ninety-three percent are hence HPV negative with no cytology. Since the start in 2015, the number of cytology specimens decreased 20-25 % the first three years and the last year approximately 40 %. There is a challenge with two different screening methods and three different algorithms, as cytotechnologists spend more time than before on specimen logistics and control of the screening history to ensure proper follow-up. Cytotechnologists have this competence after years of screening cytology specimens.

Conclusions: The controlled implementation of HPV primary screening in the first counties in Norway has been successful and made it possible for the laboratories to adjust. The experience of having cytotechnologists perform the HPV-testing is very good and keeps the cervical cytology competence, which we need for several years still, in the laboratory.
The many faces of lung CARCINOIDS in liquid based cytology.

Invited Speakers
Lars Övergaard

Objectives: Carcinoids constitute 5% of all pulmonary malignancies.

They are challenging to diagnose, but it is important to single out the carcinoids since their treatment differs from other types of lung tumours.

The criteria for carcinoid diagnoses are based on air-and alcohol-fixed cytology smears.

Our objective was to evaluate the use of liquid based cytology in carcinoid diagnoses.

Material and methods: The presentation is based on cases diagnosed using ThinPrep ® at Lund University Hospital between 2012-2018.

Results: 13 cases of 19 liquid based samples provided diagnostic information leading to a carcinoid diagnose.

Conclusions: Liquid based cytology can confirm carcinoides in pulmonary cytology.

Selected references
The quality of Papanicolaou staining assessed in a pilot international external quality assessment (EQA)

Invited Speakers
Irena Srebotnik Kirbis¹
Helena Gutnik¹, Margareta Strojan Flezar¹
¹ Institute of Pathology, Faculty of Medicine, University of Ljubljana, Slovenia

Objectives: To evaluate interlaboratory variability in quality of Papanicolaou staining, staining procedure and perception of staining quality.

Material and methods: All cytospins (60) were prepared from the same mixture of cells from remains on a cervical brush after conventional smears were prepared in three healthy women enrolled in cervical screening programme. Cytospins were fixed according to five different protocols (Mercofix with or without air-drying, 96 % ethanol, 70 % ethanol, air-drying) and sent to 12 laboratories with a request to stain them, assess the quality of staining and return stained slides with the staining procedure details. The panel of four assessors from two different laboratories blindly and independently assessed the quality of Papanicolaou staining using published criteria for Papanicolaou staining and scoring system from one to 10.

Results: The highest score achieved by individual laboratory for the quality of Papanicolaou staining on all five differently fixed cytospins was 8.4 ± 1.1 and the lowest 5.4 ± 0.9. The quality of Papanicolaou staining was the best in all laboratories on the cytospin fixed with Mercofix ( = 8.0 ± 0.9) and the worst on the cytospin air-dried before fixation with Mercofix ( = 5.8 ± 0.9). Significant difference between in-house assessors and external assessors scores for the quality of staining (≥ 2 scores) was found in 3/12 laboratories. Duration of Papanicolaou staining procedures varied from 18 to 80 minutes with 12 to 26 steps and staining times for haematoxylin, OG6 and EA50 from 12 seconds to 4 minutes, 2 seconds to 4 minutes and 1.5 to 6 minutes, respectively.

Conclusions: Our pilot international EQA study on cytospins prepared from the same sample clearly demonstrated great variability in all aspects of Papanicolaou staining and showed that the quality of Papanicolaou staining is affected by fixation procedure as well as by staining procedure.
The role of Cytotechnologists in transplantation diagnostics

Invited Speakers
Veronika Anić¹
Nada Tudek¹, Mariela Stipetic¹
¹ Department of Clinical Cytology and Cytogenetics, Merkur University Hospital, Zagreb, Croatia

Objectives: In Merkur University Hospital in Zagreb, Croatia, there is a long-standing, very successful tradition of transplantation medicine. Transplantation diagnostics in cytology can be divided into pre-transplantation, during transplantation, and follow-up or post-transplantation diagnostics. Cytotechnologists participate in diagnosing and monitoring the treatment success of such patients.

Material and methods: Data were collected during long-term experience in the implementation of diagnostic tests required for transplantation of solid organs (kidney, single or combined with pancreas or liver) and in patients undergoing peripheral blood stem cell transplantation (PBSCT).

Results: Each part of the transplantation process involves certain analysis. In haematological patients undergoing peripheral blood stem cell transplantation (PBSCT) procedure, we perform cell viability of fresh and cryopreserved peripheral blood progenitor cell (PBPC) grafts. Due to the fact that infections remain a major source of morbidity in immunocompromised patients undergoing PBSCT procedure we perform swabs of the mouth cavity and skin changes, peripheral blood cells and bone marrow analysis in order to follow the course and effectiveness of the treatment. Polyoma virus (PV) infection is being recognized as a cause of renal allograft dysfunction. Patients, before and after kidney or liver transplantation, undergoing immunosuppressive prophylaxis and therapy to prevent rejection, are at risk for BKV reactivation and possible nephropathy. In those patients, among many other analysis, we perform screening of the urine samples for decoy cells as screening method for BK viruria.

Conclusions: Cytology is an accurate, fast and sensitive diagnostic method in assessing and monitoring the conditions and immunologically related transplantation events in patients receiving chemotherapy and immunosuppressive therapy during transplantation process. Given the changes in cytodiagnostics by introducing new technologies, reduced and changed role of cytotechnologists, this is a very good example that cytotechnologists possessing knowledge and competencies can perform diagnostic tests in transplantation medicine.
Thyroid Cytology Slide Seminar: Thyroid FNA Cases in the Mirror of Bethesda Reporting System: Bethesda I - Unsatisfactory

Invited Speakers
Jaroslava Dušková, CSc., FIAC¹, ², ³
¹ Inst. Pathol., 1st Fac. of Medicine, Charles University, Prague
² CGOP Ltd., Prague
³ Topelex, Ltd., Prague

Objectives: The new WHO Classification of thyroid neoplasms induced revision of the Bethesda system for reporting thyroid Cytopathology. The implied risk of malignancy has not changed for Bethesda category I - Unsatisfactory. It is a category that deserves attention.

Material and methods: Illustration cases. Retrospective material analysis.

Results: Illustration cases:

1. 85 year old female with hypodense nodules in the left lobe of thyroid and cervical lymphadenopathy.
   1st FNAB – necrosis only in both thyroid and lymph nodes aspirates.
   2nd FNAB – necrosis in thyroid aspirate, undifferentiated carcinoma in the lymph node material. Died from generalisation.

2. 76 year old woman. Solitary nodules in both lobes, the left nodule with calcification.
   FNAB BI right lobe, B III/V left lobe. Total thyroidectomy – bilateral papillary carcinoma.

The analysis of our B I cases originating from both a university and a private laboratory during the years 2013-2018 shows the incidence of 20% and 17% respectively in altogether 6280 thyroid cytologies diagnosed.

The causes for BI both qualitative and quantitative at the material taker’s and pathologist’s side are analysed.

Conclusions: BI is not just an „easy“ case quickly solved in the daily pathologist’s workload.

The cause of BI must be formulated. It is highly desirable to suggest further strategy for the repeated material acquisition.
Unusual cases of uterine cervix

Invited Speakers
Vedrana Roger
1
1 Departement of Clinical Genetics and Pathology

Objectives: In most cases when we speak about cervical cancer we think of squamous cell carcinoma or adenocarcinoma. But in some rare cases there are other primary tumors that can occur in cervix, such as neuroendocrine tumors and lymphomas.

Material and methods: A liquid based Pap specimens from three different women prepared with ThinPrep. Two are from 2018 and one from 2017.

Results: Patient one, with histologically confirmed large-cell neuroendocrine carcinoma, was diagnosed cytologically with malignancy without further specification.
Patient two, with histologically confirmed small-cell neuroendocrine carcinoma, was diagnosed cytologically with atypia of unclear celltype.
Patient three, with histologically confirmed diffuse large B-cell lymphoma, was cytologically diagnosed with unsatisfactory for evaluation due to scant squamous cellularity.

Conclusions: Cytological recognition is difficult mostly because of lack of experience, since only a few cases of each of these conditions have been reported.

Selected references


Work of Cytotechnologists in Finland

Invited Speakers

Eeva Liikanen

1 Tampere University of Applied Sciences

Objectives: The aim of this study was to survey the work that cytotechnologists carry out in Finland.

Material and methods: An electronic questionnaire was planned with the Board of the Finnish Association of Cytotechnologists and an email containing the link was sent to all 107 of its members in January 2018.

Results: Just under half (45%) replied. Their average age was 51 (range 28-64), 41% had a Bachelor degree, 59% had college-level training and they had spent an average of 15 years screening cytology specimens.

All cytotechnologists screened Pap smears, 94% screened urinary and respiratory specimens, 82% screened effusions and 39% screened fine-needle aspirations. Less than half (n=22, 47%) of respondents did not sign off normal and inadequate Pap smears.

Most of the cytotechnologists (85%) had duties other than screening and they mostly included a combination of histotechnology and cytotechnology (37.5%) or just cytotechnology (17%) or histotechnology (9%). The other 15% only screened cytology specimens.

Conclusions: Cytotechnologists reported many other duties in addition to microscopy screening.
“Cytological diagnosis of malignant mesotheliomas in effusions - Advantages and limitations”

Invited Speakers
Lutz Welker\textsuperscript{1,2}
Lea I.S. van der Linde\textsuperscript{1,2}, Florian Stellmacher\textsuperscript{3}
\textsuperscript{1} Cytology Laboratory, LungenClinic Grosshansdorf, Großhansdorf;
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Objectives: Malignant mesotheliomas (MM) are rare asbestos related tumors of serous membranes, in 96.4\% of pleura. Whereas histological analysis of biopsies are well established, the suitability and accuracy of cytological diagnosis of MM remains controversial. In the earliest stages 70\% of patients develop effusions. Consequently, one of the first specimens that laboratory receives are effusions. With regard to the clinical symptoms and progress of MM the diagnosis without any surgery procedures seems to be a desirable goal.

Material and methods: The present retrospective study included 2205 consecutive analyzed effusions of 1769 patients (1136 male, average age 69.2 years; 63.4 female, average age 67.3 years) obtained from January 2011 to December 2017. Out of these patients, 144 were diagnosed with MM. Additional determinations of hyaluronic acid (HA) value were performed.

Results: Cytological evaluation stated 80 of 145 MM on the whole, thereof 70 by cytology alone and additional 10 by determination of HA values. The cytological MM diagnosis reached a sensitivity of 0.49, a specificity of 1 and a positive predictive value (PPV) of 1. Determination of HA values raised the sensitivity to 0.56 without affecting specificity or PPV.

Conclusions: Relevant cytological MM features are the appearance of a single epithelial cell lineage with mesothelial nuclear and cytoplasmic features, characteristic cell formations and cell clusters, and high number of cells. In spite of disadvantages of cytology (uncertain dignity of individual mesothelial cells, poor detection of tumor architecture and spread) it provides a number of advantages (low invasiveness, minimization of tumor cell seeding, large diagnostic area). Immunocytochemistry enables the detection of MM cell character of suspicious cells and the differentiation from malignant extra pleural tumors. The crucial influencing factor is the exfoliation rate of MM cells. Despite the low sensitivity the specificity of cytological MM diagnosis is very high and cytological MM diagnosis is as reliable as histology.
Free Communication
1. Gynecological Cytology
The limitations of organized cervical cancer screening

1. Gynecological Cytology
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Objectives: To show the limitations of organized cervical cancer screening and the influence of atrophy, infection and inflammation on results of PAP smears as well as on the number of cervicis uteri biopsies.

Material and methods: The comparison was performed on two groups of women from Zajecar district upon whom cervical cancer screenings with conventional PAP smear had been conducted. The first group was comprised of women screened in a mobile ambulance in the villages surrounding Zajecar city without previous preparation. The second group was comprised of women who came regularly to health center for gynaecology check up and depending on the results had been treated with antibiotics or hormone therapy because of an infection or atrophy before taking PAP smear.

Results: The target group included a total of 2,415 women aged between 25 and 69 screened in 2016 and 2018. The first group of 1018 women, screened in 2016, resulted with 81 (7.95%) women having suspicious PAP smear and undergoing biopsies. The second group of 1397 women, screened in 2018, had 53 (3.79%) women with suspicious PAP smear and underwent biopsies.

Conclusions: Because of infection and atrophy, treatment of women before taking PAP smears is useful for both patients and doctors and would reduce inadequate and false positive findings and consequently the number of biopsies.

Selected references
The Institute of Public Health of Serbia “Dr Milan Jovanovic-Batut”, National program of organized cervical cancer screening, Belgrade 2013

<table>
<thead>
<tr>
<th>Year</th>
<th>The number of cervical cancer screenings with PAP test</th>
<th>The number of biopsies</th>
</tr>
</thead>
<tbody>
<tr>
<td>2016</td>
<td>1018</td>
<td>81 (7.95%)</td>
</tr>
<tr>
<td>2018</td>
<td>1397</td>
<td>53 (3.79%)</td>
</tr>
</tbody>
</table>
2. HPV Cervical Screening
Abnormal cytology results and HRHPV status among sexually active young women from an urban area in Poland – prevalence, genotypes and phenotypes

2. HPV Cervical Screening
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Objectives: The aim of our retrospective study was to determine a prevalence and a distribution of a high-risk human papillomavirus (HRHPV) among sexually active women under the age of 30 (the age range between 16 and 29) with abnormal cytology results in an urban area in Poland.

Material and methods: A study group was selected among 871 sexually active women who were attending an office-based opportunistic cervical cancer screening, outside the public health system in Wrocław (Poland) between August 2015 and August 2018. Endocervical samples for HRHPV detection were collected together with material for liquid-based cytology in all patients. A real-time PCR for genotyping HPV types 16 & 18 and phenotyping 12 non-16 & non-18 types was used. All cytology samples were processed using an automated laboratory preparation.

Results: Of the 215 women with abnormal cytology, 172 were infected with at least one HRHPV genotype. The mean age in group was 25.8 years. The prevalence of HR-HPV infection among these women was 80%. Data for each cytology result are given in the following order: % of all cytology results/% of HRHPV-positive cases/% of types 16 or (and) 18+/% of types non-16 & non-18-positive for ASC-US: 10.9/71.6/54.4/45.6; LSIL: 11.9/84.6/32.9/67.1; ASC-H 0.7/100.0/66.7/33.3; HSIL: 1.0/100.0/66.7/33.3 & AGC: 0.1/100.0/100.0/0.0 were detected. The most frequent genotype was HPV 16 (38.9%).

Conclusions: Type 16 is the most common HPV genotype in the group of Polish women <30 from an urban area with abnormal cytology results. Simultaneously, in this group HPV-positivity for ASC-US & LSIL results is higher compared with women over 30. A larger study with a more representative sample would be needed to determine predominant oncogenic genotypes in described sub-region and especially in cancer cases in this young-age group.
Objectives: A shift of primary cervical screening from cytology to hrHPV detection introduce a major change in the technical and logistical infrastructure for screening. To develop real-world evidence for preferred cervical cancer screening strategies, we compared liquid based cytology (LBC) screening every 3 year (current screening modality) with high-risk human papilloma virus (hrHPV) testing every 5 year in Norway (health service study trial number 006_2014_10_RHS).

Material and methods: Between February 2015 and December 2017, 185,114 women, aged 34 to 69 years, who returned for their routine, triennial cervical cancer screening were assigned hrHPV-testing (cobas® HPV Test (Roche Diagnostics)) or LBC, based on even/odd day of birth. Cervical intraepithelial neoplasia grade 2, 3 and cervical cancer (CIN2+) was detected among 58,971 women who completed their follow-up of a positive screening test by 2017.

Results: Screening attendance by age was similar in HPV-screening and LBC-screening, being 53.6% vs 52.3% after 1st and 31.8% vs 32.4% after 2nd reminder, respectively. The proportion of screening test positives was 5.4% in LBC-screening and 6.5% in HPV-screening, and declined by increasing age. HPV16/18 were detected in 28% of hrHPV-positives. Compared to LBC-screening, we observed 40% more biopsies all over and 50% more CIN3+ in HPV-screening.

Conclusions: HPV-screening was well accepted and detected more pre-cancers, suggesting that HPV-screening should replace LBC-screening. Randomized implementation of HPV-screening allows to monitor the performance of novel technology in real-life, reassuring the overall high performance of the program and mitigating the transition. Based on the results from the randomized implementation, Norwegian health authorities have decided upon a randomized national implementation of primary HPV test from 2019 until 2022. The 3 piloting laboratories have converted to primary HPV-screening.
Evaluation of the clinical performance of the Aptima HPV assay for women aged 60-64 years for primary screening in Denmark

2. HPV Cervical Screening

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**Objectives:** Danish Health Authorities recommend HPV DNA testing as primary screening in women aged 60-64 years. However, in multiple studies the mRNA-based Aptima human papilloma virus test has shown similar sensitivity to DNA-based test methods when detecting high-grade cervical intraepithelial neoplasia. Moreover, the mRNA-based method targeting the E6/E7 oncogenes has shown higher specificity. The objective of the present study is to compare the performance of Aptima HPV mRNA assay with the cobas HPV assay routinely used in our department.

**Material and methods:** We examined 2563 Danish women aged 60-64 years. Cervical samples were collected during routine screening visits using BD SurePath™ liquid-based Pap test collection vials and analyzed using the Aptima HPV mRNA and cobas HPV assays. The comparison includes positivity rates/genotype and sensitivity/specificity against a clinical endpoint defined as a histologically confirmed diagnosis of CIN2/3+. All women with a positive HPV test were examined using liquid-based cytology, and women with ASCUS (or repeated HPV positivity) were referred to a gynaecologist.

**Results:** The HPV high-risk genotypes among the women were distributed as follows: negative using both methods, 2324 samples (91%); positive using both methods, 126 samples (5%); only Aptima-positive, 46 samples (2%, 10 were positive for HPV high-risk genotypes 16/18/45); only cobas-positive, 67 samples (3%, 11 were positive for genotypes 16/18). The examined women are currently in cytological and histological follow-up. Presently, among samples showing discrepant HPV high-risk genotyping, there is a tendency towards the Aptima HPV mRNA assay having the highest sensitivity to detect women with abnormal cytology (62% vs. 39%).

**Conclusions:** As the examined women are currently in cytological and histological follow-up, remaining data will be presented. Presently, the Aptima HPV mRNA assay seems to be a preferable choice of screening method with the potential of lowering the number of false positive HPV screening results and unnecessary colposcopy referrals.
Measuring the risk of harboring a positive HPV test in a cervical screening program. A 3-year follow up after cotest with normal cytology.

2. HPV Cervical Screening

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Objectives: To determine the cumulative risk of developing a CIN2+ lesion in HPV+/Cytology negative (AHPV+/Cyt-) patients after cotest. We also aim to determine the role of a persistent HPV infection in this group of patients.

Material and methods: After cotesting 5,053 women with LBC cytology and APTIMA® HPV (AHPV) within an opportunistic screening program, 5,9% had AHPV+/Cyt- results. 188 underwent colposcopy with biopsy. Women without CIN2+ were followed-up with a new cotest and colposcopy/biopsy according to current guidelines. Statistical risk assessment was calculated.

Results: 14.9% of AHPV+/Cyt- women biopsied at baseline had CIN2+ lesions after a 65.2% attendance to colposcopy. The mean follow-up time was 29.1 months. From 151 women with a previous biopsy, 99.3% (150) underwent a new cotest and 136 had at least a second biopsy, yielding 12 new CIN2+ lesions (8.8%). The 3-year CIN2+ cumulative risk was 23.7%. Persistent AHPV was seen in 44(29.1%) patients with an associated risk of CIN2+ lesion of 28.6% and a cumulative risk of 43.5% after a 3y FU. In contrast, the CIN2+ risk associated to the 70.9% of women who cleared the HPV infection was 0%, and its corresponding cumulative value was 14.9%. Cytology was abnormal in 75% (7 HSIL; ASCUS) of those patients with CIN2+ lesions.

Conclusions: The risk of CIN2+ in AHPV+/Cyt- women decreased 3 years after cotest (from 14.9 to 8.8%). Although most women (70.9%) cleared the infection, those with persistent HPV harbour a large 3-year cumulative risk (43.5%) for high-grade lesions. The sensitivity of cytology was 75% for CIN2+. Patient adherence, which increased from 65.2% at baseline to 90.1% during FU in this study, is essential for the correct management of these women.

Selected references

Tertiary p16/Ki67-test as a new diagnostic approach for HPV-positive and cytology-negative results in a primary HPV-based cervical cancer screening

2. HPV Cervical Screening

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Objectives: Primary HPV-based cervical cancer screening requires triage of HRHPV-positive women to identify those at HSIL+ (CIN2+) risk due to low specificity for these lesions. Secondary cytology-based algorithm predominates in clinical approach. The aim of our study was to investigate a role of double stained p16/Ki67-test (DS) to improve HSIL-detection in HRHPV-positive/NILM cases.

Material and methods: In the office-based opportunistic (outside the public health system) primary-cotesting screening, a retrospective study (08/2015-01/2019) among women aged 30–70 was conducted. In HRHPV-positive/NILM cases with a cytology-based secondary approach, DS as a tertiary test was performed. The study group was selected from 4496 women with known HRHPV status and NILM cytology result characterised by: 437 HRHPV+/296 DS results/67 DS-positive. 42 cases achieved the final study endpoints with DS and histology results (after colposcopy). LBC and DS were carried out using an automated laboratory procedures. HRHPV DNA was detected using PCR assays which phenotypes 12 non-16/non-18 types and genotypes types 16 and 18. Histological diagnosis was made according to LAST and WHO 2014 terminology and p16-testing was performed according to ASCCP/CAP 2012 recommendations.

Results: In the final study group 31 positive DS were detected including: 14 cases of HPV 16 and 18 (6 HSIL/7 negative or LSIL in histology), 17 cases of HPV non-16 and non-18 (4 HSIL/13 negative or LSIL in histology) and 1 case of primary cervical lymphoma (B-DLBCL, NOS). In 11 DS-negative group all cases were HPV 16 and 18-positive (0 HSIL/11 negative or LSIL in histology).

3. Lymph Node Cytology
Determination of cytoplasmic light chains and Bcl-2 expression improve accuracy of cytological diagnoses in B-cell lymphomas with inconclusive flow cytometric surface light chain results

3. Lymph Node Cytology

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Objectives: Ratio between surface immunoglobulin (sIg) light chains (LC) determined by flow cytometer (FC) is widely used for distinguishing B-cell lymphomas (BCL) and reactive lymphocytic proliferations (RLP). Since sIg LC ratio cannot always be determined we want to assess if cytoplasmic immunoglobulin (cIg) LC and Bcl-2 expression improve accuracy of cytological diagnoses of BCL.

Material and methods: Our study included 232 fine needle aspiration lymph node cases suspicious for BCL. sIg LCs were inconclusive in 220 cases. In all cases expression of Bcl-2 and cIg LC were determined by FC.

Results: 130 patients had lymphoma and 102 RLP. Five patterns of sIg LC staining were found: negative (69 %; 159), dual positive (20 %; 47), difficult to interpreted (6 %; 14), monotypic (2%; 5) and polypotypic (3%; 7). In the cases with inconclusive sIg LCs the percentage of lymphomas was highest in the dual positive group (92 %; 43/47) and lowest in negative group (44 %; 70/159). Morphology coupled with cIg LC determination and Bcl-2 expression was able to give a correct diagnosis in 91% of cases.

Conclusions: Determination of cIg LC and Bcl-2 expression analysis allows accurate cytological diagnosis of BCL in most cases with inconclusive sIg LC.

Selected references

4. Head and Neck Cytology (including Salivary Gland)
Pathologist-Performed Ultrasound-Guided Salivary Gland Fine-Needle Cytology reduces inadequacy rates and improves specificity and sensitivity.

4. Head and Neck Cytology (including Salivary Gland)

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**Objectives:** The aim of this study was to compare the results of ultrasound-guided salivary gland fine needle cytology (UG-FNC) performed through collaboration between pathologists and ear nose and throat (ENT) surgeons at a university hospital outpatient clinic.

**Material and methods:** This study was a retrospective review of consecutive patients in a 2-year period (January 2016 – January 2018) who underwent UG-FNC of one of the salivary glands. The UG-FNC was performed by either (i) an ENT surgeon alone or (ii) a pathologist. Medical records, cytology results, and surgical pathology results were collected and analysed with respect to the two practice situations.

**Results:** A total of 144 patients had salivary gland resections performed during the study period. Of these, 131 patients had histology and preoperative salivary gland cytology. UG-FNC was performed on these lesions a total of 177 times, with 42 being sampled twice and 4 lesions being sampled three times. Of the 177 UG-FNC performed, 118 were performed by an ENT surgeon and 59 by a pathologist.

Of the total number of salivary gland lesions sampled 42% were localised in glandula parotis, 18% in glandula submandibularis, 2% in glandula sublingualis og 38% from unspecified salivary glands.

The FNC inadequacy rate was 20% for ENT surgeon-performed UG-FNC and 3% for pathologist-performed UG-FNC.

For all lesions, UG-FNC performed by pathologists compared with an ENT-surgeon performed UG-FNC had higher sensitivity (95% vs. 71%) and specificity (96% vs. 79%).

For neoplastic lesions, UG-FNC performed by pathologists compared with an ENT-surgeon performed UG-FNC had higher sensitivity (94% vs. 64%) and specificity (96% vs. 67%).

**Conclusions:** Collaboration between ENT surgeons and pathologists concerning ultrasound-guided FNC of lesions in salivary glands results in fewer non-diagnostic/inadequate samples and improves the sensitivity and specificity of the cytological diagnosis.
5. Thyroid Cytology
Fine needle aspiration diagnosis of follicular neoplasm, Hürthle cell type is associated with higher risk of malignancy

5. Thyroid Cytology
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Objectives: To determine the risk of malignancy in cases diagnosed as follicular neoplasm/suspicious for a follicular neoplasm, Hürthle cell type (FN-H) and identify clinical features that may help in estimating malignancy in patients with FNA diagnosis of FN-H.

Material and methods: We retrospectively reviewed 56 thyroid FNA samples with the diagnosis of FN-H from January 2013 to December 2018 at our institution, which in all the histological follow-up was available. The cases were also evaluated for patient’s age, sex, and size of the nodule and histological diagnosis.

Results: Out of 56 cases, 80.4% were from women and 19.6% from men. Mean age was 48.9 years (24-73). The final histological diagnoses were benign at 39.3% (n=22) of cases and malignant at 60.7% (n=34) of cases. In benign group, 17.8% (n=10) of cases were follicular adenoma, 3.5% (n=2) lymphocytic thyroiditis and 17.9% (n=10) hyperplastic/adenomatoid nodule. The malignant histological diagnoses included 30.4% (n=17) follicular carcinoma and 30.4% (n=17) papillary carcinoma. 78.6% of nodules were neoplastic. The malignant diagnosis among female patients was 44.7% and 90.9% among male patients. The risk of malignancy was significantly higher in male patients (P=0.021). There wasn’t any correlation by means of age (P=0.543) or nodule size (P=0.420) between benign and malignant categories. Size of the nodule was significantly higher at follicular adenoma group when compared with follicular carcinomas (P=0.047).

Conclusions: Because most of the cases diagnosed as FN-H are neoplastic and a significant number are malignant, surgical excision should be performed to exclude malignancy in all thyroid nodules that FNA results are at FN-H category. Furthermore to avoid completion thyroidectomy, total thyroidectomy might also be preferred over lobectomy for FN-H cases, especially in male patients.
Rising trend of goitre in the northern India – Is it due to non usage of fortified salt?

5. Thyroid Cytology

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Objectives: 1. To study the emerging and changing trend of goitre in all thyroid swellings in a tertiary care centre.

2. And incidentally also to evaluate the prevalence of benign and malignant lesions in the thyroid.

Material and methods: Thyroid swellings shows increasing trend in our tertiary care centre inspite of a strict government legislation which allows only iodine fortified salt to be sold, bought and used as per National Iodine Deficiency Disorders Control Programme (NIDDCP) 1998.

In our study

1. 1000 Patients with thyroid swellings who came to the department of pathology G.R Medical college Gwalior over a period of 10 years (from Jan 2009 to December 2018) were studied.

2. The study included age, sex, duration, presenting signs & symptoms.

3. Diagnosis of the lesions was made using fine needle aspiration cytological smears stained with Romanowsky stains.

Results: Out of 1000 cases 115 (11.5%) were neoplastic (Benign & malignant). 85 (8.5%) cases showed thyroiditis. 45 (4.5%) cases were thyroid cyst. But the majority of 755 (75.5%) cases showed features of colloid goitre.

Male: Female ratio was 1:5.54. Mean age of presentation was 37.3 ± 14.9 years. Majority of patients presented as nodular goitre.

Conclusions: The commonest lesion was non-neoplastic lesions and was common in females. Diffuse nodular goitre was the most common non-neoplastic lesion. Papillary Carcinoma was most common malignant thyroid neoplasm. We concluded that there was a rising number of goitre patients in our tertiary care centre over the 10 years period despite the legislation of government to implement NIDDCP.

Although there is a government ruling to supply only iodized salt but the rise in the incidence of goitre suggest that people may still be using non-iodized salt from other sources and this requires further stringent regulation

Selected references

1. Mathew john. Burden of Thyroid Diseases in India. Need for Aggressive Diagnosis medicine. (medicine update; vol. 18).


Subclassification of AUS/FLUS thyroid FNA as per 2017 TBSRTC: An analysis of Risk of Malignancy of different subcategories

5. Thyroid Cytology
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Objectives: The 2017 Bethesda system for reporting thyroid cytopathology (TBSRTC) recommends subclassification of Atypia of Undetermined Significance/ Follicular Lesion of Undetermined Significance (AUS/FLUS), a heterogeneous category into six subcategories (1). Although it has been stated that this will not impact management, the change has been introduced owing to few reports documenting risk of malignancy (ROM) varying depending upon the nature of atypia. Most previous studies have subclassified AUS/FLUS primarily based on nuclear and architectural atypia. ROM and risk of neoplasia (RON) among all the 6 sub-categories remains to be evaluated.

We hypothesized that different subcategories of AUS/FLUS will have different ROM and RON.

Material and methods: All cases of thyroid fine needle aspiration reported as AUS/FLUS over last 5 years (January 2014-June 2018), and with available histology, were reviewed and subclassified into cytologic atypia (AUS-C), architectural atypia (AUS-A), cytologic and architectural atypia (AUS-C&A), Hürthle cell AUS/FLUS (AUS-H), lymphocytic atypia (AUS-L) and Atypia, not otherwise specified (AUS-NOS). ROM and RON were calculated and compared. Data analysis was done using statistical software Stata 14.0.

Results: Of 3101 thyroid aspirates, 299 (9.6%) were AUS/FLUS. Ninety-nine were subsequently operated. Eighteen were recategorized into other Bethesda categories. Remaining 81 cases were evaluated. AUS-C&A was the most frequent (63%; 51/81); others included: AUS-H (12.3%; 10/81), AUS-A (11.1%; 9/81), AUS-NOS (7.4%; 6/81), AUS-C (5%; 4/81) and AUS-L (1.2%; 1/81). RON (P=0.748) and ROM considering Non-invasive follicular thyroid neoplasm with papillary like nuclear features (NIFTP) as malignant (p=0.245) and non-malignant (p=0.494) did not vary significantly among the different sub-categories.

Conclusions: The different sub-categories of AUS/FLUS do not differ significantly in terms of associated ROM and RON. Despite being a considerably long-duration study, the number of cases in most of the sub-categories was small, which may have created a bias. A larger multi-institutional study is required to validate this finding.

Selected references
5. Thyroid Cytology

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Objectives: The aim of this study was to compare the results of ultrasound-guided thyroid fine-needle cytology (UG-FNC) performed through collaboration between pathologists and endocrine surgeons at a university hospital thyroid outpatient clinic.

Material and methods: This study was a retrospective review of consecutive patients in a 4-year period (October 2014 – October 2018) who underwent UG-FNC of the thyroid gland. The UG-FNC was performed by either (i) a surgeon alone with no rapid on-site evaluation (ROSE) of sample adequacy, (ii) a surgeon with ROSE by a pathologist, or (iii) a pathologist with ROSE by that pathologist. Medical records, and cytology and histopathology results were collected and analysed with respect to the three practice situations.

Results: A total of 456 patients had thyroid resections performed during the study period. Of these, 289 patients had histology and preoperative thyroid cytology for a total of 317 thyroid nodules with UG-FNC being performed on these nodules a total of 348 times. Of the 348 UG-FNC performed, 110 were performed by a surgeon without ROSE, 49 by a surgeon with ROSE by a pathologist and 189 were performed by a pathologist.

The FNC inadequacy rate was 13.6% for surgeon-performed UG-FNC without ROSE, 6.1% for surgeons-performed UG-FNC with ROSE, and 2.1% for pathologist-performed UG-FNC.

For neoplastic lesions, UG-FNC performed by pathologists had higher sensitivity (97.6% vs. 66.7%) and specificity (95.2% vs. 85.1%) when compared with UG-FNC performed by surgeons with ROSE. UG-FNC performed by a surgeon without ROSE was had lower sensitivity (59.2%) and lower specificity (75.1%) than both other groups.

For malignant lesions only, UG-FNC performed by pathologists had higher sensitivity and specificity than both other groups.

Conclusions: Collaboration between endocrine surgeons and pathologists with ultrasound-guided FNAC of thyroid nodules results in fewer non-diagnostic samples and improves the accuracy of the cytological diagnosis.
6. Lung Cytology and Mediastinum
Accuracy and Turnaround Time for Tumor Diagnosis of Real Time Cytology during Video-Assisted Thoracoscopic Surgery.

6. Lung Cytology and Mediastinum

Luis De Las Casas

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Objectives: The study aim is to evaluate diagnostic accuracy and turn around time (TAT) of real time cytology (RTC) in the operative room (OR) for video-assisted thoracoscopic surgery (VATS).

Material and methods: A validation study was performed for RTC in the OR for patients undergoing VATS between August 2018 and February 2019.

Diff-Quik stained imprints or scrape smears were used for RTC. A single cytopathologist performed RTC in the OR and gave immediate interpretation to the surgeon. After cytologic preparations were obtained the specimen was sent to the Lab for frozen section (FS).

Diagnosis and TAT of RTC were analyzed using final histologic diagnosis as gold standard.

Results: 50 specimens from 27 patients were reviewed; 15 (56%) males, 12 (44%) females.

Age ranged from 37 to 87 years (median: 65 years). Specimens included 21 (42%) lungs, 15 (30%) mediastinal/hilar lymph nodes, 11 (22%) chest wall/pleura, 2 (4%) mediastinum, and 1 (2%) diaphragm.

Final histologic diagnoses include adenocarcinomas, 16 (32%); benign lesions, 27 (54%); squamous cell carcinomas, 2 (4%); mesothelioma, 2 (4%); and 1 each (2%), thymoma, large cell neuroendocrine carcinoma, and metastatic hepatocellular carcinoma.

RTC deferred diagnosis for 1 sample 3. RTC correlated with final histologic diagnosis in 48 out of 49 specimens (98% accuracy).

RTC TAT ranged from 2 to 15 min (median: 4 min).

Conclusions: These results suggest that RTC could be used as an alternative to FS for tumor diagnosis, potentially decreasing VATS operative time, considering the College of American Pathologists (CAP) 20-minute TAT limit for at least 90% of FS.

Selected references


Biancosino C, Kruger M, Vollmer E, Walker L: Intraoperative fine needle aspirations - diagnosis and


Idiopathic Pulmonary Hemosiderosis (IPH) on Bronchoalveolar Lavage Cytology Complemented with Cell Block Preparation.

6. Lung Cytology and Mediastinum

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Objectives: Objective: Diffuse alveolar hemorrhage (DAH) is a rare and life threatening condition characterized by hemoptysis, dyspnea, alveolar infiltrates on chest radiograph and various degrees of anemia. DAH can occur as a primary disease of the lungs; idiopathic pulmonary hemosiderosis (IPH) or secondary to cardiovascular or systemic disease. The diagnostic value of BAL smear cytology supplemented with iron special stain on cell block preparation has been demonstrated in two case reports of IPH.

Material and methods: Patient’s and Methods: The first case was a 19 years old female, presented with progressive shortness of breath that started 2 years ago together with occasional cough with blood tinged sputum, fatigue and persistent anemia that was not corrected by blood transfusion. The second case was a 16 years old male who was improperly diagnosed at the age of five to have G6PD deficiency anemia.

Results: Result: Computed tomography scan showed ground glass opacity, nodules, reticululations and cysts. A list of differentials diagnoses was rendered in descending order that include hypersensitivity pneumonitis, NSIP, AIP and others. BAL with cell block preparation showed numerous hemosiderin laden macrophages that confirmed by cell block Pearl’s stain for iron as well as lung biopsy. Idiopathic Pulmonary Hemosiderosis diagnosis was suspected after exclusion of all secondary causes. Diagnosis of DAH was confirmed by BAL demonstrating increased numbers of hemosiderin-laden macrophages (>40% of total number of macrophages) that confirmed by Pearl’s (Prussian blue) iron stain.

Conclusions: Conclusion: The highly variable clinical presentation of PH often leads to a delay in its diagnosis as in our two cases. Due to lack of pathognomonic findings, IPH diagnosis can be conclusively established upon exclusion of all other possible secondary causes of DAH along with BAL cytology rich in hemosiderin laden macrophages complemented by cell block preparation and Prussian blue Iron stain.

Selected references
Case Report
Idiopathic Pulmonary Hemosiderosis in Adults: A Case Report
Case Report in Medicine. Volume 2012, Article ID 267857, 5 pages
Lung cancer typing - contribution of immunocytochemistry to the safety of cytological examination

6. Lung Cytology and Mediastinum

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Objectives: Following remarkable advances in molecular biology techniques, in endoscopic ultrasound biopsy procedures and in targeted therapy previously incurable lung cancer patients comes into the focus of scientific interest. More than ever a reliable morphological typing on small biopsy and cytology is an indispensable requirement. The present study aims to assess the safety of the immunocytological staining procedures compared with corresponding immunohistochemical staining of resected pulmonary tumors.

Material and methods: Between 20011 and 2016 a total of 3419 patients of the LungenClinic Grosshansdorf were investigated including 1960 patients with primary lung cancer, 174 with tumors of other primary sites, and 441 patients with finally benign lesions. 1197 fine-needle aspirations (FNA), 435 scratch biopsies or imprint specimens, 212 effusions, 116 brush biopsies and 10 other cytology specimens were performed in patients of the primary lung cancer group. The immunostaining patterns of TTF-1, Napsin, CK7, p40 (+p63), CK5/6/CK5/14, NCAM/CD56 and Synaptophysin (Syn) were matched with the corresponding immunohistological findings and final diagnosis of the tumors.

Results: In 969 primary adenocarcinomas (ADC), the final diagnosis of tumors was predicted by the TTF-1 and CK7 immunocytological findings in 86.5% and 95.8% of patients respectively. With regards to the CK5/6/CK5/14 and p40 (+p63) expression, a correct diagnosis could be achieved in 90.3% and 52.1% of the 260 patients suffering from primary squamous cell carcinomas (SQC). In 268 primary neuroendocrine carcinomas (NET), TTF-1, NCAM/CD56, and Syn corresponded with 76.9%, 85.0% and 60.3% of patients for cytological analysis.

Conclusions: Our study shows that with exception of the p40 (+p63) antibody reaction, the majority of immunocytological staining leads to results comparable to immunohistochemistry and emphasizes the high predictive value of immunocytological typing of lung carcinomas.
Small cell transformation of lung adenocarcinoma as acquired resistance mechanism to EGFR inhibitors: A case report and literature review

6. Lung Cytology and Mediastinum
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**Objectives:** Patients with non-small cell lung cancer (NSCLC) and epidermal growth factor receptor (EGFR) activating mutation can obtain significant tumor regression with EGFR tyrosine kinase inhibitors (TKI) but nearly all patients eventually acquire resistance, with progression of the disease.

Histologic transformation of EGFR mutant lung adenocarcinoma (ADC) into small-cell lung cancer (SCLC) has been described as one of the major resistant mechanisms to EGFR TKIs.

**Material and methods:** We present a case of EGFR-mutated lung adenocarcinoma with T790M mutation and small cell transformation secondary to EGFR TKIs.

**Results:** A 50 year-old female with stage IV lung adenocarcinoma (T2bN3M1b) activating EGFR mutation (exon 19 deletion) treated with TKIs. After initial response to treatment for over 2,5 years, radiologic studies demonstrated progression. In cytological and histological samples, we observed densely packed sheets of small tumor cells with finely granular chromatin, absent nucleoli and nuclear molding.

The transformation to SCLC was validated with morphology and confirmed by immunohistochemistry with expression of neuroendocrine markers (TTF1+, CD56+, synaptophysin+, Ki67 index: 90%). Small cell histology was not found in pre-treatment samples, which show conventional adenocarcinoma morphology. On a second reevaluation, 8 months later, we obtained new samples with combination of adenocarcinoma and SCLC.

According to the literature, the most common mechanism of resistance is the acquisition of a secondary EGFR (T790M) mutation. In addition, several acquired resistance mechanisms such as amplification of the MET receptor, PIK3CA mutation or small cell transformation has been described.

**Conclusions:** The transformation into small cell carcinoma is one of several acquired resistance mechanisms in lung adenocarcinomas with EGFR mutations after treatment with EGFR TKIs. We present a case with two acquired resistance mechanism: T790M mutation and small cell transformation. Recognition of small cell carcinoma transformation in acquired resistance samples is critical because this probably requires a switch into other chemotherapy regimens.

**Selected references**
What is the weight of an Endobronchial Ultrasound Fine Needle Aspirate and what proportion is used for Rapid Onsite Evaluation in malignant cases?

6. Lung Cytology and Mediastinum

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Objectives: 1) To establish the weight of Endobronchial Ultrasound fine needle aspirates (EBUS FNAs) from targeted sites including node stations and lung masses in malignant disease

2) To determine the proportion of total weight by pass and by site that is used for Rapid Onsite Evaluation (ROSE)

Material and methods: Our practice is to expel all aspirated material into a Petri dish, then triage onto slides for ROSE and into formalin and other media. Thus, we are able to weigh each aspirate and the various media before and after triage.

We cease ROSE once diagnostic material is seen; subsequent passes are wholly processed for cell block.

Before each pass, the Petri dish and two labelled slides are weighed using a calibrated balance with a sensitivity of 0.1mg. After expelling the pass into the Petri dish, the dish is reweighed. A drop of the liquid material is placed onto each weighed slide and the slides reweighed before spreading.

The measurements are recorded on a spreadsheet and the aspirate and ROSE weights calculated, together with the proportion used for ROSE.

Results: Weight of aspirate (44 passes): 105.6mg (mean) 0.3 - 207.9mg (range)

ROSE proportion by pass (17 passes with matched data): 8.3% (mean) 0.9 – 36.6% (range)

ROSE proportion by site: 1.5% (mean) 0.6 – 2.7% (range)

Conclusions: To our knowledge, there has not been an attempt to quantify aspirate amount and proportion used for ROSE in EBUS, although a previous study weighed FNAs taken by palpation¹. There is a perception that material from EBUS used for ROSE (or cytology in general) is excessive and wasteful². Data from this study demonstrates only a small percentage of EBUS material is used for ROSE.

This study is in an early phase and more data will be presented.

Selected references
2. Bracey T. A poor man’s opinion on EUS and EBUS: it’s not all necessarily ROSEy. ACP News Winter 2018: 47-49
7. Cytotechnology
Comparison of ultrafast papanicolaou stain with the standard papanicolaou stain in body fluids and fine needle aspiration specimens

7. Cytotechnology
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Objectives: Most cytology laboratories in all Gulf countries including Oman, use the standard papanicolaou (PAP) method to stain various cytological specimens. The aim of this study was to investigate the possible application of ultrafast PAP (UF-PAP) method in cytology laboratory.

Material and methods: Samples from 46 patients containing 26 body fluids and 20 fine needle aspirations (FNAs) (9 thyroids and 11 breasts) were collected. Two air dried and two wet smears from each sample were prepared and stained by UF-PAP and the standard PAP stains, respectively. Background, nuclear staining, cell morphology, and overall staining were independently reviewed by two cytoscreeners.

Results: In all cases of FNA, UF-PAP stain gave a good score for the background, nuclear staining, cell morphology, and overall staining when compared with the standard PAP method. Although the correct diagnosis was made in all cases of body cavity fluids cases except in one case, UF-PAP stain gave a fewer score in the assessment of body cavity fluid samples.

Conclusions: The findings of this study support the use of UF-PAP method in cytology laboratory with a high emphasis on FNA samples.

Selected references:
The pre-analytical phase of fine needle aspiration cytology in the Netherlands

7. Cytotechnology

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Objectives: In this new era of personalized medicine fine needle aspiration cytologic (FNA) material is becoming an important source for predictive markers. However, for interlaboratory and international reproducibility the pre-analytic phase is of utmost importance.

Material and methods: To get an indication of the variability of the pre-analytic phase for FNA in the Netherlands an inventory was made among delegates of 34 laboratories, representing about half of the laboratories in the Netherlands.

Results: Of the 34 laboratories all but one performed immunohistochemistry (IHC) on cytology material. 22 laboratories performed molecular analyses as well, whereas 10 labs send their material out for molecular investigation.

All labs used giemsa stained smears. In 15 laboratories a thin layer PAP stained method was also used whereas in 6 PAP-stain on smears was performed.

All labs used a technique to make blocks of part of the material. However, the techniques to make blocks were very different. Most used techniques were Cellient (11) and agar (14), but also formaline Cellient and formaline cytoblocks were made and fixation was sometimes done with cytorich red (CRR) or methanol.

In all laboratories by preference the blocks were used to perform immunohistochemistry but in 4 laboratories immunohistochemistry was also performed on unstained smears and in 2 laboratories also on PAP-stained smears.

Molecular analysis in 13 of 22 laboratories was performed on smears only, whereas 10 labs also used cellblocks.

Conclusions: This inventory shows the heterogeneity of the pre-analytic phase for cytology. Although for immunohistochemistry cell-blocks seem to be used universally, this is not the case for molecular analysis. The large diversity in the preparation techniques for the cellblocks and especially the diversity in the fixation involved might be a reason for suboptimal results with these blocks, not only for molecular analysis but also for predictive IHC markers.
8. Breast Cytology
Cytomorphological criteria of columnar cell lesions in breast fine needle aspirates

8. Breast Cytology
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Objectives: To present characteristic cytologic features of fine-needle aspirates from columnar cell lesions in the breast.

Material and methods: Columnar cell lesions are seen as mammographic microcalcifications of powdery or crushed stone type, usually in mammography screening. Most microcalcifications are investigated by stereotactic vacuum biopsies, but some may also be seen by ultrasound examination. Incidental cases have work-up with fine-needle cytology. The presenter has seen a handful of such cases, and the cytomorphological findings will be presented.

Results: The cytomorphology mirror the histological findings and show a wide range of features including monolayer sheets, transition from monolayer to aggregates, palisading strips, part of tubular structures, micropapillary groups and microcalcifications. Nuclear changes are discreet, myoepithelial cells may be present, but are few in number.

Conclusions: The cytomorphology mirror the histological continuum that can be seen. The differential diagnoses include low grade DCIS and proliferative borderline or benign lesions.
Immunocytochemical evaluation of ER receptor, PR receptor, and Her-2 in breast cancer cell blocks and corresponding tissue blocks

8. Breast Cytology
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Objectives: Immunohistochemistry (IHC) is a routinely performed method to demonstrate estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor 2 (HER2) in surgical breast cancer specimens but not on cell block (CB) of fine-needle aspiration (FNA). The aims of this study were to evaluate the expression of ER, PR, and HER2 using immunocytochemistry (ICC) on CB and compare with the corresponding tissue blocks as gold standard as well as to compare with other similar studies.

Material and methods: Forty-eight breast carcinoma CB specimens with their corresponding tissue blocks were identified. ICC on CB for ER, PR, and HER2 was performed and compared with tissue blocks. Sensitivity, specificity, positive predictive value (PPV), and negative predictive value were measured for each receptor. The degree of agreement between CB and tissue blocks was calculated using Cohen's kappa (κ) test.

Results: ER results showed 67.7% sensitivity, 94.1% specificity, 95.5% PPV, and a moderate agreement (κ =0.588). PR results showed 50% sensitivity, 90% specificity, 87.5% PPV, and a fair agreement (κ =0.368). HER2 results showed 58.3% sensitivity, 100% specificity, 100% PPV, and a moderate agreement (κ =0.539).

Conclusions: The results of this study confirm the wide variations that occur between CB ICC and tissue block IHC in the detection of ER, PR, and HER2 in breast cancers. In comparison with other studies, we report a low sensitivity and high specificity rates for ER, PR, and HER2 in FNA CB. Further studies are recommended.

Selected references


Not all you see is what it seems to be: breast nodules that were metastases from ovarian carcinoma.

8. Breast Cytology

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Objectives: Metastatic lesions of the breast from an extramammary origin are rare events¹. On the other hand, ovarian cancer rarely metastasizes to the breast².

Material and methods: A female patient, 80 years old, followed regularly by internal medicine, performed routine mammography, which showed two oval nodules at the superior inner quadrant (SIQ) (the largest one was irregular, with 9x8x6mm and the other regular, with 9x7mm) of the left breast. In the right breast, there was a 10mm nodule at the superior outer quadrant (SOQ). Two bulky left axillary adenopathies, the largest with lobed contour, with 35x33x27mm and the other 27x20mm. Meanwhile, a Positron Tomography with 18 F-FDG was performed and showed "Right adnexal malignant neoplastic lesion of high metabolic degree, with extensive ganglionic, peritoneal, pleuropulmonary and muscular metastasis.". After these findings, we hypothesized: two distinct lesions (ovarian and breast cancers) or related lesions (metastatic breast carcinoma), the last one as the most expected.

Results: Fine needle aspiration (FNA) of the 3 nodules revealed an invasive carcinoma, with aggregates of epithelioid atypical cells, with higher nuclei/cytoplasm ratio. However, the mammography showed well delimited nodules and we hypothesized primary breast carcinoma or metastasis from ovarian, due imagiological findings.

It was performed FNA from axillary lymph nodes, which revealed the same neoplasia which immunohistochemical study revealed positivity for estrogen and progesterone receptors and negativity for Her2. Ki67 30-40%. In addition, strong and diffuse positivity for PAX8, P16, WT1 and P53; negativity for GATA3 and mamoglobin were found.

Simultaneously, breast biopsies were done, which showed infiltrative epithelial neoplasia, composed of the same type of cells previously described, with the same IH profile.

Conclusions: As a final diagnosis, we concluded that the breast nodules were bilateral metastases by Mullerian carcinoma, probably with high-grade serous carcinoma. Usually, breast metastasis are carcinomas (ovary, lung, gastrointestinal and genital tracts), melanomas or sarcomas¹.

Selected references

SWISS CHEESE DISEASE (JUVENILE PAPILLOMATOSIS) FINE NEEDLE ASPIRATION: A Case Report: CYTO-HISTOLOGIC FINDINGS.

8. Breast Cytology
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Objectives: Objective: Juvenile papillomatosis (JP) of the breast is a distinct clinico-pathologic entity that warrants accurate cytologic prediction on fine needle aspiration findings. JP has family history of breast cancer that ranged from 28% to 58% and 5% in patients).

Material and methods: Method and Material: A 17 Y-O female patient presented to Cytology Division, at National Cancer Institute, Cairo University, with defined mobile right breast unlike Fibroadenoma mass along with radiology finding of defined inhomogeneous pattern. Fine needle aspiration was done using 24 gauge needle. Aspiration findings showed partly cystic and partly solid aspirate. The aspiration material was divided into Diff Quick and Papanicolaou stained smears for cytologic evaluation.

Results: Results: Smears showed cellular material featuring papillary clusters, fronds, and sheets and aggregated of hyperplastic ductal epithelial cells intermixed with myoepithelial cells together with focal apocrine changes and focal mild atypical cytomorphic changes. The background showed proteinaceous hemorrhagic colloid looking material entangling strips and loose groups of bland ductal cells together with hemosiderin laden macrophages and cell debris. Cytologic diagnosis was rendered as papillary neoplasm and a list of differential diagnoses was suggested that included JP (likely), duct papilloma and complex Fibroadenoma with degenerative changes. Subsequent excision biopsy showed defined circumscribed but not encapsulated mass. Cut sections showed clusters of microcystic spaces and intervening solid tissue. Histology showed typical JP findings.

Conclusions: Conclusion: FNAC findings of JP are reliable and reproducible for proper patient’s management which should be followed by careful long term follow-up for patient’s family and patients themselves.

Selected references

9. Effusions Cytology
Cell block immunocytochemistry of in Peritoneal effusion cytology and FNA for enables accurate diagnosis for institution of neo-adjuvant chemotherapy

9. Effusions Cytology
Radhika Srinivasan

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Objectives: High Grade Serous carcinoma [HGSC] is the most common type of ovarian cancer. Clinically, it presents as an adnexal mass with or without ascites. We evaluated the role of effusion cytology and Fine needle aspiration cytology (FNAC) in making a diagnosis of HGSC based on cytomorphology and cell block immunocytochemistry (CB-ICC).

Material and methods: Retrospective analysis of all cases of metastatic adenocarcinoma of putative ‘ovarian’ origin in effusion cytology and FNAC, between June 2016-18 was performed. CB-ICC was performed for CK7, CK 20, PAX8, WT1 and p53. Clinical data and serum CA-125 levels was recorded. Histopathology of resected surgical specimen (n=34) following interval debulking surgery was evaluated for the probable site of origin and chemotherapy response scores (CRS).

Results: A total of 61 cases were evaluated comprising 56 peritoneal fluid cytology and 5 FNA samples. CA-125 levels ranged from 113-10,000 IU/ml. Peritoneal cytology showed high cellularity in 54% and tumor cells showed papillaroid clusters in 65%, moderate pleomorphism in 71%, prominent nucleoli in 80%, cytoplasmic vacuoles in 89% and frequent mitosis in 40% cases. CK7, PAX8 and WT1 showed diffuse strong or moderate positivity in all cases. p53 immunohistochemistry showed mutation type positivity in 82% cases and null type in 18% cases, where strong diffuse p16 positivity was observed. Histopathological correlation was obtained in 34 cases who underwent interval debulking surgery. The probable site of origin was fallopian tube in 15, tubo-ovarian in 18 and peritoneum in 1 case respectively. Chemotherapy response score was assessed and score 1, 2 and 3 was seen in 14, 15 and 5 cases respectively.

Conclusions: An accurate diagnosis of high grade serous carcinoma of ‘ovarian’ origin is feasible on effusion cytology and FNA by employing cell block immunocytochemistry and should be mandatory before institution of neoadjuvant chemotherapy.
Characterization and ratio of malignancy of the effusions: descriptive analysis of 4176 specimens

9. Effusions Cytology
Jordi Temprana-Salvador
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Vall Hebron Barcelona University Hospital. Pathology Department.

Objectives: Cytological examination of serous effusions is of great importance because it allows the diagnosis of advanced stage cancer, inflammation or other pathologies. We aim to characterize and classify fluids, calculate the ratio of malignancy and describe the diagnoses of malignancy found.

Material and methods: 4176 samples of pleural, pericardial and ascitic effusions are reviewed, between June 2014 and January 2019.

Results: 4176 specimens: 2236 (53.54%) correspond to pleural fluid; 1679 (40.21%) to peritoneal fluid and 261 (6.25%) to pericardial fluid. Out of the total of effusions, 2236 (53.54%) correspond to inflammatory samples, 797 (19.09%) to malignancy, 663 (15.88%) are negative for malignant cells without other consideration and 376 (9.00%) show marked mesothelial hyperplasia. Only 70 (1.68%) are classified as inconclusive atypia and 34 (0.81%) are nondiagnostic.

Ratio of malignancy: pleural fluids 421 (18.83%); ascites 321 (19.12%); and pericardial 55 (21.07%). Diagnoses of malignancy are: Adenocarcinoma NOS 559 (70.49%), Clear cell adenocarcinoma 2 (0.25%), Papillary adenocarcinoma 1 (0.13%), Carcinoma NOS 104 (13.11%), Merkel cell carcinoma 1 (0.13%), Signet ring cell carcinoma 2 (0.25%), Squamous cell carcinoma 7 (0.88%), Transitional carcinoma 4 (0.5%), Infiltrating Ductal Carcinoma 8 (1.01%), Endometrioid Carcinoma 4 (0.5%), Hepatocarcinoma 1 (0.13%), Pseudomyxoma peritonei 1 (0.13%), Lobular carcinoma 1 (0.13%), Neuroendocrine carcinoma 24 (3.03%), Serous cystadenocarcinoma 12 (1.51%), Germinoma 1 (0.13%), Lymphoma NOS 33 (4.16%), Diffuse large B-cell lymphoma 3 (0.38%), Burkitt lymphoma 3 (0.38%), Follicular lymphoma 2 (0.25%), Melanoma 4 (0.50%), Mesothelioma 10 (1.26%), Multiple myeloma 4 (0.5%) and Atypical Teratoid Rhabdoid Tumor 2 (0.25%).

Conclusions: 72.76% of the metastases correspond to adenocarcinoma. Metastases are 79.3 times more frequent than primary tumors (mesothelioma). The ratio of malignancy in the three types of effusions is similar (between 18-21%), the most common diagnosis being inflammation. Cytology of serous effusions is a useful tool to diagnose and typify malignant effusions.
Metabonomic profiling of malignant peritoneal effusion with 1H-NMR

9. Effusions Cytology

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Objectives: Effusions are one of the most suitable biospecimens for investigations in the field of metabonomic profiling, however especially human malignancies in this setting remain a poorly investigated field¹². Here we present the preliminary metabonomic results of malignant ascitic effusion (mAE) in ovarian cancer (OC), hepatocellular carcinoma (HCC), and non-neoplastic effusions (bAE). Effusions are one of the most suitable biospecimens for investigations in the field of metabonomic profiling, however especially human malignancies in this setting remain a poorly investigated field¹². Here we present the preliminary metabonomic results of malignant ascitic effusion (mAE) in ovarian cancer (OC), hepatocellular carcinoma (HCC), and non-neoplastic effusions (bAE).

Material and methods: So far our study consists of 49 samples, comprising 20 mAE from OC, 9 mAE from HCC and 20 bAE. Storage of the samples has been done according to a protocol described in an our previous study¹, and profiling was carried out with ¹H-NMR spectroscopy. After spectra acquisition principal component analysis was applied to classify the effusions.

Results: The multivariate statistical analysis (PCA and PLS-DA) performed on the ¹H-NMR spectra data showed a clear differentiation for the metabolic profiles between mAE and bAE in OC; this was mainly due to triglycerides, beta-hydroxybutyrate (BHB), acetate, acetoacetate, lactate, pyruvate and glucose. In particular, all the mentioned metabolites increased in mAE, while glucose decreased. Conversely, in mAE from HCC the metabolites responsible for the different metabolic profiles appear to be others than those identified as main discriminators in OC.

Conclusions: Our preliminary data suggest how metabonomic profiling could be efficient in classifying effusions. In particular, ¹H-NMR spectroscopy could be useful in effusions with low cellularity or in cases where morphology and/or other classical ancillary tests failed to achieve a diagnosis. However, due to the low number of samples collected till now, these preliminary results need to be confirmed in the future.

Selected references


10. Pancreatico/biliary and Liver Cytology
Diagnostic yield of EUS-guided tissue acquisition in pancreatico-biliary lesions - 4 year experience from a non-HPB centre in the United Kingdom

10. Pancreatico/biliary and Liver Cytology
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Objectives: Endoscopic ultrasound guided (EUS) tissue acquisition is an extensively used investigation to confirm the histopathological diagnosis in pancreatico-biliary (PB) lesions. The joint advisory group (JAG) suggests an adequacy rate of more than 75% for EUS-fine needle aspiration cytology (FNAC) in PB lesions (1). The aim of this study was to determine the diagnostic performance, adequacy of tissue acquisition and safety of EUS-FNA in PB lesions in non-hepatopancreatico-biliary (HPB) centre in UK.

Material and methods: We carried out a retrospective audit of all patients (identified from endoscopy and pathology database) who underwent EUS-FNA between January 2015 and December 2018. Data collected included patient demographics, cross-sectional imaging, cytopathological diagnosis (Panc 1 to 5 based on European cytopathology classification of pancreaticobiliary terminology), treatment modality, complications and 30-day procedure related mortality. Final diagnoses were confirmed from EUS-FNAC, surgical resection specimen or cross-sectional imaging discussed in a MDT (if histology negative).

Results: A total of 152 patients (mean age 66.9 years, 53.9% females) underwent 161 PB EUS-FNAC procedures. 135 patients had a final diagnosis of cancer. 144 (89.4%) samples were deemed adequate for analysis. Sensitivity, specificity, overall accuracy, positive predictive value (PPV) and negative predictive value (NPV) were 87.7%, 100%, 88.9%, 100% and 46.7% respectively (PANC3 included as false negative in patients with cancer as final diagnosis). These figures improved to 94.2%, 100%, 94.8%, 100% and 66.7% respectively when PANC3 was excluded from false negative category in the cancer group. There were no procedure-related complications or mortality.

Conclusions: This study confirms that EUS-FNA of PB lesions can be highly effective and safely carried out in a non--HPB centre. Overall adequacy of tissue acquisition is in compliance with JAG guidelines. Our diagnostic yield is in keeping with the published literature. NPV is relatively low because EUS-FNA has been carried out to confirm suspected cancer on cross-sectional imaging.

Selected references
1. JAG (Joint advisory group) on gastrointestinal endoscopy guidance-JAG summary guide to quality and safety indicators.

2. Dumonceau JM et al. Indications, results and clinical impact of endoscopic ultrasound (EUS) - guided sampling in gastroenterology: European Society of gastrointestinal endoscopy (ESGE) clinical guideline
Insulinoma-Associated protein 1 (INSM1) as a useful marker for determining grade for Pancreatic Neuroendocrine Tumors.

10. Pancreatico/biliary and Liver Cytology
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Objectives: Pancreatic neuroendocrine tumors (PanNET) are categorized into 3 grades, G1, G2, and G3 based upon their mitotic count and Ki-67 index. Each grade has differing prognostic outcomes and accurate grading on limited material, including evaluating Ki-67 index, continues to be a challenge (1, 2). In our prior study, we demonstrated that insulinoma-associated protein 1 (INSM1) is a robust marker in differentiating PanNET from other solid tumors of the pancreas. In this study, we investigate the utility of INSM1 in grading PanNET in cell blocks (CB) and surgical resections (SR).

Material and methods: A search for PanNET from 2000-2018 in our pathology database identified 48 samples (20 CB and 28 SR) with adequate cellular material. These were further separated based upon grade as high (n= 2 CB and n= 3 SR), intermediate (n= 3 CB and n= 8 SR), and low grades (n= 15 CB and n= 17 SR) based upon their final pathology report and Ki-67 level. Immunohistochemical (IHC) staining for INSM1 (SantaCruz Biotech., C-8, 1:100) was performed as described previously [3] and quantified using an H-score system (score: 0-300).

Results: 100% of PanNET (48/48 total: 20/20 CB, 28/28 SR) demonstrated nuclear INSM1 staining. The average H-scores for INSM1 staining of PanNET, were 175/300 and 248/300 in CB and SR respectively. The H-scores exponentially decreased with increasing tumor grade, with low (G1), intermediate (G2), and high grade (G3) tumors showing average INSM1 H-scores of 185/300 and 286/300, 252/300 and 216/300, 73/300 and 115/300 in both CB and SR respectively (Figure 1).

Conclusions: IHC with INSM1 plays a role in both identifying and potentially grading PanNET with lower expression of INSM1 correlating with higher grades.

Selected references
Figure 1. Staining pattern of INSM1 in PanNET with decreased staining in higher grade PanNET.

<table>
<thead>
<tr>
<th>Grade</th>
<th>Surgical Resection (SR)</th>
<th>Cell Blocks (CB)</th>
</tr>
</thead>
<tbody>
<tr>
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11. Pediatric Cytology
Diagnostic accuracy of fine needle aspiration cytology for salivary glands tumors in paediatric patients.

11. Pediatric Cytology
Immacolata Cozzolino
Marco Montella, Andrea Ronchi, Giorgio Toni, Giovanna Ferrara, Giuseppe Colella, Renato Franco
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Objectives: Salivary gland (SG) diseases are rare in paediatric population and incidence, clinical features, treatment and behaviour of these lesions are not fully known. Salivary gland tumours (SGT) are very uncommon in young patients, as only about 5% of all SGT arise in paediatric age. The role of the preoperative fine-needle cytology (FNC) in case of SG diseases is controversial. The purpose of this study is to investigate the accuracy of cytological diagnosis and potential clinical utility of FNC in paediatric SG diseases.

Material and methods: The criteria of inclusion in this study were: 1) patients younger than 19 yrs; 2) major and minor salivary glands lesions; 3) inflammatory, hamartomatous and neoplastic; 4) salivary gland lesions submitted to FNC. At the time of our study, the files of the Vanvitelli University contained 34 cases of salivary gland lesions correspond to these criteria in the 1999-2018 period. Salivary gland FNC (SG-FNC) was performed by a cytopathologist or a head&neck surgeon; ROSE for cellularity adequacy was assessed when SG-FNC was performed by cytopathologist.

Results: The series include 16 males and 18 females, with a median age of 14 yrs. The targeted anatomical sites were: 15 left parotids; 16 right parotids; 2 left submandibulars; 1 right submandibular. ROSE was performed in 19/34 cases. The FNC samples were adequate and representative in 32/34 cases; inadequate in 2 cases. The definitive cytological diagnosis was performed in 29/34 cases. Cytological diagnosis was inconclusive in 3 cases (1 myoepithelial cell neoplasm, 1 basaloid cell neoplasm, 1 possible schwannoma). In 19 cases, including 1 inadequate case and 2 inconclusive cases, surgical resection was performed, and histological examination confirmed the cytological diagnosis in 18 cases.

Conclusions: Preoperative FNC of SG diseases is a simple, useful and accurate tool in diagnosing benign from malignant lesions and in planning appropriate clinical management in paediatric population.

Selected references
12. Urine Cytology
**CellDetect® improves Bladder cancer diagnosis in comparison to standard cytology and Urovysion-FISH**

12. Urine Cytology
Keren Gueta-Milshtein
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3 Department of Pathology, Bnai Zion Medical Center, Haifa

**Objectives:** Standard urine cytology is the common test intended to detect bladder cancer however suffers from low performance. Urinary markers for bladder cancer are being developed to replace standard cytology but until now none presented high-performance, reproducibility, and cost-effectiveness to become a routine test. CellDetect® is a histochemical staining platform allowing color discrimination alongside morphological examination for differentiation between normal and cancer cells that present high performance throughout all cancer grades and stages. The following study was aimed to compare CellDetect® performance to that of standard cytology and Urovysion-FISH.

**Material and methods:** Voided-urine samples were obtained from subjects under routine surveillance for bladder cancer. Cytocentrifuge smears were prepared for each specimen, separately stained with either Pap stain, CellDetect® or U-FISH test. The smears were blindly reviewed by a pathologist, and the performance of all three techniques was compared to gold standard (biopsy for positive cases and cystoscopy for negative cases).

**Results:** The study included 86 patients (43 negative and 43 positives) confirmed by gold standard. The diagnostic accuracy of each test was computed based on determined readings and showed: CellDetect® correctly identified 89% of the positive cases while standard cytology and U-FISH identified only 47% and 62% of the cases, respectively. The specificity of CellDetect® was 83%, standard cytology 95% and U-FISH 78%. 12 cases were categorized as atypical by standard cytology; within these cases, CellDetect® identified 3 true positive cases and 3 true negative cases, resulting in 100% sensitivity and specificity.

**Conclusions:** CellDetect® outperforms both urine cytology and U-FISH sensitivities in the detection of bladder cancer. By utilizing CellDetect® detection rates of existing bladder cancer increase and performance in ruling out malignancy is good. Moreover, by reducing the number of atypical read-outs, CellDetect® contributes to a more precise choice of treatment for patients. Thus, using CellDetect® will aid in a confirmatory diagnosis of Bladder cancer.
Does “The Paris System” make a difference to rate of atypia in urine cytology?

12. Urine Cytology

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Objectives: The Paris system (TPS) was conceived at the 18th International academy of cytology congress in Paris May 2013 to standardise terminology for reporting urinary cytopathology. TPS diagnostic categories include: 1. Unsatisfactory, 2. Negative for high grade urothelial carcinoma, 3. Atypical urothelial cells, 4. Suspicious for high grade urothelial cells and 5. High grade urothelial cells present (1).

There is wide interobserver variability in reporting atypical urothelial cells. Nationally the rate of atypia in urine cytology varies among institutions and ranges from 2-30%. Atypia at our institution prior to the introduction of TPS was 9.4% with individual pathologist atypia rate ranges from 4-27%.

TPS was introduced to standardise terminology for urine cytology reporting from June 2017 with an objective to reduce the atypia category.

Material and methods: A retrospective SNOMED search of departmental database was done to identify urine samples received between June 2017 to December 2018. The departmental atypia rate as well as the distribution of atypia category amongst reporting cytopathologists was calculated. The results obtained were compared with the initial audit done before the introduction of TPS.

Results: A total of 4874 urine samples were received during this period. 6.2% (300) samples were reported as atypia, compared to previous audit which showed an atypia rate of 9.4% (293). There was a reduction in this rate by 3.2%. Individual pathologist atypia rate range also reduced to 2-22% compared to the pre TPS range of 4-27%.

Conclusions: This audit shows that "The Paris System" for reporting urine cytology can be used objectively to reduce the atypia rate as well as interobserver variability and hence aid in patient management decisions.

Selected references


Two year of experience using the Paris System in atypical urine cytology samples

12. Urine Cytology

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Objectives: The Paris System for Reporting Urinary Cytology (TPS) focuses on the diagnosis of high-grade urothelial carcinoma (HGUC) and provides criteria with which to define the category of atypical urothelial cells (AUC) (1). The aim of the present study is to clarify the risk of malignancy for the cytological data classified as AUC according to the TPS.

Material and methods: The study was conducted over a period of 2 years from January 2016 to December 2018. All the cytological urine specimens examined during this period were retrieved from the database. For diagnostic purposes, voided urine samples from three consecutive days were examined from each patient before giving the final report. All samples were processed using ThinPrep T5000 method (Hologic Inc., Marlborough, MA, USA) and stained with Papanicolaou stain.

Results: A retrospective 2-year analysis of urine cytology was carried out and a total of 233 cases classified as AUC were evaluated. Overall, 699 urine cytology smears of three consecutive days obtained from 233 patients were assessed. The patient's age ranged from 30 to 98 years with a mean age of 73 years and the female-to-male ratio was 1:3. A period of 1 year was allowed between the cytological follow up and histopathological specimens. Correlation with histopathology was available for 69 specimens. Histology revealed HGUC in 5.6% and low grade urothelial carcinoma (LGUC) in 24% of the cases. Negative and positive urine cytologies represented 66.4% and 4%, respectively after cytological follow up (table 1).

Conclusions: In our experience, the risk of detecting a biopsy-proven HGUC following an AUC diagnosis was 5.6% and there was less frequent then described in TPS (ranges from 8.3 to 37.5%). Expert pathologist dedicated and liquid based cytology could be the reason for this discrepancy (2). Moreover in about one quarter of the cases AUC category identifies LGUC.

Selected references
14. Soft Tissue and Bone Cytology
Synovial Sarcoma in the left masseter muscle, a diagnostic challenge.

14. Soft Tissue and Bone Cytology

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Objectives: Synovial sarcoma represents 5 to 15% of all soft tissue sarcomas. It is rarely located on the head and neck region and there are only few reports of its cytological features.

Material and methods: We report the case of a 28-year-old male presenting with a 3.9cm lesion in the left masseteric region, clinically suspected to be an abscess. Fine-needle aspiration biopsy was performed.

Results: The smear was hypercellular, composed by a biphasic cellular population. Clusters of spindle cells, with numerous mitotic figures and focal areas of necroptosis, as well as less numerous, loose clusters of epithelioid cells. Its location near the parotid gland raised the possibilities of a myoepithelial or a epithelial-myoeplithelial carcinoma; however, the cytological features and the negativity for S-100, p63 and CK8/18 supported the hypothesis of a mesenchymal neoplasm, favoring synovial sarcoma. The lesion was removed and the diagnosis of biphasic synovial sarcoma was made based on the histological examination and subsequently confirmed with the SS18-SSX translocation by fluorescence in situ hybridization. After six months of chemo- and radiotherapy the patient has currently no evidence of disease.

Conclusions: This case gave us the opportunity to study the cytological features of synovial sarcoma, in an infrequent location and highlights the importance and the efficacy of cytology as a diagnostic procedure in characterizing even subsets of sarcomas, and helping to choose the most adequate surgical procedure.

Selected references
15. Molecular and Immunocytochemistry
Current practices in immunocytochemistry among European cytopathology laboratories – preliminary results of the EFCS on-line survey

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2 Anatomic Pathology Service, Portuguese Institute of Oncology Francisco Gentil, Lisbon EPE, Portugal

Objectives: An update and comprehensive overview about current practices for immunocytochemistry (ICC) on cytological samples in European laboratories was conducted under the auspices of EFCS.

Material and methods: A survey with 19 questions about ICC including: type of cytological slides used for ICC, positive/negative controls, participation in external quality assessment, platforms and protocols used for ICC, number of ICC per year and organization of ICC in daily work was created using open source web application 1KA. An invitation to the survey participation was conducted via EFCS officers to European cytological laboratories.

Results: A total of 216 laboratories from 24 European countries responded to the survey by the end of January 2019. More than half of laboratories (155/216, 72 %) perform ICC on a single type of cytological preparation such as cell blocks (80/216, 37 %), cytopins (39/216, 18 %), smears (20/216, 9 %) or LBC slides (16/216, 7 %), while 56/216 (26 %) laboratories use a combinations of two, three or four different cytological preparations for ICC, no data about preparations used for ICC were received for 5/216 (2 %) laboratories. Majority of laboratories (185/216, 84 %) perform ICC using one of the automated platforms and 74/216 (34 %) laboratories use optimized and validated ICC protocols. Cytological preparations as positive controls are used in 63/216 (29 %) laboratories, negative controls are regularly used in 39/216 (18 %) laboratories and in external quality assessment (EQA) participate 91/216 (42 %) laboratories. The number of ICC per year ranges between 100 and 5000 in majority of laboratories (112/141, 80 %). In more than half of responding laboratories (86/158, 54 %) Biomedical scientist responsible for immunohistochemical (IHC) laboratory is taking care for ICC on cytology samples.

Conclusions: ICC is extensively performed in European laboratories on differently prepared cytological samples using automated platforms mostly without recommended quality assurance measures.
EGFR, ALK and PDL1 testing for lung carcinoma on cytological samples

15. Molecular and Immunocytochemistry

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Objectives: In this era of personalised medicine with advances in lung cancer treatment optimal use of all diagnostic material for molecular mutational testing is most important. Testing is undertaken in stage 3 or 4 lung cancer patients with a good performance status prior to Multi disciplinary team discussion at our centre.

1. To find out sample adequacy for molecular mutational testing (EGFR mutation, ALK translocation and PDL1 status) in cytologic material.

2. Suitability of Endoscopic -Ultrasound guided Transbronchial needle aspiration (EBNA-TBNA) biopsies for PDL1 testing following a change in practice of sample fixation in formalin at collection.

Material and methods: A retrospective SNOMED search of our departmental database was done to identify all cytology specimens related to lung cancer submitted for molecular testing between June 2017 and February 2018. The cytology samples were further analysed for the type of molecular testing undertaken.

Results: Thirtyfour cytological samples were submitted for molecular testing out of a total cohort of 130 specimens (bronchial, lung, lymphnode and other metastatic sites) in the above stated time period.

20 (62%) - adenocarcinoma, 8 (23%) - squamous cell carcinoma, 5 (14%) - Non-small cell carcinoma (NOS) 5 (15%) 1 (2.9%) - Small cell carcinoma and mixed carcinoma.

Sample Types: EBUS-TBNA: 29 (85%), Pleural fluid: 2 (5.8%), Pericardial fluid - 2 (5.8%) and FNA neck node - 1 (2.9%).

EGFR mutation testing was done on 25 cases. The sample was adequate in 23 (92%). 3 (12%) tested positive.

ALK translocation testing was done on 26 cases. All samples were adequate for testing (100%). 2 (7.6%) tested positive.

PDL1 testing was done on 30 cases. The sample was adequate in 28 (93%). 25 (89%) tested positive.

Conclusions: 1. Cytology samples (EBUS, pleural and pericardial fluid) provide adequate well-preserved material for molecular mutational testing in lung carcinoma.

2. PDL1 testing can be carried out effectively on EBUS-TBNA samples following formalin fixation.
16. Training and Teaching
Implementation of Diagnostic Medicine ABCDE Algorithm in our clinical practice: challenges and advantages.

16. Training and Teaching

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¹ “G. Papanicolaou” General Hospital, Department of Pathology, Division of Cytopathology, Thessaloniki, Greece

Objectives: Diagnostic Medicine ABCDE Algorithm is a generic approach to medical image interpretation. It consists of a preliminary step “Correct” and five consecutive steps A: Available information, B: Body, C: Cancer, D: Diagnosis, E: Exhibit.

Material and methods: Its implementation in our everyday practice revealed certain challenges and advantages.

Results: The “Correctly labelled” preliminary step helped us on several occasions to avoid interpreting a slide with a different number than the request form. The “Correctly prepared” step urged us to resolve the problem we had with the stain in order to achieve an optimal image preparation technique. Step A was the most challenging one. To be able to have all A: AVAILABLE INFORMATION for patients, we’ve sent a written request to the Directors of Clinics asking to provide a full clinical history of patients on the request form. During B: BODY, we make clear where the specimen comes from and we apply adequacy criteria. Step C: CANCER is about morphology and if necessary, the use of ancillary techniques, to answer the following questions: Is it normal? Is it a neoplasm? Is it malignant? What type? Making the diagnosis in our mind is Step D: DIAGNOSIS while writing down the report is Step E: EXHIBIT. Steps B, C, D, E are part of a standard diagnostic approach for Cytopathologists, but the sequence of it proved to be a valuable guideline that can save up time, especially during difficult cases.

We also use the ABCDE algorithm when writing case reports; it’s a logical way to present a patient case and helps easily navigate through information that needs to be shown.

Conclusions: In conclusion, the Diagnostic Medicine ABCDE Algorithm is a generic standard approach which is not limited to specific specimens; it’s a valuable guideline that can lead to faster diagnosis with fewer mistakes in everyday practice.

Selected references


Figure 1. A Systematic Approach to Radiology Image or Pathology Slide Diagnosis.
17. Digital Cytology
17. Digital Cytology

Emilio Madrigal
Raza Hoda, Elena Brachtel

Massachusetts General Hospital

Objectives: Digital images of non-cytology breast specimens have previously been shown to be suitable for manual or automated biomarker quantitative evaluation.1-2 With breast fine needle aspiration biopsy (FNAB) as a reliable diagnostic test for breast lesions,3 the feasibility of digitizing and analyzing their corresponding formalin-fixed paraffin-embedded (FFPE) cell blocks (CBs) has not been studied. We developed automated biomarker analysis workflows with trained machine learning (ML) classifiers and applied it to a cohort of breast FNAB CBs.

Material and methods: The anatomic pathology laboratory information system from our institution was queried for breast FNABs spanning 9 years (2009 – 2017) resulting in 1931 exported reports. Slides of cases with concurrent CBs were retrieved (n=92) and digitized at approximately 40x magnification. The image analysis workflow was developed in QuPath,4 and consisted of stain color vector estimation, cell detection with 83 computed features, interactive training of tumor vs. non-tumor ML classifiers based on various user-defined classes (tumor, stroma, blood, immune cells, and artifact), and setting stain intensity thresholds.

Results: Sixteen FFPE CBs of breast FNAB resulted as “positive for malignancy” including hematoxylin and eosin (H&E), hormone receptor (ER, PR) and HER2 immunohistochemistry were successfully scanned at a pixel width and height of 0.2214 μm. Custom workflows for H&E tumor detection and ER quantification were scripted and applied to the entire cohort; analysis measurements were exported. The H&E workflow identified an average of 15,435 (109 – 57,660) tumor nuclei encompassing a mean area of 33,311,051 μm². The ER analysis identified an average of 9,761 (32 – 42,975) tumor nuclei in an average area of 34,595,107 μm², of which 72.6% were considered positive for ER [Table 1].

Conclusions: Our preliminary data demonstrates a practical approach to generate measurable ancillary marker data. Adaptation of an automated biomarker analysis has the potential to improve cytopathologists’ efficiency and provide objective measurements that may impact therapeutic selection.

Selected references
4. Bankhead P, Loughrey MB, Fernández JA, Dombrowski Y, McArt DG, Dunne PD, McQuaid S,

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Our Experience in Applying Artificial Intelligence and Cloud Computing in Cervical Cancer Screening in Rural China

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Objectives: Internet technology played an important role in reinventing various medical procedures, facilitating quick access to medical services and care in rural China. We applied artificial intelligence and cloud computing for slide analysis with the goal to bridge countrywide lack of pathologist, particularly in rural areas. Our results showed that this approach has the potential to make cervical cancer screening available for every woman in China and compensate for a country-wide lack of adequate personnel.

Material and methods: Samples were collected from 800,000 women from Hubei Province. Slides were prepared, stained with DNA specific stain and Pap stain (both stains on the same slide) for automated analysis. Scanners Screening in different locations across the province. After scanning all collected data were uploaded for cloud computing using secure networks and protocols to ensure patient confidentiality. The slides were also sent to the Landing Central Laboratory for analysis in a “conventional way.”

Results: From 2016 till today we conducted feasibility and validation studies using this approach and processed over 1 million slides. We will present data collected From January 11, 2018, to June 7, 2018, from 82 counties (16 cities) of Hubei Province that participated in a cervical cancer screening organized for rural women. A total of 370,096 women were screened. Among those 16,074 were diagnosed as positive, with a positive rate of 4.34%. Out of that number 5767 required further follow up with biopsies. CIN2 and CIN3 accounted for 19% of the total number of biopsies and invasive cancers accounted for about 1.6% of the total number of biopsies.

Conclusions: We found two major advantages using this approach, we had a higher detection rate compared to some conventional methods, and on measuring accessibility and economic value of cloud computing we found that the cloud allowed accurate and fast analysis even in the absence of local expert pathologists.

Selected references
2. Automated Quantitative Cytology Imaging Analysis System in Cervical Cancer Screening in Shanxi Province, China, Dong Y. and all, 2017 Cancer and Clinical Oncology; Vol. 6, No. 2; DOI:10.5539/cco.v6n2p51
18. Others
Cytomorphology study of Cerebrospinal Fluid in leptomeningeal infiltration using an unconventional processing technique.

18. Others
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Objectives: Cerebrospinal fluid is the mainly specimen used to demonstrate the presence of leptomeningeal infiltrations associated with primary tumors. Therefore, the use of specific methods such as the cytomorphological study with a Suta Chamber supplemented with biochemical studies is essential to carry out an accurate diagnosis that allows the clinician an adequate management of the patient.

Material and methods: Study based on a field design, of a prospective type corresponding to a descriptive research, based on the collection and analysis of twenty three (23) samples of cerebrospinal fluid from patients who presented meningeal infiltrations of tumor processes, between the months January-September 2015.

Results: The age groups with the highest incidence of meningeal pathologies were those ages between 6-11 and 42-47, representing 17.39% of each of the cases studied. It was possible to demonstrate that the incidence of meningeal infiltrations was higher in male patients, representing 52.17% of the samples.

A cytomorphological classification was carried out based on the epithelial lineage, in which a predominance of neoplasms of epithelial lineage was observed (43.4%), followed by primary tumors of the central nervous system (23%).

Conclusions: Non-conventional methods, such as the accelerated gravitational sedimentation technique using the Suta chamber, allowed an impeccable processing of the cerebrospinal fluid during the study, providing a perfectly preserved cytological preparation. The cytological diagnoses in the study of cerebrospinal fluid are strictly compatible, therefore, the cytomorphological criteria must be clear for each pathological entity present in order to be able to make an accurate cytomorphological diagnosis that guides the final diagnosis.

Selected references

15. Arnaldo Tabares O. Infiltración Meningea por Leucemia a células plasmáticas y por Linfoma de Hodgkin. Servicio de Neurocirugía. Hospital Central de las Fuerzas Armadas, Caracas, Venezuela.
18. Universitario Ciencias Neurológicas Pedro BarrièMaz Univ Santiago De Compostela, Meningiomas, 1 De Ene. De 2002
22. Keith L. Moore Elsevier España, 2013 Embriología clínica
The one thousand faces of hepatocellular carcinoma

18. Others
Fátima Ramalhosa¹
João Fraga¹, Graça Fernandes¹
¹ Serviço de Anatomia Patológica dos Hospitais da Universidade de Coimbra – Centro Hospitalar e Universitário de Coimbra

Objectives: Usually, hepatocellular carcinoma (HCC) metastasizes to lung, abdominal lymph nodes and bone in advanced tumour stages¹.

Material and methods: We present middle aged men, with no history of other pathologies, came to the emergency service with dyspnoea, low back pain and asthenia lasting for one week, worsened in the last 2 days. Accordingly to the patient, in the last 3 months, he lost around 10 kg, associated with a lack of appetite. Imaging analysis was done to clarify the respiratory symptoms. X-ray showed homogeneous opacity extending from the right hemidiaphragm, likely to be pleural effusion. Computed Tomography (CT) Scan showed a globular liver, with a subcapsular nodule of 29 mm in the segment V, and adenopathies in both renal arteries and celiac trunk, the largest with 18x22 mm. Moderate peritoneal effusion in the pelvic excavation. Lytic lesion with 13 mm in the left paramedian slope of the vertebral body of L1.

A pleural aspiration was done, simultaneously started with levoﬂoxacin empirically.

Results: We performed cell block which showed discohesive cells, enlarged, increase of nucleus/cytoplasm ratio, nuclei pleomorphism and hyperchromasia. Immunohistochemistry (IH) was performed for different cell markers. At first, calretinin and berEp4 were done, in order to clarify the origin of the lesion. BerEp4 was positive and Calretinin negative, which excluded both mesothelioma and lymphoma. Further studies, showed TTF1, CK7 and CK20 negativity, which excluded pulmonary and gastric carcinoma. Hepatocellular carcinoma diagnosis was confirmed with three markers: HepPar1, arginase and AFP.

Conclusions: Accordingly, with the literature, the pleural effusion related with metastatic carcinoma of unknown primary lesion in masculine gender is caused by pulmonary carcinoma (50%), leukaemia/lymphoma (20%) and gastrointestinal carcinoma (7%)². However, because of our findings in CT scan, we should consider also mesothelioma, lymphoma and hepatocellular carcinoma. Our final diagnosis was pleural infusion due to hepatocellular metastases.

Selected references
Invited Speakers
#FNAFriday: How Cytopathologists Learn, Teach and Share Knowledge on Twitter

Invited Speakers
Eduardo Alcaraz-Mateos¹
Jordi Temprana-Salvador², Tania Labiano³, ldaira J. Expósito-Afonso⁴, Lara Pijuan⁵, Zubair Baloch⁶, Maren Y. Fuller⁷, Valerie A. Fitzhugh⁸, Emilio Madrigal⁹, Vighnesh Walavalkar¹⁰, Xiaoyin “Sara” Jiang¹¹

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Objectives: Twitter is a social media network used by medical professionals. Hashtags are used to organize posts by topic. #FNAFriday is a hashtag created by one of the authors to share educational cytopathology cases. This study aims to review the tweets tagged with #FNAFriday to assess impact/usefulness on the cytopathology community.

Material and methods: The Twitter search engine was used to review all tweets tagged with #FNAFriday (April 2015-February 2019). The following data were collected: author, date, pictures, stains, ancillary techniques, subspeciality, comments, retweets, and likes, among others.

Results: A total of 349 original tweets with 11256 Likes, 7099 Retweets, and 1208 Comments (L/RT/C) were collected from 47 accounts. The most active users were @edusqo with 120 Tweets (2843L/1794RT/311C), @Sara_Jiang with 92 Tweets (3707L/2038RT/472C), and @aakasharmand with 33 Tweets (1119L/763RT/96C). The most represented subspecialties were endocrine (77 Tweets, 321L/1675RT/243C), gastrointestinal (42 Tweets, 1298L/895RT/129C), and head/neck (42 Tweets, 1048L/768RT/106C). The average number of pictures was 3.22/Tweet and the most frequent stain was Papanicolaou (49.86%), followed by Romanowsky-type stains (42.98%), and hematoxylin and eosin (18.91%). Cell block (14.04%), histologic correlation (10.03%), immunohistochemistry (8.60%), molecular tests (2.01%), gross pictures (4.58%) and radiologic pictures (3.4%) were also part of the content. Most cases (269, 63.7%) included the diagnosis and a smaller number (57, 13.5%) presented cases in an unknown/quiz format. A progressive increase in interaction was observed over the years (Figure 1).

Conclusions: The #FNAFriday hashtag has been adopted by the cytopathology community with robust engagement and continued growth in usage.

Images (mostly the classic Papanicolaou and Romanowsky stains) are central to these posts, and correlation with gross findings, radiology, and ancillary studies is included.
The #FNAFriday hashtag has been an effective way for cytologists to share knowledge and helps bring awareness of the fine needle aspiration technique as an effective method for diagnosis.
Poster
1. Gynecological Cytology
A five-year retrospective follow-up study of cervical cytological specimens with LSIL, LSIL-H, ASC-H

1. Gynecological Cytology
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Objectives: The conventional Pap smears display sometimes undeniable features of 'low grade squamous intraepithelial lesion' (LSIL) associated with some cells suspicious for, but not diagnostic of 'high grade squamous intraepithelial lesion' (HSIL), in which the last recommended terminology is LSIL+ASC-H. It is known that LSIL has usually a relatively lower prevalence of high-grade lesion on following biopsy, but 'atypical squamous cells, cannot exclude HSIL' (ASC-H) is associated to an increased risk of a higher-grade lesion on biopsy even if it is not a conclusive interpretation.

Material and methods: In the current study, we assessed cervical biopsies in cases of LSIL, LSIL-H, and ASC-H diagnosed based on Bethesda 2001 standards from January 1, 2014 to December 31, 2018 in our laboratory.

Results: Of 256 cytological smears that were processed during the study period in women aged between 17-73 years, we diagnosed 66.01% as LSIL (median age 33.59 years), 5.47% as LSIL-H (median age 37.64 years), and 28.52% as ASC-H (median age 41.24 years). Information regarding the biopsy follow-up was available for 43%, 48%, and 51% of our cases. Even the percentage of histological HSIL associated with LSIL/ASC-H (41%) was higher than rates of LSIL (12%) and not significantly different from ASC-H (40%), the LSIL/ASC-H was more often associated with a conclusive histological diagnosis of HSIL than ASC-H (28% vs. 9%).

Conclusions: According to our results, the diagnosis LSIL/ASC-H must acquire a special attention in the Bethesda System. Along with the opinion of other cytologists, we recommend that all patients with cytological features of LSIL/ASC-H to be followed-up by the same method as ASC-H. In our experience regarding the communication with the clinicians, it’s better to use the terminology "LSIL-H (cannot exclude a higher grade)" in place of "LSIL+ASC-H".
A sample abstract for ECC 2019

1. Gynecological Cytology

Andreas Andersson

Objectives: Objectives
Material and methods: Material and methods
Results: Results
Conclusions: Conclusions
Selected references
Selected references
Atypical glandular cells in cervical cytology and reflex HPV testing: prediction of high grade cervical lesions

1. Gynecological Cytology
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Objectives: The finding of atypical glandular cells (AGC) in cervical cytology is important in the diagnosis of high-grade cervical lesions, cervical carcinoma and extra-cervical cancer. This study evaluates the diagnostic significance of AGC and reflex human papillomavirus (HPV) testing in the detection of high-grade cervical lesions or worse.

Material and methods: During the period from 2014 to 2017 altogether 161,594 conventional cytology samples were analysed and 432 (0.27%) were diagnosed as AGC. A total of 92 cases of AGC with reflex HPV Hybrid capture 2 test were identified. Histological diagnosis of high-grade cervical lesion or worse was considered as positive outcome. The negative follow-up cytology up to five years or negative histology was considered as negative outcome. The AGC was subdivided in three categories: AGC unspecified (AGC-NOS); AGC, endocervical origin, unspecified (AGC-ES-NOS); AGC, endocervical origin, favour neoplastic (AGC-EC-FN).

Results: 38% of AGC had histology with disease rate of 24%; 18.5% with CIN2+/AIS, 3.3% with cervical cancer and 2.2% with endometriai cancer. In the category of AGC-NOS the disease rate was 26.7% with one case of endometrial carcinoma, 17.7% in AGC-ES-NOS and 63.6% in AGC-ES-FN with one case of endometrial carcinoma. Overall, 31.5% of AGC were HPV positive. The disease rate was 68.9% in HPV positive group and 1.6% in HPV negative group. The sensitivity and specificity of HPV test was 95.2% and 87.3%, respectively, with PPV of 68.9% and NPV of 98.4%. One out of three invasive cervical adenocarcinoma was HPV negative case of mucinous cervical adenocarcinoma of gastric type.

Conclusions: The results demonstrate that combining AGC cytology with reflex HPV test raises detection of high-grade cervical lesions or worse from 24% to 68.9%. The high NPV value of HPV test can help in reducing the number of invasive diagnostic procedures.
BD SurePath™ Direct to Slide Processing Validation: An Liquid-based Cytology Method for Laboratories with Limited Resources

1. Gynecological Cytology
Ryan Callaghan
Ramona Nelson, Dondrea Purnell, Lisa Allen, Didier Morel, Adriann Taylor, Clark Whitehead

Becton Dickinson and Company

Objectives: To expand LBC cytology screening into regions of the world with limited resources, we developed a manual LBC processing method that incorporates the cell enrichment advantages of BD SurePath™, using only a modified slide holder and common tabletop centrifuge. This simplified method lowers the cost to produce a high quality LBC slide. We report here the validation of rare event detection and slide quality features of this Direct to Slide processing method.

Material and methods: Slides were produced by utilizing modified slide holders that hold a slide and standard Settling Chamber. 1ml of density reagent was placed onto each settling chamber. 2mls of a BD SurePath specimen was then layered on top of the density reagent. The apparatus were centrifuged for 2 min at 200xg, decanted, washed with alcohol and manually stained following standard Papanicolaou methods. A rare event pool was constructed by spiking cultured SiHa cells into a pool of normal cervical samples at a concentration of 1/6,025. 384 individual BD SurePath cervical specimens were processed to evaluate slide quality features.

Results: All 48 slides produced from the rare event pool had at least 2, and an average of 5 SiHa cells on each slide. 384/384 individual slides were scored as optimal or acceptable for Stain Quality, Cellular Preservation and Cellular Distribution (95% CI (99%, 100%)). For Total Cellularity, 383/384 slides were scored as having > 5,000 cells per slide (95% CI (98.5%, 100%)).

Conclusions: The data validate that high quality LBC slides can be produced from this manual BD SurePath Direct to Slide method using only modified slide holders and a common tabletop centrifuge. This method retains the SurePath advantages of decreasing inflammatory and red blood cells while enriching the population of diagnostically relevant cells. Laboratories with limited resources may benefit from incorporating this manual LBC method.
Cervical cancer screening and monitoring of treated women in Zajecar district from 1996-2018

1. Gynecological Cytology

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**Objectives:** To show the frequency of cervical cancer, age structure, treatment methods used at Zajecar gynaecology department in the period 1996-2018 and the importance of cervical cancer screening in the early diagnosing of this malignant disease to preserve women’s reproductive health and life.

**Material and methods:** A retrospective research and medical documentation analysis in the period 1996-2018 at Zajecar gynaecology department. Data was analysed statistically and presented in graphs and tables.

**Results:** During the past 23 years, 60,168 cervical cancer screenings with PAP tests were conducted. During that period, 636 of those women were treated of cervical cancer by operation, operation and irradiation, and irradiation only. There were 384 (60,38%) women who were treated of preinvasive and microinvasive cervical cancer, 326 (84,90%) of whom were diagnosed thanks to the screening. Of these 384 women, 238 (37,42%) were operated by conisation as they were younger, while 146 (22,96%) being older, underwent hysterectomy. Invasive cervical cancer was treated with 127 (19,97%) women who were operated and irradiated after the operation. Irradiation only was applied with 125 (19,65%) women who could not be operated on, and 62 (49,6) of them were over 61 and did not undergo cervical cancer screening in the last 5 years.

**Conclusions:** Serbia has a very high cervical cancer incidence rate 29,1 per 100,000 women and Zajecar district is leading with incidence of 62,3. In Zajecar, cervical cancer screening with conventional “PAP smear” is the only option. There is no liquid based cytology or HPV testing. Colposcopy is the answer to the lack of equipment. Reduced number of doctors resulted in reduction of cervical cancer screening which led to diagnosing less preinvasive and microinvasive cervical cancer. And the number of women with invasive cervical cancer is constantly increasing.

**Selected references**

Cytohistologic discrepancies of Papanicolaou test and HPV status: Analysis of 502 cases.

1. Gynecological Cytology

**Ji Hye Kim**

Misung Kim, Hye jeong Choi

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2 University of Ulsan College of Medicine, Ulsan, South Korea

**Objectives:** Infections with human papillomavirus(HPV) in Papanicolaou tests have been reported but the histologic correlation and clinical meaning remains debatable. The present study aimed to evaluate characteristics of woman with a discrepancy between cytology and HPV status.

**Material and methods:** We retrospectively searched our pathology database based on HPV infection for patient exhibiting negative for malignancy or atypical squamous cells of undetermined significance(ASCUS) in their initial Papanicolaou test results between January 2014 and December 2015. Follow-up Papanicolaou tests with concurrent HPV testing results were recruited. The cytological samples were liquid-based Papanicolaou smears, classified according to the 2014 Bethesda system. The HPV status was identified using the liquid bead DNA microarray.

**Results:** Total 502 cases during the study period were recruited. 76 cases had a previous record of LSIL/cervical intraepithelial neoplasia 1(CIN1) (30 of 76 cases) and high-grade squamous intraepithelial lesion(HSIL) or invasive carcinoma (46 of 76 cases). Despite papanicolaou test results, histologically 14 cases were newly confirmed LSIL/CIN1 (7 of 14 cases) and HSIL or invasive carcinoma (7 of 14 cases) showing cytohistological discrepancy. A year later, we followed up on 199 cases and found progression to LSIL and ASC-H in 12 cases and 1 case, respectively. The cytohistological discrepancy was demonstrated again in 12 cases exhibiting negative for malignancy (2 of 12 cases) and ASCUS (10 of 12 cases). They were diagnosed as LSIL/CIN1 (5 of 12 cases) and HSIL or invasive carcinoma (7 of 12 cases) in biopsy.

**Conclusions:** Monitoring women with HPV infection is the best way of selection of women who should be treated and monitored in order to prevent cervical cancer. In case of cytohistological or HPV discrepancies, a careful review of the HPV status and the degree of cytologic atypia should be performed. Longer follow-up will clarify the role of HPV testing in patient care.

**Selected references**

Cytological aspects of adenosquamous cervical carcinoma. Presentation of a case in a 28-year-old patient

1. Gynecological Cytology
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Objectives: To describe the cytological findings in a case of an adenosquamous carcinoma in Pap smear, cervical brushing and liquid-based cytology.

Material and methods: A 28-year-old woman with no history of interest, studied by Pap smear and diagnosed of HSIL with features suspicious for invasion. Afterwards, a second cytologic study with cervical brushing and cervical biopsy were performed, which led to a diagnosis of adenosquamous cell carcinoma.

Results: In all samples, abundant atypical cells highly suspicious for malignancy were present. The first cytology examined was the Pap smear. Although there was no agreement if it was a squamous or glandular lesion, the presence of isolated cells with dense and well defined cytoplasm were decisive to make the diagnosis of HSIL with features suspicious for invasion. Later, endocervical brushing showed abundant groups with endocervical features, with presence of cylindrical cells and groups with feathering and acini formation. Isolated squamous cells were hardly found. Concomitant biopsy showed an adenosquamous cell carcinoma, confirmed by immunohistochemistry. Glandular component was positive for CK7, CEA, estrogen/progesterone receptors, while the squamous component was positive for CK34betaE12 and p63. Proliferative index was 85%. Intense p16 positivity was observed in both components. Liquid based cytology was performed previously to conization. The glandular component was also predominant in the cytology and HPV16 was detected by PCR. Clear keratinization was never found.

Conclusions: Due to its low prevalence, identification of cervical adenosquamous cell carcinoma in cytologic specimens can be challenging. When facing hypercellular malignant appearing samples in which the squamous or glandular nature of the atypical cell is difficult to establish, the possibility of this unusual neoplasm must be kept in mind. Histological examination and adjuvant immunohistochemical techniques allow to confirm the diagnosis and to exclude other possibilities, mainly glandular invasion in the context of high-grade intraepithelial squamous neoplasm or pure squamous or glandular carcinoma.
DEALING WITH GLANDULAR PATHOLOGY IN CERVICOVAGINAL SCREENING. ACCURACY AND UNCOMMON CASES.

1. Gynecological Cytology
Carlos Herrero
Patricia Rivera, Concepción Chinchilla, María Victoria Lorenzo, Raquel Revenga, Ana Luz Martín, Elena Escudero, José Luis Seco, Frank Diego Montoya, Sandra Campillos, Ricardo Castillo, Beatriz Jiménez-Ayala
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Objectives: To highlight the small incidence of glandular lesions in PAP smears, review their cytological characteristics and show some unusual cases to try to improve our diagnostic skills.

Material and methods: Review of 491,850 PAP smears in a six-year period (2013-2018) to find all specimens that were diagnosed as AGC (NOS, endocervical, endometrial, favor neoplastic included), AIS, endocervical or endometrial adenocarcinomas. Histologic or clinical correlation was done when available.

Results: We found 117 cytologies (0.024%) with any type of glandular abnormality. 94 samples were diagnosed as AGC (NOS 16 cases, endocervical 44, endometrial 25, favor neoplastic 9), 3 as AIS and the remaining 20 smears were diagnosed as adenocarcinomas (5 NOS, 8 endocervical and 7 endometrial adenocarcinomas).

While most of the cases had typical cytological features, some showed a complex differential diagnosis. Follow up was performed when available and in most of the cases confirmed the presence of glandular lesions. In fact, almost all the endometrial AGC with follow up turned out to be endometrial adenocarcinomas.

Conclusions: Glandular lesions are not as common in the screening routine practice as squamous lesions are. The lack of practice and the peculiar aspect of glandular lesions make their recognition quite hard. In our experience most cases were well classified as glandular, without confusion with squamous cell atypia. Besides, we have confirmed a good accuracy for the distinction between endocervical and endometrial origin.
Large Cell Neuroendocrine Carcinoma of the Uterine Cervix detected by HPV Primary screening with cytology triage- a case study

1. Gynecological Cytology
Stephen Burrows
1 Manchester University NHS Foundation Trust

Objectives: Large Cell Neuroendocrine Carcinoma of the Cervix (LCNEC) is a rare aggressive tumour, accounting for less than 0.1 % of all cervical cancers. The cytological features are not widely recognized in Liquid Based Cytology (LBC) preparations in view of its rarity. The objective of the case study is to describe the cytological features in an LBC preparation of a LCNEC detected by HPV primary screening with cytology triage.

Material and methods: A routine cervical screening sample was taken on a 32 year old female and this was HPV primary screened on the Roche Cobas 8800 HPV testing platform. This poster describes the review findings of HPV test result, cytological features, and subsequent histological findings.

Results: The Surepath LBC sample was tested for the presence of HPV, with HPV type 18 being detected. In line with the England HPV Primary Screening pilot protocol a LBC slide was produced from the residual material. The Surepath sample contains abundant three-dimensional balls of cells. The clusters are of uniformly large cells, possessing moderate amounts of eosinophilic cytoplasm. Nuclei are monomorphic, round to oval with variable nucleoli present. Some groups display chaotic architecture with pseudo-rosette formation, and some possess palisading nuclei. In view of the unusual cytological features the slide was reported as showing "borderline changes in squamous cells" and the woman was referred for colposcopic assessment. Cervical punch biopsy and subsequent large loop excision of transformation zone (LLETZ) confirmed the presence of a high grade solid tumour with focal areas of glandular differentiation and cribriform architecture. Immunohistochemistry showed positivity for AE1/AE3, CD56, Synaptophysin and p16; these results are consistent with large cell neuroendocrine carcinoma of cervical origin.

Conclusions: Precise cytological interpretation is important for a diagnosis of large cell cervical neuroendocrine carcinoma, as early diagnosis is crucial for improved survival and therapeutic options differ from other common cervical cancer types.

Selected references

Spectrum of neuroendocrine carcinomas of the uterine cervix, including histopathologic features, terminology, immunohistochemical profile, and clinical outcomes in a series of 50 cases from a single institution in India Rekhia et al Ann Diag Pathol 2013 Feb; 17 (1); 1-9

Twelve cases of neuroendocrine carcinomas of the uterine cervix; Cytology, histopathology and discussion of their histogenesis Shuxia Li Acta cytologica 2013; 57; 54-60

Smear cytological features of large cell neuroendocrine carcinoma of the uterine cervix in pregnancy: A case report and review of the literature Busra ersan Erdam et al Diagnostic Cytopathology 2018; 1-5

142
Smear Cytology findings of Large Cell Neuroendocrine Carcinoma of the Uterine Cervix
Naoto Kuroda et al Diagnostic Cytopathology (41), No. 7
Liquid based cytology to conventional cytology as a screening tool to cervical abnormalities. Our experience

1. Gynecological Cytology
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Objectives: Cervical cancer screening programs have significantly decreased the incidence and mortality of this disease worldwide mainly in developed countries. Pap - Test has been considered the most cost effective method of prevention and detection of cervical cancer so far, either using the conventional collection technique or the liquid based method. In the present study, we evaluate and compare retrospectively the efficacy of liquid based cytology with the conventional cytology to the diagnosis of cervical abnormalities in relation to the patients age.

Material and methods: 10,903 Conventional cervicovaginal smears from women from January 2000-2009 (A period) and 14,207 liquid based (ThinPrep) cervical specimens from January 2010 -2018 (B period) were evaluated. The smears were submitted from patients attending the inpatients and outpatients of our hospital and the Bethesda system 2014 for reporting was used.

Results: Conventional Pap smear detected 0.56% (62/10,903) cervical abnormalities classified as 48.38% (30/62) ASCUS, 51.61% (32/62) LSIL and 3.2% (2/62) AGC-NOS. LBC cytology revealed 3.79 % (539/14,207) cervical abnormalities comprising 41.37% (223/539) ASCUS, 53.61% (289/539) LSIL, 3.71% (20/539) HSIL, 1.66% (9/539) AGC-NOS, and 3.34% (18/539) malignancies. 1.29% (7/539) Squamous cell carcinomas and 1.85% (10/539) adenocarcinomas and 0.18% (1/539) metastatic ones were included in the malignant cases. The distribution of these lesions showed wide range of incidence from second to eighth decade.

Conclusions: Liquid based cytology offers greater sensitivity in detecting cervical abnormalities mainly regarding LSIL, HSIL and malignancy when compared with conventional Pap-Test smear cytology regardless of the patient’s age.

Selected references


P16/Ki-67 Immunostaining in the Triage of Young Women with LSIL and borderline Cytology

1. Gynecological Cytology

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Objectives: Management of young women under 30 years of age with low or borderline Pap cytology is very challenging. Possible overtreatment in this age group can lead to complications in future pregnancies. In this prospective study we assessed the performance of dual p16/Ki67 immunostaining in women under 30 years with repetitive low grade and borderline cytology.

Material and methods: Our study group consisted of 64 patients under 30 years who had at least two consecutive low grade or borderline Pap test results. Histologic examination was performed in 54 patients and 10 patients were followed only by cytology, which was normal. Conventional Pap cytology specimens were destained and p16/Ki-67 immunostaining was performed retrospectively. The results were compared with histology results or cytology follow-up in cases with no biopsy.

Results: Among 64 cases in the study there were 24 (37.5%) with CIN2+, 23 (35.9%) with CIN1 diagnosis, 7 (10.9%) with normal histology and 10 (15.6%) with normal cytology follow up. Dual p16/Ki67 staining was positive in 19 (79.2%) of CIN2+ cases, 13 (56.5%) of CIN1 cases and in 3 (42.9%) of normal histology cases, respectively (chi-square = 10.125, p = 0.006). 8 out of 10 patients (80%) with normal cytology follow up had negative p16/Ki67 reaction.

Conclusions: Dual p16/Ki67 staining is a useful additional tool in detection of patients at risk for high grade lesions. Besides, predictive value of negative reaction can be helpful in avoiding overtreatment, especially in very young patients.
The usage of liquid-based Pap test and the expression analysis in the residual media for postmenopausal women

1. Gynecological Cytology

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Objectives: The goal of this study was to analyze the efficiency of liquid-based Pap test CellPrep and the possibility of precision diagnosis precancerous lesions and cancer based on the expression of the 24-gene panel mRNA measurement by quantitative PCR in the residual media for postmenopausal women.

Material and methods: 40 postmenopausal women had subsequent histological examination after cytodiagnosis by Pap test CellPrep. mRNA expression of the 21 genes (Ki-67, STK-15, CCNB1, CCND1, MYC, MYBL2, P16INK4A, PTEN, BIRC5, BCL2, BAG1, TERT, NDRG1, ESR1, PGR, HER2, GRB7, MGB1, MMP11, CTSL2, CD68) and 3 house-keeping genes (GUSB, HPRT1, B2M) was measured by quantitative PCR in the Pap test CellPrep media.

Results: The histological examination was classified into following clinical groups: normal epithelium/benign tissue changes (17 patients), LSIL (3 patients), HSIL (16 patients), squamous cell carcinoma (2 patients), endocervical adenocarcinoma (2 patients). We observed that 3 cases of LSIL and 1 case of HSIL were false positive and the specificity for the Pap test CellPrep was 66.67 % . The sensitivity of the Pap smear was 90%. We discovered that the combined evaluation of the 24 gene expression panel allows, according to the discriminant analysis, to carry out the correct differentiation for all five groups in 95% of cases. The correct classification is mainly influenced by the estimation of the expression of 5 genes.

Conclusions: The introduction of the expression analysis of 24-gene panel by quantitative PCR to the Pap test CellPrep may lead to more accurate detecting for severe cervical lesions.
2. HPV Cervical Screening
CERVICAL CANCER SCREENING WITH HUMAN PAPILLOMAVIRUS TEST. RESULTS IN WOMEN 30 TO 35 YEARS OLD

2. HPV Cervical Screening
Belen Lloveras 1
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Objectives: Cervical cancer screening programs based on Human Papillomavirus (HPV) detection are being established as an alternative to cytology due to a higher sensibility in detecting preneoplastic lesions. However, the starting age is controversial and there are few results in the literature. Our objective was to collect data from women 30 to 35 years old in our health area applying HPV screening and cytology triage.

Material and methods: A HPV-based cervical cancer screening opportunistic programme started on April 2018 in the Hospital del Mar health area. A gradual implementation plan based on age groups is ongoing. Until December 2018, 812 women 30 to 35 years old were included in this protocol based on HPV testing and triage by liquid based cytology. Colposcopy was indicated when HPV16 and/or HPV18 (HPV16/18) were positive and/or cytology diagnosis was LSIL or worse.

Results: Among the 812 women tested, 180 (22.2%) were HPV+ and 105 (58.3%) of those had cytological abnormalities (84 ASCUS/LSIL, 21 ASCH/HSIL). HPV16/18 were detected in 52 women (28.8%). Eight out of thirteen cytologic HSIL (61.5%) were HPV16/18 while 30.9% of ASCUS/LSIL tested positive for HPV16/18. Among the 75 normal cytologies 11 (1.5%) were HPV 16/18 positive. Forty-three cases had biopsy accounting for 12 HSIL (75% HPV 16/18) and 19 LSIL (57.8% HPV 16/18). The positive predictive value for HSIL of a HPV+ test with positive triage cytology was 35%.

Conclusions: The rate of HPV positive women in this population was high (22.2%) and 12 histologic HSIL were diagnosed after triage with cytology. These results show the applicability of HPV based screening in women on the 30 to 35 years age group (VPP=35%). The referral area covered by the hospital has a population with high prevalence of cervical lesions therefore these results might differ in other areas.
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WNL = Within normal limits; LSIL = Low-grade squamous intraepithelial lesion; HSIL = High-grade squamous intraepithelial lesion; SIL = Squamous intraepithelial lesion (non-gradable); INSUF = Insufficient for diagnosis; NILM = Negative for intraepithelial lesion or malignancy; ASC-US = Atypical squamous cells of unknown significance; ASC-H = Atypical squamous cells, cannot rule out high-grade lesion; LSIL-H = LSIL+ASC-H
**E6/E7 HPV mRNA expression in women with abnormal smears referred to a Colposcopy University Department for further management. The Greek Experience.**

2. HPV Cervical Screening  
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**Objectives:** Knowledge of carcinogenesis mechanisms and HPV impact has led to the implementation of biomarkers for evaluation of the risk for high-grade lesions and further personalized management as only a small proportion of “pathological results” yields a potentially severe lesion. The aim of this study is the comparison between HPV-mRNA and HPV-DNA testing for the detection of high-grade lesion in women with abnormal cytology or suspicious clinical image that are referred to our Colposcopy Clinic.

**Material and methods:** 420 referred women 21-65 yrs with cytology ASCUS(55), LGSIL(263), ASC-H(11), HGSIL(86), SCC(3) and Adeno-Ca(2) were subjected to Thin-prep Pap-test sampling and colposcopy. HPV-related biomarkers were analyzed with genotyping DNA-Test (Clart2-Genomica) and HR-HPV-mRNA-test (APTIMA-Hologic). The predictive value of the biomarkers was studied for the CIN2+ detection.

**Results:** From 420 women, 102 were histologically confirmed as CIN2(+). HR-HPV-mRNA-test was positive in 89/102 and HPV DNA-Test in 90/102. In CIN2(-) cases (n=318), HR-HPV-mRNA-test was positive in 62/318 and HPV-DNA-Test in 128/318. HR-HPV-mRNA-test had 87.2% Sensitivity (SV), 80.5% Specificity (SP), 59% Positive Predictive Value (PPV) and 95.1% Negative Predictive Value (NPV) while HPV-DNA-Test had 88.2%, 59.7%, 41.3% and 94% respectively. From 3 SCC, 1 was negative in both HR-HPV-mRNA-test and HPV-DNA-Test and the other 2 had positive HR-HPV-mRNA-test test but only one had positive HPV-DNA-Test. Both AdenoCa cases (1 endocervical endometrioid-type and 1 endometrial serous) were negative for HR-HPV-mRNA-test and HPV-DNA-Test, although endocervical endometrioid-type Ca is HPV-related.

**Conclusions:** In abnormal cytology (mainly low-grade), HR-HPV-mRNA-test (APTIMA) offers similar SV and NPV, but higher SP compared to HPV-DNA-Test in terms of CIN2+ detection, improving PPV. So, it is of great value, combining lifestyle studies, for personalized management, risk stratification and avoidance of unnecessary interventions. It should be noted that some HPV-related cervical cancers were negative for HPV-DNA-Test or HR-HPV-mRNA-test or both in terms of primary screening. In any case, higher study sample is required.

**Selected references**
Status of Cervical Cytology as a National Cancer Screening Test in Korea -Analysis by National Health Insurance Data Base-

2. HPV Cervical Screening

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Objectives: Since 1999, Korea has been conducting cervical cytology as National Cervical Cancer Screening Program. The target age has been expanded from the 40 years older age to work insured 30 years older women in 2008, to all 30 years older women in 2011, and to all 20 years older women in 2016. National Cancer Screening Cervical Cytology (NCSCC) has been in quality control since 2004 in the Korean Society for Cytopathology (KSC). The purpose of this study was to investigate the incidence changes of cervical cancer/precancerous lesions according to the expansion of subject and the effectiveness of cervical cytology as a mass screening method.

Material and methods: With National Health Insurance DB, all those who had NCSCC from 2005 to 2016 were selected. The incidence of cervical cancer/precancerous lesion were statistically processed. We collect data from people who presented for obstetrics & gynecology (OBGY) in the current or next year after NSCSS. We compared the diagnosis of OBGY with NCSCC result.

Results: The incidence of SCC and HSIL is steadily decreasing, while LSIL have increased in 2008, 2011 and 2016. The increase of LSIL incidence occurred when the target age was expanded. Annual ASC rate of NCSCC were sustained for less than 5%. The ASC/SIL began to increase from 2012 and ranged to around 5%. The result of NCSCC correlates well with the diagnosis of OBGY. The specificity of NCSCC maintained value of over 0.95. The sensitivity of NCSCC, which was 0.49 in 2005, has continued to improve, reaching 0.78 in 2016.

Conclusions: Even though, the incidence of LSIL increased as the subject age expanded, the incidence of cervical cancer would be reduced by early detection effect. The sensitivity of NCSCC has been increasing steadily and it seems to play the role of mass screening test more effectively under control of KSC.
3. Lymph Node Cytology
Cytologic findings of a peripheral T-cell lymphoma, lymphoepithelial variant (Lennert lymphoma) in fine needle aspiration: A case report

3. Lymph Node Cytology
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Objectives: Background:
Lymphoepithelioid lymphoma is a variant of peripheral T-cell lymphoma with histologic features that highlights proliferation of epithelioid histiocytes admixed with small-sized lymphoid cells. The cytologic features of this variant has been rarely described in the literature.

Material and methods: Results: Case Report:
A 66-year-old male with a history of a pulmonary squamous cell carcinoma presented with weight loss and multiple lymphadenopathy in the cervical and inguinal area. Due to his previous history of lung cancer, metastatic lymphadenopathy was concerned. Fine needle aspiration cytology was performed in the inguinal lymph node, which displayed well-formed aggregations of epithelioid histiocytes on the background of small lymphoid cells. Granulomas themselves had small lymphoid cells embedded in them. These lymphoid cells displayed mild nuclear atypia. Necrosis was not found. Chest and abdominal computed tomography revealed multiple, prominent lymph nodes, measured up to 2.3cm, in both axillae, supraclavicular regions, mediastinum, porta hepatitis, portocaval, paraaortic, aortocaval, iliac and superficial inguinal chains. Excisional biopsy was performed in the cervical lymph node. Histologically, the lymph node was diffusely effaced with clusters of epithelioid histiocytes and surrounding small-sized lymphoid cells. The small lymphoid cells were positive for CD3 and CD5, and Ki-67 proliferative index was 60% in the most active area. Histologic features and immunoprofile confirmed the diagnosis of a peripheral T-cell lymphoma, lymphoepithelial variant (Lennert lymphoma).

Conclusions: Conclusion:
Cytologic features of a peripheral T-cell lymphoma, lymphoepithelial variant (Lennert lymphoma) is distinctive from other lymphomas. However, the distinctive pattern which displays aggregates of epithelioid histiocytes can be misleading as benign granulomatous lesions. The possibility of a peripheral T-cell lymphoma, lymphoepithelial variant (Lennert lymphoma) should always be considered when examining a specimen with abundant epithelioid histiocytes.
FISH-study in the diagnosis of non-Hodgkin's lymphomas on cytological material

3. Lymph Node Cytology
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**Objectives:** Determine the possibilities of using FISH research for the diagnosis of non-Hodgkin's lymphomas on cytological material.

**Material and methods:** A total of 513 patients with non-Hodgkin's lymphomas were examined. 82 patients with non-Hodgkin's lymphomas were given a FISH study. Probes Bcl2 (t (14; 18)), MALT1 (t (11; 18)), CCND1 (t (11; 14)), c-MYC (t (8; 14)), ALK (t (2; 8)), Bcl6 (t (3; 14)).

**Results:** In 82 (16%) patients with non-Hodgkin's lymphomas, a FISH study was required to determine the morphological variant. Cytological material was used in 21 (4%) patients with non-Hodgkin lymphomas. Small lymphocytis lymphoma was established in 1 patient, lymphoma from cells of the marginal zone in 4 patients, follicular lymphoma in 28 patients, lymphoma from mantle cells in 5 patients, lymphoplasmacytic lymphoma - 1 patient, B-cell large cell lymphoma - 23 patients, Burkitt's lymphoma - 19 patients, T anaplastic large cell lymphoma - 1 patient.

**Conclusions:** Cytological material is suitable for conducting FISH studies to determine the morphological variant of non-Hodgkin's lymphomas.
4. Head and Neck Cytology (including Salivary Gland)
"The Milan System for the cytological diagnosis of the salivary gland. Our experience and correlation with the histological result"

4. Head and Neck Cytology (including Salivary Gland)
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Objectives: The Milan system for the cytological diagnosis of the salivary gland which was published in 2018 has recently been implemented in our centre.

Our objective is to determine the degree of agreement between the cytological diagnosis according to the Milan system and the definitive histological result, as well as establishing the risk of malignancy of each diagnosis.

Material and methods: Ultrasound-guided fine needle aspirations (FNA) which had been carried out by pathologists between May 1st, 2016 and May 31st, 2016 were compiled. The compiled cases were classified as per the cytological diagnosis following the Milan system. Those cases which involved subsequent surgery were set aside and a comparison between the cytological and histological diagnoses of these cases was carried out.

Results: 162 cytological studies using ultrasound-guided FNA were compiled.

The cytological diagnoses were as follows: No diagnoses – 2,5% (Milan 1); Non-neoplasm – 30,3% (Milan 2); AUS – 7,4% (Milan 3); Benign neoplasm – 51,2% (Milan 4A); Suspected malign neoplasm – 3,1% (Milan 5); and Malign – 5,6% (Milan 6).

78 of the 162 studies went on to subsequent surgery where the histological diagnoses were as seen in the table.

Cohen’s kappa coefficient was found to be 0.43 thus showing a moderate alignment between the cytological and histological diagnoses.

Conclusions: Fine needle aspiration (FNA) is a simple and highly efficient technique, which in many cases enables doctors to diagnose with confidence and increases its efficiency when they are performed by pathologists.

The Milan System consists of a uniform international approach for classifying and reporting salivary gland FNA samples.

The alignment between the cytological and histological diagnoses was moderate (Cohen’s kappa coefficient 0.43). The risk of malignancy (ROM) of this study matches to a large degree the data found in the literature where larger differences were found in Milan levels 2 and 5.

Selected references
Mody D. R., Thrall M.J., Krishnamurthy S.; Diagnostic Pathology: Cytopathology; ELSEVIER; 2018.
Song S.J. et al.; *The utility of the Milan System as a risk stratification tool for salivary gland fine needle aspiration cytology specimens*; Cytopathology; 2019

Karuna V. et al.; *Effectuation to Cognize malignancy risk and accuracy of fine needle aspiration cytology in salivary gland using "Milan System for Reporting Salivary Gland Cytopathology": A 2 years retrospective study in academic institution*; Indian J Pathol. Microbiol.; 2019

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Application the Milan system for reporting salivary gland cytology (MSRSGC) and risk stratification in a tertiary center in India over a 2-year period

4. Head and Neck Cytology (including Salivary Gland)

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Objectives: The objective of the present study is to evaluate the diagnostic accuracy of salivary gland FNA and to apply the proposed Milan system for reporting salivary gland lesions. The study also aims to elucidate the clinical applicability and potential benefits of the Milan system

Material and methods: A retrospective audit of 210 FNA specimens of salivary gland lesions reported in the year 2017 and 2018 was performed. A correlation with the follow up histopathology was done wherever it was possible. The aspirates were then categorized according to Milan system as follows: nondiagnostic, non neoplastic, atypical, benign neoplasm, neoplasm of uncertain malignant potential, suspicious for malignancy or positive for malignancy. Furthermore, the risk of malignancy (ROM) was calculated for all diagnostic categories.

Results: A Total of 210 salivary gland aspirates were evaluated : 4.2% were nondiagnostic, 60% indicated non neoplastic lesions, 35.8% indicated neoplastic lesions. Histopathology was available for 32% cases (15.2%). FNA had a sensitivity of 80% and a specificity of 98% with an overall diagnostic accuracy of 92% for differentiating malignant from benign tumors. The overall risk of malignancy (ROM) was 14% for the noneoplastic category, 80% for atypical category, 2% for benign neoplasm category, 30% for the NUMP category and 98% for the positive for the malignancy category.

Conclusions: Salivary gland FNA continues to show high diagnostic accuracy and is thus helpful for guiding management. Milan system for reporting salivary gland lesions is a valuable tool that can help standardize reporting and stratify cases preoperatively. The management is not standardized for the category, Salivary Gland neoplasm of uncertain malignant potential, as clinical information plays an important role in planning surgical procedures at an individual basis. Though using Milan system resulted in better communication between cytopathologist and clinician resulting in better patient care, further such studies are required to define appropriate management and predict ROM appropriately.

Selected references
Cytologic diagnosis of mucosal leishmaniasis in nasal brushing: A case report

4. Head and Neck Cytology (including Salivary Gland)

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Objectives: INTRODUCTION: Leishmaniasis is caused by the protozoan leishmania parasite that spreads through sandfly vectors bite. The three main clinical forms of the disease, in descending order of frequency, are cutaneous, visceral (kala-azar) and mucocutaneous leishmaniasis, with the second considered potentially lethal if untreated. Although multiple epidemiological factors contribute to finally developing the disease, leishmaniasis shows specific geographical distribution with Greece bearing very low disease burden¹. We report the rare incidence of diagnosing leishmaniasis by nasal cytology.

Material and methods: CASE REPORT: PATIENT HISTORY: A 55-year-old Greek woman, urban dweller, admitted to our hospital complaining of breathing difficulty. Patient had a prior history of mucocutaneous leishmaniasis, 2 years ago, properly treated, and systemic lupus erythematosus under corticosteroid treatment. Rhinoscopy revealed a mass in the right nasal cavity and brushing smears were prepared. Imaging and serological tests were, also, conducted.

Results: CASE REPORT: CYTOLOGY: Cytology showed an abundance of histiocytes and macrophages, often with intracytoplasmic inclusions suggestive of Leishman-Donovan bodies, and suppurative granulomatous reaction in the background. Appropriate therapy was implemented and the lesion disappeared.

Conclusions: Diagnosing a rare infection such as leishmaniasis may require multiple methods including molecular testing, histology, cytology and culture². In our case, nasal cytology confirmed the disease recurrence in the fastest and most accurate way, ascertaining its valuable role to everyday clinical practice³.

Selected references
1. https://www.who.int/news-room/fact-sheets/detail/leishmaniasis
HPV positive squamous cell carcinoma in the tonsil in FNA: A case report.

4. Head and Neck Cytology (including Salivary Gland)

Charalabia Psachoulia

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Objectives: Human papillomavirus type 16 was recognized as a risk factor for oropharyngeal squamous cell cancer, were tonsil and base of tongue cancer dominate. HPV positive tonsil squamous cell cancers present increasing frequency noticed in >70% of men and have better clinical outcome than corresponding HPV negative cancers. In the present study, a case of HPV positive squamous cell cancer in FNA of tonsil is presented.

Material and methods: A 53 years old male was referred to our hospital for difficulty in swallowing. During physical examination enlargement of the left tonsil was detected. A fine needle aspiration (FNA) was performed and the conventional and THINPREP specimens taken were Papanicolaou and May-Grümwald stained.

Results: Cytologic examination revealed the presence of numerous small lymphocytes intermingled with abundant, isolated, round or polygonal cells with hyperchromatic and pyknotic nuclei, cytoplasmic keratinisation. In addition ghost cells and necrosis were noticed. Positive P16 immunocytochemistry was found. The morphological picture, the immunoprofile and the patient’s medical history allowed the diagnosis of squamous carcinoma of the tonsil (TSCC) correlated with HPV virus.

Conclusions: We wish to emphasize the role of P16 immunocytochemistry in squamous cell carcinoma of the tonsil in FNA as a helpfull adjunct for detecting the HPV status. P16 tumor expression is considered a good marker of HPV status and is an important prognostic factor in young men with TSCC, more chemo- and radiosensitive.

Selected references


Metastatic Pleomorphic Dermal Sarcoma Diagnosed by Fine Needle Aspiration Cytology

4. Head and Neck Cytology (including Salivary Gland)

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Objectives: Atypical fibroxanthoma (AFX) and pleomorphic dermal sarcoma (PDS) are likely to represent part of the spectrum of the same cutaneous neoplasm and distinction between both entities is important because of the more aggressive behavior of the latter (metastatic rate of 10%). To our knowledge, cytological presentation of a metastatic PDS has been only rarely documented.

Material and methods: We describe the case of an 86 year-old-female presenting with a submandibular swelling and a complex medical history, including a resected AFX two months before. Direct fine needle aspiration cytology was performed with a 25G-needle and unstained slides were prepared for ancillary studies.

Results: Smears were highly cellular on H&E and Diff-Quik slides, and an acute inflammation background was found together with necrosis and atypical spindle cells tadpole-like with variable pleomorphism. Immunohistochemistry was performed on slides to rule out metastatic carcinoma (cytokeratin) and melanoma (S100 and MelanA). Only vimentin and CD68 (focal) were positive. A diagnosis of metastatic AFX/PDS was made and cervical lymphadenectomy was performed, confirming the diagnosis.

Conclusions: Since the clinical presentation (superficial, ulcerating mass, usually on the sun-exposed areas of the head and neck region of the elderly) and morphology (admixture of spindle, histiocytic-like, xanthomatous and multinucleated cells with marked pleomorphism and mitotic rate) are the same, just some features favor the diagnosis of PDS (notorious extension beyond the dermis into subcutis, necrosis and vascular invasion).

In the present case, a diagnosis of AFX was rendered based on lack of necrosis and vascular invasion while subcutaneous infiltration was moderate, being this latter aspect a controversial issue, and making a comment advisable when diagnosing AFX due to its potential aggressive behavior.

Selected references
Pleomorphic adenoma with bizarre myoepithelial cells: A diagnostic challenge on cytologic smear

4. Head and Neck Cytology (including Salivary Gland)

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Objectives: The diagnosis of salivary gland tumor with myoepithelial component on fine needle aspiration (FNA) materials is challenging because myoepithelial cells have a wide cytomorphologic spectrum. The authors describe another phenotype of benign myoepithelial cells in pleomorphic adenoma of parotid gland. Myoepithelial cells were bizarre, severely pleomorphic, causing an erroneous diagnosis of myoepithelial carcinoma arising in the pre-existing pleomorphic adenoma (carcinoma ex pleomorphic adenoma). Such myoepithelial cells were observed in subsequent surgical tumor resection, but compatible with pleomorphic adenoma with degeneration rather than malignant feature.

Material and methods: A 55-year-old female was found to have a mass of right parotid gland. FNA showed hypercellularity, containing loosely cohesive fragments of spindle-shaped myoepithelial cells admixed with some small nests of epithelial cells. Moreover, there were occasional bizarre cells possessing severely pleomorphic nuclei with hyperchromasia and coarse clump chromatin interspersed with sheets of spindle cells (figure). The cytologic diagnosis was ‘suspicious for carcinoma ex pleomorphic adenoma’.

Results: Right total parotidectomy was performed. The pathologic diagnosis was pleomorphic adenoma with complete resection. In addition, the bizarre cells were positive for p63, compatible with myoepithelial cells. They distributed throughout the mass, and showed severely pleomorphic nuclei and coarse clump to smudging chromatin. Such cells also stream from the ductal elements into the occasional hyalinized stroma associated with extravasated red blood cells and hemosiderin pigments, the features of which were suggestive of degenerative change. They were positive for p16 and rarely positive for p53. Ki-67 showed a nuclear proliferation rate of about 1%. On the basis of the combined light microscopic and immunhistochemical findings, no evidence supports malignant change.

Conclusions: It is worth noting that myoepithelial cells can vary greatly in appearance and may show bizarre, severely pleomorphic nuclei in benign tumor. This detailed description of the cytopathology will aid in the recognition of the morphologic spectrum this tumor.

Selected references
Rhabdomyosarcoma of Parotid Gland

4. Head and Neck Cytology (including Salivary Gland)
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Objectives: Rhabdomyosarcomas (RMSs) are a group of soft-tissue malignant tumors which derive from primitive skeletal muscle tissue that mainly affect children and adolescents. RMSs are very rare in adults, where they are usually located in the extremities.

The head and neck region is the most common axial site in children, and these RMSs are divided into three subgroups: orbital, parameningeal, and nonorbital nonparameningeal that including paratid region.

Here we report a RMS case in parotid region which diagnosed with fine needle aspiration (FNA) cytology.

Material and methods: A one-year-old boy admitted to hospital with swelling in parotid area. In ultrasonographic examination; a 3 cm diameter hypoechoic mass was detected in the left parotid gland. FNA was applied with on site cytological evaluation. 4 conventional, 1 liquid based (Thinprep®) slides and a cell block were prepared. PAP and MGG used for staining of slides.

Results: In cytomorphological evaluation, a cellular tumoral lesion was seen which composed of large, polygonal, undifferentiated cells, multinuclear giant cells and with a less proportion rhabdomyoblast-like cells. There was anisonucleosis and cellular pleomorphism. In immunocytochemical study; a strong staining was seen with Desmin in tumor cells, a weak-intermediate staining with Myogenin in immature forms, while there were no staining with Pan CK, CD3, CD20 and S100. Ki 67 proliferation index was 40-45%. With all cytomorphological and immunophenotypic features case was diagnosed as RMS.

Conclusions: Four main subtypes of RMS are recognized in the current WHO classification: embryonal, alveolar, pleomorphic, and spindle cell/sclerosing. The embryonal and alveolar forms occur mainly in children. Because the distinction between its two principal forms, embryonal and alveolar, has significant prognostic implications, and cytomorphology is usually insufficient for this distinction, ancillary diagnostic methods, especially cytogenetic and molecular studies were required. In our case; additional studies have not been performed in our center. Therefore, no distinction could be made.

Selected references
SALIVARY GLAND FINE-NEEDLE ASPIRATION CYTOLOGY (FNAC) 
RECATORIZATION ACCORDING TO MILAN SYSTEM AND HISTOLOGICAL CORRELATION

4. Head and Neck Cytology (including Salivary Gland)

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Rubén Benages Álvarez, Susana Ubalde Rizos, Sara González Moya, Marc Montes Miño, Francesc García Fouz, Jordi Temprana-Salvador, Carme Iglesias Felip, Santiago Ramón y Cajal Agüeras, Carmen Dínarés Fernández, Margarita Alberola Ferranti

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Objectives: The Milan System (MS) is a guide to classify the salivary gland lesions in six categories that are associated with a malignancy risk and therapeutic attitude: non-diagnostic (ND) (I), non-neoplastic (NN) (II), atypia of undetermined significance (AUS) (III), benign neoplasm (BN) (IV A), salivary gland neoplasm of undetermined malignant potential (SUMP) (IVB), suspicious for malignancy (SM) (V) and malignant (M) (VI), that allows the unification of criteria and standardization of cytological reports.

The aim of this study is review salivary gland FNAC between 2015 and 2018 in our hospital and reclassify them according to the MS, then evaluate the sensibility, specificity and malignancy risk and its histological correlation.

Material and methods: We select 150 salivary gland FNAC and their histological specimens. These samples were reclassified according to the MS. Histological correlation was made.

Results: Our results were: ND 8,72%, NN 6,05%, AUS 0,67%, BN 57,05%, SUMP 2,01%, SM 2,01% and M 23,49%. The malignancy risk in our series was: ND 30%, NN 17%, AUS 0% (only one case), BN (IV A) 1,3%, SUMP (IV B) 25%, SM 100% and M 85%. 10 cases were reclassified in ND and 2 cases in SM (excluded this study). The correlation with biopsy was concordant in 94,93% (131/138) and discordant in 5,07% (7/138). The sensibility was 85,02%, specificity 94,85%, positive predictive valor 86,08%, negative predictive valor 94,11%, false negative 9,93% and false positive 4,34%.

Conclusions: The MS provides more precise guidelines that facilitate the reclassification of samples. The ND category has increased in our case. The subcategory IVB allows discriminating better SUMP. Our risk of malignancy was similar to MS. This new system aims to unify cytological diagnosis throughout these categories, improves understanding of reports and allows a better clinical management of patients.

Selected references
**Sclerosing polycystic adenosis of the parotid gland: A rare tumor**

4. Head and Neck Cytology (including Salivary Gland)

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³ Department of Radiology, Marmara University, School of Medicine, Istanbul, Turkey

**Objectives:** Sclerosing polycystic adenosis (SPA) is extremely rare reactive/neoplastic inflammatory lesion of salivary glands, first described by Smith et al in 1996. It occurs most commonly in major salivary glands, usually slow growing and clinically asymptomatic well circumscribed mass. SPA is relatively common in female and reported cases have a wide age distribution.

**Material and methods:** We report a case 63-year-old woman who complained painless swelling on right parotid gland region. The USG guided FNA was performed on 3x2.5 cm solid mass.

**Results:** The cytological smears were hypocellular but there were diverse cells: Apocrine cells, histiocytes, tightly cohesive clusters of ductal cells, large epithelial cells with granular cytoplasm, and vacuolated cells in a proteinous background with lymphocytes. In addition, the cell block consisted of cells that showing tubular and glandulary pattern. It was reported as "atypia of undetermined significance" by Milan System for Reporting Salivary Gland Cytopathology with a differential diagnosis including benign cystic lesions, cystadenoma, ductal cyst, pleomorphic adenoma and Wharthin tumor. The patient underwent superficial parotidectomy and it was signed out as SPA.

**Conclusions:** Since SPA is rare entity and has diverse morphological features, correct and definite diagnosis is very difficult. Therefore it is important to know this lesion.

**Selected references**
Marques RC, Felix A. Invasive Carcinoma arising from sclerosing polycystic adenosis of the salivary gland. Virchows Arch 2014:
DOI 10.1007/s00428-014-1551-4
Secondary salivary gland tumors diagnosed by FNA

4. Head and Neck Cytology (including Salivary Gland)

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\(^1\) Charles University, Faculty of Medicine in Pilsen, Czech Republic
\(^2\) Biopptical laboratory ltd, Pilsen, Czech Republic
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\(^4\) Fimlab Laboratories, Tampere, Finland
\(^5\) Tampere University, Faculty of Medicine and Health Technology, Tampere, Finland

Objectives: Fine-needle aspiration (FNA) is an important diagnostic tool in the evaluation of salivary gland nodules. The incidence of salivary gland malignancies is less than 5 per 100 000. Secondary tumors of salivary glands are even more uncommon and represent only 5% of all malignancies of this localization. The aim of the study was to examine the cytological and clinical features of secondary tumors diagnosed by FNA.

Material and methods: A series of 30 secondary tumors from two university hospital pathology department is presented.

Results: The study population consisted of 30 cases (13 males and 17 females) with mean age 64.7 years (age range 41-96 years). The most common site of the metastasis was parotid gland (21 cases) followed by submandibular gland (9 cases). The primary malignancy was known in 18 cases at the time of FNA diagnosis. The most common primary sites were skin of head and neck area (8 cases) followed by lung (5 cases), tonsil (4 cases), kidney (2 cases), breast (2 cases) and malignant melanoma, gastrointestinal tract and soft tissue 1 case of each. The immunocytochemistry was performed on cell blocks in 18 cases.

Conclusions: FNA is a reliable technique in the diagnosis of salivary gland secondary malignancies. The knowledge of the personal history of malignancy is essential for the quick diagnosis, because appropriate targeted immunocytochemical panel allows the effective use of the cell block material, which is usually of limited amount.
4. Head and Neck Cytology (including Salivary Gland)

Charalabia Psachoulia

Eirini Mpotà, Maria Kardàri, Charalabia Psachoulia, Christina Chondrogianni

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Objectives: Thymomas are rare, indolent, epithelial mediastinal tumors that represent fewer than 1% of all mediastinum’s neoplasms and are characterized by slow growth and capacity to spread locally. Distant metastases of these tumors are rare and the incidence of extrathoracic metastases is estimated to be 3-6%.

Material and methods: A 59 years old female was referred to our hospital for investigation of a pelvic mass. Eleven years ago a diagnosis of thymoma was histologically confirmed by core biopsy of an anterior mediastinum’s mass detected by CT scan. Thymectomy was performed as well as partial pleurectomy because of pleural and pericardial lesions detected during operation. Adjuvant chemotherapy and radiotherapy was followed for four cycles. Afterwards, in during routine CT scan followed, recurrent metastatic pulmonary lesions were seen and lobectomy as well as adjuvant radiotherapy was followed. A year ago, a pelvic mass was found in the CT scan. A FNA of the abdominal mass was performed and smears taken were stain with the Papanicolaou stain.

Results: Cytologic examination showed a biphasic pattern composed of epithelial tumor cells and numerous small lymphocytes. The epithelial tumor cells shows mild nuclear atypia, fine chromatin with inconspicuous nucleoli, slightly nuclear contour, scant to moderate cytoplasm with delicate or dense texture. Positive immunocytochemistry for CD3, CD5, TDT was detected in the lymphoid population as well as cytokeratins cocktail (AE1/AE3) in the epitheloid tumor cells. The morphological picture, the immunoprofile and the patients medical history allowed the diagnosis of metastatic thymoma.

Conclusions: We emphasize that the correct diagnosis of metastatic thymoma can be made in fine needle aspirates when the morphological and immunocytochemical results are taking into account and may contribute to the patient’s therapy.

Selected references

5. Thyroid Cytology
Audit of thyroid cytology reporting categories over a six year period at South Tees NHS Hospitals Trust, United Kingdom

5. Thyroid Cytology

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¹ 1. The James Cook University Hospital, Middlesbrough TS4 3BW.
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Objectives: Thyroid cytology is important in the management of thyroid nodules. The thyroid reporting system most commonly used in the United Kingdom is the British Thyroid Association (BTA)/ Royal College of Pathologists (RCPPath) Thy1-5 system which includes Thy1/Thy1c (Non-diagnostic), Thy2/Thy2c (Non-neoplastic), Thy3 (possible neoplasm; Thy3a- cytological and architectural atypia, Thy3f- follicular neoplasm), Thy4 (suspicous of malignancy) and Thy5 (malignant).

1. To calculate the proportion of reporting categories within the department in comparison to RCPPath data¹
2. To determine the positive predictive value (PPV) of Thy3 to Thy5, where follow up histology was available¹

Material and methods: A retrospective search into the departmental reporting system database (SNOMED) was performed to identify thyroid cytology samples received from January 2013 to December 2018. In addition, follow up histology specimens were also analysed for samples classified from Thy3 to Thy5.

Results: A total of 1363 cytology specimens were received. 172/250 of Thy3 (68.8%) which included 14/36 (39%) of Thy3a and 158/214 (74%) of Thy3f, 26/35 (74.3%) of Thy4 and 17/19 (89.5%) of Thy5 samples had follow up histology available. Table below depicts proportion of Thy1-5 categories within the department in % with RCPPath data in parentheses.

Conclusions:

1. The data shows that our departmental proportion of Thy2 to Thy4 categories are in keeping with RCPPath data. The proportion of Thy1 category is higher which could suggest inter-individual variability in sampling due to skills and experience. This also includes category Thy1c which is lesion dependent. Proportion of Thy5 category was lower compared to RCPPath data. This could suggest under-categorising the malignant cytology specimens.
2. PPV of our Thy3a and Thy4 category is higher which again could indicate under-categorising malignant specimens.

This data should lead on to examining individual rates of inadequate sampling and cytological classification with focused training.

Selected references
1. RCPath Guidance on reporting of thyroid cytology specimens Jan 2016.  
   https://www.rcpath.org

<table>
<thead>
<tr>
<th>RCPath categories</th>
<th>Departmental Data in % (RCPath Dataset)</th>
<th>PPV % (RCPath dataset)</th>
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<tr>
<td>Thy1/1c</td>
<td>27.7 (18 – 22)</td>
<td>N/A</td>
</tr>
<tr>
<td>Thy2/2c</td>
<td>50.0 (42 – 51)</td>
<td>N/A</td>
</tr>
<tr>
<td>Thy3a</td>
<td>2.6 (5 -10)</td>
<td>28 (17)</td>
</tr>
<tr>
<td>Thy3f</td>
<td>15.7 (14 -16)</td>
<td>33.5 (upto 40)</td>
</tr>
<tr>
<td>Thy4</td>
<td>2.6 (2 – 4)</td>
<td>96.4 (upto 68)</td>
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<tr>
<td>Thy5</td>
<td>1.4 (5 -10)</td>
<td>89.5 (upto 100)</td>
</tr>
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</table>
Audit of Thyroid Fine Needle Aspiration Cytology Reporting – Conclusions of a Three-Year Review at a Tertiary Referral Centre

5. Thyroid Cytology
Sona Appukutty¹
Anna Paterson¹, Maria O'Donovan¹, James Chan¹, Adam Duckworth¹, Alison Marker¹
¹ Department of Histopathology, Addenbrooke's Hospital, Cambridge, UK

Objectives: To (i) calculate the proportion of thyroid fine needle aspiration (FNA) cytology cases placed in each Thy category and compare this with UK RCPPath standards, (ii) establish the double-reporting rate for Thy3 cases, (iii) determine the proportion of Thy3-5 cases discussed at a multidisciplinary team (MDT) meeting, and (iv) calculate the malignancy rates for cases in each Thy category that subsequently underwent surgery.

Material and methods: All thyroid FNA samples reported at Addenbrooke’s Hospital, Cambridge between January 2015 and December 2017 were included. The data was discussed with the broader MDT and individual clinical cases reviewed to determine likely causes for deviations from expected standards and to identify areas for practice change.

Results: 630 thyroid FNA samples were received and categorised as shown in Table 1. 90% of Thy3a and 82% of Thy3f samples were double-reported, whilst an MDT discussion was documented in 72% Thy3a, 83% Thy3f, 73% Thy4 and 77% Thy5 cases. The percentage of cases in each category found to be malignant, if resection was undertaken, was in line with RCPPath standards (Table 1).

Conclusions: The Thy2/Thy2C category was used less than would be expected, likely reflecting local clinical practice where cases are often not sampled if they appear benign on clinical and radiological assessment, and have no symptomatic cystic component. The Thy1/Thy1C category was used more than expected, following clinical review of these cases, the provision of more clinical information in the future may allow a subset of these to be classified as Thy2/Thy2C. The Thy3a category was used more, and the Thy3f category slightly less than expected, however the risk of malignancy determined on subsequent histology met RCPPath standards, whilst the nature of the malignancies identified showed that all Thy3f cases, and two thirds of Thy3a cases, had a follicular architecture, suggesting that these categories had been used appropriately.

<table>
<thead>
<tr>
<th>Thy Category</th>
<th>Cases (%)</th>
<th>RCPPath Standard (%)</th>
<th>Malignancy rate if resected (%)</th>
<th>RCPPath Standard (%)</th>
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<tr>
<td>Thy 1/1C</td>
<td>43%</td>
<td>18-22%</td>
<td>9%</td>
<td>0-10%</td>
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<tr>
<td>Thy2/2C</td>
<td>18%</td>
<td>42-51%</td>
<td>5%</td>
<td>0-3%</td>
</tr>
<tr>
<td>Thy3a</td>
<td>22%</td>
<td>5-10%</td>
<td>15%</td>
<td>5-15%</td>
</tr>
<tr>
<td>Thy3f</td>
<td>11%</td>
<td>14-16%</td>
<td>18%</td>
<td>15-30%</td>
</tr>
<tr>
<td>Thy4</td>
<td>2%</td>
<td>2-4%</td>
<td>90%</td>
<td>60-75%</td>
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<tr>
<td>Thy5</td>
<td>4%</td>
<td>5-10%</td>
<td>100%</td>
<td>97-100%</td>
</tr>
<tr>
<td>Not appropriate</td>
<td>1%</td>
<td>-</td>
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Table 1: Percentage of thyroid FNAs categorised into each Thy group and the proportion of resected cases found to be malignant in each Thy group, both compared to the UK RCPPath standard.
Cytohistological Correlation of Thyroid Samples With Intermediate or Malignant Categories of The Bethesda System for Reporting Thyroid Cytopathology

5. Thyroid Cytology

Ana Caeiro¹
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¹ Hospital Professor Doutor Fernando Fonseca
² Instituto de Anatomia Patológica, Faculdade de Medicina da Universidade de Lisboa

Objectives: Correlate thyroid cytology samples diagnosed as Folicular Lesion of Undetermined Significance (FLUS), Folicular Neoplasm (FN), Suspicious for Malignancy (SM) and Malignant (MGN) with histological thyroid samples.

Material and methods: A retrospective study from 2014 to 2018, totalizing 1474 thyroid fine-needle aspiration cytology (FNAC), from which 119 were categorized with intermediate or malignant categories on cytology and had a correspondent surgical specimen. The cytological samples were categorized according to The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC), adapted from Portuguese Society of Cytology.

Results: The distribution of the 119 samples by TBSRTC was: 53,8% FLUS, 15,1% FN, 18,5% SM and 12,6% MGN. The distribution of these categories per histological diagnosis was as in table 1.

From the FLUS cytology samples, 34,4% were identified as nodular hyperplasia and 20,3% as papillary carcinoma in subsequent histology. 33,3% of FN cases were classified as papillary carcinoma and 11,1% as follicular carcinoma. Regarding the SM and MGN categories, 59,2% and 99,9% were recognized as malignant respectively.

Conclusions: The percentage of malignancy recommended by TBSRTC is: FLUS 5-15%, FN 15-30%, SM 60-75% and MGN 97-99%. In this study the malignancy risk is higher in FLUS (23,4%) and FN (50%), and within recommended values in SM (59,2%) and MGN (99,6%).

All FN cytology cases with the diagnosis of papillary carcinoma (on the surgical specimen) were follicular or oncocytic variants. All cases were previous to the WHO 2017 classification of NIFTP.

Since 2016 in our laboratory, thyroid specimens were prepared exclusively with liquid based-cytology (LBC), which could have an influence in our results (differences in cellularity of the specimens, “new” morphological evaluation, etc.)

Recently described entities with low malignant potential (namely NIFTP), could bring a change to the published TBSRTC categories and different observation methods (LBC vs direct smear) may bring difficulties when comparing results between different Hospitals.

Selected references


Table 1 - Distribution of the cytological categories per correspondent histological diagnosis

<table>
<thead>
<tr>
<th>Cytological diagnosis</th>
<th>Histological diagnosis (%)</th>
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<tr>
<td></td>
<td>Nodular Hyperplasia</td>
</tr>
<tr>
<td>FLUS (n=64)</td>
<td>34,4%</td>
</tr>
<tr>
<td>FN (n=18)</td>
<td>5,6%</td>
</tr>
<tr>
<td>SM (n=22)</td>
<td>13,6%</td>
</tr>
<tr>
<td>MGN (n=15)</td>
<td>0,0%</td>
</tr>
</tbody>
</table>

*Includes well-differentiated tumor of uncertain malignant potential.
Diagnosing medullary thyroid carcinoma on liquid-based cytology

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George Kazamias, Emmanouil Mastorakis
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2 Cytopathology Department, "Venizeleio" General Hospital, Heraklion, Crete, Greece

Objectives: Medullary thyroid carcinoma is an infrequent neuroendocrine malignancy derived of parafollicular (C) cells. This report aims to highlight the role of liquid-based cytology and immunocytochemistry for the diagnosis of this entity.

Material and methods: Our hospital archives were searched for FNA-diagnosed cases of medullary thyroid carcinoma, histologically confirmed. All patients underwent ultrasound-guided FNA and the aspirated material was processed by the liquid based cytology (ThinPrep®) method. Whenever necessary, immunocytochemistry was performed from the residual material on ThinPrep® slides. An FNA diagnosis was rendered according to the TBSRTC.

Results: Seventeen patients (9 females and 8 males), aged from 18 to 80 years (mean age 63.2 years), were selected for this report. Of them, fifteen out of seventeen underwent solely a thyroid FNA, one both thyroid and lymph node FNAs, and one only a lymph node FNA. Out of the total thyroid aspirations, 12/16 were reported as positive or suspicious for malignancy (Bethesda Categories Malignant and Suspicious, respectively), 3/16 as atypical (Bethesda Category AUS) and 1/16 as benign (Bethesda Category Benign). Medullary carcinomas were mostly presented as hypercellular samples composed of loose clusters and single cells of plasmacytoid or spindle morphology, granular “salt and pepper” chromatin, and well-preserved cytoplasm. Immunocytochemistry was positive for calcitonin expression in all confirmed cases; mCEA was also positive while thyroglobulin and CK19 were negative whenever applied. Two out of the 3 atypical (AUS) cases and the benign case proved to be microcarcinomas in histology. The lymph node FNAs were both diagnosed as “positive for malignancy, consistent with metastatic medullary thyroid carcinoma” by using cytomorphology and immunocytochemistry. One of them was a familiar form of this cancer in an 18-year-old male.

Conclusions: Liquid-based cytology is an effective modality that underscores the unique cytomorphologic and immunophenotypic criteria of medullary thyroid carcinoma.

Selected references
Diagnosing unsuspected parathyroid lesion in Fine Needle Aspiration (FNA) of thyroid nodule using Sure Path cytology: A case series in Eastern Canada.

5. Thyroid Cytology

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Sara Afraz

Memorial University of Newfoundland, Eastern Health, Laboratory Medicine; St.John’s, Canada

Objectives: Thyroid nodules are commonly evaluated by FNA. Due to the anatomic proximity, parathyroid lesions are getting sampled more often. Parathyroid chief and oxyphil cells mimic thyroid follicular cells and oncocyes/Hurthle cells, respectively. Clinical and radiologic findings are often nonspecific and overlapping. These lead to the possibility of misdiagnosing parathyroid lesions as thyroid follicular neoplasm/ suspicious for follicular neoplasm (Bethesda IV category), resulting in surgical resection (lobectomy). Therefore, correct diagnosis in the FNA material is of utmost importance.

Material and methods: We report six cases of parathyroid lesions diagnosed in the FNA of clinically and radiologically suspected thyroid nodules between 2017 and 2018 in Eastern Health, St.John’s, Canada. We used Sure Path cytology and relevant immunohistochemistry on the unstained cytology slides or cell block.

Table 1. describes these six cases with brief demography, relevant clinical, radiologic findings and follow up surgical pathology and other corroborative findings.

Results: Careful cytomorphologic analysis (small monotonous cells, round nuclei, finely granular chromatin, fragile cytoplasm and lack of colloid) raised the initial suspicion of parathyroid over thyroid origin. Immunohistochemical stains (Chromogranin, Calcitonin, TTF-1, Thyroglobulin, CD4), used on liquid based cytology or cell block, helped to clinch the diagnosis. Follow up investigations (radiology, serum Parathormone, calcium) were correlated, when available.

Conclusions: In this study we demonstrate that FNA can be successfully used to diagnose parathyroid lesions; which prevents unnecessary surgical intervention and reduce mortality. In certain case, the cytology diagnosis initiates the work up for hyperparathyroidism.

Selected references

3. Lieu D. Cytopathologist-performed ultrasound-guided fine-needle aspiration of parathyroid lesions. Diagn Cytopathol 2010; 38: 327-332
Follicular thyroid - type carcinoma arising in struma ovarii - a case report

5. Thyroid Cytology

Iris Fabijanić1

Ivana Šamija-Projić1, Lada Škopljanac-Mačina1, Goran Vujić2, Damir Babić1

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2 Department of Gynecology and Obstetrics, University Hospital Centre Zagreb, School of Medicine, University of Zagreb, Zagreb, Croatia

Objectives: Struma ovarii is a rare ovarian germ cell tumor, usually asymptomatic with an unknown risk of malignant transformation. In addition, there is currently no established diagnostic and therapeutic approach for malignant struma ovarii. Thus, the diagnosis can be very challenging on cytology samples. We present a case of metastatic follicular thyroid-type carcinoma in a patient with a history of estrogen-receptor (ER) positive breast cancer and tamoxifen therapy.

Material and methods: Magnetic resonance imaging revealed a polypoid mass within the endometrial cavity and a multicystic mass with solid components in the left adnexa suggestive of ovarian neoplasm. A total abdominal hysterectomy with bilateral salpingo-oophorectomy, lymphadenectomy, omentectomy and appendectomy were performed. Cytological examination of peritoneal washing revealed monotonous population of atypical cells arranged in follicular pattern. The immunocytochemistry was positive for TTF-1, Thyroglobulin, and negative for EA (BerEP4), EMA, MOC-31, CD117, CK20, WT-1, Calcitonin, PR and ER.

Results: Based on this findings, cytological diagnosis was consistent with metastatic follicular thyroid-type carcinoma. A diagnosis was subsequently confirmed by histopathology - follicular thyroid-type carcinoma arising in struma ovarii. The presence of infiltration by tumor cells into the surrounding extra-ovarian tissue and metastasis supported the diagnosis of malignancy. Postoperative ultrasound of the neck revealed a normal thyroid gland. Furthermore, thyroid scan with TC99 as well as thyroid function tests were normal. At 1-year follow-up, the patient had an unremarkable course with no clinical, radiological or biochemical evidence of disease recurrence.

Conclusions: Struma ovarii is a rare tumor that is difficult to accurately characterize preoperatively and sometimes difficult to distinguish from thyroid cancer arising in struma ovarii histologically. Further investigation and studies with more cases are needed to establish diagnostic and therapeutic approach for this rare entity.

Selected references


5. Thyroid Cytology
Ana Caeiro
Cátia Ribeiro, António Alves
1 Hospital Professor Doutor Fernando Fonseca
2 Instituto de Anatomia Patológica, Faculdade de Medicina da Universidade de Lisboa

Objectives: Determine the distribution of diagnostic categories, applying The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC), adapted from Portuguese Society of Cytology, using direct smears (DSM), liquid based-cytology (LBC) and both simultaneously.

Material and methods: A retrospective study was made, totaling 1474 thyroid fine-needle aspiration cytology (FNAC), collected from 2014 to 2018. The diagnoses were classified according to adapted TBSRTC: Nondiagnostic (ND), Benign (BG), Folicular Lesion of Undetermined Significance (FLUS), Folicular Neoplasm (FN), Suspicious for malignancy (SM) and Malignant (MGN).

From 2014 to 2018 samples were prepared using exclusively smears or LBC (ThinPrep®) and others LBC and DSM simultaneously. Samples were stained with MayGrünwald-Giemsa and Papanicolaou stains.

Rapid on-site evaluation (ROSE) was made in all the samples prepared using DSM.

Results: The distribution of various categories from 1474 evaluated thyroid nodules was: ND 17,9%; BG 58,8%; FLUS 16,7%; FN 2%; SM 2,3% and MGN 2,2%.

The percentage of samples per preparation method was: DSM 27,2%; LBC and DSM 37,6%; LBC 35,2% and the distribution of the diagnostic categories per different methods was as follows in table 1.

Conclusions: The percentage of ND was lower in DSM and BG was higher. The number of FLUS was higher in LBC, and lower in DSM. The diagnosis of FN and MGN decreased with LBC and SM remained stable.

ROSE was applied in every FNAC with DSM, which may have influenced the presented results.

The increase in FLUS in exclusive LBC samples can be a result of an unusual morphology that requires a learning curve to identify colloid and not over diagnose minor nuclear changes. In some cases, additional clinical information would have favored a BG diagnosis. Also the introduction of the “noninvasive follicular thyroid neoplasm with papillary-like nuclear features” diagnosis (WHO), could have influenced this increase.

The decrease in FN and increase in ND could be a result of a decrease in the cellularity, noted in the LBC samples.

Selected references

<table>
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<th>Diagnostic category (adapted TBSRTC)</th>
<th>Cytological Methods (%)</th>
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</thead>
<tbody>
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<td></td>
<td>DSM</td>
</tr>
<tr>
<td>Nondiagnostic (ND)</td>
<td>10,7</td>
</tr>
<tr>
<td>Benign (BG)</td>
<td>65,6</td>
</tr>
<tr>
<td>Folicular Lesion of Undetermined Significance (FLUS)</td>
<td>14,0</td>
</tr>
<tr>
<td>Folicular Neoplasm (FN)</td>
<td>3,2</td>
</tr>
<tr>
<td>Suspicious for malignancy (SM)</td>
<td>2,2</td>
</tr>
<tr>
<td>Malignant (MGN)</td>
<td>3,7</td>
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</table>
Thyroid microcarcinoma – our experience

5. Thyroid Cytology
Ljubica Fustar Preradovic1
Ivan Svagelj1, Drazen Svagelj1,2
1 Department of Pathology and Cytology, General Hospital Vinkovci, Vinkovci, Croatia
2 School of Medicine, Josip Juraj Strossmayer University of Osijek, Croatia

Objectives: Thyroid microcarcinoma is defined as a malignant thyroid tumor measuring 1 cm or less in maximal diameter. Papillary microcarcinoma (PTMC) is the most common form of thyroid microcarcinoma. Ultrasound (US) with ultrasound-guided fine-needle aspiration biopsy (US-FNA) is he preferred modality for diagnose PTMC.

Material and methods: Between January 2013 and December 2017, a total of 164 patients who underwent total thyroidectomy with the diagnosis of papillary thyroid carcinoma were included in this study. Study material was obtained by UG-FNA performed by a clinical cytologist (interventional cytologist) who also performed cytomorphological analysis of the material obtained. Prophylactic central neck dissection was performed routinely in all cases. Patient demographics, tumor properties, lymph node metastasis, preoperative neck ultrasonography findings and surgical outcomes were analyzed. A total of 164 patients were enrolled into the study. Patients were allocated into 3 distinct groups with tumor size >10 mm (group 1, n=83, 50.6%), with tumor size 5 – 10 mm (group 2, n=75, 45.7%) and with tumor size ≤5 mm (group 3, n=6, 3.66%).

Results: Preoperative neck ultrasonography showed clinically suspicious lymph nodes at the central compartment in 38 patients (18.6%). Central neck dissection pathology interpretations revealed lymph node metastasis in 42 patients (25.6%) at group 2 and 2 patients (1.22%) at group 3. Statistical analyses also showed significant relationship between increase in the tumor size and central lymph node metastasis. Multifocal tumors are presented with metastasis in the central neck compartment in 14.02% patients. Tumor capsule invasion, multicentricity and lymphovascular invasion show significant relationship with central lymph node metastasis.

Conclusions: In view of the above, the question arises as to what kind of procedure should be done with the patients? Operation is the first method of treatment. What should be its scope? Should you choose a less aggressive procedure - lobectomy or do total thyroidectomy.

Selected references
Ultrasound guided fine needle aspiration of thyroid gland performed by pathologist.
Improving the technique.

5. Thyroid Cytology
Héctor Enrique Torres Rivas
Luis Manuel Fernández Fernández, María Paz González Gutiérrez, Karen Villar Zarra, Ana Belén Dávila Lemos, Natividad Antoraniz Álvez, Luisa Posada Mesa, Aurora Astudillo González
1 Hospital Universitario Central de Asturias
2 Hospital Universitario del Henares
3 Complejo Asistencial Universitario de León
4 Fundación Hospital de Jove

Objectives: There is a validated methodological current of work in pathology, in which the pathologist resumes his interventional role, and becomes an active agent in getting samples. Pathologists, properly trained, can use ultrasound equipment to perform fine needle aspiration (fna) of the thyroid gland, with similar or greater efficiency than non-pathologists. The intention is not to make an ecographic diagnosis, but to qualify the fna procedure.

Material and methods: All thyroid FNA were quantified since January 2017 to December 2018 at Hospital Universitario Central de Asturias (HUCA). They were cataloged depending on where they were made (Pathology Department or Radiology Department). Pathologist performed ultrasound guided fna plus rapid on site evaluation (ROSE). The Bethesda classification was implemented.

Results: At mentioned period, 33 thyroid fna were performed by Radiologist, and 1253 thyroid fna were performed by pathologist (see table 1).

Conclusions: Almost all the ultrasound guide fna of the thyroid gland were performed by pathologist at pathology department. Data showed the high diagnostic performance that is reached when it is the pathologist who is responsible for performing ultrasound guided fna of thyroid gland. It seems obvious that the diagnostic superiority lies in the fact of combining ultrasound guided fna and ROSE. This work methodology allows to reduce the number of passes required, reduce the number of insufficient samples, decrease diagnostic time, decrease patients waiting lists, and lower the costs. Several scientific paper support the use of ultrasound equipment by trained pathologists. HUCA pathology department and Hospital Universitario del Henares pathology department are pioneers in Spain in this work methodology.

Selected references
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6. Lung Cytology and Mediastinum
A case of atypical carcinoid pulmonary origined assessed by bronchoscopic brushing cytology and biopsy

6. Lung Cytology and Mediastinum

Ji-Eun Kwon1

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2 Department of Pathology Incheon Saint Mary’s hospital, Incheon, South Korea

Objectives: Clinical presentation: A 55-year-old man presented with a 6-month history of intermittent cough. He has been suffered from epigastric soreness due to gastroesophageal reflux disease diagnosed since 4-years, and 3 pack-year of smoking history was presented. On his chest X-ray, a 1.8 cm size hilar haziness in right middle lobe area was identified, and subsequent computerized tomography also was perform. No lymphadenopathy or systemic symptoms were present, and complete blood count, liver and kidney function were within normal limits.

Material and methods: To diagnose the hilar lesion, bronchoscopic brushing cytology and biopsy were performed.

Results: Cytologic findings: The bronchoscopic brushing cytology showed rosette-like clustering tumor cells, which showed small and round nuclei, scant cytoplasm, finely granular chromatin and inconspicuous nuclei characteristically. However, nuclear molding, tumor apoptosis, mitosis, and necrosis were not observed (Fig 1). The biopsy demonstrated monotonous small and round tumor cells arranged around the capillaries mimicking adenocarcinoma of papillary pattern. These cells were positive for CD56, but negative for TTF-1, Napsin-A, p63, and the Ki-67 labelling index was 5% in the immunoshistochemical stain studies (Fig 2), which made the diagnosis possible the atypical carcinoid pulmonary origined.

Conclusions: Discussion: Very rare reports discussing the cytologic features of atypical carcinoid pulmonary origined proven by biopsied method are found in the literature [1-3], and we are eager to share our valuable experience.

Selected references
Fig. 1: Tumor cells showing typical clustering around bronchiolar epithelial cells assessed by bronchoscopic brushing cytology. (b) The clustered tumor cells (upper half) characteristically demonstrate small and round to oval cells with hyperchromatic, monomorphic nuclei that appear in contrast to the rosette-like bronchiolar epithelial cells (lower half) (Papanicolaou, ×100). (c) The specimen-like clustered tumor cells without nuclear molding, anaplasia, nuclei, and vacuoles (Papanicolaou, ×400).

Fig. 2: The bronchoscopic biopsy demonstrating relatively monotonous tumor cells also shown in brushing cytology with neuroendocrine differentiation. (a and b) The tumor cells arranged around the capillaries mimicking adenocarcinoma of papillary pattern (H&E, ×100). (c) The perifocal strong positivity of tumor cells consistently showing neuroendocrine differentiation to their histologic characteristics (H&E). (d) Myoepithelial cells diagnosed based on focally Ki-67 labeling index of the tumor cells (Ki-67).
Comparison of PD-L1 expression in paired CytoLyt-fixed cell blocks and biopsies with Non-Small Cell Lung Cancer.

6. Lung Cytology and Mediastinum

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Hans Brunnström1, Despoina Violidaki1
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Objectives: Diagnosis of lung tumors is based mainly on endoscopic procedures that often yield cytological material as the only diagnostic specimen. Cell blocks (CB) prepared from cytological material allow the performance of diagnostic and predictive markers that are essential in the diagnostics of lung cancer. However, different fixation methods of the cytological material could affect the results of these markers. We investigated the expression of PD-L1 in CytoLyt-fixed CB with Non-Small Cell Lung Cancer (NSCLC) by comparing it with paired biopsies.

Material and methods: 21 cytological specimens with paired biopsies from patients with previously untreated NSCLC were selected. Only cases with positive PD-L1 expression in the biopsies were included. In 14 cases the paired materials were from the same site and in 7 cases primary tumor was compared with concurrent lymph node metastases. CB were prepared using the Cellient® system from cytological material fixed in ThinPrep® CytoLyt. Staining was performed with PD-L1 clone 22C3 from pharmDx on a Ventana Benchmark Ultra platform. Assessment of the staining and the Tumor Proportion Score (TPS) were based on the recommendations for PD-L1 clone 22C3.

Results: In 4 of the 21 cases the PD-L1 expression in CB was negative or lower compared to the paired biopsy, resulting to different TPS. Three of the discrepant cases were observed in the high expression TPS group (≥50%). In two of these cases the compared material was primary lesion and concurrent metastasis. Other factors, like duration of fixation and presence of necrosis did not differ between the discrepant and non-discrepant cases (Table 1).

Conclusions: Lower expression of PD-L1 can be observed in CytoLyt-fixed CB leading to underestimation of TPS and thus potentially affecting the patients’ treatment options. These observations are in keep with previous studies and could, at least partially, be attributed to different fixation methods or heterogeneity between primary lesions and metastases.

Selected references
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Abbreviations: AdenoCa: Adenocarcinoma, BB: bronchial brushing, CB: cell block, EBUS-TBNA: endobronchial ultrasound guided transbronchial needle aspiration, FNA: fine needle aspiration, LN: lymph nodes, N/A: not available, NSCLC NOS: Non-Small Cell Cancer, not otherwise specified, TBB: transbronchial biopsy, TPS: Tumor Propagation Score, SqCC: Squamous cell cancer

$^3$ Matched sites of lesions: primary tumor with primary tumor, metastasis with metastasis

$^*$ PD-L1 expression defined as low (1-49%) and high (50%) in biopsy

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CONJOINED EBUS-NEEDLE ASPIRATION, ROSE, AND MULTIDIMENSIONAL FLOW CYTOMETRY. LOOKING FOR AN ACCURACY DIAGNOSTIC OF NEUROENDOCRINE LUNG TUMORS

6. Lung Cytology and Mediastinum
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² Flow cytometry
³ Pneumology department

Objectives: Endobronquial ultrasound guides transbronquial needle aspiration (EBUS_TBNA) is a very useful tool in lung cancer diagnosis. Rapid On Site Evaluation (ROSE) is a rapid, real time examination of the cytology obtained by EBUS allowing a proper specimen examination. Multidimensional flow cytometry (MFC) has a well established role in diagnostic and follow-up of hematolymphoid neoplasms. This contrasts with the widely accepted lack of utility in non hematolymphoid neoplasm.

Aim of this study is to determinate the diagnosis efficacy of conjoined EBUS,ROSE,immunocytochemistry (ICC) and MFC, to a rapid and accuracy diagnostic of neuroendocrine lung neoplasms.

Material and methods: Retrospective study , in the period 2017-2018, of the EBUS-TBNA plus ROSE suspected of neuroendocrine tumor and study of MFC. For the ROSE we use Diff-quick technique. For ICC we used Roche-Ventana panel with CD56; TTF-1; sinaptofisin and Ki67 in cell block. For MFC we used FACSC cito II (Becton-Dickinson) in needle washing.

Results: 26 neuroendocrine lung tumor were diagnosed through EBUS-ROSE-ICC-MFC. 17 were men, with 65 years of median age and 9 were women with 60 median age. Two cases were typic carcinoid. The most frecuent location was 4 mediastinum lymph node. All cases were CD56, synaptofisin and TTF-1 positive. The level of ki67 was 5% in carcinoid tumors and 80-90% in the rest. The MFC results were, in all cases, CD56, CD326 and CD81 positive with variable CD117. The average of response time was 24-48 hours.

Conclusions: Conjoined EBUS-TBNA, ROSE, ICC and MFC are a powerful and rapid diagnostic tool in the correct diagnosis of neuroendocrine tumor of the lung.

Selected references
Zhongguo Fei Ai Za Zhi. 2018 Nov 20;21(11):833-840. doi: 10.3779/j.issn.1009-3419.2018.11.05. [Value of C-ROSE During EBUS-TBNA to Obtain the Tissue Sample \( \frac{E}{S} \) in the Diagnosis of Lung Cancer].

Utility of Rapid On-Site Cytologic Evaluation during Endobronchial Ultrasound-Guided Transbronchial Needle Aspiration in Malignant and Nonmalignant Disease.

Impact of Rapid On-Site Cytological Evaluation (ROSE) on the Diagnostic Yield of Transbronchial Needle Aspiration During Mediastinal Lymph Node Sampling: Systematic Review and Meta-Analysis.
Cryptococcus neoformans infection in non-immunocompromised patients: A case report

6. Lung Cytology and Mediastinum
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² Department of Cytology, General Hospital of Corfu, Greece.

Objectives: Cryptococcus neoformans infection is an important disease globally. It is extremely rare among people with healthy immune system and quite common in immunocompromised patients. Moreover, it can be systemic involving the lymph nodes and visceral organs, particularly the lungs, as well as the central nervous system (meninges). A case of Cryptococcus neoformans infection diagnosed in bronchial washings of a healthy patient is presented.

Material and methods: A 32-year old non-immunocompromised female was presented to the hospital complaining of fever, cough and breath shortness. No use of immunocompromised drugs was referred to her clinical history. Bronchial washing was performed and the specimen taken was prepared using the ThinPerp technique. The prepared cytological slides were stained with Papanicolaou stain.

Results: The cytological examination of bronchial washings revealed the presence of pale, brownish-pink round yeasts with a thick, sharply demarcated mucoid capsule and a teardrop budding, either phagocytised by macrophages or remaining free in the background. In addition, positivity to Mucicarmine and PAS-D followed strains confirmed the fungal identification. Based on the above findings the cytological diagnosis was consistent with Cryptococcus neoformans infection and was confirmed by the subsequent diagnosis of the lung biopsy.

Conclusions: The role of exfoliative cytology in the diagnosis of Cryptococcus neoformans infection in non-immunocompromised patients is very important and contributes significantly to the patient’s treatment.
Endobronchial Ultrasound-guided Transbronchial Needle Aspiration and role of cytopathologists in diagnosis of diseases with mediastinal involvement.

6. Lung Cytology and Mediastinum

Tania Labiano

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² Pulmonology Department, Complejo Hospitalario de Navarra, Spain
³ Medical Oncology Department, Complejo Hospitalario de Navarra, Spain
⁴ Navarrabiomed, Spain

Objectives: TBNA-EBUS is a minimally invasive and effective procedure used to diagnose lung cancer, non-pulmonary origin metastases and other inflammatory diseases with mediastinal involvement. The aim of this study is to describe our results and highlight cytopathologists role in the decision making process of these diseases with broad differential diagnostic that requires completely different treatment.

Material and methods: From September 2017 to December 2018, a retrospective analysis of consecutive patients (N=188;126M/62F, x=63.7y) who underwent TBNA-EBUS was undertaken. We review clinical and pathological data. A cytopathologist was present for ROSE in 100% of procedures.

Results: A total of 98% of EBUS were valid (184/188). Forty one out of 184 patients (22.3%) were diagnosed with Adenocarcinoma (22 M/19 F). EGFR (RT-PCR), ALK/ROS1 (FISH) and PD-L1 (SP263) assessment was performed in 95% of patients with adenocarcinoma diagnosis. Four patients showed ALK rearrangement (3F/1M, x=53.5 y), one patient showed ROS1 rearrangement (67y-o M) and 4 patients showed EGFR mutations (4F, x=57.4 y 1/4 T790M resistance mutation post TKI treatment). PD-L1 was studied, showing expression of more than 50% in 23.3% of the cases. Twenty four out of 184 patients were diagnosed with SCC (6F/18M, x=75.7 y). Expression of PD-L1 was > 50% in 45.45%. Nineteen patients out of 184 were diagnosed with SCLC/LCNEC (14M/5M, x=64.42 y). Fourteen out of 184 patients presented metastatic disease of non-pulmonary origin (8 Breast Ca, 2 Melanoma, 2 CRC, 1 Endocervical Ad, 1 Unclassifiable tumor) and 2 Lymphomas. Fourteen patient out of 86 negative diagnosis (14/86, 16.3%) were diagnosed with Sarcoidosis (7M/7F, x=55.3 y; one case was infliximab-related).

Conclusions: EBUS-TBNA provides adequate cytology material for diagnosis of neoplasm of pulmonary and non-pulmonary origin and non-neoplastic diseases with mediastinal involvement. Effective feedback between health care professionals involved and cytopathologists may help cytopathologists obtain adequate tissue for diagnosis, ancillary techniques and molecular testing, frequently needed.
EXPRESSION OF Krüppel-Like-Factor 8 IN NSCLC: CORRELATION WITH PROGNOSTIC FACTORS

6. Lung Cytology and Mediastinum

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ANGELOS TSIPIS, MARIA GONIDI, ELEFTERIA HAINI, KYRIAKOS HAINIS,
EFROSSINI SALEMI, PAVLINA ATHANASSIADOU

1 MEDICAL, SCHOOL, NATIONAL AND KAPODISTRIAN UNIVERSITY OF ATHENS, GREECE

Objectives: Krüppel-Like-Factor 8 belongs to the KLF family and has various roles in the regulation of the cell cycle, proliferation and tumor genesis. The aim of the present study was to determinate the association between KLF8 expression with various clinicopathological parameters and with the expression of E-cadherin and Ki-67.

Material and methods: Imprints from 72 resected lung cancers were examined immunocytochemically with the use of KLFS, E-cadherin and Ki-67 markers. The results correlated with stage, lymphnodes status and grade of the tumour.

Results: Overexpression of KLF 8 protein was found in NSCLC samples and significantly associates with clinicopathological variables (p<0.05). High expression of KLF 8 was related with poor prognosis, and E-cadherin low expression and overexpression of Ki-67

Conclusions: This data suggested that KLF8 overexpression in NSCLC correlated with poor prognosis.
Presentation of a case of lung carcinoid tumor

6. Lung Cytology and Mediastinum

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Objectives: This report presents the case of a 77-year-old male, non-smoker with a right lung peripheral shadow at the radiological chest examination.

Material and methods: FNA of the mass was performed with a CT scan guidance and the material after being fixed to Cytolite, was processed through the Cytological method of Thinprep and the Papanikolaou stained smears were followed by an immunocytochemical control.

Results: In the microscopic examination, the material was cellular, with solitary cells and clusters, small sized, uniform cells, without atypia, oval-round shaped nuclei, finely granular nuclear chromatin (salt and pepper), nonpycnotic nuclei and regular nuclear membrane.

Immunocytochemistry showed: CD56 (+), Chromogranin (+), TTF1 (+), CK 5/6 (-). These morphological and immunocytochemical findings are compatible with the diagnosis of carcinoid tumor of the lung.

Conclusions: Carcinoid lung tumor can be diagnosed cytologically by FNA, fact that is very important for the preoperative evaluation.
THE ROLE OF EBUS-TBNA CYTOLOGY IN THE DIAGNOSIS OF THYMOMA TYPE A: A CASE REPORT

6. Lung Cytology and Mediastinum
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Objectives: Aspirates of thymomas are distinguishable from other lesions and FNA is a proven method for investigating mediastinal masses. A thymoma is classified in six subtypes according to morphology of epithelial cells and the ratio of lymphocytes and epithelial cells.

Material and methods: We reported a case of thymoma diagnosed via EBUS-TBNA by sampling mediastinal lymphonode in a women 53 years old with a mediastinal mass 10 cm in size.

Results: The smears show a cellular neoplasm composed predominantly of spindle shaped cells with mild atypia and lymphocytic components. Immunocytochemistry reveals the tumor cells to be positive for Ber EP4. Based on cytomorphology and ICH we diagnosed this case as atypical type A thymoma.

Conclusions: EBUS-TBNA cytology in combination with ICH plays a role in the subtyping of thymomas.
Two cases of Pneumocystis Jirovecii in HIV-positive patients.

6. Lung Cytology and Mediastinum
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Objectives: The utility of Cytology for triage and diagnosis of Pneumocystis jirovecii in HIV patients.

Material and methods: Case 1: 60 year-old female, recently diagnosed with Human Immunodeficiency Virus (HIV) presenting with visual disturbances, photophobia and headache. Chest X-ray showed bilateral patchy bibasal infiltrates and high-resolution computed tomography (HRCT) scan showed ground glass opacities, consolidation in the right and left upper lobes of the lungs, and bilateral pleural effusions.

Case 2: 55 year-old male with known history of HIV, previously treated with anti-retroviral therapy, presenting with progressive dyspnoea and weight loss. HRCT showed bilateral mid-zone ground glass opacities. Patients underwent bronchoscopy with bronchioalveolar lavage for cytology and microbiology.

Results: Both cases showed foamy and flocculent alveolar casts containing 4-6 micrometer cysts with a ‘cup and saucer’ appearance on Diff Quik (DQ) stain, which was more pronounced on the Papanicolaou (PAP) stain. A silver stain (Gomori-methenamine silver stain) highlighted these cysts brilliantly, showing them to be consistent with Pneumocystis jirovecii-associated pneumonia (previously called Pneumocystis carinii pneumonia or PCP). Pneumocystis jirovecii was identified in the sputum of both cases and by polymerase chain reaction (PCR) performed on the bronchioalveolar lavage in Case 2.

Both patients were commenced on intravenous and oral antibiotics with subsequent commencement of a prophylactic antibiotic. Resultant resolution of the infection was demonstrated on follow-up tests.

Conclusions: Pneumocystis jirovecii is a fungus, which was initially identified in immunocompromised children and young adults. However, today it is a commonly diagnosed opportunistic lung infection in HIV patients who may not necessarily present with respiratory symptoms (Case 1)¹. The mainstay of diagnosis is through a BAL specimen and these organisms are sometimes difficult to identify with routine cytological stains but are demonstrable with a silver stain, and by the sensitive PCR method². The main differential is red blood cell ghost cells.

Selected references
Value of bronchoalveolar lavage in Pulmonary Langerhans Cell Histiocytosis

6. Lung Cytology and Mediastinum
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Objectives: Pulmonary Langerhans cell histiocytosis (PLCH) is a rare histiocytic interstitial lung disease, characterized by abnormal infiltration of Langerhans cells (LCs) with lung parenchyma destruction.

Material and methods: PLCH is normally diagnosed from resected lung specimens. Cytological diagnostic methods such as bronchoalveolar lavage (BAL) used as a less invasive technique through bronchoscopy. In our laboratory between January 2017 and December 2018 we studied BAL fluid on 1675 patients and the 27 of them were diagnosed with PLCH.

Results: The diagnosis based at the increase of Langerhans cells identified immunocytologically by their reaction with the monoclonal antibody CD 1a and the polyclonal antibody S-100 (S-100 protein).

Conclusions: In conclusion BAL cytology can be a useful approach to diagnosis of PLCH and assessment of disease activity.
7. Cytotechnology
Comparative Study in hematic and high mucus content specimens: conventional technique versus Citospin Collection Fluid® procedure.

7. Cytotechnology
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Elisa López Bua, Maria Cruz García Díaz, Sara Márquez del Pozo, Nadia Espejo Herrera, Núria Baixeras González, Alba Zanca Càlix, Cristina Márquez López, Maria Pilar López Callau, Eider Rodiño Díaz, Nerea Gálvez Aguilar, Roger Llatjós Sanuy, Isabel Català Costa
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Objectives: Hematic and high mucus content (HMC) samples are difficult to process, since this elements prevent from selecting adequate material to obtain optimal smears. A variation of the conventional procedure (CP) was designed in our department to facilitate the processing: the addition of Citospin Collection Fluid® (CCF®) -as an hemolitic and mucolitic agent- before centrifugation. Our aim is to demonstrate the new method’s efficiency and perform a comparative study with the CP.

Material and methods: The study included samples from serous cavities and bronchial aspirates (BA). The samples were random distributed into two test tubes, one processed according to the CP and the other one by new method. The CCF® was added to the samples before the centrifugation in a ratio 1:1 and centrifuged at 1500 RPM/15’ (revolutions per minute). As a result of the process, we obtained four slides: 1-2 processed by CP and 3-4 by CCF®. The evaluated parameters were: amount of cellularity, quantity of eritrocites and mucus in a scale from 1 to 3.

Results: 68 samples were collected: 44 hematic BA (64.7%), 12 HMC BA (17.6%), 5 ascitic effusions (7.4%), 5 pericardial effusions (7.4%) and 2 pleural effusions (2.9%). Convention procedure: lower cellularity (scale 1) in 44.1% of samples; CCF® procedure: higher cellularity (scale 3) in 48.5% of samples (p=0.08). The samples processed with CP with less amount of eritrocites represented the 30.9% of the total, whereas processing with the new method the samples with less quantity of red blood cells were the 38.2% (p<0.001).

Conclusions: The CCF® added method used in hematic and HMC samples facilitate the technicians procedure to obtain better quality smears, aboiding repetitions and diagnosis delays.
Performance of the Genesis® processor for cervical cytology and HPV testing: workflow and test results

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Miet Van Herck², Birgit Weynand²
¹ Imelda Hospital, Bonheiden, Belgium
² University Hospital Leuven, Leuven, Belgium

Objectives: To compare the Genesis (Hologic)® versus the T2000 (Hologic)® processor for slide preparation and versus manual aliquoting for HPV in terms of
- workflow
- cytology quality and HPV results
- the ease of use, chain of custody and ergonomic considerations

Material and methods:
100 routine cervical cytology samples were assessed in Imelda hospital, both on the Genesis® in 'slide prep mode' and on the T2000(R) to produce duplicate cytological slides from the ThinPrep® vial. 100 routine samples for HPV testing were processed in UZ Leuven in duplicate both on the Genesis® in 'aliquot mode' and by manual aliquoting of the sample into an Aptima® transfer tube. The workflow measurement included the average time to prepare one specimen (TAT), the number of specimens prepared in an 8 hour workshift (throughput) and the hands-on time per 8 hour shift (HOT). Operators (2) and processor shifted after 50 samples. Slide quality measurement included cellularity, cell spot border and equal cell distribution. Reproducibility of cytological diagnosis was assessed blinded in 10 NILM and 10 pathological slides. The HPV result comparison included concordance of positivity rate and invalids.

Results: The workflow results are presented in table1. As well for slide preparation as for HPV sample aliquoting, the Genesis® processor for slide preparation yielded longer TAT and lower 8-hour throughput. However, using the Genesis® significantly reduced the operator-time variability and HOT (over 40% decrease). The slide quality was highly reproducible after Genesis® or T2000® preparation: cellularity 98%, spot border 100%, cell distribution 96%. Cytological diagnosis was in perfect agreement with both processors. HPV results were reproducible in 85% of samples. There was no inter-operator variability.

Conclusions: For similar cytological and HPV results, the Genesis® processor generates more walk-away time, which is perceived positively in a routine laboratory workflow, despite longer TAT.

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8. Breast Cytology
BREAST CANCER IN YOUNG WOMEN

8. Breast Cytology

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Objectives: Breast cancer is one of the most common human neoplasms in women. About 1 in 8 women will develop breast cancer before the age of 85, with the incidence to increase with age. It is estimated that about 30% of newly diagnosed cancers in women in USA will be breast cancers. In women age 40 or younger the risk is low, under than 7%. However by the time a lump can be felt, the cancer is often advanced, due to the density of breast tissue. Fine Needle Aspiration of breast lesions is routinely performed in our Hospital. We reviewed our archives and found the cases of breast cancer in women under forty years old.

Material and methods: One thousand two hundred twenty four aspirates, in breast palpable lesions, were performed during the last ten years. The aspiration was done using 21-gauge needle. Conventional and Thin Prep smears were prepared. Estrogen and progesterone receptor studies were part of the routine work up of breast cancer as well as other markers (HER-2/neu, p53, E-Cadherin, Ki-67).

Results: Three hundred and six of the cases were carcinomas, whereas twelve in women under forty (mean age 38.2). In nine of these patients, Cytology correctly diagnosed eight ductal and a tubular carcinoma. In three cases the cytologic diagnosis was suggestive of carcinoma and the excisional biopsy revealed a lobular, a ductal and a mixed carcinoma.

Conclusions: Most young women consider themselves too young to have breast cancer but it can happen at any age, so a young woman must be aware of the disease. A breast lesion, in this age group, must be identified using mainly Ultrasound and FNA, which are simple and well tolerated diagnostic tools. However breast MRI is recommended for younger women when a family history of breast cancer or other risk factors exist.
Fine Needle Aspiration Biopsy is a Valuable Diagnostic Test in Women Age 70 Years or Older with Breast Lesions

8. Breast Cytology

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Objectives: Advancing age is a major risk factor for developing breast carcinoma. Few studies, however, focused on diagnostic approaches or treatment regimens for the cohort of older women diagnosed with breast carcinoma.1,2,3 Fine needle aspiration biopsy (FNAB) can be used as a diagnostic tool for women of all ages. Breast FNAB in combination with ancillary testing may be helpful in older patients with comorbidities that make additional diagnostic or surgical procedures clinically challenging.

Material and methods: Cytology reports from women who underwent FNAB for predominantly palpable breast masses at Massachusetts General Hospital from 01/2009 to 07/2018 were retrieved and retrospectively evaluated. Clinical data, cytologic diagnoses, histologic outcomes, and immunohistochemical stains of hormone receptors (ER/PR) done on cell blocks were recorded. FNABs were classified as non-diagnostic, benign, atypical, suspicious for malignancy or malignant. Risk of malignancy (ROM) was determined by histology (705 cases) or clinical outcomes (1105 cases).

Results: A total of 1810 breast FNABs were sampled from 1612 patients, of which 305 (16.9%) cytology samples were from 271 patients age ≥70 yrs and 1505 (83.1%) were from 1341 patients age <70 yrs. For breast FNABs in patients age ≥70 yrs, complete sensitivity, specificity and non-diagnostic rate was 89.0%, 84.1%, and 6.9%, respectively, compared to 79.9%, 85.8% and 5.3% for patients age <70 yrs. For age ≥70 yrs, ROM for FNABs in non-diagnostic, benign, atypical, suspicious and malignant categories was 42.9%, 5.7%, 71.4%, 100% and 100%, in contrast to 13.8%, 4.0%, 36.5%, 84.2% and 98.9% for patients <70 yrs. Cell blocks provided hormone receptor status in 50 patients age ≥70 yrs as the primary diagnostic modality.

Conclusions: FNAB is minimally invasive while maintaining high sensitivity and specificity in older women ≥70 yrs. Additional diagnostic procedures can be spared by using FNAB cell blocks for hormone receptor testing, allowing for initiating of treatment, as indicated by clinical condition.

Selected references
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Hydrogel based marker (Hydromark®) in fine needle aspiration (FNA) of axillary adenopathy. Recognition and prevention of possible errors.

8. Breast Cytology

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Objectives: HydroMARK® is a tissue marker based on polyethylene glycol hydrogel with a central metal coil used in ultrasound-guided biopsies to facilitate localization of the lesion in subsequent surgery. To date, there are few cases in the literature that describe the cytological findings related to the use of this marker.

We aim to discuss the cytological characteristics, differential diagnoses and the importance of recognizing the findings associated with hydrogel.

Material and methods: 60-year-old woman with infiltrating ductal carcinoma of the left breast. FNA of left axillary lymph nodes previously marked with HydroMARK® was performed during ultrasound evaluation of the response to neoadjuvant chemotherapy for the breast carcinoma. Five smears and one cell block were obtained. Two smears were stained with DiffQuick and three were stained with Papanicolaou.

Results: Smears showed haematic background with abundant presence of finely granular violaceous material with some separate dense globules on Diff-Quick® stained smears; and pink-blueish material with separate dense globules on Papanicolaou stained smear. Abundant multinucleated giant histiocytes and mononucleated histiocytic proliferations, with occasional accompanying lymphoid cellularity, were present.

The groups of histiocytes in a gel background mimic mucinous carcinoma, posing it as a differential diagnosis. Morphologically, some histiocytes can be confused with epithelial cellularity, therefore immunostaining for GATA3 was performed and resulted negative. The differential diagnosis also includes myxoid material, silicone, hyaluronic acid, ultrasound lubricating gel and even chondroid material.

Conclusions: Hydrogel material can be overlooked or confused with other substances and mimic fat necrosis or vacuoles of cryptococcosis in FNA of the breast. Immunocytochemical techniques may be required to rule out malignancy and it is especially important to review the clinical history, in order to avoid diagnostic errors.
9. Effusions Cytology
A rare mimicker of malignancy in pleural fluid: reactive histiocytic proliferation

9. Effusions Cytology

Deniz Tolga
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Objectives: Pleural effusion (PE)s are one of the most common material a cytopathologist deals with. Various types of malignancies or benign diseases may lead to fluid accumulation within the pleural spaces. Detecting malignancies is the primary role of the cytopathologist in PEs. However, due to variations in cell populations and amount, this may be challenging. Atypical mesothelial or histiocytic proliferations are the most problematic cases with many features overlapping with malignancy. Hereby, we present cytological features of a reactive histiocytic proliferation.

Material and methods: A 80-year-old man, followed up with chronic obstructive lung disease and congestive heart failure; with a medical history of coroner by-pass operation in 2002, presented with shortness of breath. PA chest X-ray revealed right sided pleural effusion. For symptomatic relief, a total of 2000 cc pleural drainage was made with a week interval, using pleurocan catheter, which was found to be transudate. Part of the pleural fluid was spared for ARB and tuberculosis culture, which both were negative.

65 cc macroscopically clear effusion fluid was sent to cytopathology. Both conventional and liquid based cytology methods were used for preparation, 5 slides and 3 cell blocks were obtained.

Results: The cytologic material was predominantly composed of cells with eccentrically located nuclei and vacuolated cytoplasm accompanied by sparsely scattered mesothelial and inflammatory cells. Ancillary studies was performed to the cell block. Intracytoplasmic mucin was not detected with mucarmin stain. Immunocytochemical stains revealed diffuse strong cytoplasmic staining with CD68 in the signet cell mimicker cells while the epithelial / mesothelial markers were negative (PanCK, CK7, CK20, Ber-EP4, TTF-1, PSA, PSAP, calretinin). The case was reported as reactive histiocytic proliferation.

Conclusions: Reactive histiocytic proliferation is uncommon in PEs. Atypical histiocytes resembling especially signet ring cell adenocarcinoma, may be challenging for the cytopathologist. Awareness of this difficult entity and performing ancillary studies is important for accurate diagnosis.

Selected references
Disseminated Strongyloidiasis diagnosed in ascitic fluid cytology

9. Effusions Cytology
Subitha Kandamuthan

Objectives: Strongyloidiasis is a parasitic infection caused by intestinal nematode Strongyloides stercoralis. Disseminated disease can give rise to gastrointestinal, pulmonary, neurological and dermatologic manifestations. We describe a case of disseminated strongyloidiasis in a chronic alcoholic patient which was diagnosed by ascitic fluid cytology.

Material and methods: The case is that of a 66 year old male who was a chronic alcoholic and smoker. The patient was admitted to the hospital with two episodes of haematemesis and melena. On physical examination the patient was drowsy. He was emaciated with bilateral pedal edema and abdominal distension. There was no organomegaly. Stool examination for parasitic ova and cysts were found to be negative. Ultrasonogram of abdomen showed normal sized liver with coarse parenchyma with bilateral pleural effusion and ascites.

Results: A wet mount microscopic examination of the ascitic fluid showed multiple live motile parasitic larvae which were identified as larvae of Strongyloides stercoralis [Figure 1a]. Papanicolaou and Leishman staining of smears of the ascitic fluid showed the filariform larvae in a background of inflammatory cells composed of neutrophils and eosinophils [Figure 1b,1c]. With a diagnosis of Strongyloidiasis patient was started on Albendazole. A skin biopsy was taken from the purpuric lesion on the abdominal flank which demonstrated filariform larvae in the dermis confirming a diagnosis of disseminated strongyloidiasis [Figure 1d]. Unfortunately the patient died on the second day of diagnosis.

Conclusions: Strongyloides stercoralis infection has a higher prevalence in tropical and sub tropical regions worldwide. Prevalence in India was found to be 6.6% in community-based surveys and 11.2% in hospital-based surveys[1]. Screening pts for asymptomatic strongyloides infection is crucial to prevent hyperinfection syndrome. Among the available tests, serologic tests are the most reliable and most sensitive in population from strongyloides endemic areas. Anyone at high risk for dissemination should be screened before starting immunosuppressive therapy.

Selected references
Immunocytochemical expression D2-40 in cases of adenocarcinomas in contrast to expression in cases of reactive mesothelial cells in serous effusions

9. Effusions Cytology
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3 Koliou Angeliki Cytology department of General Hospital Snt Andreas, Patras (Greece)
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Objectives: The microscopic appearance of reactive mesothelial cells have a broad spectrum of characteristics, ranging from normal to atypical presentation. Atypical cells show variation in nuclear size, contour, and coarse chromatin and prominent nucleoli. Since mesothelial cell atypia is difficult to be distinguished from metastatic adenocarcinomas it is necessary to evaluate further suitable immunostains. The purpose of this study is to evaluate the expression of D 2-40 in cases of adenocarcinomas in comparison to cases of reactive mesothelial cells of serous effusions

Material and methods: We examined 50 specimen of serous effusions of males and females patients. The average patients age was 65 years. 25 of the specimens was adenocarcinomas and 25 was mesothelial cells reaction. The recommended liquid-based sample preparation protocol was followed for each specimen. The immunohistochemistry method with polymer detection system (biotin-free) was performed on destained cytologic smears for the detection of D2-40. Membranous immunoreaction of D2-40 was considered as positive if present in >10% of stained cells.

Results: Immunostaining in D2-40 was demonstrated in 92% (23/25) of cases with reactive mesothelial cells but none (0/25) in cases of adenocarcinomas.

Conclusions: The expression of D2-40 can be determined in serous effusion cytology and D2-40 may be a useful marker in differential diagnosis of reactive mesothelial cell proliferation versus adenocarcinomas.
Malignant pleural effusion due to small cell carcinoma of the lung: A cytomorphological study of 23 cases

9. Effusions Cytology
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Objectives: To evaluate the frequency and cytomorphological features of small cell carcinoma of the lung (SCCL) in pleural effusions samples.

Material and methods: We analyzed 1902 pleural effusions samples received in our Department between 2008 and 2018. The cytological characteristics of those diagnosed as SCCL were reviewed.

Results: The 313 cases of the malignant group (16.45%) included 224 cases of primary lung carcinomas (71.57%), 3 cases (0.9%) of mesotheliomas, 24 cases (7.7%) of extrapulmonary metastatic carcinoma, 20 cases (6.4%) of hematologic neoplasms and 12 cases (3.8%) of other neoplasms (sarcoma, melanoma).

Of the 224 cases due to primary lung carcinomas, 23 (10.27%) were diagnosed as SCCL. 20 cases (86.9%) were males with a mean age of 69.8 years (range 50-84). The cytological samples were predominantly hypercellular (13 cases, 56%) and the background was mostly hemorrhagic (16 cases, 69.6%). Small-to-medium size neoplastic cells appeared in groups in 21 cases (91.3%), 86.9% of which (20 cases) included cords, rows and aggregates in crescents. Associated inflammation was mainly lymphomonocytic (16 cases, 69.6%) and of moderate intensity (13 cases, 56.5%); its characteristics did not depend on tumor cell density. Atypical mitoses and/or apoptosis were seen in 20 cases (86.9%). The distinctive nuclear features of SCC were easily identified in all; only in 2 cases (8.7%) cytoplasms were more abundant than expected. Spindle-cell morphology was not observed. All cases were immunoreactive for neuroendocrine markers.

Conclusions: The occurrence of pleural effusions in the clinical course of small cell carcinoma of the lung is an infrequent event. Although the different preservation conditions of the neoplastic cells in fluids and their tendency to degenerate rapidly may hinder their recognition, some of the most characteristic features of this neoplasm are retained. Immunocytochemical stains allow to confirm the diagnosis and exclude other neoplasms, mainly lymphoproliferative disorders and small blue round cell tumors.

Selected references
Metastatic tumors that are seen rarely in effusions

9. Effusions Cytology

Deniz Tolga

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Objectives: Although it varies according to localization, age and sex, some malignancies cause malignant effusion more frequently; such as lung, breast, ovary, gastrointestinal tract carcinomas, and lymphoreticular malignancies. Also another malignancies may cause malignant effusion. Metastatic tumors, which are seen rarely in effusions, cause different diagnostic difficulties.

The aim of this study is to analyze the cytomorphological and immunocytochemical features of rare metastatic tumors in effusions and to investigate the effectiveness of preparation methods and clinical data in terms of reaching diagnosis.

Material and methods: 3200 effusion cytology were analyzed retrospectively between 2010-2018. Rare metastases in malignant effusion cases were included to the study. Demographic data and clinical information were obtained from pathology reports. The slides were prepared by conventional method ± liquid based cytology techniques. Conventional method ± plasma-thrombin methods were used to prepare the cell blocks.

Results: 38.4% of the effusions were malignant effusion. 36 cases with rare malignant tumor metastasis in effusions were included in the study. The number of pediatric / adult cases was 2/34. Pediatric cases were 15 and 16 years-old. The mean age in adults was 53.7 (range: 20-88).

Distribution of cases according to determined primary tumor: various sarcomas (2 rhabdomyosarcoma): 7, lobular carcinoma of the breast: 7, clear cell carcinoma of the ovary: 2, nasopharyngeal carcinoma: 2, malignant melanoma: 2, renal cell carcinoma: 2, plasma cell tumor: 2, thymic malignancy: 2, various other malignancies: 10 cases.

Tumors observed in the pediatric age group were Ewing's sarcoma and adrenocortical carcinoma metastasis.

5 of 7 sarcoma metastases were detected in pleural effusion while all of the lobular carcinoma metastases were detected in peritoneal effusion.

Conclusions: Although rare metastatic malignancies in effusions create some diagnostic difficulties; identification of malignancy features by a rigorous cytomorphological analysis, classification of malignancy with the support of clinical history and immunocytochemical studies, provide accurate diagnosis.

Selected references

Myelomatous peritoneal and pleural effusion - A rare initial presentation of multiple myeloma mimicking carcinomatosis with raised serum CA 125

9. Effusions Cytology
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Objectives: Multiple myeloma (MM) is a neoplastic disease of plasma cells accounts for 2% of all malignancies worldwide, most commonly involving the bone marrow and causing local destruction of the bones along with anaemia, kidney failure, hypercalcemia, infections and paraproteinemia. Malignant effusions are rare initial presentation of MM without any bone symptoms. We report a case of multiple myeloma, IgG kappa, ISS stage III, presented initially as myelomatous pleural and peritoneal effusion.

Material and methods: A 45 year old female admitted with complaint of abdominal distension, loss of appetite and weight with no history of bone pain. There was no palpable lymphadenopathy or hepatosplenomegaly. Complete blood examination revealed only anaemia with raised serum CA125 (221.2 U/mL). High Resolution CT (HRCT) chest showed bilateral moderate pleural effusion with passive segmental collapse of right lung. Computerized tomographic (CT) abdomen revealed moderate ascites with diffuse peritoneal and omental thickening and no obvious adnexal lesion. Hence, possibility of peritoneal carcinomatosis was given elsewhere. Repeat cytologic examination of ascitic and pleural fluid was done in our centre. Smears were stained with May Grunwald-Giemsa and Haematoxylin-Eosin which showed presence of atypical plasmacytoid cells. Immunohistochemistry was done on the cell block which showed CD138 and MUM1 positive tumor cells. Later, PET scan showed diffuse involvement of bone marrow without any lytic lesions.

Results: A complete review of world literature showed only 7 cases of plasmacytic ascites diagnosed at initial presentation 1 while plasmacytic pleural effusion presents only in 0.8% cases secondary to disease 2

Conclusions: To our knowledge this is the first case of MM presented initially with both myelomatous peritoneal and pleural effusion, raised serum CA125 and diagnosed by fluid cytology. This case emphasise the importance of cytomorphological examination, utility of immunostains in fluid and considering the possibility of raised CA125 in neoplasms other than primary peritoneal or adnexal epithelial tumors in females.
Selected references
p40 is more specific than p63 in distinguishing between squamous cell carcinoma and adenocarcinoma in effusion cell blocks

9. Effusions Cytology
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Objectives: While squamous cell carcinoma (SCC) is one of the leading causes of cancer-associated death globally, it rarely causes malignant effusions. Distinguishing between SCC and adenocarcinoma in effusion cytology can be challenging. p63 and p40 have been frequently used to support the presence of squamous differentiation in both histologic and cytologic specimens. However, similar results have been rarely reported in cytologic preparations of effusion fluids. This study was designed to evaluate the diagnostic value of p63 and p40 immunoreactivity to distinguish SCC from adenocarcinoma in malignant effusions.

Material and methods: p63 and p40 immunocytochemical staining was performed on 30 cell block specimens, including 10 malignant effusions with SCC and 20 malignant effusions with adenocarcinoma. Any degree of nuclear staining was considered positive.

Results: Of the 10 SCC cases, 100% were positive for both p63 and p40, and most cases showed diffuse staining (>25% of tumor cells). Expression of p63 and p40 was found in 4 (20%) and 2 (10%) of 20 adenocarcinoma cases, and the extent of staining was all focal (≤25% of tumor cells). For distinguishing SCC from adenocarcinoma in malignant effusions, the p63 reactivity was found to be 100% sensitive and 80% specific with a positive predictive value of 71% and a negative predictive value of 100%. The sensitivity of p40 for SCC was 100%, the specificity was 90%, the positive predictive value was 83%, and the negative predictive value was 100%.

Conclusions: Although p63 and p40 are both useful markers for the diagnosis of SCC in malignant effusions, p40 is more specific than p63 in distinguishing between SCC and adenocarcinoma.

Selected references
Papillary renal cell carcinoma in pleural effusion: A review of the literature based on a case report

9. Effusions Cytology
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Objectives: The appearance of malignant effusions secondary to renal cell carcinoma is a rare event, with a prevalence of 1-2.2% in largest series published. Cytologic evaluation plays an important role in the recognition of renal origin, especially in papillary cases frequently confused with pulmonary adenocarcinoma or epithelioid mesothelioma. Often, immunohistochemistry serves as an important adjunct in confirming the diagnosis.

Material and methods: We present the clinicocytological details of papillary renal carcinoma in a pleural effusion and review the frequency of this neoplasm and its potential diagnostic pitfalls.

Results: We present a case of 64 year-old male with left radical nephrectomy diagnosed of Type 2 papillary renal cell carcinoma 21 months previously. In the follow-up, lymph node enlargement, pulmonary bilateral lesions and pleural effusion were interpreted as progression. Thoracentesis was performed, and the cytomorphologic features reveal a very cellular effusion with papillary groups, scattered macrophages and isolated small psammoma bodies. Cytologically, the cells display moderate amounts of cytoplasm with enlarged nuclei with prominent nucleoli. Immunohistochemistry performed on cell-block slides was diffusely positive for PAX8 and BerEP4 and TTF1, Calretinin, WT1 were negative.

Conclusions: Malignant pleural effusions are mostly related with lung, breast or ovarian adenocarcinomas but the cytological cases due to renal cell carcinoma are rare and most commonly occur in patients with papillary and clear cell tumors. Nowadays, cytologic evaluation has an important role in the recognition of renal origin and also in the diagnosis of new entities of renal cell carcinoma. When facing markedly cellular effusions with papillary groups propose differential diagnosis with epithelioid mesothelioma and papillary renal cell carcinoma. A immunohistochemical panel with calretinin, WT1, BerEP4, TTF1, PAX8 or PAX2 will detect most cases of metastatic RCC.

Selected references
Pleural effusion involvement by myeloid sarcoma: cytology importance for diagnosis of rare disease presentation

9. Effusions Cytology

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Objectives: Myeloid sarcoma (MS) is a rare disease that can present as an isolated extramedullary leukemic tumor, concurrently with or at relapse of acute myeloid leukemia¹. It presents as a tumor mass of myeloid blast, with or without maturation, occurring at an anatomical site other than bone marrow. MS has to be differentiated from an infectious process, carcinoma or malignant lymphoma. Cytology evaluation allowed the proper identification of neoplastic etiology².

Material and methods: We present a case of a 30 years-old male patient, with toracalgia, cough, nocturnal sweating and weight loss of 3 kg in the last 3 months. Thoracic computerized tomography revealed an anterior mediastinum mass measuring 6.3x8.3x15.8cm with lobed contours that was punctured by FNAC.

Results: Cytological evaluation showed, after cell-block realization, a sample constituted massively by blastic bulky cells, with high nuclei/cytoplasm ratio, with large, rounded nuclei, with granular chromatin, and positivity to LCA, myeloperoxidase, lysozyme and CD43, Ki-67 of 90%, and negativity for CD20, PAX5, CD3, CD10, TdT, ALK and CD163. Hystology of transthoracic biopsy of this mass corroborated the immunohistochemical (IHC) profile of this neoplasia.

Conclusions: It was diagnosed a pleural effusion involvement by a lymphoproliferative neoplasia with immunohistochemical profile that favored a myeloid sarcoma. Cytomorphological and immunohystochemical examination was crucial to diagnosis and allowed a quick oriented therapeutic initiation. MS may occur almost at any site in the body and cytological evaluation is an excellent first step to diagnosis²,³. In our case, the typical cytological appearance of cells, IHC profile and histological correlation helped us to conclude the final diagnosis.

Selected references
Potential diagnostic pitfalls in hydrocele fluid cytology: A case report.

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Objectives: INTRODUCTION: Hydrocele is the excess fluid accumulation in the cavity of tunica vaginalis. Both benign and malignant causes of hydrocele have been documented with the former outnumbering the latter. The commonest malignant causes of hydrocele are testicular tumors, while metastases to the testes occur extremely rare with hydrocele1, 2.

Material and methods: CASE REPORT: PATIENT HISTORY: A 54-year-old male presented to the urology clinic complaining of painless, scrotal swelling. Physical examination was suggestive of left non-communicating hydrocele and scrotal ultrasonography confirmed it. Patient had a previous history of radical cystectomy, four years ago, for high grade urothelial carcinoma. Due to past history of malignancy, hydrocele fluid was aspirated and delivered to our cytopathology department.

Results: CASE REPORT: CYTOLOGY: Cytologic examination showed several, single, atypical cells with large, irregular, hyperchromatic, occasionally mutilobated nuclei, and moderate amount of basophilic cytoplasm. In Giemsa stain fine vacuolation of the cytoplasm was noted. Although the features were highly suggestive of malignancy, immunocytochemistry was negative for HBME-1, CK20 and GATA3 and positive for CK20. Cytology report mentioned severe atypia of unknown origin. Serological tests were within normal limits. The patient was submitted to left orchiectomy and ipsilateral tunica layers excision. Histologic diagnosis was negative for malignancy and revealed chronic erosive serositis of the tunica layers.

Conclusions: CONCLUSIONS: In our case, highly reactive mesothelial cells masquerading malignancy in hydrocele fluid of a patient with history of urothelial carcinoma could have led to a false positive diagnosis. Inconclusive immunocytochemistry was helpful. Keeping in mind that tunica vaginalis is susceptible to metaplasias3, cytopathologists should be aware of pitfalls when diagnosing hydrocele fluid based on cytomorphological features alone.

Selected references
1. World Health Organization Classification of Tumours. Pathology and Genetics of Tumours of the Urinary System and Male Genital Organs. Lyon, France: IARC Press; 2004
Primary Low Grade Serous Papillary Carcinoma of Peritoneum in Ascitic Fluid: A Case Report

9. Effusions Cytology

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Objectives: Primary peritoneal serous carcinoma, especially psammocarcinoma, is very rare. Morphologically, it resembles its ovarian counterpart; in order to diagnose it, clinical and histopathological findings should be taken into consideration.

Material and methods: We describe case of a 48-year-old woman admitted to our hospital with a grade 3 ascites. A CT scan of the abdomen and pelvis revealed multiple cystic lesions dispersed throughout the peritoneum. The patient underwent an ascitic fluid paracentesis. The cytologic findings included many papillary groups of glandular cells, some of them with mild cellular atypia as anisonucleosis with some nuclei with coarsely granular chromatin and accompanied by psammoma bodies in the background. Total hysterectomy with bilateral salpingo-oophorectomy and omentum removal were performed.

Results: The histologic findings corresponded to a primary peritoneal, especially a low-grade serous carcinoma with abundant (70%) psammoma bodies (psammocarcinoma), confirming the initial cytologic diagnosis. Immunohistologically, ER, WT-1, CK7, Ca-125 were indicative of a serous carcinoma. The tumor was restricted at the peritoneal surface of both ovaries and fallopian tubes, as the uterine serosa and omentum, without invasion of underlying organs. Based on the criteria proposed by the Gynecologic Oncology Group, “primary” peritoneal carcinoma should present: involvement of extraovarian sites, normal sized ovaries without invasion and histological findings similar with epithelial ovarian cancer.

Conclusions: Psammocarcinoma is a very rare variant of primary peritoneal serous carcinoma. The cytological and histopathological findings are similar and the differential diagnosis between low-grade and borderline serous carcinoma is very difficult. Especially, the presence of psammoma bodies into the ascitic fluid is the only characteristic finding in cytology. So, any standardized management of primary peritoneal serous carcinoma has not been established, due to its rarity.
T-Lymphoblastic lymphoma presents extensive emperipolesis in pleural effusion. A case report

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Objectives: Emperipolesis, defined as the presence of a viable intact cell or cells within the cytoplasm of another cell, has been reported in several diseases. Rosai-Dorfman disease is most famous, where intact lymphocytes are found in histiocytic cytoplasms. It has been rarely described in cases of malignant lymphoma. Here, we report a case of T-lymphoblastic lymphoma of the mediastinum, first manifested as pleural effusion with prominent emperipolesis on cytology samples.

Material and methods: A 45 year-old male complained of cough, dyspnea on exertion and pleuritic chest pain on the right side. No fever or weight loss was reported. Chest CT showed a large mass in the anterior mediastinum extending from manubrium to diaphragm and massive pleural effusion of both sides. Thoracentesis was performed and a ThinPrep® (Hologic, Marlborough, MA, USA) Papanicolaou stained slide was obtained.

Results: Cytologic examination showed a cellular specimen consisting of numerous small-sized mature-appearing lymphocytes and mesothelial cells. Lymphoid cells demonstrated high N/C ratio and minimal irregularity of nuclear membrane. Occasional apoptotic bodies are present. Histiocytes frequently showed intact lymphocytes in cytoplasms, with numbers up to 15. Immunohistochemistry on additional ThinPrep® slides that lymphoid cells showed positivity for CD56 and CD3. Histologic examination of the mediastinal mass confirmed the diagnosis of T-lymphoblastic lymphoma

Conclusions: Although the incidence of emperipolesis in lymphomatous effusion is very rare, it is important to aware the possibility of lymphoma when emperipolesis is found in fluid cytology specimen

Selected references
The role of cytology in the diagnostic work-up of patients with pleural effusions

9. Effusions Cytology

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Objectives: To establish the main cause of malignant and non malignant pleural effusions

Material and methods: Patients were selected from our electronic database. Clinical history of each patient and data of pleural fluid cytology, immunocytochemistry and other laboratory and imaging findings (for negative cases) have been analyzed.

Results: The pleural fluid of total number of 81 patients were examined. Positive cytology was observed in 22 patients (18 woman 81,8 % and 4 man 18,2% ). Age distribution was 49-84 for women and 57-68 for men. Primary site of malignant effusions for women was breast in 8 patients(44,4 %), ovary in 4 patients(22,22 %), lung in 2 patients(11,1%). Mesotelioma was diagnosed in 4 women (22,2 %) For men the cause of malignant effusion was lymphoma in 2 cases, small cell carcinoma in 1 case and lung adenocarcinoma in 1 case.

In 59 cases cytology was negative.

In cases with negative cytology (59 patients) follow up showed false negative 5 cases, all were cases of lung carcinoma.

In 54 patients clinical history and imaging, laboratory investigations were analysed. The principal cause was bacterial infection (40 patients 67.7%), mycobacterial infection (5 patients 8,5%), pulmonary embolism (4 patients 6,7%), congestive heart failure (10 patients 16,9%) .

Conclusions: Cytologic examination with immunocytochemistry of pleural effusions is the easy and effective technique for the diagnosis of malignant disease.

The most frequent primary metastatic site in our study was breast carcinoma for woman.

The main cause for non malignant fluids was bacterial infection.

Selected references
1. Art and science of cytopathology R. De May 1996
10. Pancreatico/biliary and Liver Cytology
Coexistence of Acinar Cell Carcinoma and Solid-Pseudopapillary Neoplasm of the Pancreas Diagnosed by FNAC: the First Case Report

10. Pancreatico/biliary and Liver Cytology

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Objectives: Acinar cell carcinoma (ACC) of the pancreas is a rare malignant epithelial neoplasm composed of cells resembling morphologically acinar cells. Solid-pseudopapillary neoplasm (SPN) is also an uncommon, low-grade malignant neoplasm. The aim of this report was to present an unusual case of coexistence of ACC and SPN diagnosed by endoscopic ultrasound-guided fine needle aspiration cytology (EUS-FNAC).

Material and methods: We report a case of a 52-year-old woman who presented with abdominal pain for 10 days. MR imaging and endoscopic ultrasonography revealed two masses in the body (1.2 cm) and tail (5.0 cm) of the pancreas. EUS-FNAC for two separate masses in the body and tail was performed. Cytological smears were stained with hematoxylin-eosin and Papanicolaou stains. Cell block preparations were performed.

Results: The smears from the body mass showed an acinar pattern of monomorphic polygonal tumor cells. The background was clean and occasional single cells. The tumor cells had eccentrically placed, uniform, round to oval nuclei with coarse chromatin and single prominent nucleolus. The cytoplasm was abundant and granular. Immunohistochemical stain on the cell block showed positivity for α1-antitrypsin. Cytological findings were compatible with ACC. The smears from the tail mass showed papillary architecture of tumor cells, with delicate fibrovascular cores. The background was clean and loose clusters or single cells were present. The tumor cells had uniform, round to oval nuclei with finely granular chromatin and inconspicuous nucleoli. The nuclear membrane was smooth, slightly indented or grooved. The cytoplasm was scant to moderate. Extracellular hyaline globules were observed. The immunohistochemistry showed nuclear positivity for β-catenin. Cytological findings were compatible with SPN. Distal pancreatectomy was performed. Histological examination revealed coexistence of ACC and SPN of the pancreas.

Conclusions: To our knowledge, this is the first case of coexistence of ACC and SPN of the pancreas diagnosed by EUS-FNAC.
Pancreatic giardiasis in a patient with diffuse large B-cell lymphoma.

10. Pancreatico/biliary and Liver Cytology
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Objectives: Infestation of duodenal giardia is a common cause of infectious diarrheal disease in developing and industrialized areas of the world, however pancreatic involvement by Giardia lamblia is a rare entity, with few cases reported in the literature.

We aim to present a case of pancreatic giardiasis in a patient with history of diffuse large B-cell lymphoma.

Material and methods: 56-year-old male patient with history of enolism and diffuse large B-cell lymphoma in remission and latent tuberculosis infection, with 3-month history of weight loss, abdominal pain, vomiting and recurrent pancreatitis. An endoscopic ultrasound scan revealed a 13.5 mm space-occupying head-neck lesion with a solid, heterogeneous appearance and a regular outline.

Two 22G needle passes were performed and collected in ThinPrep, obtaining a cytological extension that was stained with Papanicolaou.

Results: In the smear, a necrotic background with groups of ductal cells with reactive changes was observed.

There were abundant trophozoite-like structures measuring 12-15 μm in maximum dimension, with a pear-shaped characteristic appearance, or sickle-shaped on edge. Two “mirror image” nuclei were often noted in the thick portion of the organism opposite to the flagella, which were situated the narrower part.

Conclusions: This parasite usually results in opportunistic infections in immunosuppressed patients. The abundance of trophozoites in the aspirate makes duodenal contamination unlikely as a cause. The association between Giardia lamblia and cancer is not completely known.
POSSIBILITIES OF CYTOLOGIC DIAGNOSTICS OF METASTATIC DAMAGES OF THE PANCREAS ON THE MATERIALS EUS-FNA

10. Pancreatico/biliary and Liver Cytology

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Objectives: To define possibilities of an immunocytochemistry exam of material EUS-FNA in differential diagnostics of metastatic damages of the pancreas.

Material and methods: In the period from 2016 to 2018 in The Moscow Clinical Scientific Center, 299 patients with solid tumors of the pancreas were performed. Among them, adenocarcinoma of pancreas was diagnosed in 96.3% (n=288), in 3.7% of cases (n = 11) secondary lesion of pancreas was not excluded. All patients underwent endosonography using contrast enhancement and fine-needle biopsy.

Results: Of 11 cases, metastatic pancreatic lesions were morphologically verified in 4 patients (36.4%). In two cases, metastasis of renal cell carcinoma (RCC) were diagnosed, in one - metastases of small cell lung cancer (mcl), in one - metastasis of colorectal cancer. Of the total number of patients with solid tumors, patients with secondary pancreatic lesions were 1.3%.

Conclusions: Use of an immunocytochemistry method on material EUS-FNA allows to define a tumor histogenesis that is a necessary condition of the choice of treatment tactics.

Selected references
Rapid On Site Evaluation by cytotechnologist in Fine Needle Aspiration of the pancreas guided by endoscopic ultrasound. Performance in our institution

10. Pancreatico/biliary and Liver Cytology

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**Objectives:** Rapid On Site Evaluation (ROSE) improves the suitability and precision of fine needle aspiration (FNA) to obtain a diagnostic sample of pancreas guided by endoscopic ultrasound. In this study we aim to compare the performance of pancreatic FNA with and without ROSE by cytotechnologist in our institution.

**Material and methods:** A total of 207 patients with pancreatic FNA were diagnosed in our institution between 2010 and 2018.

We categorized the diagnoses into Positive, Negative and Insufficient, and compared FNA with and without ROSE.

**Results:** Out of the 207 cases, 62 (29.95%) were positive, 27 (13.04%) were insufficient and 118 (57.00%) were negative.

ROSE was performed in 64 cases, 30.92% of the total. In those cases, the number of insufficient FNAs was 4 (6.25%), while in cases without a cytotechnologist performing ROSE, the number of insufficient cases was 23 (16.08%), finding statistical significant differences in both groups.

The most frequent diagnosis of malignancy is adenocarcinoma (13.30%), while the overall rate of atypia/suspicious for malignancy is low, 7.34%. Four neuroendocrine tumors and one non-Hodgkin B lymphoma stand out as infrequent diagnoses in our series.

**Conclusions:** As has been previously reported in the literature, our experience of 8 years confirms that having a trained cytotechnologist for rapid in situ evaluation of pancreatic FNAs, supposes a drastic decrease in the rate of insufficient cases.
12. Urine Cytology
Celldetect could be the new staining platform technology for the detection of bladder cancer.

12. Urine Cytology
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Objectives: Cystoscopy and urine cytology especially the liquid-based cytology remain the standard for detection and monitoring bladder malignancy. However the detection of low-grade malignant cells in urine presents an issue, so the idea of using an additional non-invasive method could be beneficial for bladder-cancer patients. Celldetect is a novel staining platform technology that enables discrimination between malignant and benign cells using a plant extract and generic dyes. We performed the new technique in order to confirm its possible role in screening programs.

Material and methods: Voided urine was collected from 60 patients undergoing follow-up or presented with hematuria. The samples were processed by Thin Prep 2000 into two smears for each sample, that were stained by Celldetect technique and Papanicolaou method. The pap slides were classified according to the Paris reporting System and subsequently the Celldetect slides were categorized as positive or negative. Catheter urine were excluded. The results were compared to biopsy or cystoscopy reports.

Results: Out of 28 of the Celldetect-negative group 7(7/28) patients had suspicious results by liquid-based cytology (Category 3 atypical urothelial cells). The cystoscopy was negative for 5 of them but two of them had positive biopsies. All 32 Celldetect-positive patients had positive cystoscopy/biopsies. Nuclei of malignant cells stained purple-red by Celldetect method. Clusters of inflammatory cells from 9 cases featured purple-red nuclei. Umbrella or reactive cells of 5 patients exhibited pinkish staining.

Conclusions: the present data shows that Celldetect staining technique could be used as an adjunct test to urine cytology for diagnosis and particularly the follow-up of patients with history of urothelial carcinoma or hematuria. Celldetect is able to detect early stage tumors and raise the sensitivity for low-grade urothelial carcinomas.
Cytological changes in patients who underwent combined transurethral resection of the bladder tumor and Bacillus Calmette-Guérin therapy

12. Urine Cytology
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Objectives: Transurethral resection of the bladder tumor (TURBT) is an effective method of treating non-muscle invasive bladder cancer. Therefore, it is the most common procedure performed by urologists worldwide. Combining TURBT with BCG therapy (Bacillus Calmette-Guérin) significantly reduces recurrences and risk of invasion. During BCG therapy patients are required to do urine cytology; however, cytologists often have difficulty assessing specimens from those patients because of inflammatory changes and reactive changes in cells. The purpose of the study was to demonstrate the changes in cells in urine cytology obtained from patients after transurethral resection of bladder tumor combined with BCG therapy.

Material and methods: The study was retrospective, and the material for testing was archival specimens from patients who underwent combined TURBT/ BCG therapy. The study group consisted of 104 patients, 81 men and 23 women. Urine specimens were centrifuged, fixed in alcohol, stained with hematoxylin and eosin, and re-evaluated under the microscope.

Results: The study has shown that significant changes in cell nuclei occur repeatedly, which include the marginalization of chromatin, prominent nucleoli, fine chromatin, squamous cells without nuclei, sheets of metaplastic cells, and numerous granulocytes.

Conclusions: Cytologist ought to be able to distinguish BCG instigated changes from atypical cells and avoid classifying cells as AUC (atypical urothelial cells in accordance with The Paris System for Reporting Urinary Cytology).

Selected references
Cytological diagnosis of Human Papilloma Virus infection in male urethral brushing: A case report

12. Urine Cytology
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Objectives: Genital, urethral and anal HPV infections in men are the most common sexually transmitted diseases. Urethral HPV infections are relatively uncommon and their prevalence is still unknown. Urethral warts are preferentially found on the external meatus of the urethra or to the proximal urethra. Few reports exist regarding the potential role of urethral brushing cytology in the detection of HPV infection. A case of HPV infection diagnosed in urethral brushing is presented.

Material and methods: A 63-year old male with microhematuria was subjected to cystoscopy and found to have an ulcerated lesion in his urethra. An urethral scraping smear was taken with a cytobrush and fixed in ThinPrep specimen. The cytological smears prepared, were stained with the Papanicolaou stain. In addition, an HPV DNA test, was processed with the Roche cobas HPV test.

Results: The cytological examination of urethral brushing smear showed the presence of a few squamous cells with enlarged hyperchromatic nucleus, large sharply demarcated perinuclear clear zone and rare parakeratosis (miniature superficial squamous cells with dense orangeophilic or eosinophilic cytoplasm). The cytological diagnosis was consistent with HPV urethral infection. Furthermore, the urine’s HPV DNA test was positive for the low risk type 6 and the high risk type 18 HPV.

Conclusions: We wish to emphasize the contribution of urethral brushing cytology and HPV subtyping in the clinical detection of HPV infection in male, as well as its potential role on the patient’s follow up for malignant transformation.
Immunocytochemical expression of epidermal growth factor receptor (EGFR) and its correlation between low and high grade transitional cell carcinomas

12. Urine Cytology
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Objectives: Epidermal growth factor receptor (EGFR) is a member of the erb tyrosine kinase family reported to be overexpressed in a variety of solid malignancies and is considered as a marker of poor prognosis. A berrant EGFR expression has been documented in urothelial carcinoma and has proven useful as an ancillary diagnostic aid for urinary bladder tumors. This study was conducted to investigate the correlation of EGFR expression with grade of transitional cell carcinomas.

Material and methods: 36 patients, 21 with low grade and 15 with high grade urothelial carcinoma provided voided urine samples. The majority of cases were male. The average age of the patients was 68 years (41-86). The recommended Thin-Prep sample preparation protocol was followed for each urine specimen. The immunohistochemistry method with two steps polymer detection system (biotin-free) was performed on Thin-Prep cytologic smears for the detection of EGFR. Cytoplasmic immunostain of EGFR was considered as positive in >10% stained cells.

Results: Diffuse positive expression of EGFR was noted in 10 cases (66.6%) of high grade and 9 (42.8%) of low grade urothelial carcinomas.

Conclusions: Our study showed that EGFR positively is strongly associated with high-grade transitional cell carcinomas and has been proposed as a usefull marker for tumor grading. Meanwhile wide-spread expression of EGFR in high-grade urothelial cell neoplasms makes this molecule a good target for antigrowth factor or gene therapy.
OUR EXPERIENCE FROM A NOVEL URINE CYTOLOGY STAIN FOR BLADDER CANCER DETECTION

12. Urine Cytology

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Objectives:

CellDetect Bladder is a urinary marker for early detection of bladder cancer in cytological urine specimens. It is a histochemical stain combining color and morphology to distinguish between normal and neoplastic cells.

Material and methods: A total of 62 Voided urine samples were collected from patients with history of urine hematuria or suspicious of malignancy.

Samples were processed into smears using liquid based cytology (Cell solutions- Menarini Italy).

Each sample was stained, one slide with Papanicolaou stain and another slide was stained with the method of CellDetect (Zetiq Company).

Results: Cytologically 28 cases were negative for malignancy, 24 were diagnosed with atypia or atypia with inflammation and 16 were positive for malignancy. CellDetect stain (red/purple the nucleus of suspicious for malignancy cells) was positive in 12/20 (60%) of smears with atypia and in 4/4(100%) with atypia and inflammation, in 8/20 (40%) cytologically negative smears and in all 16(100%) positive for malignancy smears.

Conclusions: Our findings show that CellDetect improved the standard cytological diagnosis especially those with cytological suspicious diagnosis for malignancy.
Our institution experience with the Paris System for Reporting Urinary Cytology with review of discordant cases

12. Urine Cytology

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Objectives: The Paris System for Reporting Urinary Cytology (TPS) is designed to standardize the criteria and terminology used in urinary tract cytology reporting. The aim of this study was to evaluate the impact of implementing TPS and to analyse the correlation of high grade urothelial carcinoma histological diagnosis with its previous urine cytology samples, in order to assess its reproducibility in our department.

Material and methods: A total of 26 discordant urinary tract cytological specimens (Thin Prep technique), with follow-up of high grade urothelial carcinoma histological diagnosis over a 2-year period, were blindly reviewed and reclassified according to TPS criteria, by two pathologists, one resident and one senior. Several cytological parameters were evaluated and correlation with surgical follow-up diagnoses was performed using Chi-Square analysis.

Results: In a total of 248 urinary tract cytologies, discordance was found in a total of 26 (10.5%) cytologies. These cytological samples were selected for review. From the initial 25 false negative diagnosis, 7 (28%) were reclassified as non-diagnostic, 2 (8%) as atypical and 2 (8%) as high grade urothelial carcinoma. The false positive case was reclassified as negative for high grade urothelial carcinoma. The inter-rater reliability evaluated using Cohen’s Kappa was 0.705 when using TPS categories, and 0.877 when using non-diagnostic, benign or malignant/suspicious for malignancy categories.

Conclusions: Our institution experience with TPS showed a 63.7% sensitivity and 99.4% specificity for high grade urothelial carcinoma. False positives and false negatives were mostly related with contaminants (18.02%) and low cell viability (53.5%). Applied correctly TPS criteria led to a reduction in 44% of false negatives in this sample.
urine cytology and molecular detection of polyomavirus in plasma for patients with renal transplant

12. Urine Cytology

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Objectives: Urine cytology and quantitative real-time polymerase chain reaction (RT-PCR) are currently used in polyomavirus detection in patients with renal transplant. The cytopathic effect of polyomavirus in urinary tract epithelial cells is recognized microscopically by the presence of decoy cells in urine. The aim of the study is to measure the sensitivity and the significance of urine cytology of BKV in comparison with quantitative RT-PCR test that measures the viral load in plasma in kidney transplant patients at Sultan Qaboos University Hospital (SQUH).

Material and methods: This study was a retrospective study. Urine cytology data and quantitative RT-PCR results from 2014-2017 were retrieved from the track-care system at SQUH. Then data was analyzed using cross tabulation (2x2). A comparison was done to evaluate sensitivity, specificity, positive predictive value and negative predictive value of urine cytology.

Results: Data of 47 renal transplant patients was retrieved within the period from 2014 to 2017 at SQUH. Among those 29 were males and 18 females, with age ranging from 16 to 68 years old. A total of 89 urine cytology samples were analyzed in parallel with plasma quantitative RT-PCR. Compared with RT-PCR, the sensitivity, specificity, positive predictive value, and negative predictive value of urine cytology were 86.6%, 67.5%, 35.1% and 96.1% respectively as shown in the Table below. While the accuracy was 70.7%.

Conclusions: Urine cytology is useful screening method and noninvasive procedure to detect BK reactivation due to cost effectiveness, and short processing time required. Our study concluded that urine cytology is reliable and sensitive screening test to detect polyomavirus for early diagnosis of PVAN. It is compatible to plasma quantitative RT-PCR. Our results are comparable with what has been published in the literature. Suggested future study is to compare BK virus detection in urine samples by urine cytology and Real Time PCR.

Selected references

USEFULNESS OF THINPREP SYSTEM® IN DETERMINATION OF CYTOPATHIC VIRAL EFFECT (DECOY CELLS) IN NON-TRANSPLANTED PATIENTS

12. Urine Cytology

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Objectives: To determine the usefulness of Thinprep® system in the determination of viral cytopathic effect in non-transplanted patients studying the presence of Decoy cells in urinary samples, highlighting the benefits of this method for molecular biology techniques.

Material and methods: 1383 urinary cytologies from 1st April 2018 to 31st January 2019 were studied by optical microscopy in slides processed with Thinprep® system. In samples with Decoy cells in non-solid organ transplanted patients PCR was performed with RealStar® BKV PCR Kit to study BK virus and RealStar® JCV PCR Kit to study JC virus. Demographic data and comorbidities of selected patients were collected.

Results: 63 patients were positive for Decoy cells. The average age was 68.20 years. 56 (88.88%) were males and 7 (11.11%) were females. Diabetes Mellitus was present in 21 (33.33%) patients. Urothelial tumor in 25 (39.68%) patients. Other tumors in 8 (12.70) patients. Only 5 (7.94%) patients presented chronic renal failure. Correlation with PCR was 90.48% for polyomavirus, matching most of them with JC virus.

Conclusions: Thinprep® system allows to detect cytopathic effect of polyomaviruses in cytology and to perform molecular technics with the same specimen.

Our results show that Decoy cells are frequent in non-solid organ transplanted patients, specially on those with diabetes mellitus or with urothelial/non urothelial tumors.

Selected references

13. CNS Cytology
Cerebrospinal fluid cytology: infection arise in neoplastic context

13. CNS Cytology

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Objectives: Cryptococcus neoformans is the most common microorganism found in cerebrospinal fluid (CSF) cytology and causes life-threatening infections in immunocompromised hosts¹. As the leading cause of fungal meningoencephalitis, Cryptococcus species typically reach the CNS through hematologic dissemination of organisms derived from primary pulmonic infections².

Material and methods: We present a 64 years old male patient with confusional syndrome and aphasia with three days of evolution, in the context of follicular lymphoma, in treatment with rituximab. Cranial computerized tomography ruled out acute or expansive vascular lesions and after administration of contrast, there were no areas of anomalous, parenchymal or extra-axial neoplastic involvement. Lumbar puncture was performed.

Results: Cephalospinal fluid cytological smear revealed numerous polymorphonuclear cells, lymphocytes and monocytes with marked activation signals. Periodic Acid Schiff (PAS) technique showed multiple spherical extracellular structures, some of them located in the cytoplasm of macrophages. It was not observed B cell lymphocytes.

Conclusions: Microbiology confirmed the presence of this fungus in CSF. In this case, cytologic examination of the CSF rapidly exclude lymphoproliferative neoplastic involvement and oriented clinical treatment to infection disease cause by immunosupression due to chemotherapy. Direct microscopic examination of clinical specimens remains an inexpensive and rapid means of morphologic identification of cryptococcal species². In-hospital acute mortality from cryptococcal meningitis continues to remain high, ranging from 30-50%, even with antifungical therapy, so a proper diagnosis is imperious to be fast³.

Selected references

Double-Hit B cell Lymphoma infiltrating central nervous system. Cytologic features, immunocytochemistry characteristics and review of the literature.

13. CNS Cytology

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Objectives: We present the case of a woman with the diagnosis of non-Hodgkin B cell Lymphoma double-hit with infiltration to central nervous system and review of the literature.

Material and methods: Woman of 60 years old with diagnosis of non-Hodgkin B cell lymphoma double-hit (MYC rearrangement and t(14;18) (q32.3q21.3) with infiltration to multiple organs. Starts with intense headache and neurologist under the suspicion of probable involucrum, decide to make neuroimaging with MRI and CT, the results do not show image evidence associated to hematological disease in CNS. Lumbar puncture was performed and the cytological examination of cerebrospinal fluid was evaluated using hematoxylin and eosin stain, the slides showed tumor cells. Immunocytochemistry was performed with C20, CD5, BCL2, C-MYC and Ki 67. (See table 1)

Results: Table 1

Inmunocytochemistry.

CD20 POSITIVE

CD5 POSITIVE EN T REACTIVE LYMPHOCYTES

BCL2 POSITIVE (90%)

C-MYC POSITIVE (70%).

Ki 67 POSITIVE (85%).

Intrathecal therapy based on methotrexate was administered.

Conclusions: Despite the wide array of biomarkers that have been tested for the detection of CNS involvement in haematological malignancies, their use has not become widespread in clinical practice. Patients with CNS involvement by lymphoma have decreased overall survival. Double/triple hit lymphoma is recognised as a distinct entity within the heterogeneous group of high grade B-cell lymphomas, accounting for between 5 and 10% of cases of diffuse large B-cell lymphoma. Diagnosing double hit lymphoma requires identification of translocations of MYC and BCL2 and/or BCL6. In this presentation we demonstrated the cytological and immunocytochemistry characteristics of a double-hit B cell lymphoma infiltrating CNS, its outcome is very poor with R-CHOP and intensified regimens. Though cerebrospinal fluid seems to play a
critical role in the evaluation of the CNS, its role in the evaluation of CSF specimens remains a subject of controversy and its a challenge for the cytopathologist.

Selected references
14. Soft Tissue and Bone Cytology
Diagnosis of metastatic clear cell sarcoma in pleural effusion cytology

14. Soft Tissue and Bone Cytology

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Objectives: Clear cell sarcoma (CCS), also known as malignant melanoma of soft parts, is a rare type of sarcoma. It typically occurs in the tendons or aponeuroses of the distal extremities of young adults. Diagnosis of metastatic CCS using pleural effusion cytology is difficult, especially without history of primary site. There were only three cases reported in the literature. Herein, we described a case with metastatic CCS presenting as pleural effusion and cytologically masquerading malignant melanoma.

Material and methods: Immunohistochemistry (IHC) and reverse transcription polymerase chain reaction (RT-PCR) followed by sanger sequencing were used to diagnose the cytology and histopathology specimens.

Results: A 52-year-old male presented to our hospital with dyspnea for one month. Chest computed tomography revealed right pleural-based tumor with massive pleural effusion. Effusion cytology showed clusters of malignant cells with pleomorphic nuclei, prominent nucleoli and frequent mitoses. By IHC study of cell block, the tumor cells were immunoreactive to S-100. The initial diagnosis of pleural effusion is malignancy, suspicious for melanoma. The subsequent biopsy of the pleural-based tumor exhibited short fascicles and nests of rounded to plump ovoid epithelioid cells divided by delicate fibrous septa. The cytomorphology of tumor cells was similar to the cytology. Translocation between exon 7 of EWSR1 gene and exon 5 of ATF1 gene was detectable by RT-PCR followed by sanger sequencing. Tracing back to the past history, the patient had toe tumor diagnosed as "giant cell tumor" after excision four years ago at local clinic. Review of the toe tumor pathology was consequently done. The immunoprofiling was the same as the pleural tumor. The final diagnosis was metastatic clear cell sarcoma with type 2 EWSR1-ATF1 gene fusion.

Conclusions: Due to overlapped cytomorphology, histopathology and immunohistochemistry between melanoma and clear cell sarcoma, molecular tests such as RT-PCR or fluorescence in situ hybridization was essential for accurate diagnosis.

Selected references

Molecular study

Lane 2: EWSR1 exon7 - ATR1 exon5
Lane 3: EWSR1 exon7 - ATR1 exon6
Lane 4: EWSR1 exon7 - ATR1 exon5
Lane 5: EWSR1 exon7 - ATR1 exon6
Lane 6: EWSR1 exon7 - CEBPA exon7
Lane 7: GAPDH
Lane 8: FUS
Lane 9: FUS
Imprint cytological Features of Metastatic Atypical Meningioma Mimicking Soft Tissue Sarcoma: A Potential Diagnostic Pitfall

14. Soft Tissue and Bone Cytology
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Objectives: Atypical meningioma is defined as WHO grade II meningioma, with a high recurrence rate. Distant metastasis occurs rarely. Metastatic atypical meningioma can be often mistaken for soft tissue sarcomas due to its rarity and overlapping cytologic features with other soft tissue sarcomas. The aim of this report was to present cytomorphological features of metastatic atypical meningioma and literature review.

Material and methods: We report a case of a 74-year-old man who presented with the right flank pain for three months. He had a history of atypical meningioma 4 years ago. Chest CT scan revealed the right 8th rib mass. The right 8th rib mass (8.5 x 5.8 x 5.5 cm) was excised. Imprint cytology of the excised mass was performed. Cytological smears were stained with Papanicolaou and hematoxylin and eosin stains.

Results: The smears showed cohesive clusters or sheets of spindle or polygonal-shaped tumor cells. The tumor cells have ovoid to elongated nuclei with coarse chromatin and prominent nucleoli, with mild to moderate amount of eosinophilic cytoplasm. Broad cytoplasmic processes were present. Nuclear grooves were seen. Intranuclear pseudoinclusions were present. Whorl formation was found. Mitotic figures were occasionally observed. Necrotic debris was seen in the background. On immunohistochemical stain for the surgical specimen, the tumor cells were positive for EMA and cytokeratin (AE1/AE3), while the tumor cells were negative for S100 protein, CD34, smooth muscle actin, desmin, TLE-1, and STAT6. Histologic findings were compatible with metastatic atypical meningioma.

Conclusions: We present cytological features of an unusual case of metastatic atypical meningioma. The presence of well-formed whorls, grooved nuclei, and intranuclear pseudoinclusion would be helpful for differential diagnosis between metastatic atypical meningioma and cytologically mimicking soft tissue sarcomas.
Role of cytology and immunocytochemistry in the diagnosis of clinically suspicious malignant soft tissue neoplasms

14. Soft Tissue and Bone Cytology

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Objectives: Soft tissue neoplasms (STNs) are uncommon tumors and present with difficulties in diagnosis and classification. Assigning a pathologic type or grade to an individual sarcoma as a means of predicting clinical behavior is often difficult. Though FNAC is used as a first line investigation in primary evaluation tumors like breast, thyroid, lymph nodes and others, its use in diagnosis of STNs is still debated. The objective of this study was to assess the diagnostic role of cytology and immunocytochemistry (ICC) in the diagnosis and sub typing STNs.

Material and methods: The study included 60 cases of clinically suspected malignant STNs in which FNA was done from swellings, smears were prepared and sample was also procured for cell-block preparation. ICC was performed on cell block sections (51/60) and cytosmears (25/60) using commercially available antibodies to the following antigens: Vimentin, Desmin, SMA, S-100 protein, CD 34, Pan CK, LCA, Synaptophysin, CD 99, CD 56; Other special markers as FLI-1, TLE, Myo D1, and MDM2 etc

Results: The study included 60 cases of clinically suspected malignant STNs. Males were more commonly affected with maximum cases presenting in fifth to sixth decade. Histopathological follow-up was available in 49 cases with a cytological diagnostic accuracy of 93.33%, sensitivity of 95.12% and specificity of 75% to differentiate between benign and malignant. Spindle cell tumors (47%) accounted for the majority of the cases in our series. Sub-typing on cytomorphology alone was possible in 28.20% cases, which further increased up to 64.10% with application of ICC.

Conclusions: The present study concluded that FNAC can be used as a reliable diagnostic tool for preoperative triaging of benign and malignant STNs with excellent sensitivity, specificity, and accuracy, and if supplemented with ICC, sub-typing is possible in good number of cases.
15. Molecular and Immunocytochemistry
Comparative study between cytology and One Step Nucleic Acid Amplification (OSNA) in the analysis of lymph node metastases in papillary thyroid cancer

15. Molecular and Immunocytochemistry

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Objectives: Selective biopsy of sentinel node with intra-operative analysis using OSNA is a widely validated molecular technique for the study of lymph node metastases (LNM) in breast cancer. OSNA stratifies the result into 3 categories: negative (includes: absolute negative and isolated tumour cells -ITC-), micrometastasis -mM- and macrometastasis -MM-. Our group has proposed its use in papillary thyroid carcinoma (PTC). We compare OSNA and concomitant cytological imprinting in a wide series of lymph nodes from lymphadenectomies in patients with PTC.

Material and methods: 572 lymph nodes have been included. In all of them, a cytological study (CS) was performed by means of two imprints (stained with DiffQuick® and Papanicolaou). The entire lymph node was analysed by OSNA. Both cytological stains are compared between them and in relation to the molecular technique.

Results: 572 nodes corresponded to 42 patients (67% women, age 49±15y). Mean number of nodes per patient was 13.6±8. There were no significant differences in the method of cytology staining. CS detected 125 (21.8%) LNM, whereas OSNA found 187 (32.6%) [p<0.001]. In 500 (87.4%) nodes there was concordance between both techniques. Discordant samples: 5 were CS positive and OSNA negative; 67 CS negative and OSNA positive. In the first case, 4 of the 5 samples were classified as ITC by OSNA and only one was absolutely negative in the molecular study. 57 of the 67 nodes with negative CS and OSNA positive corresponded to mM, with the remaining 10 being MM.

Conclusions: OSNA technique shows high sensitivity to detect and quantify LNM in PTC. Results obtained with CS exclusively are excellent -especially for the MM- while preserving morphology. The greatest discordance of OSNA and cytology is observed in the group of mM (negative CS-OSNA positive) and ITC (positive CS-OSNA negative). In the latter case, the discrepancy is due to a mismatch of thresholds between a qualitative technique (CS) and a quantitative one (OSNA) than to a true discordance.
**Hedgehog signaling is controlled by Rac1 activity**

15. Molecular and Immunocytochemistry

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**Objectives:** Hedgehog (Hh) signaling critically regulates cell fate and proliferation in development and disease. The nuclear translocation of transcriptional factor family Glis (Gli1-3) is indispensable for Hh signaling activation; however, the mechanisms governing Glis nuclear translocation are not well understood.

**Material and methods:** We performed transfection and qRT-PCR, nuclear and cytoplasmic separation, western blots, lentivirus-mediated gene knockdown, CRISPR/CAS9-mediated gene knockout, adenovirus-mediated gene overexpression, co-immunoprecipitation, GST-pull down assays, dual-luciferase report assays and immunofluorescence to explore the molecular mechanisms *in vitro*, and performed H&E staining, immunohistochemistry, immunofluorescence, co-immunoprecipitation and western blots to study biological effects using transgenic mice *in vivo*.

**Results:** Here, we report that Glis translocation in response to Hh requires the small GTPase Rac1 inactivation. We show that upon binding of Hh to its receptor Ptch1, receptor Smo dissociates from Ptch1 and binds to Vav2, resulting in decreased phosphorylation level of Vav2, which further inactivates Rac1. The role of Rac1 depends on regulation of phosphorylation levels of kinesin II family member 3A (Kif3A) with shortened primary cilia length, which in turn regulates intraflagellar transport 88 (IFT88) protein degradation and subsequently dampens the SuFu-Glis complex formation in primary cilia, resulting in release of Glis from primary cilia and translocation into nucleus. Moreover, in *GFAP-Cre;SmoM2*+/− transgenic mice cerebellum that develop medulloblastomas caused by hyper-activation of Smo, Vav2 phosphorylation is inhibited and Rac1 is inactivated; genetic ablation of *Rac1* in the mice embryonic limb bud ectoderm (*Prx1-Cre;Rac1f/f*) promotes Hedgehog signaling activation through Glis translocation.

**Conclusions:** Together, these results uncover Rac1 inactivation and subsequent Glis translocation as a hitherto uncharacterized mechanism controlling Hedgehog signaling and may provide additional targets for therapeutic intervention of this important pathway.
Immunocytochemical phenotyping of canine lymphoma

15. Molecular and Immunocytochemistry
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Objectives: To investigate the value of methanol fixed cytospins prepared from fine-needle capillary (FNC) samples suspended in cell medium for immunocytochemical (ICC) phenotyping of canine lymphoma.

Material and methods: Ten untreated dogs with a cytomorphological or histomorphological diagnosis of peripheral lymphoma were included in the study. FNC samples of enlarged peripheral lymph nodes were immediately suspended in cell medium. Several cytospins were prepared in the next 12 to 24 hours. After fixation and storage of cytospins in methanol at 4°C for up to a month, ICC was performed using two different antibodies to CD3 (polyclonal rabbit anti-human CD3 antibody, DAKO; monoclonal mouse anti-human CD3 antibody, clone LN10, Novocastra) and to CD20 (polyclonal rabbit anti-human CD20 antibody, Invitrogen; monoclonal mouse anti-human CD20 antibody, clone L26, DakoCytomation). Cytospins from a hyperplastic canine lymph node served as the positive control.

Results: Based on ICC, and in concordance with the cytomorphological or histomorphological diagnosis, eight canine lymphomas were determined as B-cell lymphomas and two as T-cell lymphomas.

Both antibodies against CD3 proved to be suitable for labelling canine neoplastic and normal T lymphocytes, whereas only the rabbit anti-human CD20 polyclonal antibody (Invitrogen) reacted with canine neoplastic and normal B lymphocytes.

Conclusions: The cell medium used in this study enabled collection of all cells obtained by FNC biopsy, easy and safe transportation of samples and preparation of high quality cytospins for ICC. Methanol fixation preserved both antigens as well as the cell morphology of canine lymphoid cells.
Loss of mitochondrial DNA and death in U937 cells treated with gemcitabine

15. Molecular and Immunocytochemistry

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Objectives: Gemcitabine is an anticancer drug used to treat a wide range of tumors by inhibiting the extension reaction of DNA strand. Although some chemotherapeutics are known to induce nuclear DNA damages, few investigations address mitochondrial (mt) DNA. Since gemcitabine inhibits ribonuclease and results in the depletion of cellular pool of deoxyribonucleotides, this model predicts that the synthesis of mtDNA is reduced. In the present study, we address the relationship of loss of mtDNA and death in U937 cells treated with gemcitabine.

Material and methods: Human myeloid leukemia cell line, U937 cells were treated with various concentrations of gemcitabine. Number of mtDNA copy was quantified using Human mitochondrial DNA monitoring primer set by real time-PCR using nuclear DNA as a standard. Designed primers of mtDNA and nDNA were two targets for ND1 and ND5, and for SLCO2B1 and SERPINA1, respectively. Relative quantification of mtDNA is determined from difference in Ct values for mtDNA and nDNA. Cell viability was measured using MTT assay.

Results: When the U937 cells incubated with gemcitabine, the viability were decreased in a dose- and time-dependent manner. The number of mtDNA copy was significantly decrease compared to nDNA after addition of gemcitabine at 24 h, but not by nocodazole which is inhibitor of cell cycle. Although the treatment with gemcitabine quickly stopped the cell cycle, the reduction of copy number was distinct from cell cycle arrest.

Conclusions: From these results, we conclude that cell death was resulted in through the loss of mtDNA induce by gemcitabine. We believe the possibility that the target of mtDNA by anticancer drug become the new strategy for cancer treatment.
STAGES OF THE CYTOLOGIC DIAGNOSTIC EXAM IN PATIENTS WITH GASTRIC CANCER ON MATERIALS OF PERITONEAL WASHOUTS

15. Molecular and Immunocytochemistry
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Objectives: To develop an algorithm of a cytologic diagnostic exam in patients with gastric cancer on materials of peritoneal washouts.

Material and methods: The analysis of 365 surgical materials in patients with gastric cancer, who treated in the department of high-tech surgery from June 2016 to December 2018 was performed. The average age of the patients was 64 years (32 to 86 years), of them 178 women and 187 men. Cytological preparations were made by a liquid technique on Cyto-Tek cyto centrifuge. The immunocytochemical study was performed with following monoclonal antibodies panel: Ber-EP4, CEA, EMA, CK7, CK20.

Results: As a result of 365 study, free tumour cells (CY+) were found in 133 (36%) cases. In 229 (63%) of patients, no tumour cells in peritoneal washout were found (CY-), three material (1%) were not informative, due to pronounced degeneration of cellular elements. The group of positive patients was divided according to the clinical importance on CY+ low and CY+ high, on the basis of the following signs: signet ring cells, ≥50 cancer cells, and ≥5 cell clusters. The groups are allocated by results the immunocytochemical study. The specificity of cytological diagnosis was 98%, and the sensitivity was 96%, the overall accuracy of the method reached 98%.

Conclusions: The developed algorithm allows to reveal diagnostic significant number of free tumors cells with high precision and the minimum economic expenses.

Selected references
16. Training and Teaching
NUCLEAR AND CYTOPLASMATIC ABNORMALITIES AS A TOOLS FOR EVALUATING GENOMIC INSTABILITY

16. Training and Teaching
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Objectives: A variety of pollutants (physical and chemical agents) as a cancerogens can cause the nuclear and cytoplasmatic abnormalities (NCA) in the human cells. Lymphocytes, with nuclear or cytoplasmatic abnormalities, are easily detected and suitable cells as biomarkers in human biomonitoring. Also, the lymphocytes are the most sensitive cells to ionizing radiation, as physical genotoxic agents. These types of abnormalities have been associated with mitotic and chromosomal instability. The occurrence of chromosomal damage can be evaluated by counting micronuclei and other NCA are indicative of apoptosis. The aim of this study was to evaluate and confirm the NCA in lymphocytes as a biomarker of DNA miss repair complexes and DNA damage on medical workers exposed to ionizing radiation.

Material and methods: Using the micronucleus (MN) assay as a test in genotoxicology we analyzed the lymphocyte with their abnormalities as a formation of micronucleus, nucleoplasmic bridges, nuclear buds, vacuolated nucleus, karyolysis and karyorrhexis, vacuolated cytoplasm, binucleated cells, apoptotic cells etc.

Results: Cytological analyses confirmed that the mean of MN frequencies and other NCA are much more present in the subjects with long time exposere than in other subjects. The abnormalities corelated with their age, sex, genetic constitution, adverse habits such as tobacco smoking and alcohol consumption. Various factors, including cell culture conditions (medium, incubation time) and population variables such as dietary habits may cause more or less NCA.

Conclusions: The present paper reviews the origin, fate and scoring criteria of MN that serves as a biomarker of exposure to genetic toxins and risk of cancer. We recommend using NCA in peripheral blood lymphocytes as a tools for evaluating genomic instability. This study suggests the needs for application of other cytogenetic techniques or trenings which allow easy detection of the rate of chromosome rearrangements and the origin of the chromosome instability.
Objectives: Non-gynaecological cytology is a quick, easy and reliable method for the diagnosis or exclusion of malignancy. Over the last few years in our institution we have noted that the number of non-gynaecology cytology specimens has been steadily decreasing with a paradoxical increase in the number of tissue biopsies received.

Material and methods: We wanted to know how often and in which specimen types ancillary techniques to further the diagnosis were employed with what success. We conducted an audit to investigate the use of immunohistochemistry, either on cytological material and cell blocks, and the correlation with subsequent tissue diagnosis, where available.

Results: 2889 cases from 2018 were interrogated mainly for the use of immunohistochemistry; referral cases and those without immunohistochemistry were excluded. In <5% of cases immunohistochemical studies were requested, the vast majority on fluids. 69 cases went on to have a tissue biopsy: 61 cases were concordant; 6 cases were found to be discordant with the subsequent histology.

Conclusions: Little use of immunohistochemistry in our institution on limited types of samples, mainly secondary to all FNA samples being received as air-dried smears with no additional material available.
17. Digital Cytology
Implementation of the Telecytopathology for the rapid on site evaluation of fine needle aspiration performed in the Endoscopy Suit

17. Digital Cytology
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Objectives: We present our experience in the implementation of telecytopathology (TCP) for the rapid on-site evaluation (ROSE) in the endoscopy unit and comparing the diagnostic concordance between ROSE and the definitive cytological diagnosis before and after using TCP.

Material and methods: Using a streaming system (Huawei TE30) in real time via Ethernet, an experienced cytotechnologist shows the most representative smear fields stained with Diff-Quik through the microscope (LeicaDM750) with a camera (Leica ICC50W) attached and connected to a screen (ThinkvisionT22i-10, Lenovo) from the ecoendoscopy suit to a monitor located at the cytopathology department where the cytopathologist confirms the preliminary diagnosis.

Results: The TCP allows the cytology visualization by the cytotechnologist and the cytopathologist each on their monitor, as well as being able to communicate each other for the selection of fields to observe to assess the sample adequacy. Once the rapid diagnosis is made, the cytotechnologist communicates the result to the endoscopist.

From December 2018 to nowadays, 129 samples with TCP, 104 (80.6%) EBUS and 25 (19.4%) EUS have been analyzed and compared with 128 samples evaluated with conventional microscopy (CM), 102 (79.7%) EBUS and 26 (20.3%) EUS. The preliminary diagnosis of negative for malignant cells (NMG), positive for malignant cells (PMC), adequate sample/differed diagnosis (AD) and pending cell block (PCB) was performed in 67.4%, 28.7%, 0.8% and 0.8% by TCP and in 67.2%, 15.6%, 2.4% by CM. The overall concordance between the preliminary and final diagnosis was 98.4% by TCP and 96.9% by CM (Fisher test p = 0.447).

Conclusions: The telecytopathology is an useful method for ROSE in both EBUS and EUS obtaining results comparable to conventional ROSE without TCP. The installation and management of the system is simple and makes possible on-site evaluations at remote locations, it also allows cytopathologists to manage their time more efficiently.
Is Digital Cytology a viable method for screening ThinPrep® Pap Test Slides?

17. Digital Cytology
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Objectives: While whole slide imaging is becoming more common in pathology, it is still primarily being used for research and teaching purposes in cytology. Several challenges make existing whole slide imaging solutions impractical for current clinical use with cytology. The 3-dimensional nature of cytology slides increases the scan time, storage requirements and complexity of scanning. Screening also takes longer on a whole slide image (WSI) compared to screening using a microscope. Despite these challenges, there are many benefits such as the potential for remote review, consultation, image analysis algorithm development, education and training. This study compares the accuracy and efficiency for slides read digitally using a gallery of images + WSI versus with a microscope.

Material and methods: 158 ThinPrep® Pap test glass slides (95 NILM, 63 ASCUS+) with adjudicated diagnoses were digitally scanned at 40x magnification (0.25μm / pixel) with multiple z-planes and merged into a single focal layer. For each slide, a gallery of representative cell images was created and presented with the corresponding whole slide image. Full review of the WSI was not required. Glass slides were manually read using a standard light microscope. Six cytotechnologists completed both arms of the study.

Results: Preliminary results show that accuracy screening manual slides was 91.9% and screening digital slides was 91.5%. On average, time to make a diagnosis on a glass slide was 202 seconds and time to make a digital diagnosis was 74 seconds.

Conclusions: Preliminary results from this pilot study suggest it may be possible to review gynecological cytology slides efficiently and accurately using a digital platform with a gallery of cell images from a merged focus WSI. While additional research is needed, efficiency gains may impact workload limits and be of future benefit as the cytotechnologist workforce continues to decline.
18. Others
Cytological diagnosis of malignant melanoma on subcutaneous lesion of the inner femoral surface by FNA. Report of a case.

18. Others
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Objectives: It is more usual to first diagnose a malignant melanoma by a dermal biopsy. We report a case of a patient who presented with a subcutaneous lesion in the inner femoral surface and diagnose as malignant melanoma by FNA.

Material and methods: A 62-years old man presented to the Surgery Department for a palpable subcutaneous lesion in the inner femoral surface. He underwent FNA. The sample was processed during the conventional method and the liquid-fase cytology technique using the Papanicolaou stain and May Grumwal Giemsa. Microscopy examination revealed many neoplasmatic cells. These cells appear single or in small clusters, they vary in size and shape from round to fusiform. The nuclei are large, round to oval and they are slightly hyperchromatic with irregularly distributed chromatin. Nucleolai are single and usually prominent. A few of cells had intranuclear cytoplasmic pseudoinclusions and rare of them were binucleated. The immunocytochemical expression was positive for HMB 45 in a small number of neoplasmatic cells and was negative for SMA, desmin, laminin.

Results: cell morphology and immunocytochemical expression proved to be helpful for diagnosis.

Conclusions: It is possible to diagnose a malignanr melanoma cytologically from an FNA sample of a subcutaneous lesion and to provide useful informations to the clinicians for further management of the patient.
FocalPoint GS System for anal cytology and relation of high risk results of anal cytology in women with cervical HPV infection

18. Others
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Objectives: To analyse the adequacy of results obtained by automated analysis through FocalPoint GS Imaging System (FPGS) (BD, Burlington, NC) for anal samples and to evaluate concomitant presence of abnormalities of anal and cervical cytology.

Material and methods: After written consent, anal samples (liquid-base) were collected from the anal canal of women with history of cervical cytology positive for HPV, during their consultation for colposcopy. SurePath samples were analyzed by the FocalPoint GS Imaging System (FPGS) (BD, Burlington, NC), which selects fifteen suspected areas and classifies them in 5 quintils according to probability of dysplasia.

Results: Forty women participated in this study, with a median age of 40 y/o (range 23-57). Previous cervical cytology showed HSIL in 10 (25%), ASC-H in 22 (55%), ASCUS in 7 (15,5%) and LSIL en 1 (2,5%). Anal cytology was successful in 33 cases and the upper quintil (more risk of dysplasia) was diagnosed in 4 (12,5%), 2nd quintil in 6 (18,2%), 3rd quintil in 7 (21,2%), 4th quintil in 6 (18,2%) and 5th quintil in 10 (30,3%). Of 30 women with high risk results in cervical cytology (HSIL and ASC-H), 8 also had high risk alterations on anal cytology (upper 2 quintils) (p=0,62). There was no relation between higher risk results on anal cytology and practice of anal intercourse.

Conclusions: The use of FocalPoint GS system for anal cytology was feasible, and in our study we did not find a relation of higher risk in anal cytology compared to higher risk of cervical cytology.
Reliability of esophageal brush cytology in the follow-up of eosinophilic esophagitis

18. Others

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**Objectives:** To explore the reliability and feasibility of esophageal brush cytology as a less aggressive diagnostic test alternative to biopsy in the follow-up of patients with eosinophilic esophagitis (EoE).

**Material and methods:** Esophageal brushing liquid-base cytology was performed in addition to manometry, endoscopic examination and esophageal biopsies in 53 patients (95\% male, age range 10-62 years, average age 43) with a diagnosis of EoE as established by most recent consensus guidelines. In cytologic samples the degree of inflammation and the eosinophil count, including total number per sample and number per high-power field (hpf), were assessed.

**Results:** Among the group of 22 patients (41.5\%) that fulfilled histological criteria of EoE in the follow-up biopsy, eosinophils were identified in brushing cytology in 9 (41\%). These cases accounted for 90\% of the total brushing cytologies in which eosinophils were identified (10 cases). The total number of eosinophils per sample in these cytologies was ≥ 15 in 8 cases (88.9\%, 6 of them (75\%) with counts >100/sample ) and <15 in 1 (11.1\%). The average number of eosinophils/hpf was of 15.5 (range 1-51); the count was ≥ 15 in 4 (44.4\%) and <15 in 5 cases (55.6\%). 49 patients (92.5\%) show esophageal inflammation in cytology, 32 (60.4\%) slight and 17 (32.1\%) moderate. Five patients (8.8\%) show no inflammation. In four cases (7\%) candidiasis, not detected by histology, was found.

**Conclusions:** The presence of eosinophils in esophageal brushing cytology was highly suggestive of EoE in our study, with a specificity of 96.77\% (90.55\%-100\%, CI 95\%) and a positive predictive value of 90\% (71.41-100\%, CI 95\%). However, sensitivity was low (40.91\% [20.36\%-61.45\%, CI 95\%]). Four-quadrant brushing sampling analogous to endoscopic biopsies could improve the sensitivity of this procedure. More studies are needed in order to establish, if possible, counting criteria for the cytological evaluation of this entity.

**Selected references**