

decreased from 60 and 35 to 40 and 20 in fertile and infertile groups after vitrification, respectively. It is concluded that vitrification impairs fertilizing capacity of sperm in fertile and infertile men.

P 275

SECONDARY TUMORS TO THE PANCREAS. STUDY OF 17 CASES WITH FINE NEEDLE ASPIRATION CYTOLOGY

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Introduction: Secondary tumors to the pancreas are unusual and in most cases part of an advanced metastatic disease. They account for 3-16% of all pancreatic malignancies. They present as solitary or multiple lesions, sometimes with clinical and radiological features that can be confused with a primary pancreatic adenocarcinoma. Fine needle aspiration cytology (FNAC) is a quick, low-expensive technique that allows a rapid identification of metastatic carcinoma and the patient's management in pre-surgical diagnostic phase.

Purpose: To report the clinical and cytological findings of secondary tumors to the pancreas observed in the Department of Pathology of Verona University.

Methods: Between January 1990 and December 2004, we studied 17 secondary tumors to the pancreas diagnosed by FNAC.

Summary: The mean age of patients was 62 years. 10 patients were males (59%) and 7 females (41%). In 16 cases (94%) FNAC was performed with ultra-sonography guide on outpatients and in 1 case FNAC was performed during abdominal laparotomy for suspected pancreatic adenocarcinoma.. The metastatic lesion involved the head of the pancreas in 41% of cases (7/17) and the body-tail in 35% (6/17). In 24% of cases (4/17) the anatomic site of pancreatic metastasis was not available. Radiological imaging showed in 94% of cases (16/17) a suspicious solitary mass and in a single case multiple lesions; the diameter of suspicious nodules ranged from 2 to 8 cm. The histotype of metastasis was renal cell carcinoma, classic type: 35%(6/17), large bowel carcinoma: 18%(3/17), gastric carcinoma 6%(1/17), breast carcinoma: 4%(1/17), prostate carcinoma: 6%(1/17), carcinoma of the lung:6%(1/17). In 24% of cases the primitive site of neoplasia could not be assessed. The clinical history was available in all cases of renal cell carcinoma with a mean interval of 13 years between the primitive renal carcinoma and the pancreatic metastasis.

Conclusion: Most secondary tumors to the pancreas occur as a solitary lesion, both on the head and tail of pancreas, simulating a primary pancreatic carcinoma. The most frequent metastatic tumor to the pancreas is renal cell carcinoma, often many years after the primary renal tumor. FNAC allows a distinction between primary pancreatic cancer and secondary tumors and represents an important diagnostic tool in the follow up of oncologic patients.

P 276

VIRAL MARKERS IN CYTOLOGIC AND HISTOPATHOLOGIC PREPARATIONS OF CERVICAL LESIONS

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Consistent epidemiologic evidence had demonstrated that human papillomavirus (HPV) infection plays a central role in

the development of cervical cancer precursors and invasive cervical cancers. Over 20 types of HPV are associated with cervical intraepithelial lesions and cervical carcinomas. The purpose of our study was to analyze viral markers expression in cervical lesions. 105 Pap smears and 84 biopsies were selected as they exhibited viral cytopathic effects. We performed immunohistochemical staining for HPV on both types of preparations, in cases with SIL diagnosis. Cytologic aspects were: koilocytosis associated with dyskeratocytes, and parabasal cells (\pm atypia), low grade squamous intraepithelial lesion (L-SIL) and high grade squamous intraepithelial lesion (HG-SIL). Histology revealed chronic cervicitis, condyloma acuminatum, L-SIL and H-SIL. HPV was detected in 61.1 % of smears, exhibiting a double localisation: nuclear and cytoplasmic and in 65.71 % of biopsies, with a nuclear localisation and an enhanced expression within superficial layers of condylomas. Concluding, HPV may be evidenced in cytologic and histologic preparations from cervical lesions, the negative preparations being attributed to (co)existence of other infectious agents or to the limited sensitivity of the immunohistochemical method.

P 277

COMPARATIVE STUDY OF CONVENTIONAL AND LIQUID-BASED CYTOLOGY IN A POPULATION WITH OPPORTUNISTIC CYTOLOGICAL SCREENING

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Background: In the last decade the conventional Pap smear (CPS) has frequently been replaced by the liquid-based cervical cytology (LBC) in the routine clinical practice due to improved technical quality and increased efficacy in detecting squamous intraepithelial lesions (SIL) and cervical cancer by LBC.

Aims: The purpose of this study is to report the preliminary results of the performances of LBC versus those of CPS, in a country with an opportunistic cytological screening and a high incidence of cervical carcinoma.

Methods: This comparative retrospective study is an initial part of a wider project, anticipated to assess the validity of the new method and its impact on clinical practice. It includes two groups of women referred to a tertiary centre for gynaecologic care, in different six-month periods. In each group, the Pap smear was taken by one of the methods. Data from unselected, consecutive series of 3,261 LBC tests and of 4,102 CPSs were analysed. Cytological diagnoses were classified using the Bethesda system. The differences in specimen adequacy and the rates of atypical squamous cells (ASC) category between the two groups were compared using χ^2 test. The histological correlation with histopathological findings could be established in 901 patients with the LBC and in 1,408 with the CPSs. Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and diagnostic accuracy of the findings were evaluated as well.

Results: The improvement in the specimen adequacy for LBC was documented by the more than fivefold reduction of the proportion of unsatisfactory specimens (0.4% for LBC vs. 2.1% for CPS, $P < 0.001$). The rate of χ^2 satisfactory but limited by χ^2 was also reduced by 40% for LBC ($P < 0.001$). However, the rate of ASC category was similar in both methods (0.9% for LBC vs. 1.2% for CPS). The performances of LBC for detection of cervical SIL or cancer lesions versus those of CPS, such as sensitivity (94% vs. 79%), PPV (85% vs. 69%) and NPV (90% vs. 66%) were significantly different ($P < 0.001$). There was no statistically significant difference in

specificity between the methods (65% vs. 62%, $P < 0.15$). Ultimately, the diagnostic accuracy was higher in LBC than in CPS (84% vs. 73%, $P < 0.001$).

Conclusions: Our preliminary results suggest that the implementation of LBC significantly improved the technical quality of the smears and resulted in statistically significant improvement of the diagnostic value of the Pap test.

P 278

IMMUNOPHENOTYPING OF CSF SPECIMENS BY SLIDE BASED LASER SCANNING CYTOMETRY JASTANIA Raid, BOERNER Scott, GEDDIE William

Background:

In cerebrospinal fluid (CSF) cytology analysis of cell surface markers is frequently required for diagnosis or subtyping of lymphoproliferative disorders, but insufficient cells may be present for flow cytometry. Laser scanning cytometry provides a simplified and inexpensive means of immunophenotyping paucicellular samples. We have assessed our experience with this method in CSF samples requiring immunophenotyping over an 18-month period.

Materials and Methods: Cyto centrifugation and rapid Giemsa stain were used for immediate assessment of 521 consecutive CSF samples. Slide based immunophenotyping by Clatch's method was attempted in 60 samples that showed some increase in lymphoid cells. Twenty-five cases had a history of previous hematolymphoid malignancy, and in 12 HIV positive patients neurologic decompensation led to suspicion of CNS lymphoma. 37 samples contained 10,000 cells or less, with the remainder ranging up to 150,000 cells total. In 12 samples with over 60,000 cells a full immunophenotyping panel of 20 antibodies was performed. In the samples with fewer cells at least kappa and lambda light chains and CD19 were initially assessed. By repetitive re-staining of the unfixed cells on the slide more surface antigens could be documented. After fixation and permeabilization intracellular markers could be assessed. All analyses were performed in the cytology laboratory using an LSC (Laser Scanning Cytometer - Compucyte®)

Results: 43 cases showed a reactive immunophenotype and a diagnosis of negative or chronic inflammation was rendered. The 17 remaining cases were diagnosed as suspicious for lymphoma (7) or malignant lymphoma (10). In 3/7 suspicious cases the LSC result was supportive of, but not conclusive for, lymphoma. In the remainder it was not contributory. In the 10 definite lymphoma cases the LSC immunophenotype confirmed B-cell clonality or an abnormal T-cell phenotype consistent with the clinical and morphologic diagnosis.

Conclusions: Slide based cytometry using Clatch's method is useful in the evaluation of CSF samples containing a hematolymphoid population suspicious for lymphoma, allowing specific reactive or malignant diagnoses in a number of cases containing inadequate cells for flow cytometry.

P 279

UTILITY OF CYTOLOGIC IMPRINTS IN INTRAOPERATIVE DIAGNOSIS

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Intraoperative diagnosis has been traditionally based on frozen histologic sections because provide architectural information, cytologic smears allow us to evaluate morphology on well preserved cells, without the well-known artifacts associated with freezing. Both methods thus complement each other. However, utilizing only the cytologic technique is possible if performed by experienced hands in special situations.

Material and Methods

We study 416 consecutive specimens submitted for intraoperative biopsies over a two-year period, Imprint cytologies were done on all specimens, and frozen sections were performed on 384 of them. The diagnoses obtained from both methods were recorded and compared with the final diagnosis yielded by the definitive histological sections.

Results

9 of 416 cases (2.2%) were misdiagnosed by IC, and there were diagnostic errors in 8 / 384 (2.1%) cases by frozen sections. Diagnostic accuracy was 97.8% for imprint cytology and 97.9% for frozen sections. Six cases of 416 (1.4%) were misdiagnosed by both imprint cytology and the frozen section. The accuracy rate of both methods used jointly was 98.5%.

Conclusions

Intraoperative imprint cytology is a quick, easy-to-perform, and low-cost technique that provides morphological details on intact cells without the artifacts resulting from freezing. It is therefore a highly valuable adjunct to the frozen section.

The benefits obtained from the use of imprint cytology are unquestionable, particularly under the following situations and considerations: 1) assessment of small specimens; 2) necrotic lesions; 3) to prevent cryostat contamination in infectious processes; 4) to facilitate identification of micro-organisms; 5) no special equipment is required; 6) to preserve material for special studies (ultrastructure, immunohistochemistry, etc.); 7) for the assessment of some specimens for which it is not possible to perform frozen sections, such as bone specimens; 8) it shows cytogenetic characteristics (e.g. parathyroid tissue, cytological type of lymphoma); and 9) experience is gained in cytology.

P 280

THE IMPORTANCE OF P-16'S EXPRESSION CORRELATED WITH KI-67 IN ESOPHAGEAL INTRAEPITHELIAL HPV-INDUCED SQUAMOUS LESION

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BACKGROUND The esophageal malpighian epithelium may be the siege of HPV infections, thus determining intraepithelial squamous lesions, similar to those found at the level of the uterine cervix. P-16 is a cyclin-dependent kinases inhibitor and a key regulator of the G1-S, and check point in the cell cycle. The over-expression of P-16 marker has been observed in CIN and invasive carcinomas, associated with high-risk hPV types.. Ki-67 is a nuclear antigen expressed in g1-S-G2-M phases, also being an aggressivity indicator.

DESIGN A number of 31 esophageal biopsies and 8 esophagectomy specimen material was formalin-fixed, paraffin embedded , HE stained and analysed immunohistochemically using specific antibodies against Ki-67 and P-16 by ABC immunoperoxidase procedure. Out of those 31 biopsies, 7 were showing HPV-induced low grade intraepithelial squamous lesion ,and 18 were showing high grade intraepithelial squamous lesions (CIN II-6, CIN III-9, CIS-3) and 6 cases of invasive carcinoma.

RESULTS :Our study demonstrates that HSIL-s have uniform and diffuse nuclear staining (Ki-67+) 100% (24 cases) and (P-16+) 80% (19 cases) throughout all cell layers of the lesion. None of our 7 LSIL cases showed (+) Ki-67 in the superficial layer. In the lower third of the epithelium our LSIL cases showed positive staining for both P-16 and Ki-67 but the upper third of the epithelium showed no such staining for either of them.

CONCLUSION Although the classical morphological features are the main diagnostic mean in the case of HPV