Pulmonary adenocarcinoma histology

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Modified WHO classification of adenocarcinomas

This review will focus on morphologic diagnosis of adenocarcinoma, pointing to areas where improvements for diagnosis are possible, show the prognostic relevance of the different patterns. Reviews on how to handle specimens for predictive analysis of NSCLC have recently been published (1,2).

Interestingly, the WHO classification of lung cancer is based on resection specimen examination, while only about 10-20% of patients with lung cancer are amenable to surgery. In most cases the diagnosis of lung cancer is performed on cytology and small biopsy samples obtained by various means (2). The recent multidisciplinary classification of lung adenocarcinoma extends the classification to such small samples (3). In the morphological classification, primarily of surgically resected tumors, 5 invasive tumor patterns are recognised: adenocarcinoma with lepidic growth pattern, acinar pattern, papillary pattern, micropapillary pattern, and solid pattern with mucin. When the lesion is purely lepidic and 3 cm or less in diameter, adenocarcinoma in situ (AIS) is diagnosed. Such a lesion is considered a non-invasive carcinoma with a 100% five year survival rate.

Distinction of well differentiated adenocarcinomas from well differentiated squamous cell carcinomas is generally good, but less easy when the carcinomas are poorly differentiated. Given that most cases of pulmonary adenocarcinoma show mixed morphology in relation to the five major histological patterns, evidence shows that a predominant pattern can be reproducibly identified with high concordance amongst pathologists in resection specimens (4). However, in this study partly designed to uncover differences in interpretation, recognition of the adenocarcinoma in situ pattern is more problematic, though kappa values are fair. This area may be improved by having more precise definitions and subsequent better education on interpretation of existing terminology, and/or additional markers of invasion. The prime confounder is using a two dimensional histological section to diagnose a lesion with a complex three dimensional architecture, an issue that is not adequately addressed within current definitions. In addition, also other factors contributed to the variation in interpretation: (I) a stromal component was considered as tumour-related stroma with fibroblasts (also called desmoplastic stroma), while others considered the same feature benign scarring/fibroelastosis; (II) the presence of elastin was variably weighed as representing native alveolar wall by some pathologists but not by others; (III) inflammation in alveolar walls implied invasive disease to some; (IV) although there was good agreement between pathologists in cases with a prominent micropapillary component, there was variation in interpretation between what some interpreted as focal micropapillary component and tangential cutting of both lepidic and true papillary structures; (V) some pathologists interpreted a mucinous lepidic component as being invasive, based upon the reasonable assumption that elsewhere in the tumour an invasive component with scarring is highly likely, while others interpreted the image in itself as non-invasive. It is therefore notable that much of the interobserver variation stems from interpretation based on operator experience and opinion and improved definitions and better education on their usage are required to reduce interobserver variability. Although within one educational setting high agreement is feasible (5-7), differences in interpretation do still occur.

Resection artefact and classification of adenocarcinoma

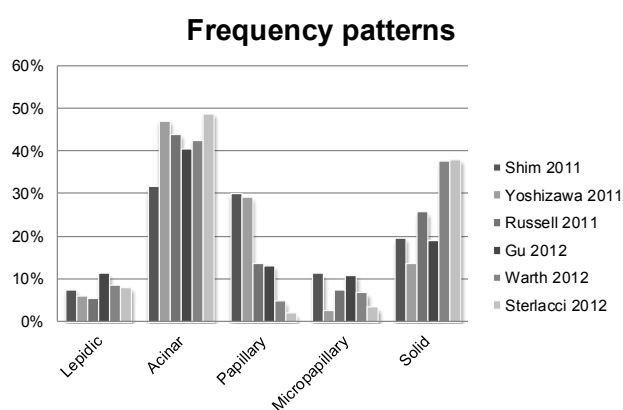Surgical removal and pathological handing of resected lung

Frequency patterns

Figure 1 The frequency (%) of histologic patterns described in different studies

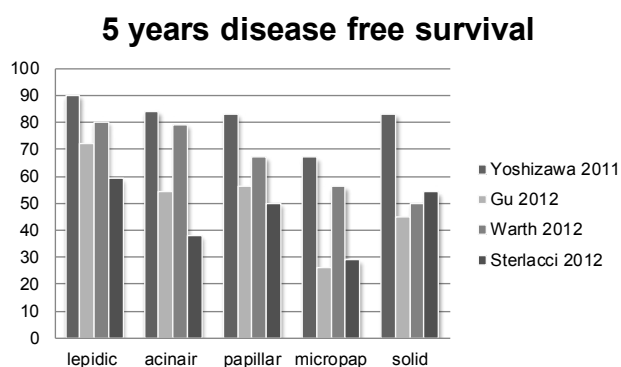## 5 years disease free survival

Figure 2 The 5 years disease free survival (%) is shown for the 5 major histologic patterns. Data of Sterlacci et al. were kindly provided by the authors

tissue may have a compressive effect on the alveolar lung tissue, distorting its architecture. To facilitate a pulmonary resection and improve access in the thoracic cavity, the lung is usually deflated during segmentectomy, lobectomy and pneumonectomy. Especially during video-assisted thoracoscopic surgery (VATS) procedures, visualization of the surgical field is seriously hampered by a ventilated lung. The effect of surgical atelectasis on morphology has not been examined in depth with respect to lung adenocarcinomas.

In histologic sections the deflation effect of surgical atelectasis induces 'lack of empty space' equivalent to air *in vivo*. If the open biopsy or resection specimen is not subsequently perfusion fixed, either through the bronchus, or by injection through the visceral pleura, this artefact

remains, as is common in daily practice. This surgical atelectasis is a prime confounder in reproducibility of adenocarcinomas subtypes, as this gives rise to a misleading morphologic appearance of a papillary pattern, while actually a lepidic pattern is present (8). It should be recognised that, in specimens where the alveolar tissues are compressed or deflated, a pseudopapillary pattern may be evident where tumor cells appear to line central fibrovascular cores. These cores lacking a myofibroblastic stroma but showing discontinuous elastic fibres and an obvious lepidic pattern at the edge are clues to the correct diagnosis of AIS. In addition, pigmented alveolar macrophages in acinar lumen point to a re-existing alveolar structure (9).

Frequency of patterns and prognosis

Since the multidisciplinary classification of pulmonary adenocarcinoma several studies examined the frequency of occurrence of the patterns and also the relation with prognosis either by 5 years overall survival or by 5 years disease free survival. So far 6 studies described the frequency of occurrence of the different patterns (10-15), see *Figure 1*. The most frequently diagnosed pattern is the acinar pattern. The second is the solid by some and papillary pattern by others. The biggest range in frequency is noted for papillary, micropapillary and solid pattern. This variation may be explained by differences in interpretation of the patterns.

Grouping the patterns into well differentiated (lepidic), moderately differentiated (acinar and papillar) and poorly differentiated (micropapillar and solid) reveals for these studies a prognostic difference. For the 5 years disease free survival the data of 4 studies are shown in *Figure 2* (10,13,15).

Remarkably for all patterns there is at least a difference in prognosis of 30% between these 4 studies. Moreover, the paper of Yoshisawa *et al.* shows a high DFS for all patterns being the lowest for (67%) for the micropapillary pattern (15), in contrast to the others. Although there may be several reasons to explain these prognostic differences, one of these may be that a high fraction of cases are called predominant papillary or acinar carcinoma which might be called lepidic by other pathologists, introducing a kind of stage shift with improved prognosis.

Biomarkers for diagnosis of adenocarcinoma

As only 10-20% of lung cancers are amenable for resection,

more diagnosis of lung cancer are made on biopsies and cytology specimen. These are frequently obtained in cases with higher stage. In smaller samples, the chance is higher that a well differentiated area is lacking compared to resection specimen, explaining the difficulty of distinguishing adenocarcinoma from squamous cell carcinoma in biopsies (2). This morphological grey zone in biopsies is called non-small cell lung carcinoma nototherwise specified (NSCLC-NOS) and may comprise 20-40% of the biopsy specimen (3). The help of additional stains reduces this number to 4-6%, with NSCLC-NOS 'favour adenocarcinoma' or 'favour squamous cell carcinoma' as useful terms in daily practice. The stains in this context are TTF1, Napsin A and mucin for adeno-differentiation and p63/p40 for squamous cell carcinoma (16-27). In the application of p63 only a strong staining in >80% of the basally located tumor nuclei points to squamous differentiation, as p63 is negative in better differentiated part of squamous cell carcinoma and sometimes weakly positive in adenocarcinomas (24). Napsin A and TTF1 stain adenocarcinomas, with an approximately similar frequency with slightly better performance for TTF1 in some (28), and Napsin A in other studies (17,27). In diagnosis of NSCLC on small samples liberal use of the category NOS is advocated, with subsequent use of additional stains, as the distinction between adenocarcinomas and squamous cell carcinomas forms in metastatic lung cancer the basis for treatment selection and predictive biomarker testing.

In conclusion, the accurate appreciation of tumor architecture in lung adenocarcinoma has important biological and clinical implications. Interobserver variation may explain differences in frequency of subtype and prognosis. In diagnosis of small samples immunohistochemistry is frequently helpful in the distinction of adenocarcinomas from squamous cell carcinomas.

Acknowledgements

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