Title: BASELINE PREDICTORS OF NEGATIVE AND INCOMPLETE PLEURAL CYTOLOGY IN PATIENTS WITH SUSPECTED PLEURAL MALIGNANCY - DATA SUPPORTING 'DIRECT TO LAT' IN SELECTED GROUPS

Authors: Selina Tsim[1], Sarah Paterson[2], Douglas Cartwright[1], Christopher J Fong[1], Laura Alexander[3], Caroline Kelly[3], Jayne Holme[2], Matthew Evison[2], Kevin G Blyth[1,4]

Authors' affiliations:

1. Glasgow Pleural Disease Unit, Queen Elizabeth University Hospital, Glasgow, UK
2. Respiratory Medicine, Manchester University NHS Foundation Trust, Wythenshawe Hospital, North West Lung Centre, Manchester, UK
3. Cancer Research UK Clinical Trials Unit, University of Glasgow, Glasgow, UK
4. Institute of Infection, Immunity & Inflammation, University of Glasgow, Glasgow, UK

Corresponding Author: Dr. Kevin G Blyth, Glasgow Pleural Disease Unit, Queen Elizabeth University Hospital, Glasgow, G51 4TF, UK, kevin.blyth@ggc.scot.nhs.uk

Conflicts of interest

All co-authors declare no conflicts of interest relevant to this manuscript.

Word counts – Abstract (295/300), Text (3652/5000)

Keywords: Pleural effusion; Mesothelioma; Cytology; Lung Cancer; Malignant Pleural Effusion

ABSTRACT

Objectives

Negative effusion cytology is more common in certain forms of Malignant Pleural Effusion (MPE) and results in pathway delay. Local Anaesthetic Thoracoscopy (LAT) is extremely sensitive and safe but cannot be offered to all. A stratified pathway, including ‘Direct to LAT’ in selected cases could enhance patient experience but requires reliable baseline predictors of unhelpful cytology, including both negative (no malignant cells) and incomplete results (malignant cells identified by predictive markers failed), since pleural biopsies will be required in the latter for optimal management. This retrospective analysis of a prospective multi-centre study, sought to identify baseline features for pathway rationalization.

Materials and Methods

363/638 (57%) of patients recruited to the DIAPHRAGM study (ISRCTN10079972) were included. Prospective data, including final diagnoses, asbestos exposure and fluid cytology results were supplemented by retrospective Computed Tomography (CT) and predictive marker reports. Independent predictors of negative and incomplete cytology were determined by multivariable logistic regression. Contingency tables were used to assess diagnostic value of cytology in associated phenotypes.

Results

238/363 (66%) patients were diagnosed with MPE (18 tumour types). Fluid cytology was negative in 151/238 (63%) and independently associated with

asbestos-exposure (Odds Ratio (OR) 5.34) and a malignant CT (OR 2.25). When both features were recorded the sensitivity and negative predictive value of fluid cytology were 19% (95% CI 11 – 30%) and 9% (95% CI 4 – 20%)), respectively. Cytology was incomplete in 34/238 (14%), i.e. 47% of positive cytology cases) but was not associated with any baseline feature. ORs for incomplete cytology in Ovarian, Breast, Renal and Lung Cancer were 83, 22, 21 and 9, respectively.

Conclusion

Negative cytology is extremely likely in patients with asbestos exposure and a malignant CT report. A ‘Direct-to-LAT’ approach may be appropriate in this setting. No baseline predictors of incomplete cytology were identified.

INTRODUCTION

Malignant Pleural Effusion (MPE) is common and often causes incapacitating breathlessness, requiring emergency hospitalization. Although MPE management can usually be generalized, precise tumour sub-typing and molecular profiling is required for treatment planning. [1] Detailed diagnostics also need to be completed quickly, in patients who may be physically debilitated, using the minimum number of invasive tests in combination with active palliation of symptoms. Pleural fluid aspiration (or thoracentesis) is a simple early investigation and allows transudative or infective causes to be identified rapidly. However, the diagnostic yield of fluid cytology is relatively low (averaging 60%) and varies considerably by tumour type. [2] In Malignant Pleural Mesothelioma (MPM), histological biopsies are recommended in all patients, when clinically relevant, by recent guidelines [3,4]. In the era of personalised medicine, pleural biopsies may also be required in patients with 'positive' effusion cytology, if the necessary predictive markers for that tumour [5],[6] cannot be performed. Therefore, the definition of 'negative' or at least 'incomplete' pleural cytology might reasonably be extended to include cases in whom predictive markers cannot be assessed, since pleural biopsies will be required to make an optimal treatment plan.

Current pleural disease guidelines advocate an unstratified diagnostic approach to MPE, whereby thoracoscopy is only offered if fluid cytology is negative (i.e. no malignant cells are seen). [3] In our experience, this is frequently associated with pathway delay, particularly in patients who are subsequently diagnosed with MPM, [3,4] and in those in whom additional

predictive markers are needed. However, the actual time patients spend on pleural pathways has, to our knowledge, never been prospectively recorded so this issue is likely under-appreciated.

Pathway rationalization for patients with suspected MPE is difficult because reliable predictors of unhelpful cytology, based on data available at the start of the process, have not been defined. Arnold *et al* recently highlighted the variable performance of effusion cytology (defined as the absence of malignant cells) based on the nature of the underlying cancer [2], However, this information, based on the outcome of the diagnostic process, cannot be easily used to stratify patients at first presentation. This is particularly the case in those where the predominant abnormality is a pleural effusion, without overt evidence of a primary cancer elsewhere.

The primary objective of this study was to define, for the first time, baseline clinical predictors of 'negative' effusion cytology (no malignant cells seen) and 'incomplete' effusion cytology (including patients with positive cytology, in whom predictive markers have failed). In these groups, a stratified pathway, including proceeding 'Direct to LAT' might be appropriate. Our secondary objective was to report diagnostic pathway length in patients with suspected MPE, since this has not been prospectively reported before and this information would influence the potential impact of 'Direct to LAT'.

MATERIALS AND METHODS

Study Design

This retrospective study was designed and reported per the Standards for the Reporting of Diagnostic Accuracy Studies (STARD) statement, 2015. [7] A limited retrospective dataset was collected to supplement data recorded in the prospective, multi-centre study: DIAPHRAGM (Diagnostic and Prognostic Biomarkers in the Rational Assessment of Mesothelioma (ISRCTN 10079972)). [8] Ethical approval was granted by the West of Scotland Research and Ethics Service (reference 13/WS/0240). DIAPHRAGM tested the diagnostic performance of several MPM biomarkers in an 'intention-to-diagnose' suspected MPE population recruited from 23 sites across UK and Ireland.

Importantly, the eligibility criteria used in DIAPHRAGM (see next section) selected the population most relevant to the objectives of the current study, i.e. patients in whom the presenting pleural effusion was identified as the primary diagnostic target. The inclusion criteria used did not preferentially select for MPM, since neither asbestos exposure nor radiological evidence of this (e.g. pleural plaques) were inclusion criteria. The study also incorporated robust diagnostic assessment, including access to thoracoscopy in all centres, and mandatory follow-up of benign diagnoses for 12 months to mitigate against potential false negative pleural sampling results [8].

Study Selection Criteria

Consecutive patients with suspected MPE recruited to DIAPHRAGM in Glasgow and South Manchester were potentially eligible. These sites were 2/23 sites involved in DIAPHRAGM, but contributed 73% (466/638) of the total

DIAPHRAGM study population. They were selected for this analysis because retrospective data collection was most feasible in these two large cohorts. DIAPHRAGM inclusion criteria were: suspected pleural malignancy (defined by a unilateral pleural effusion or pleural mass lesion); sufficient fitness for diagnostic sampling (pleural aspiration as a minimum); written informed consent. DIAPHRAGM exclusion criteria were: an inter-costal chest drain in-situ or within the preceding 3 months. DIAPHRAGM patients were excluded from the current study in any of the following instances: inadequate CT imaging (defined as non-contiguous high-resolution or non-contrast CT) to allow baseline classification; pleural thickening or mass only therefore no fluid samples attempted; diagnosis clearly benign based on clinical features and biochemistry or microbiological results (e.g. pleural transudate in the context of heart failure or bacterial pleural infection).

Data Collection

The majority of study data reported, including patient demographics, pleural fluid biochemistry, microbiological and cytology results, pleural histology results, asbestos exposure history and final diagnosis, were recorded prospectively within DIAPHRAGM. The final diagnosis recorded was based on strict criteria established in the DIAPHRAGM protocol, and was histological where possible. [8] However, since CT scans were acquired and reported as part of routine clinical activity in DIAPHRAGM, all CT reports were reviewed retrospectively by respiratory physicians (ST and SP). Reports were classified as malignant or benign based on previously reported criteria, [9] generating a dichotomous CT outcome (Malignant CT or Benign CT) for use as a candidate

predictor variable. CT reports were considered malignant if they increased the level of pre-CT suspicion of MPE, based on terms such as ’suspicious of malignancy’ and ’probable malignant effusion’. CT reports without such terms were classified as benign, replicating methods reported previously [9].

Pleural Cytology and Predictive Marker Assessment

Pleural cytology and predictive marker results were recorded retrospectively using electronic records. Pleural cytology was recorded as ‘positive’ if the final cytology report confirmed the presence of malignant cells, and ‘negative’ if no malignant cells were identified. Cytology was recorded as ‘incomplete’ if malignant cells were reported, predictive markers were indicated for that cancer based on national/international guidelines [10], but these tests failed or could not be performed. Cytology was recorded as ‘complete’ if malignant cells were seen and all indicated predictive markers were performed.

Diagnostic Pathway Length

A range of pathways intervals were recorded, including times (in days) from:

- referral from primary care to outpatient review in respiratory clinic
- first outpatient respiratory clinic (or date of emergency admission) to thoracentesis
- thoracentesis to authorised cytology report
- authorised cytology report to pleural biopsy
- overall time from first outpatient review (or emergency admission) to final diagnosis

Statistical Analysis

Data are presented as mean (± standard deviation (SD)) or median (± Interquartile Range (IQR)) depending on distribution. Binomial logistic regression was used to determine the association between clinical predictor variables and the outcome variables 'negative pleural cytology' and 'incomplete pleural cytology' in patients with a final diagnosis of MPE. Baseline clinical predictor variables and final diagnosis cell type variables were included in separate models. Only major cell types with known recommended predictive molecular markers were included in the incomplete pleural cytology model. The following clinical predictor variables were treated categorically: asbestos exposure history, a malignant CT report, current/ex-smoker status, a history of current/previous malignancy, emergency presentation, low serum albumin (<35g/l), gender, low haemoglobin (<14g/dl), high white cell count (≥8.2 $x10^9$/l) and final diagnosis (cell type). Age and pleural fluid volume were treated as continuous variables. Predictor variables associated with a p value <0.2 were included in a multivariable logistic regression model with the same outcome variable, after testing for co-linearity. Effect sizes were reported as Odds Ratios (OR).

Any baseline clinical predictors that demonstrated statistical significance on multivariable analysis were combined to produce clinical phenotypic sub-groups. The diagnostic sensitivity and negative predictive value (NPV) of pleural fluid cytology based on these phenotypic sub-groups were then compared using 2 x 2 contingency tables. Differences in diagnostic pathway times and pleural fluid volumes were compared using Mann-Whitney test. A p value ≤0.05 was considered statistically significant in multivariable logistic

regression and contingency tables. Analyses were performed using SPSS v22 (IBM, New York, USA).

RESULTS

Screening and Case Selection

466 patients were potentially eligible for the study, having been recruited consecutively to DIAPHRAGM in Glasgow (n=404) and South Manchester (n=62) between December 2013 – December 2016. Screening and eligibility assessment outcomes are summarized in Figure 1. 363/466 (78%) cases were eligible and included.

Study population

Median age was 74 years (IQR 65 – 80), 69% (n=263) were male, 27% (n=104) had a prior history of malignancy and 82% (n=312) were current or ex-smokers. 42% (n=161) were asbestos exposed and 42% (n=159) had a malignant CT report. The median volume of pleural fluid sent for cytology examination was 75 (IQR 40 – 120) ml. 141/379 (37%) were diagnosed with benign pleural disease and 238/379 (63%) patients were diagnosed with MPE, see Table 1. Of the patients diagnosed with MPE, 39% (n=92) had MPM and 61% (n=146) had secondary pleural malignancy, see Table 1.

Primary Objective

Baseline Predictors of Negative Pleural Fluid Cytology

276 of the total population (n=363 (76%)) had negative pleural fluid cytology. 151/238 (63%) patients with proven MPE had negative fluid cytology. The

proportion of patients with positive fluid cytology differed depending on the underlying tumour type (see Table 2).

On univariable analysis, a history of asbestos exposure, (OR 5.95 (95% Confidence Interval (CI) 2.86 – 10.57, p <0.0001), a malignant CT (OR 2.07 (95% CI 1.21 - 3.54), p=0.008) and male gender (OR 3.57 (95% CI 2.04 - 6.23), p <0.0001) were associated with an increased likelihood of negative pleural cytology (see Table 3). Current or ex-smokers (OR 0.52 (95% CI 0.3 - 0.93), p=0.026) and patients presenting as an emergency (OR 0.53 (95% CI 0.31 - 0.91), p=0.021) were associated with a reduced likelihood of negative pleural cytology. The volume of pleural fluid sent was not significantly associated with negative pleural cytology results (OR 0.999 (95% CI 0.996 - 1.002, p=0.371). In a subgroup analysis of patients who had a final diagnosis of secondary pleural malignancy only (n=146), the volume of pleural fluid was similarly not associated with the likelihood of negative cytology results (OR 0.995 (95% CI 0.990 – 1.001), p=0.09). Patients with a final diagnosis of NSCLC (OR 0.31 (95% CI 0.17 – 0.55, p <0.0001)), breast cancer (OR 0.08 (95% CI 0.02 – 0.35, p=0.001)) and ovarian cancer (OR 0.18 (95% CI 0.04 – 0.93, p=0.041)) had a reduced likelihood of negative cytology on univariable analysis (see Table 3). Patients with a final diagnosis of mesothelioma had a significantly increased likelihood of negative cytology on univariable analysis (OR 63.44 (95% CI 15.05 – 267.54, p <0.0001), see Table 3).

On multivariable analysis, asbestos exposure (OR 5.34, (95% CI 2.71 -10.52), p <0.0001) and a malignant CT (OR 2.25 (95% CI 1.25 - 4.06), p=0.007) were

the only baseline clinical variables to retain independent predictive value for negative fluid cytology results (Table 3). This multivariable regression model was constructed using asbestos exposure, malignant CT, positive smoking status, emergency presentation and low serum albumin. Male gender was not included in the multivariable regression model due to co-linearity with asbestos exposure. On multivariable analysis, mesothelioma (OR 31.3 (95% CI 6.71 – 145.97, p <0.0001) and breast cancer (OR 0.11 (0.02 - 0.54), p=0.007) retained independent predictive value for negative pleural cytology (Table 3).

The relative sensitivity and negative predictive value (NPV) of pleural cytology based on 4 clinical phenotypes based on the presence of these independent predictive factors (‘malignant CT and asbestos exposed’’, ‘benign CT and asbestos exposed’, ‘malignant CT and not asbestos-exposed’ and ‘benign CT and not asbestos-exposed’) is reported in Table 4.

Baseline Predictors of Incomplete Fluid Cytology

Assessment for predictive markers was recommended by international guidelines in 73/87 (84%) of cases with malignant pleural cytology (Table 2). Pleural cytology specimens were insufficient for these analyses in 34/73 (47%), equivalent to 14% of the total MPE cohort (n= 238). Pleural biopsies were acquired in 22/34 (65%) incomplete cases.

On univariable analysis, no variables were associated with an increased probability of incomplete cytology. Asbestos exposure (OR 0.37 (95% CI 0.16

– 0.85, p=0.019) and male gender (OR 0.4 (95% CI 0.2 – 0.8, p=0.01) were associated with a reduced likelihood on univariable analyses but these features failed to demonstrate any subsequent independent association (see Table 4).

Subsequent diagnoses of Ovarian (OR 83.09 (95% CI 11.26 – 613.12), p <0.001), Breast (OR 22.43 (95% CI 4.86 – 103.55), Renal (OR 21.25 (95% CI 3.39 – 133.14), p=0.001) and Non-small cell Lung Cancer (NSCLC) (OR 9.7 (95% CI 3.14 – 29.97), p <0.001) were all strongly associated with an increased likelihood of incomplete cytology in multivariable analyses. Marker assessment was unsuccessful in all cases of Ovarian Cancer with positive cytology (n=6), but was successful in 10/14 (71%) Breast Cancer cases.

There was no difference in the volume of pleural fluid sent in complete versus incomplete cytology cases (median volume 100 (IQR 75 – 126) ml versus 125 (IQR 65 – 345) ml, p=0.55).

Secondary Objective: Diagnostic Pathway Times

The median time from GP referral to first outpatient respiratory appointment (or emergency admission) was 14 (IQR 9 – 17) days. The median time from initial outpatient review (or emergency admission) to thoracentesis was 5 (IQR 1 – 16 days). The median time from thoracentesis to authorised cytology report was 7 (IQR 4 – 9) days. The median time from authorised cytology report to pleural biopsy was 14 (IQR 8 – 32) days. The total median time from

first outpatient appointment (or emergency admission) to final diagnosis was 26 (IQR 14 – 48) days.

Patients with negative pleural cytology had a significantly longer time to diagnosis than patients with positive pleural cytology (median 30 days (IQR 20 – 53) versus median 13 (IQR 9 – 17) days respectively, p <0.0001).

DISCUSSION

In this retrospective analysis of a large prospective multi-centre study, negative pleural cytology, defined by the absence of malignant cells was independently associated with a history of prior asbestos exposure (OR 5.34 (2.08 – 10.52), p <0.0001) and a malignant CT report (OR 2.25 (1.25 – 4.06), p=0.007), based on previously used definitions [9]. The diagnostic sensitivity and NPV of fluid cytology were therefore extremely low in patients with both of these features (sensitivity 19% (95% CI 11 – 30%), NPV 9% (95% CI 4 – 20%). On this basis, proceeding ‘Direct to LAT’ may be appropriate in patients exhibiting this phenotype. The low utility of fluid cytology in this setting is likely due to the high prevalence of Mesothelioma (MPM, 64%) in this group, of whom cytology was positive in no patients. This is consistent with an OR for negative cytology in subsequently diagnosed MPM of 63.44 (95% CI 15.05 – 267.54, p <0.0001).

In contrast, we could not identify any baseline characteristics that reliably predicted incomplete cytology, defined as visible malignant cells but failure of essential predictive markers. In the precision oncology era, this is a major

constraint to further pathway rationalization. Nevertheless, given the extremely high OR associated with certain cancers, particularly Ovarian (OR 83.09 (95% CI 11.26 – 613.12), p <0.001), and to a less extent, Breast (OR 22.43 (95% CI 4.86 – 103.55), Renal (OR 21.25 (95% CI 3.39 – 133.14), p=0.001) and Non-small cell Lung Cancer (NSCLC) (OR 9.7 (95% CI 3.14 – 29.97), p <0.001), 'Direct to LAT' may still be appropriate in patients with a high pre-test probability of these tumour types (e.g. based on gross radiological findings).

Fluid Cytology Yield

The average diagnostic yield of fluid cytology reported here in patients with secondary pleural malignancy (60%) is within the range reported in previous studies (40 – 67%). [2] [4] Interestingly, pleural fluid volume was neither a predictor of negative pleural cytology, nor was it associated with a higher rate of failed predictive marker testing. These findings are concordant with some earlier studies [24],[25] but discordant with Swiderek *et al* and Rooper *et al*, in which lower performance was reported with volumes below 60mls and 75mls respectively. [26],[27]. This may reflect improvements in cytological sensitivity since these papers were published (in 2010 and 2014, respectively) since the median volume of fluid in this study was around this threshold (75 (IQR 40 – 120) ml).

The absence of any cytological diagnoses of MPM in this study reflects current UK practice and recent recommendations in national guidelines. [21] These, in turn reflect the bland cytological appearances of MPM cells, which

make differentiation from benign reactive mesothelial cells extremely challenging. [22] Although a small number of highly-skilled, expert cytopathology centres report a cytological sensitivity of 73% in MPM, [23] the guideline view is that these data are difficult to generalize, being heavily reliant on expert opinion +/- ancillary tests.

Predictive Markers

In the era of personalised cancer treatment, predictive molecular and/or immunohistochemistry testing has become essential for optimal treatment planning. The feasibility of predictive markers based on pleural fluid has to date received relatively little attention in the literature. Rekhtman *et al* reported that EGFR and KRAS molecular testing was feasible in 126/128 (98%) cytology specimens in patients with NSCLC. However, only 23% of these specimens were pleural fluid samples. [23] This may explain the lower success rate in the current study (61% in NSCLC and 53% overall (39/73)), which also included a number of different tumour types. This mandated a subsequent pleural biopsy in 65% (22/34) of cases, delaying treatment initiation and potentially adversely affecting patient outcomes.

In Ovarian Cancer cases reported here, molecular predictive markers were possible in none of the 6/8 cases (75%) in whom cytology was positive. Clearly, the low number of cases involved in these analyses mandates caution in interpreting these data, however the OR associated with certain tumour types, particularly Ovarian, certainly warrant further studies, and in our opinion ‘Direct to LAT’ may be justified where the pre-test probability of Ovarian cancer is high, based on gross radiological features.

Pathway Length

In our cohort, the median time to diagnosis was 26 (IQR 14 – 48) days, including 7 (IQR 4 – 9) days from thoracentesis to authorised cytology report. The National Optimal Lung Cancer Pathway in England recommends pathology results being available within 3 days of sampling and results from molecular marker testing being available within 10 days. To our knowledge, there have been no previous reports regarding pathway lengths in suspected MPE, including MPM and the range of other primary cancers reported here. Recent quality improvement reports have highlighted the considerable challenges in delivering rapid turnaround times in pathology labs, although changes to processes may considerably improve these outcomes [11] without major redesign elsewhere.

Unsurprisingly, time to diagnosis was significantly longer in MPE patients in whom fluid cytology was initially negative. Previous studies report similarly prolonged pathways in patients with initially negative diagnostic tests, and associate these outcomes with multiple consecutive investigations [12-14] and treatment delay, which may in turn be associated with poorer survival, [12] [15] increased patient anxiety and poorer quality of life. [16] Repeated pleural interventions can also cause painful procedure tract metastases in MPM. [17] Pathway rationalization, including proceeding ‘Direct to LAT’ in some cases, has the potential to improve these outcomes without overwhelming the limited capacity of LAT centres or exposing patients to unnecessary risk.

Potential Clinical Implications

Our findings indicate that negative fluid cytology, defined by the absence of malignant cells, is extremely likely in asbestos-exposed patients with evidence of pleural malignancy on CT. In our opinion, it is reasonable to proceed ‘Direct to LAT’ without prior thoracentesis in this group of patients as this is unlikely to obviate the need for histological sampling. Pleural fluid biochemistry is of limited utility in this setting, since 5 – 9% of MPE present with a pleural transudate. [28],[21],[22]

In contrast, we found no reliable baseline features that predicted incomplete cytology, defined by failure of predictive marker tests. This outcome was, however, considerably more likely in patients with subsequent diagnoses of ovarian cancer, breast cancer, renal cancer or NSCLC, consistent with previous observations [2]. A ‘Direct to LAT’ strategy may therefore be appropriate in patients with a high pre-test probability of these tumour types, particularly Ovarian Cancer but this area requires further study.

Adoption of a ‘Direct to LAT’ strategy is likely to shorten the time to diagnosis and reduce the required number of pleural interventions. Based on the intervals reported here, this could save a median of 12 days by omitting the time required for thoracentesis and cytology reporting. Using such an approach would also allow earlier planning for LAT, even before the first clinic attendance, assuming pre-clinic CT imaging and a basic clinical history were available from the referral. If a ‘Direct to LAT’ strategy were adopted in selected cases, this may result in an overall increase in LAT cases and

individual centres would need to examine if such adjustments to LAT capacity were possible.

Study Strengths and Limitations

This study was performed and reported in accordance with published STARD guidelines and the majority of the data were collected within a multi-centre, prospective study. Clinically reported CT scans were used rather than blinded research-specific reporting. The diagnostic performance of CT scans in a 'real world' population is poorer than in research studies using research-specific reporting. {Tsim:2017db} Utilising clinical CT reports in this study for inclusion in the predictive models is therefore more likely to be representative of routine clinical practice. However, CT reports and predictive marker results were reviewed retrospectively introducing potential recall and omission bias. To mitigate against this, considerable care was taken to identify all available retrospective data resulting in 100% of CT reports and molecular pathology results being reviewed. Additionally, the prevalence of MPM (39% (92/238)) in our MPE cohort was relatively high. Our results are therefore less generalisable to populations with limited asbestos exposure and thus a lower prevalence of MPM. However, patients included in this study were recruited to the DIAPHRAGM study, which included deliberately broad eligibility criteria, designed to be generalisable to all patients presenting with suspected pleural malignancy. Finally, overall patient numbers used for the assessment of predictors of incomplete cytology were low, resulting in wide confidence intervals around OR estimates. A further study examining rates of successful predictive marker testing within a larger population is therefore warranted.

CONCLUSIONS

Negative pleural effusion cytology is extremely likely in patients with a history of asbestos exposure and a malignant CT report. Assuming this information is available at the start of the diagnostic process, a ‘Direct-to-LAT’ approach may be appropriate in this setting. No reliable baseline predictors of incomplete cytology were identified; however, the probability of failed predictive markers appeared higher in certain tumour types. A ‘Direct to LAT’ approach may therefore be justifiable in patients with a high pre-test probability of these tumour types, but this area requires further study within a larger patient population.

ACKNOWLEDGEMENTS

The DIAPHRAGM study was funded by the Chief Scientist Office Scotland (ETM/285).

REFERENCES

[1] A.O. Clive, B.C. Kahan, C.E. Hooper, R. Bhatnagar, A.J. Morley, N. Zahan-Evans, et al., Predicting survival in malignant pleural effusion: development and validation of the LENT prognostic score, Thorax. 69 (2014) 1098–1104. doi:10.1136/thoraxjnl-2014-205285.

[2] D.T. Arnold, D. De Fonseka, S. Perry, A. Morley, J.E. Harvey, A. Medford, et al., Investigating unilateral pleural effusions: the role of cytology, Eur. Respir. J. 52 (2018) 1801254. doi:10.1183/13993003.01254-2018.

[3] C. Hooper, Y.C.G. Lee, N. Maskell, BTS Pleural Guideline Group, Investigation of a unilateral pleural effusion in adults: British Thoracic Society Pleural Disease Guideline 2010, Thorax. 65 Suppl 2 (2010) ii4–17. doi:10.1136/thx.2010.136978.

[4] I. Woolhouse, L. Bishop, L. Darlison, D. De Fonseka, A. Edey, J. Edwards, et al., British Thoracic Society Guideline for the investigation and management of malignant pleural mesothelioma, Thorax. 73 (2018) i1–i30. doi:10.1136/thoraxjnl-2017-211321.

[5] T. Tursz, R. Bernards, Hurdles on the road to personalized medicine, Mol Oncol. 9 (2015) 935–939. doi:10.1016/j.molonc.2014.08.009.

[6] F.R. Hirsch, M.W. Wynes, D.R. Gandara, P.A. Bunn, The tissue is the issue: personalized medicine for non-small cell lung cancer, Clin. Cancer Res. 16 (2010) 4909–4911. doi:10.1158/1078-0432.CCR-10-2005.

[7] J.F. Cohen, D.A. Korevaar, D.G. Altman, D.E. Bruns, C.A. Gatsonis, L. Hooft, et al., STARD 2015 guidelines for reporting diagnostic accuracy studies: explanation and elaboration, BMJ Open. 6 (2016) e012799.

doi:10.1136/bmjopen-2016-012799.

[8] S. Tsim, C. Kelly, L. Alexander, C. McCormick, F. Thomson, R. Woodward, et al., Diagnostic and Prognostic Biomarkers in the Rational Assessment of Mesothelioma (DIAPHRAGM) study: protocol of a prospective, multicentre, observational study, BMJ Open. 6 (2016) e013324. doi:10.1136/bmjopen-2016-013324.

[9] S. Tsim, D.B. Stobo, L. Alexander, C. Kelly, K.G. Blyth, The diagnostic performance of routinely acquired and reported computed tomography imaging in patients presenting with suspected pleural malignancy, Lung Cancer. 103 (2017) 38–43. doi:10.1016/j.lungcan.2016.11.010.

[10] S. Novello, F. Barlesi, R. Califano, T. Cufer, S. Ekman, M.G. Levra, et al., Metastatic non-small-cell lung cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up, Ann. Oncol. 27 (2016) v1–v27. doi:10.1093/annonc/mdw326.

[11] S. Alshieban, K. Al-Surimi, Reducing turnaround time of surgical pathology reports in pathology and laboratory medicine departments, BMJ Qual Improv Rep. 4 (2015) u209223.w3773. doi:10.1136/bmjquality.u209223.w3773.

[12] N. Navani, M. Nankivell, D.R. Lawrence, S. Lock, H. Makker, D.R. Baldwin, et al., Lung cancer diagnosis and staging with endobronchial ultrasound-guided transbronchial needle aspiration compared with conventional approaches: an open-label, pragmatic, randomised controlled trial, Lancet Respir Med. 3 (2015) 282–289. doi:10.1016/S2213-2600(15)00029-6.

[13] E.A. Rakha, S. Patil, K. Abdulla, M. Abdulkader, Z. Chaudry, I.N. Soomro, The sensitivity of cytologic evaluation of pleural fluid in the diagnosis of malignant mesothelioma, Diagn. Cytopathol. 38 (2010) 874–879.

doi:10.1002/dc.21303.

[14] D.W. Henderson, G. Reid, S.C. Kao, N. van Zandwijk, S. Klebe, Challenges and controversies in the diagnosis of mesothelioma: Part 1. Cytology-only diagnosis, biopsies, immunohistochemistry, discrimination between mesothelioma and reactive mesothelial hyperplasia, and biomarkers, J Clin Pathol. 66 (2013) 847–853. doi:10.1136/jclinpath-2012-201303.

[15] W.-C. Tsai, P.-T. Kung, Y.-H. Wang, W.-Y. Kuo, Y.-H. Li, Influence of the time interval from diagnosis to treatment on survival for early-stage liver cancer, PLoS ONE. 13 (2018) e0199532. doi:10.1371/journal.pone.0199532.

[16] A. Segal, G.F. Sterrett, F.A. Frost, K.B. Shilkin, N.J. Olsen, A. William Musk, et al., A diagnosis of malignant pleural mesothelioma can be made by effusion cytology: results of a 20 year audit, Pathology. 45 (2013) 44–48. doi:10.1097/PAT.0b013e32835bc848.

[17] J. Walters, N.A. Maskell, Biopsy techniques for the diagnosis of mesothelioma, Recent Results Cancer Res. 189 (2011) 45–55. doi:10.1007/978-3-642-10862-4_4.

[18] A. Heilo, A.E. Stenwig, O.P. Solheim, Malignant pleural mesothelioma: US-guided histologic core-needle biopsy, Radiology. 211 (1999) 657–659. doi:10.1148/radiology.211.3.r99jn03657.

[19] N.A. Maskell, F.V. Gleeson, R. Davies, Standard pleural biopsy versus CT-guided cutting-needle biopsy for diagnosis of malignant disease in pleural effusions: a randomised controlled trial, The Lancet. 361 (2003) 1326–1330. doi:10.1016/S0140-6736(03)13079-6.

[20] P. Beckett, J. Edwards, D. Fennell, R. Hubbard, I. Woolhouse, M.D. Peake, Demographics, management and survival of patients with malignant pleural

mesothelioma in the National Lung Cancer Audit in England and Wales, Lung Cancer. 88 (2015) 344–348. doi:10.1016/j.lungcan.2015.03.005.

[21] T.E. Gonlugur, U. Gonlugur, Transudates in malignancy: still a role for pleural fluid, Ann. Acad. Med. Singap. 37 (2008) 760–763.

[22] M. Ashchi, J. Golish, P. Eng, P. O'Donovan, Transudative malignant pleural effusions: prevalence and mechanisms, South. Med. J. 91 (1998) 23–26.

[23] N. Rekhtman, S.M. Brandt, C.S. Sigel, M.A. Friedlander, G.J. Riely, W.D. Travis, et al., Suitability of thoracic cytology for new therapeutic paradigms in non-small cell lung carcinoma: high accuracy of tumor subtyping and feasibility of EGFR and KRAS molecular testing, J Thorac Oncol. 6 (2011) 451–458. doi:10.1097/JTO.0b013e31820517a3.

[24] W. Abouzgheib, T. Bartter, H. Dagher, M. Pratter, W. Klump, A prospective study of the volume of pleural fluid required for accurate diagnosis of malignant pleural effusion, Chest. 135 (2009) 999–1001. doi:10.1378/chest.08-2002.

[25] S.M. Sallach, J.A. Sallach, E. Vasquez, L. Schultz, P. Kvale, Volume of pleural fluid required for diagnosis of pleural malignancy, Chest. 122 (2002) 1913–1917.

[26] L.M. Rooper, S.Z. Ali, M.T. Olson, A minimum fluid volume of 75 mL is needed to ensure adequacy in a pleural effusion: a retrospective analysis of 2540 cases, Cancer Cytopathol. 122 (2014) 657–665. doi:10.1002/cncy.21452.

[27] J. Swiderek, S. Morcos, V. Donthireddy, R. Surapaneni, V. Jackson-Thompson, L. Schultz, et al., Prospective study to determine the volume of pleural fluid required to diagnose malignancy, Chest. 137 (2010) 68–73.

doi:10.1378/chest.09-0641.

[28] L. Ferreiro, F. Gude, M.E. Toubes, A. Lama, J. Suárez-Antelo, E. San-José, et al., Predictive models of malignant transudative pleural effusions, J Thorac Dis. 9 (2017) 106–116. doi:10.21037/jtd.2017.01.12.

FIGURE LEGENDS

Figure 1

Study flowchart summarizing eligibility assessment, study selection and results of the index (pleural fluid cytology results) and reference investigations (final pleural diagnosis). 363 of 466 potentially eligible subjects recruited to DIAPHRAGM were selected for the study